Population dynamics and lifecycle of the brown shrimp *Crangon crangon* (Caridea, L. 1758). Experimental, biochemical and theoretical aspects

Dissertation

zur Erlangung des Doktorgrades des

vorgelegt von:

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Hamburg, Dezember 2009

Genehmigt vom Department Biologie
der Fakultät für Mathematik, Informatik und Naturwissenschaften
an der Universität Hamburg
auf Antrag von Professor Dr. A. TEMMING
Weiterer Gutachter der Dissertation:
Herr Professor Dr. M. PECK
Tag der Disputation: 03. Juli 2009

Hamburg, den 19. Juni 2009

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To explain a phenomenon is to find a model that fits it into the basic framework of the theory and that thus allows us to derive analogues for the messy and complicated phenomenological laws which are true of it. (Nancy Cartwright, 1983)

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Summary

Crangon crangon supports one of the most valuable fisheries in the North Sea with up to 100 million € in annual landings value. However, until now great uncertainties exist about its population dynamics and life cycle, mainly because observed growth rates are highly variable both within and between different studies. Based on lacking age information also great uncertainties about the total mortality rate exist and furthermore the question if *C. crangon* is a protandric hermaphrodite was discussed with controversy in the past. The actual study was therefore designed to answer these questions and to gain a better understanding of the life cycle of this interesting species.

In the review section in **MANUSCRIPT 1** a complete account of published information from experimental- and cohort tracking studies is given. In a larger number of established studies very low growth rates are reported, which are not compatible with the current understanding of the life cycle. New growth experiments to investigate the hypothesis, that food quality is one main cause for the observed inconsistencies were performed and it could be shown that the availability of life food, especially copepods, is a key factor for high growth rates. Another key factor that has not been described until now was the age of the sampled cohort. In experiments with small overwintering shrimp caught in May it was not possible to produce high growth rates which is in stark contrast to experiments under identical conditions, but with shrimp sampled from the newly immigrating cohort in early summer. The key results from the successful experiments with the new cohort were very high growth rates (up to 0.5 mm /d at 25 mm length), significantly higher growth rates of females compared to males and limited variability of moult intervals compared to moult increments.

According to simulations the best correspondence from invading juveniles in spring and peak landings in September (commercial size at 50 mm) was obtained with maximum growth parameters and not with mean growth rates. This suggests that at least a part of the brown shrimp population does not experience best growth conditions in the field, although the Wadden Sea is one of the most productive areas in the world. Within **MANUSCRIPT 2** therefore different indicators (RNA/DNA, dry weight at length and caloric content) were examined for their suitability as feeding- and condition-proxy within controlled starvation laboratory experiments. The results were applied in the next step to field data collected at three different sites in the Wadden Sea over three years. The results suggested that even in the highly productive summer season at least 25% of the whole population is food limited. Furthermore there were indications that over the whole winter period about 75% of the population was starving despite opportunistic omnivor feeding of *C. crangon.*

In **MANUSCRIPT 3** dry weight at length and $RNA \cdot DNA^{-1}$ were tested for the suitability to describe growth rates of *C. crangon.* For this purpose animals used in the growth experiments described in manuscript 1 were analyzed. No correlation of growth with dry weight condition index could be determined. $RNA \cdot DNA^{-1}$

explains parts of the variability within growth rates and correlated with length growth at high temperatures, but from the determined results it was concluded that it is not a suitable growth proxy for *C. crangon.* Reasons were most probably the effect of short term variability of $RNA{\cdot}DNA^{-1}$ due to moulting, maturation and feeding intensity in comparison to the relatively long time needed to determine growth accurately.

Mortality determination in crustaceans and small fishes, is not always possible due to problems with age estimation. An alternative are length based methods but these are bound to specific basic assumptions and are only valid under nonseasonal conditions and an evaluation of these methods under seasonal conditions is outstanding. Within **MANUSCRIPT 4** therefore a review of existing methods was performed and the methods were examined according to their bias under seasonal recruitment, growth and mortality conditions. The evaluation was performed by applying the methods to simulated, artificially created length frequency distributions with known conditions. The results will not only help to improve existing and future mortality estimates but also showed that some methods totally fail for certain habitat conditions. In a species-specific approach, population specific simulated length frequency distributions of the brown shrimp *Crangon crangon* were created using available information on the seasonality of growth, recruitment and mortality.

In **MANUSCRIPT 5** the knowledge gained from manuscript 4 was applied to determine accurate mortality estimates of *C. crangon.* Therefore long term time series data from four German and Dutch surveys of half a century were analyzed. The results suggested that total mortality of *C. crangon* doubled since the 1950s towards the 1990. Later a slight decrease of total mortality can be observed. The high mortality of approximately $Z = 5$ further implied that *C. crangon* has mainly an annual life cycle.

MANUSCRIPT 6 deals with the reproductive strategy of *Crangon crangon*. Until now there is an ongoing debate as whether the brown shrimp is an obligate hermaphrodite or not. Based on samples taken in Büsum, Meldorf and Wilhelmshaven it could be shown that the population is female biased in all length classes and that large males exist in the field. This indicates that sex change is most probably not present or plays an tangential role for male shrimps. The findings were substantiated by the results of a computer simulation showing that observed sex ratios of all size classes are explainable after accounting for sex specific growth rate differences. Our simulations suggested that the fraction of eggs contributed by secondary females to the whole amount of eggs produced by one cohort of male and female shrimps, is less than 1%. Although the ability exists that male shrimps change sex the fraction is small. The reason that this possibility that has been evolutionary been selected for in the past is now not advantageous any more might be a reduction of the maximum age or changes in the population dynamics after invading into the Wadden Sea after the last ice age.

Zusammenfassung

Die Nordseegarnele *Crangon crangon* ist eine bedeutende Spezies in der Nordsee und stellt mit einem jährlichen Erlös von bis zu 100 Mio. Euro ein lukratives Ziel der Fischereiindustrie dar. Die bisher beschriebenen Wachstumsraten dieser Spezies sind sowohl innerhalb als auch zwischen verschiedene Studien hoch variabel. Wichtige Fragen bezüglich der Populationsdynamik und des Lebenszyklusses konnten daher bisher nicht ausreichend beantwortet werden. Aufgrund fehlender Altersmerkmale bestehen darüber hinaus große Unsicherheiten hinsichtlich der Gesamtsterblichkeit der Garnele und kontrovers diskutiert wird zudem die Frage ob *Crangon crangon* ein Hermaphrodit ist. Ziel der aktuellen Studie war daher die Beseitigung der genannten Unsicherheiten und eine bessere Beschreibung des Lebenszyklusses.

In **MANUSKRIPT 1** wurden beschriebene Wachstumsraten aus der Literatur (Kohortenverfolgung und Laborversuche) analysiert. In vielen anerkannten Studien wurden sehr geringe Wachstumsraten beobachtet, die nicht in das aktuelle Verständnis des Lebenszyklusses der Garnele passen. Folglich wurde in dieser Arbeit die Hypothese überprüft, ob die Futterqualität ein wichtiger Faktor ist, bestehende Unterschiede in den Wachstumsraten zu beschreiben. Es konnte gezeigt werden, dass Lebendfutter, insbesondere Copepoden, ein Schlüsselfaktor für hohe Wachstumsraten ist. Ein Weiterer wichtiger Faktor könnte im Alter der Tiere begründet sein, was in dieser Arbeit erstmals beschrieben werden konnte. Garnelen aus der Überwinterungspopulation wuchsen sehr langsam, was im starken Gegensatz zu den, unter gleichen Bedingungen beobachteten, hohen Wachstumsraten der neu eingewanderten Garnelen der Frühjahrskohorte steht. Die wichtigsten Ergebnisse der erfolgreich durchgeführten Experimente mit jüngeren Tieren waren hohe Wachstumsraten (bis zu 0.5 $mm \cdot d^{-1}$ bei 25 mm Länge), signifikant höhere Wachstumsraten der Weibchen im Vergleich zu gleichgroßen Männchen und geringere Variabilität der Häutungsintervalle, im Vergleich zu den Häutungsinkrementen.

Basierend auf Simulationen konnte das Maximum der einwandernden juvenilen Tiere im Frühjahr mit dem Maximum der kommerziellen Landungen im Herbst (>50 mm Mindestlänge) nur verbunden werden, wenn maximales Wachstum angenommen wurde. Obwohl das Wattenmeer eines der produktivsten Meeresgebiete der Welt ist, deutet dieses Ergebnis auf eine teilweise Limitierung der Population hin. In **MANUSKRIPT 2** wurden daher verschiedene Parameter ($RNA \cdot DNA^{-1}$, Trockengewicht und Kaloriengehalt) in kontrollierten Hungerversuchen auf ihre Eignung als Futter- und Konditionsindikator getestet. Die Ergebnisse wurden mit Felddaten verglichen, welche über drei Jahre an drei verschiednen Stationen im Wattenmeer gefangen wurden. Aus den Ergebnissen konnte gefolgert werden, dass selbst in der hochproduktiven Sommerphase und trotz opportunistisch omnivorer Ernährung, 25% der Tiere hungern. Im Winter scheinen es sogar 75% der Garnelen zu sein.

Basierend auf den Tieren aus den Wachstumsversuchen (Manuskript 1), wurde in **MANUSKRIPT 3** die Möglichkeit untersucht, Trockengewicht und $RNA \cdot DNA^{-1}$ als Wachstumsindikator zu verwenden. Hierbei konnte keine Korrelation zwischen dem Trockengewicht und dem Wachstum der Tiere hergestellt werden. RNA/DNA erklärte zwar einen geringen Anteil der Variabilität und korrelierte mit dem Längenwachstum der Garnelen vorwiegend bei hohen Temperaturen, stellte aber keinen geeigneten Wachstumsproxy für die Nordseegarnele dar.

Bis heute ist nicht nur das Wachstum, sondern auch die Gesamtsterblichkeit (Z) der Garnele nicht hinreichend genau bestimmt worden. Vergleichbar zu anderen Crustacea und tropischne Fischen ist eine Altersbestimmung schwierig. Die einzigen zurzeit verfügbaren Methoden sind für diese Arten basieren daher auf Längen-Häufigkeits-Verteilungen. Eine grundlegende Voraussetzung dieser Methoden ist, dass keine Saisonalität vorhanden ist. Bis jetzt steht eine umfangreiche Evaluation der Methoden, bei Anwendung unter saisonal beeinflussten Bedingungen, aus. In **MANUSKRIPT 4** wurde daher die systematische Abweichung der Sterblichkeitsschätzungen unter Annahme von saisonalem Wachstum, Recruitment und saisonaler Sterblichkeit geprüft. Die verschiedenen Methoden wurden hierfür auf erstellte (berechnete) Längen-Häufigkeitsverteilungen mit bekannten Eigenschaften angewendet. Anhand der Ergebnisse konnte gezeigt werden, dass einige Methoden unter bestimmten Lebensraumbedingungen nicht anwendbar sind. Die gewonnen Ergebnisse stellen zudem eine Möglichkeit dar bestehende und zukünftige Sterblichkeits-schätzungen zu verbessern.

Die neuen Erkenntnisse aus Manuskript 4 wurden in **MANUSKRIPT 5** angewendet um die Gesamtsterblichkeit der Nordseegarnele zu bestimmen. Die Schätzungen wurden basierend auf vier Langzeitdatenserien deutscher und holländischer Surveys durchgeführt. Mit diesen Daten konnte der Zeitraum eines halben Jahrhunderts abgedeckt werden. Die Ergebnisse zeigen, dass die Gesamtsterblichkeit der Garnelen sich innerhalb des Zeitraums von 1950 bis 1990 verdoppelt hat, wohingegen später ein leichter Rückgang zu beobachten war. Die hohe mittlere Sterblichkeit von $Z = 5.3$ impliziert, das *C. crangon* einen einjährigen Lebenszyklus aufweist.

MANUSKRIPT 6 behandelt die Fortpflanzungsstrategien der Garnele. Bis heute wird debattiert ob *Crangon crangon* ein obligater Hermaphrodit ist. In Manuskript 6 konnte anhand von Feldproben aus Büsum, Meldorf und Wilhelmshaven gezeigt werden, dass die Population in allen Längenklassen von Weibchen dominiert wird. Des Weiteren kommen große Männchen regelmäßig in Feldproben vor. Beide Beobachtungen weisen darauf hin, dass Geschlechtsumwandlungen höchstwahrscheinlich nicht stattfinden oder nur eine untergeordnete Rolle spielen. Diese Schlussfolgerung wurde durch Simulationen untermauert die zeigen, dass vorhandene Unterschiede in den Geschlechterverhältnissen verschiedener Längenklassen einzig durch geschlechtsabhängige Wachstumsraten zustande kommen können. Des weiteren zeigen die Simulationen, dass der Anteil der Eier sekundärer Weibchen an allen Eiern einer Kohorte höchstwahrscheinlich unter 1% liegt. Obwohl die Möglichkeit der Geschlechtsumwandlung vorhanden ist, ist der

tatsächliche Anteil der sekundären Weibchen sehr gering. Dass dieser von der Evolution selektierte Prozess den männlichen Garnelen keinen Vorteil mehr bring, ist vermutlich in der gesunkenen Lebenserwartung oder in Veränderung der Lebensbedingungen bei der Besiedelung des Wattenmeers nach der letzten Eiszeit, zu suchen.

Outline of publications

The following overview outlines the six publications included in this thesis and the contribution of the co-authors to the manuscripts. The overall objectives of this research was embedded in the project: „Entwicklung, Parametrisierung und Anwendung eines spezifischen Y/R Modells für die Nordseegarnele (Crangon crangon L.) zur Beurteilung des Befischungszustandes“.

Manuscript 1

Determining growth rates of the brown shrimp *Crangon crangon* (L.): review and laboratory approach.

Marc Hufnagl & Axel Temming

Marc Hufnagl performed the growth experiments and the literature research and wrote the manuscript. Axel Temming helped with the writing and the design of the experiments. The manuscript was submitted to *Marine Ecology progress series* a peer review Journal and is reviewed once.

Manuscript 2

Is *Crangon crangon* (L. 1758, Decapoda, Caridea) food limited in the Wadden Sea?

Marc Hufnagl, Axel Temming, Andrea Dänhardt and Robert Perger

Marc Hufnagl performed the samplings in Büsum and Meldorf, the starvation experiments, the analyzation and wrote the manuscript. Axel Temming helped with the writing and the design of the experiments. Andreas Dänhardt performed the samplings in Wilhelmshaven. Robert Perger did RNA/DNA measurements of a part of the field samples The manuscript was submitted to *Journal of Marine Science* a peer review Journal.

Manuscript 3

Is RNA·DNA-1 ratio and dry weight a suitable growth proxy for the brown shrimp *Crangon crangon* (L., Crustacea)

Marc Hufnagl & Axel Temming

Marc Hufnagl performed the growth experiments, the sample analysis and wrote the manuscript. Axel Temming helped with the writing and the design of the experiments. The manuscript was submitted to *Helgoland Marine Research* a peer review Journal.

Manuscript 4

An evaluation of various models for the estimation of total mortality from length frequency distribution data confronted with seasonal variability in growth, recruitment and mortality

Marc Hufnagl & Axel Temming

Marc Hufnagl performed the evaluation and wrote the manuscript. Axel Temming helped with the writing and model setup. The manuscript was submitted to *ICES Journal of Marine Science* a peer review Journal.

Manuscript 5

Estimating total mortality Z and maximum length L_∞ of *Crangon crangon* (Crustacea L. 1758) between 1955 and 2006

Marc Hufnagl, Axel Temming, Volker Siegel, Ingrid Tulp and Loes Bolle

Marc Hufnagl analyzed the data and wrote the manuscript. Axel Temming helped with the writing and the analysis. Volker Siegel, Ingrid Tulp and Loes Bolle provided the data and helped with the writing. The manuscript was submitted to *ICES Journal of Marine Science* a peer review Journal.

Manuscript 6

The brown shrimp a protandric hermaphrodite? Relevance of this sexual system for its population dynamics.

Marc Hufnagl, Axel Temming and Andrea Dänhardt

Marc Hufnagl performed the samplings in Büsum and Meldorf, wrote the model and the manuscript. Axel Temming helped with the writing and analysis. Andreas Dänhardt performed the samplings in Wilhelmshaven. The manuscript was submitted to *Marine Biology* a peer review Journal and is reviewed once.

General Introduction

Brown shrimp (*Crangon crangon*) is a decapod crustacean that is both profitable and palatable and is not only economically important but also biologically very interesting. It successfully inhabits a large variety of different habitats and occupies a key trophic position by transferring energy from the lower trophic levels (Pihl & Rosenberg 1984) to top predators such as Atlantic cod, whiting (Hamerlynck & Hostens 1993), seals (Behrends 1985) and humans (ICES 2006). Although a large amount of literature exists - in 1984 already about 560 articles (Redant 1984), basic knowledge concerning growth, mortality, maximum age and the life cycle is sparse. Thus, the objective of the actual work was to clarify these aspects with the main focus on growth, mortality and sexual system by application of longterm data series, biochemical proxies, laboratory trials, field samples and mathematical models.

C. crangon belongs to the infraorder Caridae and the family Crangonidae. The whole classification is as follows

Kingdom:	Animalia
Phylum:	Arthropoda
Class :	Crustacea
Subclass:	Malacostraca
Superorder:	Eucarida
Order:	Decapoda
Suborder:	Dendrobranchiata
Infraorder:	Caridea
Family:	Crangonidae
Genus:	*Crangon*
Species:	*crangon*

Among Decapoda, the infraorder Caridea is relatively old and separated from Anomura and Brachyura during the Carboniferous and Permian 300 million years ago (Gaten 1998). Caridea have the distinctive characteristic of a downturned bend in the third abdominal segment and both pleura of the second abdominal segment overlap their neighbours. Key characteristics for *C. crangon* are gills covered by a laterally compressed carapace. The first peraeopod (Figure 4-1) terminates in subchela (cheliped) and the Pleonite 6 (sixth abdominal segment) is smooth on the dorsal side. Familiar species are the japanese sand shrimp: *Crangon affinis* (De Haan, 1849), *Crangon alaskensis* (Lockington, 1877), stout crangon: *Crangon alba* (Holmes, 1900), *Crangon allmanni* (Kinahan, 1857), ridged crangon: *Crangon dalli* (M. J. Rathbun, 1902), California bay shrimp: *Crangon franciscorum* (Stimpson, 1856), *Crangon handi* (Kuris and Carlton, 1977), Holmes bay shrimp: *Crangon holmesi* (M. J. Rathbun, 1902), blacktailed shrimp: *Crangon nigricauda* (Stimpson, 1856), blackspotted bay shrimp: *Crangon nigromaculata* (Lockington, 1877), the sand shrimp: *Crangon septemspinosa* (Say, 1818) and the Japanese sand shrimp: *Crangon uritai* (Hayashi & Kim, 1999).

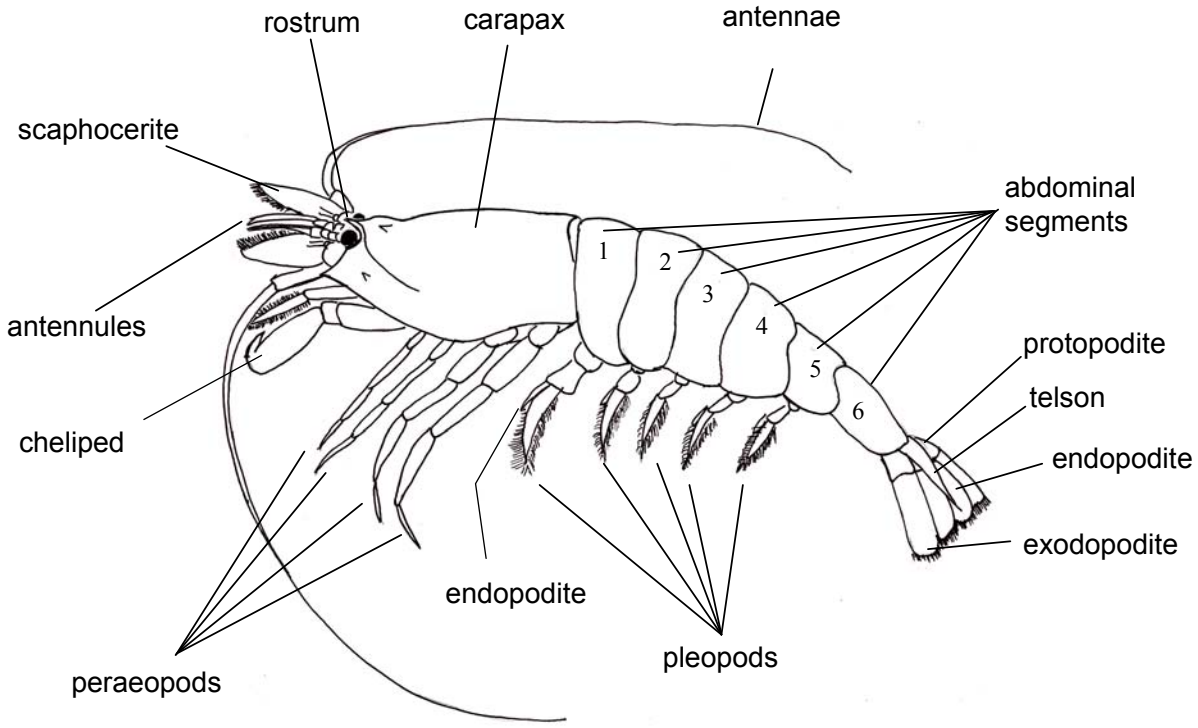

Figure 4-1: Schematic drawing of the brown shrimp *Crangon crangon*.

C. crangon is a species that tolerates a broad range of salinities (Cieluch et al. 2005, Gelin et al. 2001, Hagerman 1970) and temperatures (van Donk & de Wilde 1981, Weber & Spaargaren 1970, Caudri 1937). Water balance is mainly regulated by varying drinking rates (Che Mat & Potts 1985) which leads to higher energy consumption at extremely low and high salinities (Spaargaren & Haefner Jr. 1987). The lower salinity limit is to be set to about 5 psu. At this salinity zoeal stages and decapodids suffer high mortality (Cieluch et al. 2005) and adult shrimps are unable to reproduce (Gelin et al. 2001). The highest temperature tolerable is between 25 and 30°C where mass exodus of shrimps from tidal areas has been observed (Berghahn 1983). Apart from the low salinities and high temperatures hydrographical restrictions only exist due to low oxygen contents (Hagerman & Vismann 1995, Haefner Jr. 1971).

As a benthic species that hides from predation by burying in the ground, *C. crangon* mainly inhabits soft bottom substrates but can also be dominant on sandy shores (Beyst et al. 2001). *C. crangon* is distributed around coastal waters of Europe reaching from as far North as Iceland (Gunnarson et al. 2007) towards the Mediterranean (Labat 1977) and from the coasts of the UK (Henderson & Holmes 1987) over the Baltic (Dornheim 1969) towards the Black Sea (Luttikhuizen et al. 2008). Similar to other members of Crangonidae, *C. crangon* dwells close to the coast in shallow waters (Clifford & Moran 2006, Nakaya et al. 2004, Hanamura & Matsuoka 2003, Modlin 1980), whereas its conspecific *Crangon allmanni* inhabits sandy areas in the more offshore parts of the North Sea (Blahudka & Türkay 2002, Allen 1960), (Price 1962).

Intraspecific genetic differences have only been described for far-flung populations of *C. crangon.* AFLP (amplified fragment length polymorphism) analysis revealed three main populations in western Britain, the eastern English Channel and the Baltic Sea (Weetmann et al. 2007). Results of starch gel electrophoresis determined four regional groups North Sea, Baltic Sea, North Atlantic and Adriatic (Bulnheim & Schwenzer 1993). The most recent study was based on the comparison of cytochrome-c-oxidase fragments which also revealed four groups: Northeastern Atlantic, western Mediterranean, Adriatic Sea and Black Sea (Luttikhuizen et al. 2008). The results of these three studies show that gene flow is mainly restricted by oceanographic barriers but apart from that the population is well mixed over large areas. This is mainly based on the increased mobility of the shrimp in contrast to other benthic animals and drift of the offshore spawned offspring.

Drift also allows this mainly benthic living animal to spread over the described large area despite the fact that it is able to use tidal currents for directed locomotion (Janssen & Kuipers 1980, Al-Adhub et al. 1975, Hartsuyker 1966). Due to the high adaptability of *C. crangon,* many the habitats reached at the end of this drift phase can be, can be successfully occupied and, in most of these habitats, shrimp occupies a key trophic position. In general molluscs, polychaets, copepods and amphipods can be found in stomachs of *C. crangon* (del Norte-Campos & Temming 1998, Pihl 1985, Evans 1984, Plagmann 1939) but the shrimps are generally opportunistic omnivore. Gut fullness, peaks at night (Feller 2006), (Cattrijsse et al. 1997) where also highest activity was determined (Hagermann 1970). According to the high variety of food sources, food niche overlaps with other species are generally low (Feller 2006, Pihl 1985). At the present time it is unclear whether the growth and survival of brown shrimp is food-limited in the North Sea. It was suggested that growth rates observed in the laboratory are higher than those in the field due to food limitation (Pihl & Rosenberg 1984). In the Bristol Channel variable recruitment that results in relatively stable adult populations was observed (Henderson et al. 2006) which was suggested to result from density and resource regulation. Kuipers & Dapper (1981) discussed that based on a daily consumption of 6.7 g ash-free dry weight (ADW)·m^{-2}, food limitation would be expected if mainly macrobenthic production would be considered and competition with other consumers would be assumed. Such a bottom up control would have implications on the life cycle of the shrimp by reduction of growth performance, abundance or fecundity.

Currently, the following life stages are known and described for *C. crangon* (see also Figure 4-2). Mean length at maturity of adult female *C. crangon* is 50 to 55 mm (Oh et al. 1999). The amount of eggs that a female carries, attached to the setae of its pleopods, is length dependent and varies between 2000 (50 mm) and 10000 (80 mm) eggs (Bilgin & Samsun 2006, Redant 1977, Havinga 1930). Berried (egg carrying) females can be observed throughout the whole year but with lowest abundance during the fall (Neudecker & Damm 1992, Kuipers & Dapper 1984) where maturation of the gonads is inhibited, most probably by

hormones produced in the eye stalks (Klek-Kawinska & Bomirski 1975). The size and the amount of the eggs is dependent on the season and can be separated into winter and summer eggs. Winter eggs are bigger than summer eggs and have a higher caloric content (Paschke et al. 2004). Furthermore larvae that hatch from winter eggs are bigger (Boddeke 1982) and show a higher starvation resistance (Paschke et al. 2004). Hatch takes place together with the moulting of the mother, after 18 (20°C) to 45 (10°C) days (Redant 1978). The larvae needs between 5 and 6 days to pass through each instar and development from larvae to postlarvae (instar I - V) takes about one month (Criales & Anger 1986). The first juvenile stage is a benthic stage whereas all prior larval stages are pelagic. Juveniles of 7 to 15 mm length invade into the shallow areas of the Wadden Sea in late spring early summer (Groh 1982, Boddeke 1976) and originate most probably from winter spawned eggs (Temming & Damm 2002). The timing of the invasion seems to be mainly temperature driven and occurs later after cold winters (Beukma 1992). Juveniles < 30 mm use the shallow areas of the Wadden Sea as nursery areas, where they find shelter from predation, enough food and high temperatures for fast growth (Cattrijsse et al. 1997, Beukma 1992, Boddeke et al. 1986). Larger adults can mainly be found in deeper water and highest commercial landings are observed in fall (ICES 2007, Beukma 1992, Boddeke et al. 1986).

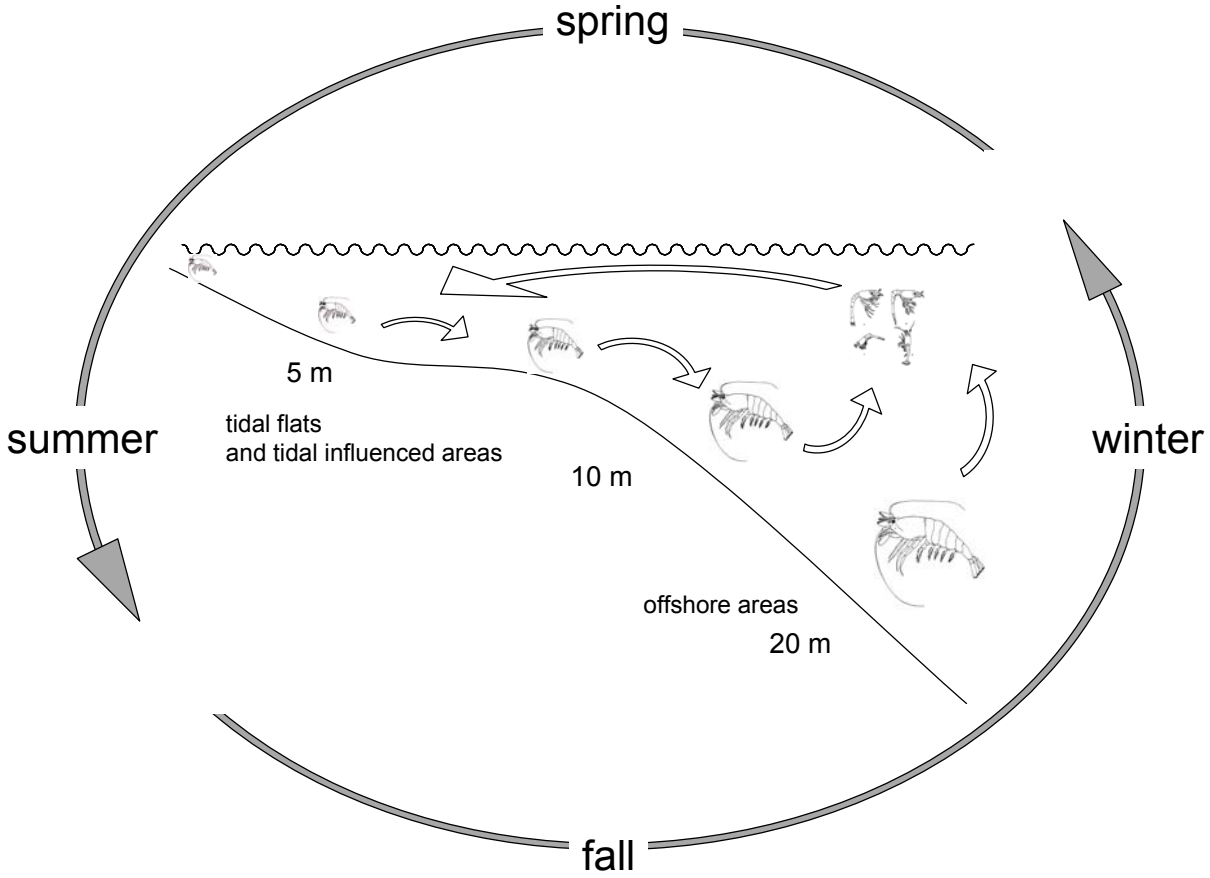

Figure 4-2: Life cycle and migration of *C. crangon* in the Southern North Sea

Previously derived growth models are so far not able to match with the described life stages and field observations (e.g. discussed in Beukema (1992) for the model

presented by Kuipers & Dapper (1984)). Therefore, there is a gap in the knowledge concerning whether or not the adults observed in autumn commercial catches originate from summer eggs, winter eggs or eggs from the summer of the previous year. There are several studies that reported growth rates and results differ largely. Observed values vary between 0 to 0.1 $mm \cdot d^{-1}$ (Oh & Hartnoll 2000, Henderson & Holmes 1987), 0.2-0.54 $mm \cdot d^{-1}$ (Beukema 1992) and 0.07-0.4 $mm \cdot d^{-1}$ (Tetard 1985). Within one size and temperature class, determined growth rate can range from nearly zero (no growth) (Edwards 1978) to 0.54 $mm \cdot d^{-1}$(Beukema 1992). The difficulties of accurate growth determination are based on the lack of age information (since Crustacean do not retain any hard parts after ecdysis (Hartnoll 2001). Only very few methods exist that allow ages of some Crustacean species to be determined. Recently, the analysis of the so called “age pigment”: lipofuscin has been employed for aging (Bluhm & Brey 2001). This pigment accumulates in the central nervous system and can be counted with a fluorescence microscope (Kodama et al. 2005, Bluhm et al. 2002, Vila et al. 2000, Sheehy et al. 1996, Sheehy et al. 1994). However, *C. crangon* has not yet been examined for this pigment, but it seems as though at least two years are required in other species to accumulate enough of this pigment to gain accurate measurements. Two other ageing methods are mentioned in Hartnoll (2001): the first method uses the radionucleids ^{226}Ra, ^{210}Pb, ^{228}Ra and ^{228}Th and the second is based on a calcified structure. The disadvantage of the radionucleid method is that it is very expensive and that it is only able to determine the time since the last moult, which is rather short for *C. crangon* (Schatte & Saborowski 2005, Meixner 1969). The second method is the examination of a calcified structure of the infracerebral organ, which lies beneath the brain. This layered structure has thus far only been observed in clawed lobsters (Nephropsidae). Each of these three aging methods: lipofuscin, radionucleids and infracerebral organ are not suitable for *C. crangon*. Furthermore, difficulties arise when age, growth and mortality are determined from data obtained from repeated field sampling of *C. crangon*. Cohort analysis requires intensive sampling and results may be biased by several factors such as spawning, growth, mortality and environmental conditions. The spawning period of *C. crangon* is rather long and larvae as well as egg bearing females can be observed nearly throughout the whole year (Siegel et al. 2008, Kuipers & Dapper 1984). Several spawning events together with variable individual growth results in cohorts with mixed age classes including slow growing earlier hatched shrimps and fast growing younger shrimps. Reasons for variable growth are multiple and include environmental influences like temperature, salinity or food supply as well as genetic factors (Weetmann et al. 2007, Beaumont & Croucher 2006, Gitterle et al. 2005), stress, pollution or diseases. Beside the problems of cohorts with mixed age at the same length, other factors bias the growth estimates like seasonal-, size-, gender- and temperature-dependent migrations (Taylor & Collie 2003, Berghahn 1983, Pihl & Rosenberg 1982, van der Baan 1975) making sampling of the whole population and all size classes at time difficult.

Laboratory studies should avoid most of the problems that occur in the field since individuals can be observed under controlled conditions, but as variability was not

only reported from field but also from laboratory experiments, it is most likely that there are factors that can not be controlled in a satisfactory manner and previous studies indicated that food quality is one important factor (Uhlig 2002). Identification and calibration of a growth proxy within the laboratory could provide a more robust growth estimates of *C. crangon* in the field compared to estimates based on cohort tracking. $RNA \cdot DNA^{-1}$ is one of the most commonly used indices for this purpose. The amount of RNA in a cell is supposed to correlate with protein synthetization as about 80% of the RNA is ribosomal RNA which is build up or degraded with the activity of the cell. DNA, since it is the carrier of the genetic information, is assumed to be constant per cell. $RNA \cdot DNA^{-1}$ has been shown to correlate with growth mainly for larval fish (Peck et al. 2003, Buckley et al. 1999, Hovenkamp & Witte 1991, Buckley 1984) but also for corals (Buckley & Szmant 2004), scallops (Lodeiros et al. 1996), copepods (Wagner et al. 1998, Dagg & Littlepage 1972) and Crustaceans (Juinio & Cobb 1994, Moss 1994a, Moss 1994b). However in some studies a correlation was not given (Lee et al. 2006, Norkko et al. 2006, Mathers et al. 1994). For *C. crangon* an examination of this proxy has not been conducted so far.

The lack of information on ages of brown shrimp not only complicates estimates of growth but also estimates of mortality and maximum age, and uncertainty in the latter, is assumed to be between one to five years (Oh et al. 1999, Tiews 1954, Lloyd & Yonge 1944, Havinga 1930, Havinga & Willer 1929). Knowing the total mortality of the shrimp could improve these estimates. Total mortality Z, the sum of fishing (F) and natural (M) mortality, can either be estimated by single determination of F and M, by application of length frequency based methods, or by estimating the quotient of production (P) and biomass (B) (Allen 1971). Due to the numerous predators of *C. crangon* and variable stomach contents of *C. crangon* in predator stomachs, an estimation of M is difficult. Further the annual mean standing biomass has to be estimated accurately, which is difficult due to uncertainties in catchability and vulnerability of the shrimp to the fishing gear. Determination of F is also difficult, at least for small sized shrimps, due to high discard volumes (Neudecker et al. 2006) and again accurate standing biomass estimates. Estimating Z as P/B would afford to sample the whole population over a long time and a large area, what is due to patchy distribution (Eriksson et al. 2005), interannual differences (Henderson et al. 2006, Siegel et al. 2005) and the before mentioned seasonal-, length- and gender-specific migrations hardly possible. The methods of choice to determine total mortality are therefore based on length frequency distributions (de Graaf & Dekker 2006, Sparre et al. 1989, Wetherall et al. 1987, Pauly 1983, Hoenig et al. 1983, Jones & van Zalinge 1981, Powell 1979, Ssentongo & Larkin 1973, Beverton & Holt 1956). Some of these methods were applied to determine total mortality of *C. crangon* in the past, but without the exact knowledge of the accuracy of the methods. A basic assumption of these methods is no seasonality in growth, mortality and spawning. Applying the methods to conditions of the brown shrimp might therefore lead to biased results that have to be quantified prior to application.

Beside growth, mortality and age, details of the sexual system of *C. crangon* are also controversial and still debated. Boddeke proclaimed that *C. crangon* is a protandric hermaphrodite, with all males changing gender after reaching a certain size (Boddeke et al. 1991, Boddeke 1966a, Boddeke 1961). To clarify this issue and to confirm Boddeke's statement (that has also been references as unpublished data) - Martens & Redant (1986) analyzed field samples to determine the amount male shrimps actually changing gender, indicated by developing oocytes. They observed up to 9% of male shrimps undergoing potential sex change but stated that this could also be an effect of malnutrition (Martens & Redant 1986). Another approach to clarify this task was performed in an experiment where single males were kept under laboratory conditions (Schatte & Saborowski 2005). After each moult external sexual characteristics of exhuvies were examined, which are the endopodites of the first and second pleopode (Tiews 1970). Sex change occurred in their trials only in one of 70 male shrimps. As shrimps were kept separated a socially induced sex change could not be examined.

Boddekes statement about sex change finally lead to the closure of the fodder shrimp fishery (undersized <50 mm shrimps caught for the production of animal food). This was based on the argument that if small shrimps are fished to extensive, there will be a substantial loss of larger females supporting the population with new recruits. Fodder shrimping was until then a major fishery and landings of undersized shrimps by far exceeded consumption shrimps landings until the 1970s. Later food consumption shrimping dominated (Schumacher 1979).

Parallel to the reduction of fodder shrimp landings, the landings of consumption shrimps has constantly increased since the 1970s. Highest landings from the whole North Sea were registered in 2005 with about 38000 t (ICES 2007, Neudecker & Damm 2006). Due to the high prices this fishery is the most lucrative fishery in Germany including approximately 100 companies directly dependent on this acquisition. In Schleswig-Holstein 18.6 Mio € from 7100 t shrimps were earned in 2007 (Anonymus 2007) and the commercial value of the whole north Sea landings sums up to about 100 million €.

Based on stronger engines, larger vessels and bigger nets the number of shrimp fishers declined since 1950 and was reduced from 300 to 100 in Schleswig-Holstein by 1994. Catch per unit effort remained about on a constant level since data collection (ICES 2007). During the last decade the traditional fleet is supplemented with bigger and more powerful trawlers from the flatfish fishery which is, to a great extent, unregulated at the moment. Due to the more powerful trawlers, a shift of fishing effort towards winter has been observed, as now also during bad weather high priced shrimps can be landed. The effects of these recent changes are not predictable due to the uncertainties that exist in the life cycle. Assuming that fall landings are based on winter egg hatched animals a reduction of the spawners biomass might significantly influence the population (Berghahn 1991).

Premised on the previous described existing uncertainties and controversies the actual work was designed to close the knowledge gaps within the life cycle and population dynamics of *C. crangon*. Based on the described restrictions of cohort tracking a laboratory approach was chosen to derive a comprehensive, temperature and length dependent growth model. To transfer laboratory growth rates to the field, and to be able to interpret field data in a better way, the applicability of dry weight and $RNA \cdot DNA^{-1}$ as a growth proxy was tested. To clarify the question whether the brown shrimp is food limited in the North Sea, the same indices were tested for their quality to determine feeding history, condition and starvation periods of *C. crangon*. In a second step the indices were applied to field samples of the shrimp to describe its nutrition. Apart from growth the second focus was set on an accurate estimate of the total mortality rates including mainly two aspects: first to evaluate whether existing mathematical methods are applicable to the conditions of the brown shrimp and second to apply the best methods to field data to determine *Z*. The third objective of the work was to determine the sexual system of *C. crangon* and whether the shrimp is an obligate hermaphrodite or not.

References

Aarnio K, Bonsdorff E, Rosenback N (1996) Food and feeding habits of juvenile flounder *Platichthys flesus* (L.), and turbot Scophthalmus maximus L. in the Åland archipelago Northern Baltic Sea. Journal of Sea Research 36:311-320

Al-Adhub AHY, Naylor, E (1975) Emergence rhythms and tidal migrations in the brown shrimp *Crangon crangon*. Journal Marine Biology Association UK 55:801-810

Allen JA (1960) On the biology of *Crangon allmani* (Kinahan) in Northumberland waters. Journal Marine Biology Association UK 39:481-508

Allen RK (1971) Relation between production and biomass. Fisheries Research Board of Canada 28:1573-1581

Anonymus. Bericht zur Situation der Nord und Ostseefischerei. Drucksache 16/1553, 1-58. 2007. Bericht der Landesregierung, Schleswig-Holsteinischer Landtag.

Basimi RA, Grove DJ (1985) Estimates of daily food intake by an inshore population of *Pleuronectes platessa* L. off eastern Anglesey, North Wales. Journal of Fish Biology 27:505-520

Beaumont AR, Croucher T (2006) Limited stock structure in UK populations of the brown shrimp, *Crangon crangon*, identified by morphology and genetics. Journal Marine Biology Association UK 86:1107-1112

Behrends G (1985) Zur Nahrungswahl von Seehunden (*Phoca vitulina* L.) im Wattenmeer Schleswig-Holsteins. Zeitschrift für Jagdwissenschaften 31:1-14

Berghahn R (1983) Untersuchungen an Plattfischen und Nordseegarnelen (*Crangon crangon*) im Eulitoral des Wattenmeeres nach dem Übergang zum Bodenleben. Helgoländer Meeresuntersuchungen 36:163-181

Berghahn R (1991) Winterfischerei Ja oder Nein? Einige biologische Grundlagen für die Beurteilung dieser Frage sowie erste Untersuchungsergebnisse. Das Fischerblatt 6:173-180

Beukema JJ (1992) Dynamics of juvenile shrimp *Crangon crangon* in a tidal flat-nursery of the Wadden Sea after mild and cold winters. Marine Ecology Progress Series 83:157-165

Beverton RJH, Holt SJ (1956) A review of methods for estimating mortality rates in fish populations, with special reference to sources of bias in catch sampling. Rapport et Procès-verbaux des Réunions, Conseil international pour L'Exploration de la Mer 140:67-83

Beyst B, Hostens K, Mees J (2001) Factors influencing fish and macrocrustacean communities in the surf zone of sandy beaches in Belgium: temporal variation. Journal of Sea Research 46:281-294

Bilgin S, Samsun O (2006) Fecundity and egg size of three different shrimp species, *Crangon crangon, Palaemon adspersus* and *Palaemon elegans* (Crustacea: Decapoda: Caridea), off SinopPeninsula (Turkey) in the Black Sea. Turkish Journal of Zoology 30:413-421

Blahudka S, Türkay M (2002) A population study of the shrimp *Crangon allmanni* in the German Bight. Helogland Marine Research 56:190-197

Bluhm BA, Brey T (2001) Age determination in the Antarctic shrimp *Notocrangon antarcticus* (Crustacea: Decapoda), using the autofluorescent pigment lipofuscin. Marine Biology 138:247-257

Bluhm B, Beyer Kerstin, Niehoff B (2002) Brain structure and histological features of Lipofuscin in two Antarctic caridea (Decapoda). Crustaceana 75:61-67

Boddeke R (1961) Sex in the brown shrimp *(Crangon crangon)*. ICES Shellfish Committee C II 1961 Nr.50

Boddeke R (1982) The occurrence of winter and summer eggs in the brown shrimp (*Crangon crangon*) and the pattern of recruitment. Netherlands Journal of Sea Research 16:151-162

Boddeke R, Bosschieter JR, Goudswaard P (1991) Sex change, mating and sperm transfer in *Crangon crangon*. In: Bauer RT, Martin JW (eds) Crustacean sexual biology. Columbia University press, New York, p 164-182

Boddeke R, Driessen G, Doesburg W, Ramaekers G (1986) Food availability and predator presence in a costal nursery area of the brown Shrimp (*Crangon crangon*). Ophelia 26:77-90

Braber L, de Groot SJ (1973) The food of five flatfish species (Pleuronectiformes) in the Southern North Sea. Netherlands Journal of Sea Research 6:163-172

Buckley BA, Szmant AM (2004) RNA/DNA ratios as indicators of metabolic activity in four species of Caribbean reef- building corals. Marine Ecology Progress Series 282:143-149

Buckley L, Caldarone E, Ong TL (1999) RNA-DNA ratio and other nucleic acid-based indicators for growth and condition of marine fishes. Hydrobiologia 401:265-277

Buckley LJ (1984) RNA-DNA ratio. an index of larval fish growth in the sea. Marine Biology 80:291-298

Bulnheim H-P, Schwenzer DE (1993) Zur Populationsgenetik von *Crangon crangon* und *C. allmanni* (Crustacea, Decapoda) im Bereich der europäischen Küsten. Zoologische Jahrbücher. Abteilung für allgemeine Zoologie und Physiologie der Tiere 97:327-347

Cattrijsse A, Dankwa HR, Mees J (1997) Nursery function of an estuarine tidal marsh for the brown shrimp *Crangon crangon*. Journal of Sea Research 38:109-121

Caudri LWD (1937) Einfluss der Temperatur und des Salzgehalts auf die Sterblichkeit von Garnelen (*Crangon crangon* L.). Archives Neerlandaises de Zoologie 3:179-196

Che Mat CRB, Potts WTW (1985) Water balance in *Crangon vulgaris*. Comparative Biochemistry and Physiology 82A:705-710

Cieluch U, Charmantier G, Grousset E, Charmantier-Daures M, Anger K (2005) Osmoregulation, Immunolocilization of Na+/k+-ATPase, and Ultrastructure of Branchial Epithelia in the Developing brown shrimp, *Crangon crangon* (Decapoda, Caridea). Physicological and Biochemical Zoology 78:1017-1025

Clifford, Sigfried A. and Moran, David. Crangonid shrimps. US Army corps of Engineers, Coastal Ecology Group and US Department of the Interior, Fish and Wildlife Service. 82(11.125) TR EL-82-4, 1-18. 2006.

Criales MM, Anger K (1986) Experimental studies on the larval development of the shrimps *Crangon crangon* and *C. allmanni*. Helgoländer Meeresuntersuchungen 40:241-265

Daan N (1973) A quantitative analysis of the food intake of north sea cod, *Gaduas morhua*. Netherlands Journal of Sea Research 6:479-517

Daan, N. ed. Data base report of the stomach sampling project 1981. Cooperative research report 164, 144. 1989.

Dagg MJ, Littlepage JL (1972) Relationships between growth rate and RNA, DNA, Protein and dry weight in *Artemia salina* and *Euchaeta elongata.* Marine Biology 17:162-170

de Graaf G, Dekker P (2006) A simple spreadsheet model to incorporate seasonal growth into length-based stock assessment methods. NAGA World Fish Center Quarterly 29:46-54

Dornheim H (1969) Beiträge zur Biologie der Garnele *Crangon crangon* (L.) in der Kieler Bucht. Berichte der Deutschen Wissenschaftlichen Komission für Meeresforschung 20:179-215

Edwards RRC (1978) Effects of water-soluble oil fractions on metabolism, growth and carbon budget in the shrimp *Crangon crangon*. Marine Biology 46:259-265

Eriksson SP, Wennhage H, Norkko J, Norkko A (2005) Episodic disturbance events modify predator-prey interactions in soft sediments. Estuarine Coastal and Shelf Science 64:289-294

Evans S (1984) Energy budgets and predation impact of dominant epibenthic carnivores on a shallow soft bottom community at the Swedish west coast. Estuarine, Coastal and Shelf Science 18:651-672

Feller RJ (2006) Weak meiofaunal trophic linkages in *Crangon crangon* and *Cracinus maenas*. Journal of Experimental Marine Biology and Ecology 330:274-283

Gaten E (1998) Optics and phylogeny: is there an insight? The evolution of superposition eyes in the Decapoda (Crustacea). Contributions to Zoology 67:223-236

Gelin A, Crivelli AJ, Rosecchi E, Kerambrun P (2001) Can salinity changes affect reproductive success in the brown shrimp *Crangon crangon*. Journal of Crustacean Biology 21:905-911

Groh, Katharina. Die Besiedelung von Lahnungsfeldern in den Nordseewatten durch Jugendstadien von *Crangon crangon* (L.). 1. 1982. Universität Hamburg.

Gunnarsson B, Ásgeirsson þ, Ingólfsson A (2007) The rapid colonization by *Crangon crangon* (Linnaeus, 1758) (Eucarida, Caridea, Crangonidae) of Icelandic coastal waters. Crustaceana 80:747-753

Haefner Jr. PA (1971) Avoidance of anoxic conditions by the sand shrimp, *Crangon septemspinosa* Say. Chesapeake Science 12:50-51

Hagerman L (1970) The oxygen consumption of *Crangon vulgaris* (Fabricius) (Crustacea, Natantia) in relation to salinity. Ophelia 7:283-292

Hagerman L, Vismann B (1995) Anaerobic metabolism in the shrimp *Crangon crangon* exposed to hypoxia, anoxia and hydrogen sulfide. Marine Biology 123:235-240

Hagermann L (1970) Locomotory activity patterns of *Crangon vulgaris* (Fabricius)(Crustacea, Natantia). Ophelia 8:255-266

Hamerlynck O, Hostens K (1993) Growth, feeding, production, and consumption in 0-group bib (*Trisopterus luscus* L.) and whiting (*Merlangius merlangus* L.) in a shallow coastal area of the south-west Netherlands. ICES Journal of Marine Science 50:81-91

Hanamura Y, Matsuoka M (2003) Feeding habits of the sand shrimp, *Crangon uritai* Hayashi & Kim, 1999, in the central seto inland sea, Japan. Crustaceana 76:1017-1024

Hartnoll R (2001) Growth in Crustacea – twenty years on. Hydrobiologia 449:111-122

Hartsuyker L (1966) Daily tidal migrations of the shrimp *Crangon crangon* L. Netherlands Journal of Sea Research 3:52-67

Havinga B (1930) Der Granat (*C. vulgaris*, Fabr.) in den holländischen Gewässern. Journal du Conseil International pour l'Exploration de la Mer 5:57-87

Henderson PA, Holmes RHA (1987) On the population biology of the common shrimp *Crangon crangon* (L.) (Crustacea: Caridea) in the Severn Estuary and Bristol Channel. Journal of the Marine Biological Association U.K. 67:825-847

Henderson PA, Seaby RMH, Somes JR (2006) A 25-year study of climatic and density-dependent population regulation of common shrimp *Crangon crangon* (Crustacea: Caridea) in the Bristol Channel. Journal of the Marine Biological Association U.K. 86:287-298

Hislop JRG, Robb AP, Bell MA, Armstrong DW (1991) The diet and food composition of whiting (*Merlangius merlangus*) in the North Sea. ICES Journal of Marine Science 48:139-156

Hoenig JM, Lawing WD, Hoenig NA (1983) Using mean age, mean length and median length data to estimate the total mortality rate. International Council for the exploration of the Sea ICES-CM-1983:

Hovenkamp F, Witte JIJ (1991) Growth, otolith growth and RNA/DNA ratios of larval plaice Pleuronectes platessa in the North Sea 1987 to 1989. Marine Ecology Progress Series 70:105-116

ICES. Report of the *Crangon* Fisheries and life history. ICES CM 2006/G:XX, 1-32. 2006.

ICES (2007) Report of the Working Group on Crangon Fisheries and Life History. International Council for the exploration of the Sea Living resources comittee:1-33

Janssen GM, Kuipers BR (1980) On tidal migration in the shrimp *Crangon crangon*. Netherlands Journal of Sea Research 14:339-348

Jones R, van Zalinge NP (1981) Estimates of mortality rate and population size for shrimp in Kuwait waters. Kuwait Bulletin of Marine Science 2:273-288

Juinio MAR, Cobb JS (1994) Estimation of recent growth rates of field-caught postlarval amercian lobsters, Homarus americanus, from RNA:DNA ratios. Canadian Journal Fisheries and Aquatic Science 51:286-294

Klek-Kawinska E, Bomirski A (1975) Ovary-inhibiting hormone activity in Shrimp (*Crangon crangon*) eyestalks during the annual reproductive cycle. General and Comparative Endocrinology 25:9-13

Kodama K, Yamakawa T, Shimizu T, Aoki I (2005) Age estimation of the wild population of Japanese mantis shrimp *Oratosquilla oratoria* (Crustacea: Stromatopoda) in Tokyo Bay, Japan, using lipofuscin as an age marker. Fisheries Science 71:141-150

Kuipers BR, Dapper R (1984) Nursery function of Wadden Sea tidal flats for the brown shrimp *Crangon crangon*. Marine Ecology Progress Series 17:171-181

Labat J-P (1977) Écologie de *Crangon crangon* (L.) (Decapoda, Caridea) dans un étang de la côte languedocienne. Vie Milieu XXVII:273-292

Lee O, Danilowicz BS, Dickey-Collas M (2006) Temporal and spatial variability in growth and condition of dab (*Limanda limanda*) and sprat (*Sprattus sprattus*) larvae in the Irish Sea. Fisheries Oceanography 15:490-507

Lodeiros CJM, Fernández RI, Bonmatí A, Himmelman JH, Chung KS (1996) Relation of RNA/DNA ratios to growth for the scallop *Euvola (Pecten) ziczac* in suspended culture. Marine Biology 126:245-251

Luttikhuizen PC, Campos J, van Bleijswijk J, Peijnenburg KTCA, van der Veer HW (2008) Phylogeography of the common shrimp, *Crangon crangon* (L.) across its distribution range. Molecular Phylogenetics and Evolution 46:1015-1030

Martens E, Redant F (1986) Protandric hermaphroditism in the brown shrimp *Crangon crangon* (L.), and its effects on recruitment and reproductive potential. International Council for the exploration of the Sea Shellfish Committee:1-15

Mathers EM, Houlihan DF, Buren LJ (1994) RNA, DNA and protein concentrations in fed and starved herring *Clupea harengus* larvae. Marine Ecology Progress Series 107:223-231

Meixner R (1969) Wachstum, Häutung und Fortpflanzung von *Crangon crangon* (L.) bei Einzelaufzucht. Berichte der Deutschen Wissenschaftlichen Komission für Meeresforschung 20:93-111

Modlin RF (1980) The life cycle and recruitment of the sand shrimp, *Crangon septemspinosa*, in the Mystic River Estuary, Conneticut. Estuaries 3:1-10

Moss SM (1994a) Growth rates, nucleic acid concentrations, and RNA/DNA ratios of juvenile white shrimp, *Penaeus vannamei* boone, fed different algal diets. Journal of Experimental Marine Biology and Ecology 182:193-204

Moss SM (1994b) Use of nucleic acids as indicators of growth in juvenile white shrimp, *Penaeus vannamei*. Marine Biology 120:359-367

Nakaya M, Takatsu T, Nakagami M, Joh M, Takahashi T (2004) Spatial distribution and feeding habits of the shrimp *Crangon uritai* as a predator on larval and juvenile marbled sole *Pleuronectes yokohamae*. Fisheries Science 70:445-455

Neudecker, T. and Damm, U. Seasonality of egg-bearing shrimp (*Crangon crangon* L.) in costal waters of the German Bight. K: 28, 1-9. 1992. ICES Shell Fish Committee.

Neudecker T, Damm U (2006) 2005: Rekord-Anlandungen an Nordseegarnelen (*Crangon crangon* L.) in Europa. Informationen aus der Fischereiforschung 53:80-81

Neudecker T, Damm U, Kühnhold W (2006) Fang, Anlandungen, Discard und Bestand der Nordseegarnele (*Crangon crangon* L.). Informationen aus der Fischereiforschung 53:82-85

Norkko J, Thrush SF, Wells RMG (2006) Indicators of short-term growth in bivalves: Detecting environmental change across ecological scale. Journal of Experimental Biology and Ecology 337:38-48

Oh C-W, Hartnoll RG (2000) Effects of food supply on the growth and survival of the common shrimp *Crangon crangon* (Linnaeus, 1758) (Decapoda, Caridea). Crustaceana 73:83-99

Oh C-W, Hartnoll R, Nash RDM (1999) Population dynamics of the common shrimp, *Crangon crangon* (L.), in Port Erin Bay, Isle of Man, Irish Sea. ICES Journal of Marine Science 56:718-733

Paschke KA, Gebauer P, Buchholz F, Anger K (2004) Seasonal variation in starvation resistance of early larval North Sea shrimp *Crangon crangon* (Decapoda: Crangonidae). Marine Ecology Progress Series 279:183-191

Pauly D (1983) Length converted catch curves: A powerful tool for fisheries research in the tropics (part I). Fish Bytes 1:9-13

Peck MA, Buckley LJ, Caldarone EM, Bengtson DA (2003) Effects of food consumption and temperature on growth rate and biochemical-based indicators of growth in early juvenile Atlantic cod *Gadus morhua* and haddock *Melanogrammus aeglefinus.* Marine Ecology Progress Series 251:233-243

Pihl L (1985) Food selection and consumption of mobile epibenthic fauna in shallow marine areas. Marine Ecology Progress Series 22:169-179

Pihl L, Rosenberg R (1982) Production, abundance, and biomass of mobile epibenthic marine fauna in shallow waters, western Sweden. Journal of Experimental Marine Biology and Ecology 57:273-301

Pihl L, Rosenberg R (1984) Food selection and consumption of the shrimp *Crangon crangon* in some shallow marine areas in western Sweden. Marine Ecology Progress Series 15:159-168

Powell DG (1979) Estimation of mortality and growth parameters from the length frequency of a catch. Rapport et Procès-verbaux des Réunions, Conseil international pour L'Exploration de la Mer 175:167-169

Price KS (1962) Biology of the sand shrimp, Crangon septemspinosa, in the Shore Zone of the Delaware Bay Region. Chesapeake Science 3:244-255

Redant, F. Konsumptie en pruoduktie van post-larval *Crangon crangon* (L.) (Crustacea, Decapoda) in de Belgische kustwateren, DEEL I. TEKST. 1978. Vrije Universiteit Brussel, Fakulteit Wetenschappen.

Redant F (1984) An indexed bibliography on the common shrimp, *Crangon crangon* (L.), complementary to Tiews, 1967. International Council for the exploration of the Sea, Shellfish Committee C.M. 1984:

Regnault M (1979) Ammonia Excretion of the Sand-Shrimp *Crangon crangon* (L.) During the moult cycle. Journal of Comparative Physiology 133:199-204

Schatte J, Saborowski R (2005) Change of external sexual characteristics during consecutive moults in *Crangon crangon* L. Helgoland Marine Research

Schumacher A (1979) On the population dynamics of the brown shrimp (*Crangon crangon* L.) off the German coast. Rapport et Procès-verbaux des Réunions, Conseil international pour L'Exploration de la Mer 175:280-286

Sheehy MRJ, Greenwood JG, Fielder DR (1994) More accurate chronological age determination of crustaceans from field situations using the age marker, lipofuscin. Marine Biology 121:237-245

Sheehy MRJ, Shelton PMJ, Wickins JF, Belchier M, Gaten E (1996) Ageing the European lobster *Homarus gammarus* by the lipofuscin in its eyestalk ganglia. Marine Ecology Progress Series 143:99-111

Siegel V, Damm U, Neudecker T (2008) Sex-ratio, seasonality and long-term variation in maturation and spawning of the brown shrimp *Crangon crangon* (L.) in the German Bight (North Sea). Helgoland Marine Research 62:

Siegel V, Gröger J, Neudecker T, Damm U, Jansen J (2005) Long-term variation in the abundance of the brown shrimp *Crangon crangon* (L.) population of the German Bight and possible causes for its interannual variability. Fisheries Oceanography 14:1-16

Spaargaren DH, Haefner Jr. PA (1987) The effect of environmental osmotic conditions on blood and tissue glucose levels in the brown shrimp *Crangon crangon* (L.). Comparative Biochemistry and Physiology 87A:1045-1050

Sparre P, Ursin E, Venema SC (1989) Introduction to tropical fish stock assessment. Food and Agriculture organization of the united nations, Rome

Ssentongo GW, Larkin PA (1973) Some simple methods of estimating mortality rates of exploited fish populations. Journal of the Fisheries Research Board Canada 24:2355-2453

Temming A, Damm U (2002) Life cycle of *Crangon crangon* in the North sea: A simulation of the timing of recruitment as a function of the seasonal temperature signal. Fisheries Oceanography 11:45-58

Tetard A (1985) Elements sur la croissance de la crevette grise, *Crangon crangon* (L.), en manche-est et sud mer du nord. ICES C.M. K:17:16pp.

Tiews K (1970) Synopsis of biological data on the common shrimp *Crangon crangon* (Linaeus, 1758). FAO Fisheries Reports 4:1167-1223

Uhlig, Nadine. Wachstum und Ernährungssituation der Nordseegarnele *Crangon crangon* (L.). -114. 2002. Institute for Hydrobiology and fishery science, Hamburg.

van Donk E, de Wilde PAWJ (1981) Oxygen consumption and motile activity of the brown shrimp *Crangon crangon* related to temperature and size. Netherlands Journal of Sea Research 15:54-64

Vila Y, Medina A, Megina C, Ramos F, Sobrino I (2000) Quantification of the Age-Pigment Lipofuscin in Brains of Known-Age, Pond-Reared Prawns *Penaeus japonicus* (Crustacea, Decapoda). Journal of Experimental Zoology 286:120-130

Wagner M, Durbin E, Buckley L (1998) RNA:DNA ratios as indicators of nutritional condition of the copepod *Calanus finmarchicus*. Marine Ecology Progress Series 162:173-181

Weber RE, Spaargaren DH (1970) On the influence of temperature on the osmoregulation of *Crangon crangon* and its significance under estuarine conditions. Netherlands Journal of Sea Research 5:108-120

Weetmann D, Ruggiero A, Mariani S, Shaw PW, Lawler AR, Hauser L (2007) Hierarchical population genetic structure in the commercially exploited *Crangon crangon* identified by AFLP analysis. Marine Biology 151:565-575

Wetherall JA, Polovina JJ, Ralston S (1987) Estimating growth and mortality in steady-state fish stocks from length frequency data. In: D.Pauly and G.R.Morgan, Editors, Length-based Methods in Fishery Research53-74

Determining growth rates of the brown shrimp *Crangon crangon* (L.): review and laboratory approach.

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Abstract

Laboratory experiments were performed to determine growth rates of different sized *Crangon crangon* at different temperatures under ad libitum feeding conditions. Mean (± sd) growth rates ranged from 0.04 ± 0.03 to 0.56 ± 0.1 mm·d^{-1} at 5 and 25°C, respectively. Beside water temperature and total length, catch date influenced growth rates, indicating animals of recent recruitment waves grow faster than overwintering shrimps of the same size. Mean ± sd individual moult intervals were determined using a marking method and varied between 8 ± 3.6 and 45 ± 7.6 days for 30 mm L shrimp at 5°C and 25°C, respectively. Variability in the observed growth rates was mainly an effect of variable increments whereas moulting occurred at regular intervals. The impacts of T (°C) and L (mm) on moult interval (mi) was best described by: $mi = 5.7066 \cdot L^{0.7364} \cdot e^{-0.09363 \cdot T}$ (r^2=0.945). A growth rate (G, mm d^{-1}) model including data from this and other studies was derived: $G = 0.02398 \cdot T - 0.000966 \cdot e^{0.09509 \cdot T} \cdot L$ (r^2 = 0.860). Applying quantile regression (0.75 percentile) a maximum growth model (G_{MAX}, mm d^{-1}) was derived that describes the growth rate of the fastest growing fraction of the population: $G_{MAX} = 0.03218 \cdot T - 0.001227 \cdot e^{0.09777 \cdot T} \cdot L$ (r^2 = 0.857). According to simulations the best correspondence from invading juveniles in spring and peak landings in September (commercial size at 50 mm) was obtained with maximum growth parameters and not with mean growth rates. The reason for this is most likely high cumulative mortality of slow growing shrimps.

Key words:

growth rates, moult interval, moult increments, maximum growth, life cycle, *Crangon crangon*

Introduction

The brown shrimp *Crangon crangon* (Crustacea, Decapoda) is one of the most common epibenthic species along the coastline of Europe (Lapinska & Szaniawska 2006, Cuesta et al. 2006, Gamito & Cabral 2003, Gelin et al. 2001, Bulnheim & Schwenzer 1993, Henderson & Holmes 1987, Pihl & Rosenberg 1984). Small, juvenile *C. crangon* mainly inhabit coastal soft bottom substrates and use the shallow intertidal parts of the Wadden Sea as nursery area (Cattrijsse et al. 1997, Kuipers & Dapper 1984) whereas adult shrimps are mainly situated in deeper water (Janssen & Kuipers 1980). Brown shrimp support a large fishery, in the Southern North Sea, with landings of up to 37000 tons in 2005 (ICES 2006). About 600 inshore twin beam trawlers are involved in the fisheries of the Netherlands, Germany and Denmark producing annual shrimp landings with a fish market value between 50 and 100 Million Euro (ICES 2007, Revill & Holst 2004).

The breeding season of *C. crangon* extends throughout the year along the Dutch and German Coasts (Kuipers & Dapper 1984) with the highest and lowest numbers of egg-bearing shrimps observed from February to July and September through October, respectively (Siegel et al. 2008, Neudecker & Damm 1992).

Over the year, two different types of eggs can be observed: summer (April to September) and winter eggs (October to March) (Havinga 1930). The two types differ according to size, dry mass, carbon content (Paschke et al. 2004) and larval hatch size (Boddeke 1982) with in all cases lower values for the summer eggs.

After hatch *C. crangon* passes up to six different planktonic zoeal stages (Criales & Anger 1986, Gurney 1982) ending in a benthic stage of approximately 5 mm total length (L). Highest abundances of post-larval and early juvenile *C. crangon* are observed within inter-tidal areas in late spring and early summer (May-June) with mean length of 10-20 mm (Temming & Damm 2002a, Beukma 1992, Boddeke 1976). Commercial catches, that mainly contain adult shrimp > 50 mm L, peak in late summer and fall (September, October) (ICES 2007, Henderson et al. 2006, Maes et al. 1998). Given the high total mortality rates reported for *C. crangon* of approximately $Z = 4.2\ y^{-1}$ (Temming et al. 1993) the peak in autumn catches can only originate from the observed juvenile peak in May, June of the same year.

Kuipers & Dapper (1984) were the first to speculate that the increase in commercial catches in the fall were associated with winter recruits. However, they assumed growth rates that were too low and did not match with the invasion of juveniles in spring (Beukema 1992). Growth rates of at least 0.23 to 0.29 $mm \cdot d^{-1}$ are required to match the sizes of shrimps (15 mm L) during the recruitment peak generally observed in May and that (50 mm) observed during the peak commercial landings in September to October. The majority of reported growth rates are much lower and lie between 0 to 0.1 mm d^{-1} (Oh & Hartnoll 2000, Henderson & Holmes 1987). Only few authors have reported high growth rates (e.g., 0.2 to 0.54 mm d^{-1} (Beukma 1992); 0.07 to0.4 mm d^{-1} (Tetard 1985)) and the results obtained are often highly variable. Given this high variability, studies that have reported high growth rates but which are based on a small number of animals (e.g., Meixner

1969, and Uhlig 2002) are difficult to interpret. In short, new experimental research is therefore recommended to produce a robust growth model.

If it would be possible to show that the autumn catch peak is build up by winter egg hatched juveniles this would not only close uncertainties in the life cycle but also have implications on the increasing winter fishery. This fishery is fueled by high prices but has been criticized for potentially decreasing catches in the fall fishery (Berghahn 1991). An increased winter fishery might decrease the breeding stock and if the winter eggs are necessary for replenishing the next generation, this might have marked implications for populations of *C. crangon*.

In small, short-lived species, generally only the fast-growing animals contribute to the spawning stock, especially if mortality rates decrease with increasing size. A meta-analysis indicated that larger fish have lower mortality levels than smaller ones (Peterson & Wroblewski 1984) and there are indications from the analysis of field data that total mortality in brown shrimps also decreases with increasing size. For small shrimps a Z of 22 (del Norte-Campos & Temming 1998) was determined whereas for large ones it ranges between 2.78 and 6.08 (Temming et al. 1993). It could be shown for larval and juvenile Atlantic herring (*Clupea harengus*), striped bass (*Morone saxatilis*) and Atlantic cod (*Gadus morhua*) that if lowest and fastest growth rates were combined with daily constant mortality rates, more than 100-fold differences in larval stage survival could result (Houde 1987). In a similar fashion, it might be assumed that mainly the fast- growing *C. crangon* survive to be captured in the fall fisheries. Therefore, the main focus of this study was to investigate the growth variability and maximum growth rate. Slow growing individuals stay longer within the size window of higher predation what directly results in a higher cumulative mortality (Cowan et al. 1996, Houde 1987).

Specific objectives of this study were to:

- investigate the influence of the food source on the growth performance,
- determine length and temperature dependent maximum growth rates of *C. crangon* under ad libitum conditions with optimal food,
- combine the new findings with additional data from the literature to develop a comprehensive model of mean and maximum growth rates of *C. crangon,*
- test the growth model by calculation of length trajectories based on observed field water temperatures,
- investigate the amount of individual variability separately for moult increments and moult duration using individual marking.

Material and Methods

Marking method and growth determination

A marking method was applied in several experiments to track individual shrimps so that individual growth rates, moult intervals and moult increments could be calculated. In these experiments, animals were marked after ecdysis with a small

colored plastic plate which was dipped into a drop of superglue and then placed on the Carapax or on the fourth segment with a pair of tweezers. Superglue was chosen because it immediately hardens in contact with water and preliminary trials showed no negative effect due to mortality and growth. A comparable method was applied by Henderson & Holmes (1987). After ecdysis, the exhuvia with the mark is released and the shrimp is not marked any more. If animals of slightly different size and known sex (can be determined from the exhuvia) are combined in one aquarium, up to 10 (5 colors, two positions) animals can be individually tracked with a high degree of certainty. The aquaria were checked daily for “naked” shrimps and were measured after calcification and hardening of the new exoskeleton. After the measurement a new mark was added. The growth rate is determined from the quotient of moult increment and moult interval.

Growth rate of unmarked shrimps was determined using a slightly different method. In these experiments animals were pre-sorted to one length (accuracy 1 mm) and placed in one aquarium. At the end of the experiment all animals were measured again and growth was determined from the elapsed time and the difference between initial and final lengths. These experiments will be referred to as “unmarked”.

In all experiments total length (L, mm) was determined from the scaphocerite to the tip of the telson to the lower mm using mm paper.

Temperature and salinity were measured in each aquarium each day. For all *C. crangon* gender was determined by the endopodite of the first pleopod and the appendix masculina at the endopodite of the second pleopod (Tiews 1970). To minimize handling stress marked animals were sexed by the exhuvia and at the end of the experiment. The unmarked animals were only sexed at the end of the trial prior to freezing. Exhuvia were not returned to the aquaria.

Preliminary feeding trials

Two preliminary trials were conducted at the Institute of Hydrobiology and Fisheries Science in Hamburg, Germany prior to the main growth experiments in July and August 2005.

In the first experiment, 60 marked animals of 18 mm size were held in group of 10 animals within six plastic aquaria (10 l, flow through, recirculation system containing 30 m³ sea water). Sand (500 - 1000 µm dried at 70°C) was provided within small basins. Each group was fed with a specific food source, either: frozen sprat (*Sprattus sprattus),* fresh mantle tissue of *Cerastoderma edule*, fresh tissue of *Littorina littorea*, frozen C. *crangon cr.*, pellet food (Dana feed) or brine shrimp (*Artemia salina*) nauplii. Growth rates were determined over 25 days for each group which included 2-3 moults. The water temperature was 15°C. For each shrimp, the mean growth rate was determined and from this mean values one growth rate for each food source (aquarium) was derived.

In the second preliminary feeding experiment, 18 marked *C. crangon* of 20 mm size were held in an enclosed recirculation system (1 m³) in a basin covered with sand. During the first 35 days animals were fed with *Cerastoderma edulis* and pellet feed following a period of 10 d where the same shrimps were fed with about 5000 (280 per shrimp) adult copepods (*Acartia tonsa)* each day. After that, feeding was switched back to pellets (Dana feed) and polychaetes (purchased from a fishing store). The water temperature was 18.2°C. For each observed moult event, the determined growth rate was plotted against time.

Growth experiments

Rearing the necessary amount of copepods was very labor intensive (4 weeks of copepod rearing for 18 shrimps) therefore this method was not deemed suitable for larger growth trials (containing 1000 *C. crangon).* The growth experiments were therefore performed at the "AWI-Biological Institute Helgoland", in the time from May 15th to August 11th 2006 (week 20-33), as life plankton is routinely caught there every day.

Sampling area

Shrimps for the growth experiments were caught on 8/9.5.2006 from the research vessel Uthörn in the Weser (58°49'N, 8°10'E), Elbe (54°02'N, 8°20'E) and Eider (54°17'N, 8°27'E) estuaries within 4 and 8 m depth using a 3 m beam trawl (salinity: 20.7 psu, surface water temperature: 14.4°C, wind: 4-5 Bft east). Animals were transported in a flow-through seawater basin to the institute.

Smaller individuals used for the experiments starting in May, as well as all animals applied in the experiments starting in July, were caught with a push net (2 mm mesh size) at the coast of Büsum (54°07'N, 8°51'E) in about 1 m water depth on May 12th (21.7 psu, 15.4°C water temp., 2 Bft NE) and July, 10th (26.2 psu, 19.2°C water temp., 5 Bft W). Transport to Helgoland was both times performed in plastic bags filled with about 30% seawater and 70% pure oxygen. This transportation method is commonly applied for live transport of aquaculture shrimps over longer time periods and is recommended for shipping shrimps on airplanes (Calado & Dinis 2008, NACA 2006, Sogbesan et al. 2006, APEC 1999, ASEAN 1998). Shrimps of size class 20 mm were only accessible in Büsum on July, 10th 2006.

Preliminary treatment

To minimize bias in the trials according to catch, sampling location or origin all animals available at a time were randomly combined in four 12°C, flow-through (9 l h^{-1}), sand filled seawater basins measuring 1.50 x 0.50 x 0.40 m). Distinguishing the origin of the shrimps was therefore not possible in the trials.

Shrimps were reared under these preliminary conditions from catch date to the beginning of the trials on May 21st. Individuals that were not used in the trials were held under these conditions and were used to replace animals that died during the trials (only marked shrimps). All shrimps were fed daily with live polychaets (*Nereis diversicolor, Nereis virens, Nereis pelagica, Lanice conchilega*), halves of the blue mussel (*Mytilus edulis*) and the green algae *Ulva lactuca*. Polychaets, mussels and algae were collected at low tide on the Northern shore of Helgoland. Polychaets were chosen because they provide an important source of poly-unsaturated fatty acids (Benzie & J.A.H. 1997). Furthermore, following the results of the preliminary experiment, live plankton, caught in the "Helgoland Reede" on working days, was also utilized. The volume towed was daily ~150 m³ with a 280 µm net and ~150 m³ with a 500 µm plankton net. As *C. crangon* mainly feed at night (Feller 2006), concentrated plankton was stored in aerated buckets at 10°C and equally distributed to all aquaria in the evening.

Feeding during the growth experiments

To determine maximum growth, experiments were performed under ad libitum feeding conditions. The food composition was the same as described before but contained no adult blue mussels since this prey type rapidly reduce water quality (degradation, toxic algae) especially at warm water temperatures. For each shrimp two polychaets were added each day. If no worm was left the following day, the ratio was doubled. Feeding on *Ulva lactuca* was visible by little holes in the leaves and by observation of the animals. Leafs were replaced after 5 days. Feeding on plankton was regularly visible by an increased locomotion of the shrimps and "catching" movements with the chelipeds.

Water supply and setup

As fresh water supply was too limited for a flow through system and as it is very costly to control the water temperature in an open system, a closed circulation setup was chosen. The whole experiment contained eight setups placed in three different temperature controlled rooms (Table 5-1) enabling growth determination of five different size classes (20, 30, 40, 50 and 60 mm) at five different temperatures (5, 10, 15, 20 and 25°C). Each circulation system (setup) consisted of one water tank (180 l) and 10 aquaria (polypropylene, 40 x 30 x 20 cm, 17 l when filled) a seawater pump (Oceanrunner 1200) and if necessary a regulated 600 W heating device. Temperature was controlled by the room temperature and the heating device. The standard deviation of the temperature was 0.3 to 0.7°C for temperatures > 5°C (Table 5-1) and only in the 5°C experiment it was 1.2°C, because the cooling had to work against high outside temperatures. The mean salinity in all experiments was 31.7 $\pm$ 0.6 psu. Each aquarium always contained only marked or only unmarked shrimps.

The aquaria within a setup were placed in two racks (of five layers if ten aquaria were included) in vertical order so that the outflow of the upper aquarium was the inflow of the lower one. Seawater from the storage tank was pumped with 30 $l \cdot h^{-1}$ in the top and the middle aquaria of each batch. To maximize water exchange in the aquaria the outlet of the upper aquarium was always placed opposite to the one beneath. To avoid systematic errors according to the position within the batch, the order of all ten aquaria was changed daily (as they had to be checked for exhuvia anyway) by aid of a set of random numbers. All aquaria contained a 1 cm layer of sand. Remaining food was removed every second day. Water in the storage tank was changed once a day with fresh temperature conditioned seawater. The seawater was pumped directly from the North Sea, filtered and UV treated.

Table 5-1: Experimental setup: Climate chamber, temperature, size class (contains *Crangon crangon* of the specified length ± 3 mm), growth determination (individually marked or unmarked animals), amount of aquaria in the setup and amount of C. crangon of the size class in the setup.

acronym		climate chamber	temperature in climate chamber	temperature in recirc. system	size classes	growth determination	amount of aquaria	amount per size class
M5M	setup 1	1	5°C	4.1 ± 1.2°C	30,50-60 mm	marked	10	100, 60
M10M	setup 2	2	10°C	10.7 ± 0.8°C	40,50,60 mm	marked	10	100,80,40
M15M	setup 3			15.1 ± 0.5°C (heating)	30,50 mm	marked	10	100,60
M20M	setup 4	3	20°C	20.8 ± 0.6°C	30,40,60 mm	marked	10	100,100,25
M25M	setup 5			25.1 ± 0.7	30,40 mm	marked	10	100,100
U10M	setup 6	2	10°C	10.4 ± 0.4°C	40 mm	unmarked	6 of 8	80
U20M	setup 7	3	20°C	21.± 0.6 °C	30 mm	unmarked	6 of 10	90
U10J	setup 6	2	10°C	10.4 ± 0.4°C	20 mm	unmarked	2 of 8	30
U15J	setup 8			15.1 ± 0.5°C (heating)	20,30 mm	unmarked	4	50, 50
U20J	setup 7	3	20°C	21.± 0.6 °C	20.30 mm	unmarked	4 of 10	80
U25J	setup 5			25.1 ± 0.3°C (heating)	20 mm	unmarked	1 of 10	20

Experimental design: marked shrimps May

In the following text acronyms for the trials as shown in Table 5-1 will be used. These acronyms include the method of growth determination (first letter: marked M and unmarked U) the temperature in the setup (5, 10, 15, 20, 25°C) and catch date (last letter May: M or July: J). Unmarked animals caught in July, reared at 15°C are therefore referred to as U15J.

Five setups were used for growth determination of marked shrimps containing a total of 50 aquaria (10 each). Following the preliminary treatment phase of six (Büsum shrimps) and 12 days (larger Uthörn shrimps), the animals were taken from the basins, marked and transferred to the aquaria.

To acclimate shrimp to the aquaria, setups 1 to 5 (marked shrimps) were first run for another ten days at field temperature (11°C). Then the temperature was gradually heated or cooled to the respective experimental temperatures within a

period of another ten days. The whole acclimatization therefore included about 30 days (6-10 days in the basins, 10 days at ambient temperature, 10 days of temperature change).

Animals that died during the experiment or were eaten by other *C. crangon* were replaced by animals held in the flow through tank under ad libitum conditions. This was only done in the aquaria with marked shrimps. As these replacing shrimps were acclimatized shorter to the temperature (one step over one day) their growth rates were only included in the evaluation if the complete moult cycle plus ten days was spent at the according temperature.

For each marked *C. crangon,* the growth rate was determined as mean of all growth rates observed for that single shrimp. From these mean values the mean growth per size class and temperature was derived.

Experimental design: unmarked shrimps May

To check whether the marking procedure influenced growth rates, experiments with unmarked animals (U10M, 40 mm and U20M, 30 mm) were conducted in parallel to the trials with the marked animals (marked 5°C May: M5M to marked 25°C May: M25M). These experiments were started 15 days later than the trials with the marked shrimps and were performed with shrimps taken from the ad libitum fed replacement stock.

In these trials, acclimatization was performed as followed. Animals were taken from the basins and acclimatized over 10 days step by step to the setup temperature. After these 10 days they were sorted according to length and groups of similar size were placed into one aquarium.

To get independent replicates from the marked shrimps (concerning the setup) these shrimps were placed in separated circulation systems (setup 6 and 7, Table 5-1). At 10°C 6 aquaria containing 80 shrimps and at 20°C 6 aquaria containing 90 shrimps were applied.

Experimental design: unmarked shrimps July

As mentioned before, no animals of size class 20 mm were caught close to Helgoland and also not during the May samplings in Büsum. Therefore the experiments with the 20 mm size class shrimps (U10J, U15J, U20J, U25J) were started in July with animals caught in Büsum on 10.7.2006. As these animals were too small for marking, they were treated as the unmarked shrimps. Additionally to the 20 mm groups, 30 mm groups were examined at 15 and 20°C.

Additional aquaria containing unmarked shrimps from the catch in July (reared at 10°C and 20°C) were added to existing setups containing the unmarked shrimps caught in May (setup 6 and 7). To determine growth at 15°C with the unmarked July animals, a new trial (setup 8) made up by 4 aquaria was installed. The 25°C

animals were added to setup 5 (M25M) as due to high mortality one aquarium could be replaced.

Literature review

A total of 33 articles was found containing information on growth, moult interval or moult increment of *C. crangon* (Table 5-5, Figure 5-4). In two cases (Oh & Hartnoll 2000, Oh et al. 1999) carapax length was transferred into total length by the relationship given in the according article. In three cases (Henderson & Holmes 1987, Labat 1977, Schockaert 1968) growth rates were extracted from scanned graphs by using a matlab routine (based on the image processing toolbox). For the conversion of wet weight growth (Edwards 1978) into length growth a length weight relationship of

$$ww = 4.0625 \cdot 10^{-6} \cdot L^{3.084} \quad (1)$$

was used which is based on own observations (German coastal waters monthly from 2005 to 2007, n = 8305, $r^2 = 0.985$), where *ww* is wet weight, and *L* is length.

From studies on toxic or stress effects only the control groups were used in our analysis (Oh & Hartnoll 2000, Lagardère 1982, Edwards 1978). Values determined by Dornheim (1969) are listed but were not analyzed as *C. crangon* in the Baltic shows lower values for L_∞ and lower growth rates than those in the North Sea.

Growth rates determined for larval *C. crangon* (Criales & Anger 1986) are added for the sake of completeness to the review but values were also not included into the analysis as only juvenile growth is examined here and larval growth might be different.

For the field data presented in Viegas et al. (2007) no monthly temperature were available therefore the data are listed in Table 5-5 but also not used in the growth model.

Growth function

The von Bertalanffy growth curve describes growth as the net effect of anabolism (left term) minus catabolism (right term)

$$\frac{dW}{dt} = H \cdot W^{\frac{2}{3}} - K \cdot W \quad (2)$$

In this equation W is weight, K is the catabolic constant, which is equivalent to % weight loss per time in starving individuals, and H is the anabolic constant which is related to food intake and synthesis of body mass. Transformation of (2) to length growth ($W \sim L^3$) leads to

$$\frac{dL}{dt} = E - k \cdot L \quad (3)$$

where L is length, k = K/3 and E represents anabolism. This equation was used with temperature modifying the constant E (see Kuipers & Dapper 1981) to calculate daily length growth of shrimp as a function of length and temperature. The model was fitted to unpublished data obtained by M. Fonds (Table 5-5)

$$\frac{dL}{dt} = a + b \cdot T - k \cdot L \qquad (4)$$

where *a* and *b* are constants and *T* is Temperature.

As metabolism not only depends upon length but also increases exponentially with increasing temperature (Gillooly et al. 2001), an exponential temperature term was included (4) with k = $c \cdot e^{d \cdot T}$.

$$\frac{dL}{dt} = a + b \cdot T - c \cdot e^{d \cdot T} \cdot L \qquad (5)$$

a, *b, c* and *d* are constants. L_∞, the mean maximum length can be calculated from equation (5) as the length where no growth occurs d*L*/d*t* = 0:

$$L_\infty = \frac{a + b \cdot T}{c \cdot e^{d \cdot T}} \qquad (6)$$

This shows that L_∞ is inversely related to k and directly proportional to the anabolism. As the exponential term increases faster than the linear term, it can directly be derived from this function, that L_∞ decreases with increasing temperature.

Model fitting to subsets of data

The growth rates of animals caught in May were significantly different from the growth rates of animals from July sampling (see later section). Combining the results of both trials in one model would therefore not be useful. Furthermore, the experiments performed with animals caught in July cover too few length classes to determine a full growth model for all temperatures and size classes based on these experiments.

Growth models were therefore determined for the following two different data sets:

Female and male growth rates from actual laboratory experiments (July unmarked) plus those work in the literature that separated growth rates according to gender.

Population growth rates derived from all actual laboratory experiments (male, female, marked and unmarked, May and July) plus literature values.

Both growth function, equation (4) with linear temperature term and equation (5) with exponential temperature term, were fitted to the data with nonlinear regression using the software SYSTAT 8.0. The quality of the model was evaluated by aid of the mean corrected r^2, the 95% confidence intervals of the parameters and the quotient of the asymptotic standard error and the parameter.

The latter value can be used as t-statistic to test if a parameter is significantly different from zero.

Only data set 2 (including all data: experimental + literature + different sexes) was used to determine the maximum growth rate of *C. crangon*. Determination of sex specific maximum growth was not possible as the number of studies that separated sex was too low.

In the present study, maximum growth (G_{MAX}) was defined as the fraction of the population growing with growth rates between the median and maximum observed growth rate. G_{MAX} was calculated by applying quantile regression (Koenker & Bassett 1978) and choosing the 75th percentile. The advantage of this method is that, similar to normal regression analysis, all data points are included.

In an alternative analysis, the mean growth rate plus one standard deviation was calculated for all size and temperature classes. In contrast to the quantile regression, only one value (the mean+sd) goes into the calculation for each temperature and size class

Application of the growth model

In this study, egg and larval development were not considered. Therefore, an initial size of 5 mm was chosen to represent the size of a post-larval shrimp (Criales & Anger 1986). When combined with daily field temperature data, equ. (5) can be used to calculate growth in the field. This was done in a stepwise fashion starting with 5 mm and adding the growth increment calculated for one day. The new length was then used to determine the new growth rate for the slightly larger animal. Negative growth rates were set to zero in the calculation. For each month, one animal with 5 mm length was started. Using this approach, length trajectories for shrimp could be generated for the whole year.

Daily temperatures were derived from temperature data measured in Büsum, as a Wadden Sea point, and the Weser estuary, as an near shore but open water point (Temming & Damm 2002b). The two geographical points were chosen as *C. crangon* dwells close to the shore but avoids low winter temperatures by migration to deeper water (Berghahn 1983, Pihl & Rosenberg 1982, Boddeke et al. 1976). From each data series the warmer of the two daily temperatures was chosen to construct one temperature data set of likely temperatures experienced by *C. crango*n. This data set can be described by a sine function:

$$T_i = 9.8933 + 7.3877 \cdot \sin\left(0.01756 \cdot \left(\mathrm{i} - 131.1761\right)\right) \qquad (7)$$

with T_i: temperature at day i and i ϵ {1...365}

Moult intervals and moult increments

From the experiments M5M to M25M individual moult intervals and increments and from the experiment U10M, U20M and U10J to U25J mean moult intervals

and increments were determined parallel to the growth rates. As the unmarked shrimps were pooled in one aquarium, moult intervals were calculated according to

$$mi = \frac{rt \cdot n}{nE} \quad (8)$$

with *rt* = run time of the trial [d], n = mean number of animals (as no animals were replaced during the experiments) during the trial and *nE* = amount of exhuvia counted during the trial. The mean moult increment for each aquarium with unmarked shrimps was then calculated from the amount of moults estimated with eq. (8) and the mean length growth (from the start to the end of the experiment).

Moult intervals from the marked and unmarked animals are once presented separately and subsequently combined with the available data from the literature (Table 5-5). From the combined data (experiments + literature) a function describing the moult interval according to temperature and length was derived.

The relation between moult interval, body length and temperature was described with the following equation:

$$mi = a \cdot L^{b} \cdot e^{cT} \quad (9)$$

where *a*, *b* and *c* are constants, *L* = length and *T* = temperature.

To determine maximum possible increments the moult increment was calculated as the product of the growth equation (5) (with the parameters for maximum growth) and the moult function (9).

Results

Preliminary trials: food effect

The result of the preliminary food trials indicated that the animals feeding on frozen *sprat* showed median growth rates of zero. Those groups of animals fed with *Littorina littorea* or *Cerastoderma edule* grew about 0.1 mm·d^{-1} and those fed *Crangon cr.*, pellets or *Artemia salina* grew about 0.2 mm·d^{-1} (Figure 5-1, left). The food source influenced the determined growth rates significantly (p=0.0057, ANOVA). During the first preliminary experiment, 20 of the 60 animals died.

In Figure 5-1, right each point represents one moult event. When fed with dry feed and cockles a mean growth rate of 0.07 ± 0.09 mm·d^{-1} was observed. During the 10 days of copepod feeding, nearly all animals moulted and, of these animals, 3 did not grow, 3 grew about 0.1 mm·d^{-1}, 5 about 0.2 mm·d^{-1} and 4 with 0.4 to 0.5 mm·d^{-1}. After the 10-d period, the 5 *C. crangon* that did not moult before moulted within 2 days after stopping the copepod feeding and showed growth rates of 0.15 to 0.5 mm·d^{-1}. The mean growth ± sd during copepod feeding was 0.21 ± 0.18 mm·d^{-1} and 0.13 ± 0.15 mm·d^{-1} afterwards (Figure 5-1, right). During the second preliminary experiment, seven of the 18 animals died.

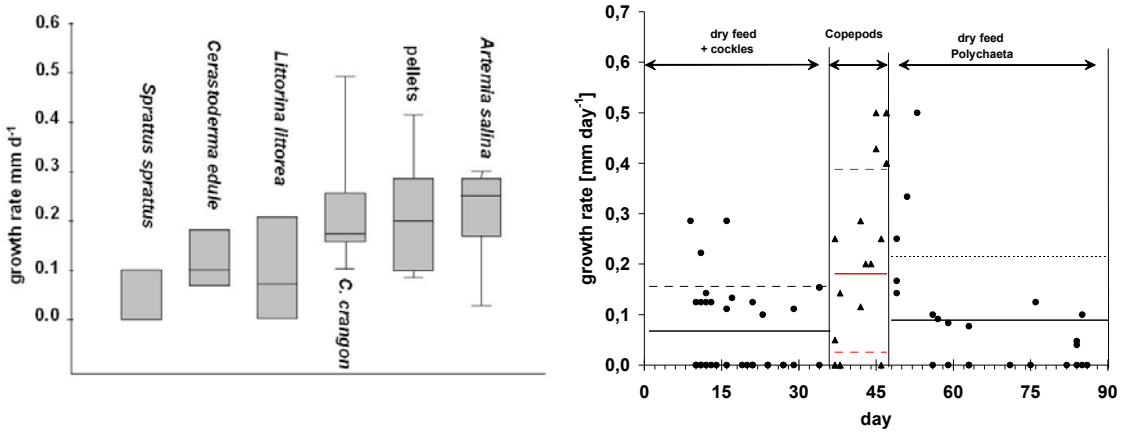

Figure 5-1: *Crangon crangon*. Observed growth rates when fed with different food. Left: Groups of C. crangon (each food source 10 animals of 18 mm total length) held at 15°C. (horizontal line = median, box = 0.25 to 0.75 percentile. Whiskers = maximum and minimum values) Right: One group of 18 animals kept at 18.2°C fed for 35 days with dry feed and frozen cockles, 10 days with live *Arcatia tonsa* and until day 90 with pellets and polychaets. Each point represents a moult event and the growth rate determined as moult increment/moult interval. Horizontal solid lines: mean, dashed line: standard deviation of growth rate.

Mortality growth experiments

Daily mortality rates were normally distributed during the whole experiment (Kolmogorov Smirnov: $p>0.05$). From the marked shrimps caught in May, 315 of 1206 survived > 60 days (mean ± sd mortality = 1.7 ± 1.4% d^{-1}). In the trials started in May with shrimps without a marking, 47 of 169 survived > 40 days (mortality = 4.9 ± 5.4% per day, mean ± sd). In those trials with the unmarked animals caught in July 215 of 241 survived over 25 days (mortality = 0.3 ± 1.0% per day, mean ± sd). A relation between mortality and sex (sex ratio ♀/♂ survivors: 2.97, sex ratio dead: 2.95), position of marking (abdominal seg. 456, carapace 435 dead) or position of the aquarium (pos1 (top): 181, pos2: 162, pos3: 168, pos4: 191, pos5 (bottom): 190) within the stage was not observed. Daily relative (percentage) mortalities between the trials (marked May, unmarked May, unmarked July) were significantly different (t-test, $p<0.001$).

Growth rates from laboratory trials

Mean growth rates were lowest at 5°C and 10°C with values < 0.1 mm d^{-1} for all size classes. At 5°C only 1 animal greater than 50 mm moulted twice. The remaining large shrimps moulted only once or not at all during 60 days, thus no growth rates could be calculated for this length class (Figure 5-2).

For the marked animals the growth rates were determined as moult increment (mm) / intermoult period (d). If more than two moult events occurred (1st moult: start point) the mean growth rate for the shrimp was calculated and used in the growth model. The growth rates for the marked as well as the unmarked animals

were plotted not over starting length but over mean length calculated from start and end length. The reason for that is the declining growth rate with total length. If the start length would be chosen as reference point the growth model would underestimate growth of small shrimps. The trials that started in July and included animals of the length class 20 mm are therefore shown in the figures as 30 mm size class due to the mean of start and end (~40 mm) length.

In general growth (moult increment) decreased with increasing body length. Also, growth rates of the unmarked animals caught in July (U10J to U25J) were always higher, not only in the 20 mm (U10J to U25J) size class but also in the comparable 30 mm size class (U20J and U20M; U15J and U20M). Besides, the animals caught in July also show a more distinct difference between the applied temperatures: the higher the temperature the higher the growth rate. The highest mean growth rates observed are those for the animals starting with 20 mm at 25°C (U25J) with 0.56 mm·d^{-1}. The lowest was 0.02 mm·d^{-1} for the 60 mm length class at 10°C (M10M Figure 5-2).

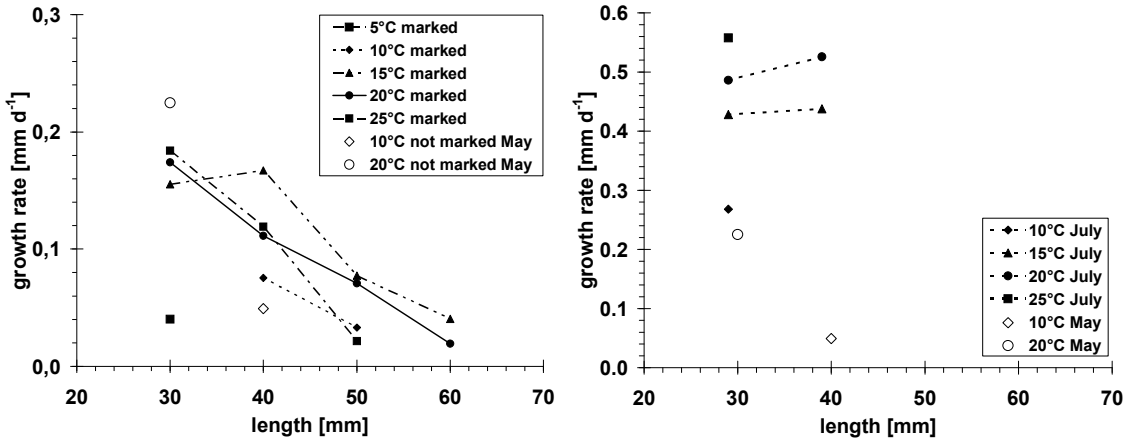

Figure 5-2: *Crangon crangon.* Mean growth rates observed in laboratory trials. Left: results of experiments M5M to M25M performed with marked and U10M, U20M performed with unmarked shrimps caught in May. Right: Mean growth rates observed in trials U10J to U25J, U10M and U20M.

The Kolmogorov-Smirnov-test for normal distribution showed that the growth rates for each experiment and each size class were normally distributed ($p>0.05$). Growth rates of the marked (M20M) and unmarked (U20M) animals caught in May were not significant different ($p = 0.144$, t-test). Significant differences were determined between growth rates of July shrimps and either marked (M20M, U20J: $p < 0.001$, t-test) and unmarked (U20M, U20J: $p < 0.001$, t-test) May shrimps. Marked animals caught in May and reared at 15°C also grew with significant different rates than the unmarked shrimps caught in July (M15M, U15J, $p<0.001$, t-test).

Table 5-2: *Crangon crangon* mean (± sd) growth rates observed in the growth experiments at different temperatures.

	May marked					May unmarked		July unmarked			
	M5M	**M10M**	**M15M**	**M20M**	**M25M**	**U10M**	**U20M**	**U10J**	**U15J**	**U20J**	**U25J**
	5°C	10°C	15°C	20°C	25°C	10°C	20°C	10°C	15°C	20°C	25°C
30 mm	0.04 ± 0.03		0.16 ± 0.08	0.17 ± 0.09	0.18 ± 0.09		0.23 ± 0.18	0.27 ± 0.06	0.43 ± 0.10	0.49 ± 0.15	0.56 ± 0.10
40 mm	0.03	0.08 ± 0.05	0.17 ± 0.08	0.11 ± 0.08	0.12 ± 0.11	0.05 ± 0.05			0.44 ± 0.09	0.53 ± 0.12	
50 mm	0.03	0.03 ± 0.03	0.08 ± 0.05	0.07 ± 0.04	0.02 ± 0.04						
60 mm		0.01 ± 0.01	0.04 ± 0.00	0.02 ± 0.02							

Within the experiments including marked May animals, no significant differences between males and females within the size and temperature classes were obvious ($p>0.11$, t-test). Within the trial with the unmarked animals, from July (U15J to U25J) significant differences between male and female growth rates were determined (t-test, $p = 0.001$). Females showed faster growth than males (Figure 5-3). At 10°C not enough animals reached a size where gender could be accurately determined.

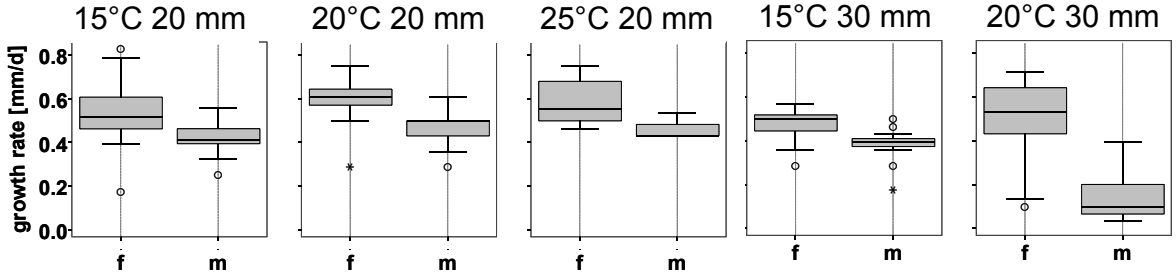

Figure 5-3: *Crangon crangon*. Growth rates [mm d^{-1}] of male and female shrimps derived from laboratory experiments with unmarked animals caught in July. Groups according to start length and temperature. (horizontal line = median, box = 0.25 to 0.75 percentile. Whiskers = maximum and minimum values, crosses: outliers = 1.5 to 3 times box length, circles: extreme values: more then 3 times box length)

Literature review

Of the 33 articles reporting growth of *C. crangon*, (Table 6) 16 studies determined growth by field cohort tracking and 14 used laboratory trials. Two articles only reported moult intervals.

Highest mean growth rates of 0.57 mm d^{-1} were observed by Dalley (1980) at 20°C for a group of animals growing from a mean length of 16.45 mm to a mean length of 22.18 mm within 10 days (Figure 4). Similarly, Uhlig (2002) observed mean growth rates of 0.52 (male) and 0.56 mm d^{-1} (female) for a group of 20 mm shrimp reared between15.9 and 17.1 °C.

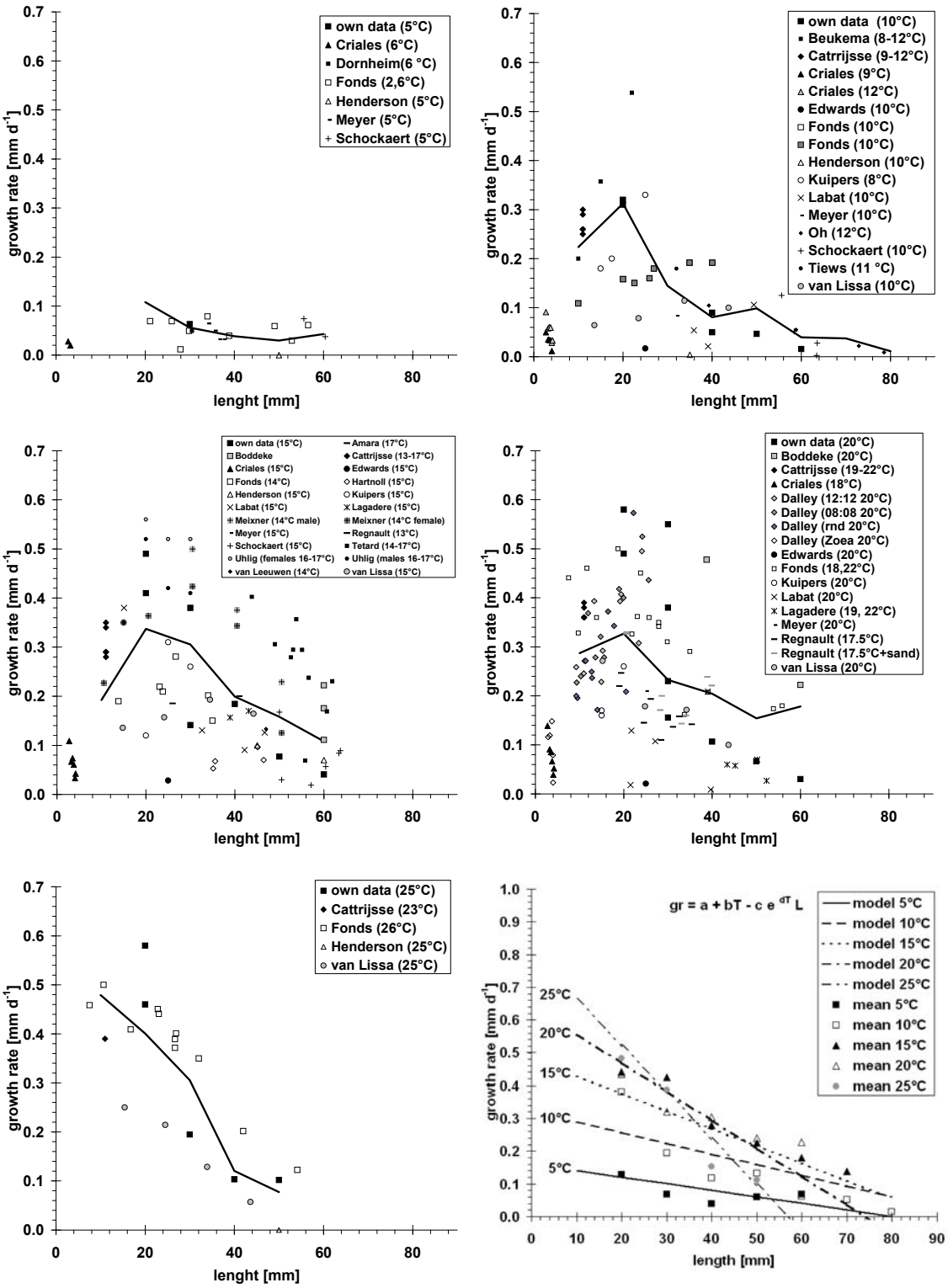

Figure 5-4: *Crangon crangon*. Growth rates obtained from literature review and own data observed in the laboratory plotted according to length and temperature. From top left to bottom left: 5, 10, 15, 20, 25°C. Bottom right: Results obtained by quantile regression. Dots represents the 0.75 percentile of the all data (own data plus literature) lines indicate the model fitted to all data (equation (4), parameters see Table 3).

Most published studies examined growth rates at temperatures between 10 and 20°C where observed growth rates are highly variable and growth rates of nearly zero are regularly reported (Figure 4). Growth rates are low for the smallest and greatest length classes and peak for *C. crangon* between 20 and 30 mm length (Figure 5-4). Growth variability was high both between and within studies and varied between 0 and 0.6 or 0.7 at 15 and 20°C, respectively. Dalley, Kuipers & Dapper or Schockaert observed growth rates that differed by more then 0.2 $mm \cdot d^{-1}$ within one temperature and size class.

Splitting the data according to cohort and laboratory experiments did not indicate any difference with regard to either mean growth rates or growth variance. Likewise variability of growth rates and mean growth rates did not change if groups of less reliable data were omitted from the analysis. Such groups referred to:

Cohort analyses where only a few samples were included.

Cohort analyses where an uncertain function for net selectivity was used.

Cohort analyses where cohorts could not be distinguished properly.

Laboratory trials that used only one food source or where the feeding regime was not described.

Growth models

Fitting equation (5) to the different data subsets, which were derived from the laboratory trials and the literature data, in all cases a confidence interval for the parameter *a* that included zero was determined and the parameter was not significantly different from zero (ASE, see material and methods). That parameter was eliminated, and the function was re-applied to the data. The resulting parameters and r^2 (mean corrected r^2) values are listed in Table 5-3. It can be seen that both growth function (linear and exponential temperature term) describe the data quite well, when applied to the whole dataset (literature + experiments) but higher r^2 values were calculated for equ. (5) ($r^2 = 0.860$, exponential T) than for equ.(4) ($r^2 = 0.695$, linear T). All remaining estimated parameters were significantly ($p = 0.01$) different from zero.

The parameters obtained from fitting (5) (growth function with exp. T) by quantile regression (0.75 percentile), or to mean + standard deviation were almost identical. For both models the parameter values were included in each others confidence intervals.

The amount of literature studies that separated growth rates according to gender was low and therefore, in this case, application of quantile regression was not useful. Additionally, the number of animals per size class and at each temperature was too low to determine a standard deviation for all categories. Sex-specific parameters in Table 5-3 therefore represent mean and not maximum growth rates.

As sex specific growth rates of marked May animals were not significantly different these results were not included in the sex specific growth model.

The results showed that growth rates of female shrimps are generally about 0.1 mm·d^{-1} (parameter b in Table 5-3 and Figure 5-3) higher than those of male shrimps resulting also in a higher L_∞ at the same temperature.

Table 5-3: Parameter estimates, 95% confidence intervals and mean corrected r² values for equation (3) and (4) fitted to the growth rates derived from laboratory experiments and literature research. Line 1: Equ. 3 fitted to the mean values of all data. Line 2: Equ. 4 fitted to the mean values of all data. Line 3. Equ. 4 fitted to the mean values + standard deviation. Line 4: Equ. 4 fitted by quantile regression (75 percentile). Line 5: Equ. 4 fitted to mean male growth rates. Line 6: Equ. 4 fitted to mean female growth rates.

data set	values	Equ.	a	95% max min	b·10³	95% max min	c·10³	95% max min	d	95% max min	r²	k (10°C)	L_∞ (10°C)
2 (lit.+own)	mean	(4)	0.1957	0.1032 0.2883	8.107	11.87 4.355	3.632	5.102 2.162			0.695	1.33	76.20
2 (lit.+own)	mean	(5)	-		23.98	20.80 27.16	0.9659	1.340 0.5920	0.09509	0.081 0.109	0.860	0.91	95.93
2 (lit.+own)	mean + sd	(5)	-		34.92	30.29 39.55	1.301	18.55 7.460	0.09805	0.0836 0.113	0.862	1.27	100.69
2 (lit.+own)	75 perc.	(5)	-		32.18	36.44 27.92	1.227	1.716 0.7380	0.09777	0.083 0.112	0.857	1.19	98.66
1 (lit.+own)	male	(5)	-		31.80	20.10 43.51	1.802	3.507 0.097	0.09529	0.052 0.138	0.641	1.71	68.05
1 (lit.+own)	female	(5)	-		45.59	37.12 54.06	2.287	3.175 1.399	0.09929	0.079 0.119	0.888	2.25	73.86

Application of the model

Most shrimps recruiting with 5 mm in January to April reach 50 mm before September (Figure 5-5, middle) when based on maximum growth (Table 5-3, line 4) and coastal temperatures (7). If however the trajectories are calculated with the model fitted only to the mean values (Figure 5-5, left) the 50 mm limit is reached one month later.

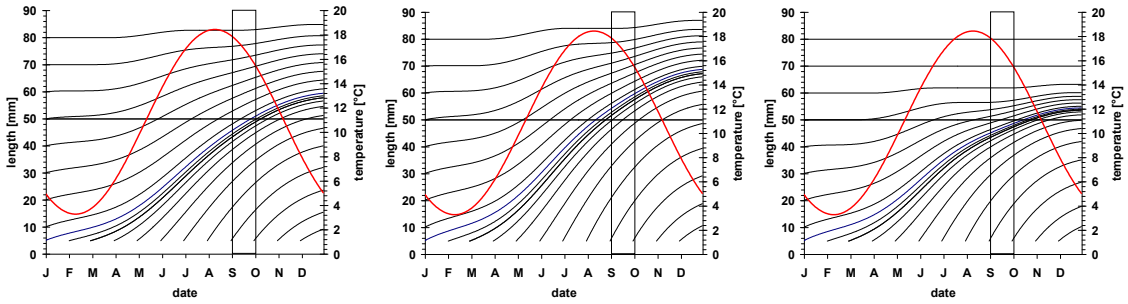

Figure 5-5: *Crangon crangon*. Calculated length trajectories based on parameters derived for the different subsets and including water temperature of the shallow North Sea at Weser light ship and Büsum (sinuid function). Left: Model using mean growth rates. Middle model assuming maximum growth (0.75 quantile regression). Right: Growth trajectories determined for male *C. crangon* Animals start with 5 mm on each 1st of the month.

Using at last the parameter set calculated for mean male shrimp growth rates reveals length trajectories below those calculated for the whole population (mean values). Males hardly reach 50 mm until November (Figure 5-5, right). Female shrimp length trajectories are not shown here but are comparable to the "maximum growth" length trajectories.

Moult intervals and moult increments

In the laboratory experiments, moult intervals decrease with increasing temperature and increase with increasing body length (Figure 5-6, Table 5-4). The longest mean (±sd) moult intervals were observed for the 30 mm size class at 5°C with 43 ± 8 d, the shrimps of the 50 mm size class only moulted once or not at all at 5°C and therefore no moult intervals could be determined for this size class. Differences in moult intervals between marked males and marked females could not be detected. A statistical analysis comparing the moult intervals of the three experiments (marked May, unmarked May and unmarked July) was not possible as the determination of the moult interval of the unmarked shrimps according to (8) only provides one value for each aquarium. For the marked animals on the opposite a moult interval was determined for each shrimp. The intervals are shown in Figure 5-6 and are in the 30 mm class comparable to the moult intervals observed for the marked shrimps although growth rates are different.

Fitting (9) to the moult intervals obtained from the experiment data and literature values (Table 5-5) leads to

$$mi = 5.7066 \cdot L^{0.7364} \cdot e^{-0.09363 \cdot T}$$

with a mean corrected r² of 0.945 and all parameters (Table 5-3) significantly different from 0 (p=0.01). The 95% confidence intervals for parameter *a* were 2.656 to 8.758 for *b* 0.604 to 0.869 and for *c* -0.106 to -0.081.

The mean moult increments observed in the experiments with the marked *C. crangon* (M5M to M25M) were between 0.57 ± 0.73 mm and 2.7 ± 0.9 mm (Table 5-4). Individual increments ranged from zero growth in all size classes and at all temperatures to maximum values of 5 mm observed at 10 and 15°C in the 40 mm size class. The mean moult increments of the unmarked animals caught in July varied between 3.6 and 7.5 mm (Table 5-4). The highest values were again observed in 15°C experiment (Figure 5-7). It was impossible to determine if 0 (zero) increments were present since individuals were not tracked.

The theoretical maximum moult increment calculated as the product of the growth function (5) the parameters obtained for maximum growth by quantile regression and the moult interval function (9), reveals that moult increments must be greatest for 30 to 40 mm long *C. crangon*. Increments peak with 6 mm at 10°C for a 40 mm shrimp (Figure 5-7) whereas the smallest moult increments are predicted for a temperature of 25°C. Yet (mean) moult increments close to 8-9 mm were observed for 20 mm size July shrimps in the present study also including 25°C (Figure 5-7 left).

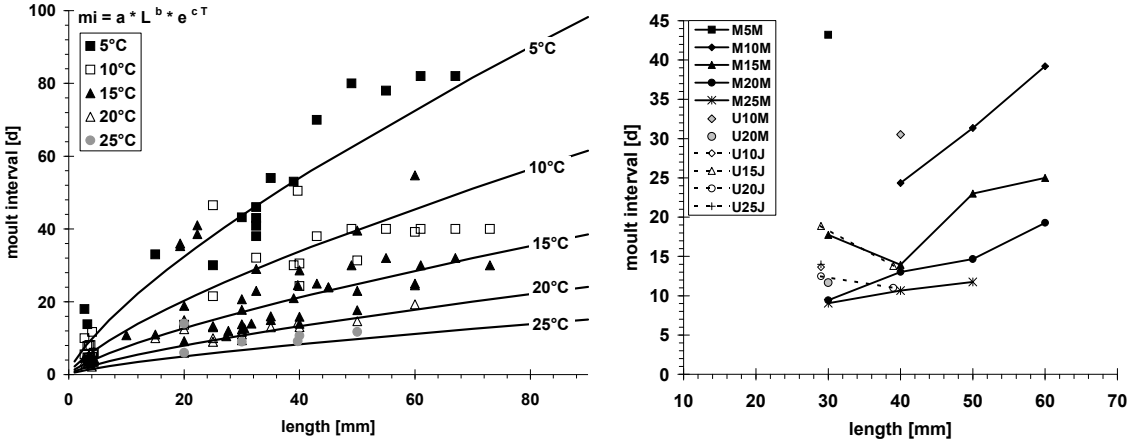

Figure 5-6: *Crangon crangon* moult intervals. Left: Mean moult intervals observed in the laboratory trials combined with moult intervals described by other authors (dots) and equ. (8) (lines) fitted to these data. Right: Moult intervals observed in the growth experiments.

Table 5-4: *Crangon crangon* mean moult increments and standard deviation observed in the growth experiments at different temperatures.

	May marked					**May unmarked**		**July unmarked**			
	M5M	**M10M**	**M15M**	**M20M**	**M25M**	**U10M**	**U20M**	**U10J**	**U15J**	**U20J**	**U25J**
	5°C	10°C	15°C	20°C	25°C	10°C	20°C	10°C	15°C	20°C	25°C
30 mm	2.7 ± 0.9		2.31 ± 1.36	1.35 ± 0.92	1.81 ± 1.18		2.25 ± 1.83	3.58 ± 0.91	8.21 ± 1.85	6.95 ± 1.59	7.54 ± 1.42
40 mm		2.06 ± 1.37	2.45 ± 1.54	1.2 ± 1.17	1.00 ± 1.14	1.48 ± 1.06			6.11 ± 1.19	5.64 ± 1.29	
50 mm		1.47 ± 1.04	1.73 ± 1.26	1.00 ± 0.94	1.19 ± 1.29						
60 mm		0.75 ± 0.43	1.00	0.57 ± 0.73							

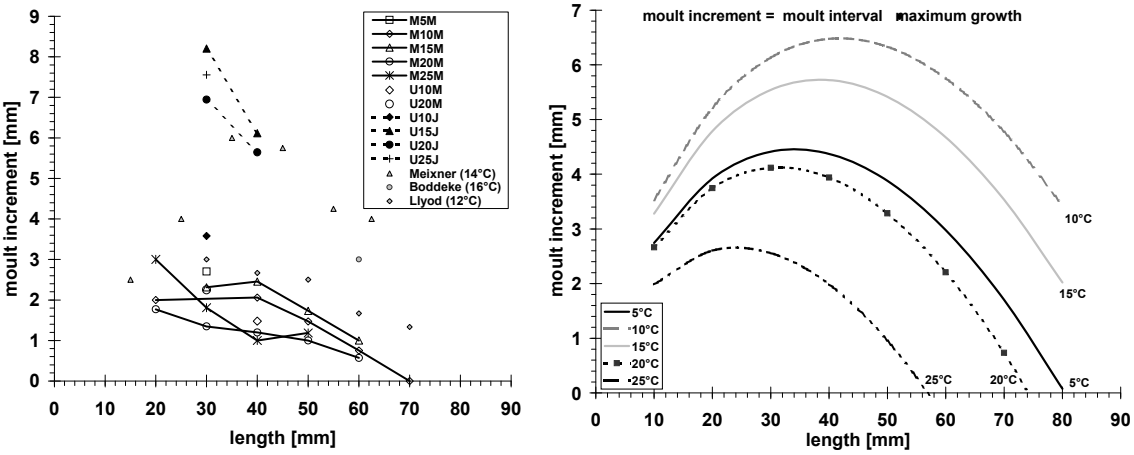

Figure 5-7: *Crangon crangon* moult increments. Left: Mean moult increments observed in the laboratory trials. Right: Theoretical moult increment calculated as the product of the moult interval equ, (9) and the maximum growth (0.75 quantile regression, equation (4), parameters Table 3).

Discussion

Laboratory experiments of the actual study

Within the experiments M5M to M25M, U10M and U20M, only low growth rates were observed with many shrimps showing moult increments of 0 mm, in contrast to the experiments that used shrimps caught in July. Moreover, mortality was significantly higher in the trials that used May shrimps compared to July shrimps. Growth rates were not significantly different according to the experimental setup (marked or unmarked) but according to the catch month (May, July). It can therefore be excluded that handling stress or the marking procedure reduced growth in the trials with the marked animals as it was also low in the control groups with unmarked animals. A marking method similar to the one used in these experiments was applied by Henderson & Holmes (1987) who used Loctite adhesive comparable to the glue used in this trial. They also observed no differences in moult rates or mortality between marked and unmarked shrimps.

Further the experimental setup was the same in all trials, and May and July shrimps were partially reared in the same circulation systems with the same food. It can therefore be excluded as source for growth differences. Further the experimental design was able to produce growth rates comparable to the highest values observed by other authors. At 10°C only Beukema (1992) observed higher growth rates and at 20 and 25°C the growth rates observed in this experiment are comparable to those determined by Labat(1977), Meixner (1969), Uhlig(2002), Dalley (1980) and Fonds (pers. comm.).

Differences in collection procedure can be ruled out since comparable size classes in July and May were both caught with a push net at the same location (Büsum) and transported in the same way to Helgoland. Haul duration might increase mortality (Gamito & Cabral 2003) but haul duration was equally short in May and July.

According to the previous section only the catch date is left to explain growth differences of May and July shrimps. The catch date determines two factors of the experimental setup: the age and the starting conditions of the shrimps. May caught shrimps are approximately 6 month older at the same size than those caught in July. July shrimps most likely originate from the winter egg production and were starting their juvenile life around April. It has however so far not been demonstrated that the mere age difference can explain the poor growth performance of the older individuals. A reason might be irreversible non-genetic adaptation as described by Kinne (1962). An animal once adapted to conditions (in this case low temperature and low food) is not able to change its adaptation once it is established.

As a second factor related to the catch date, the fitness or energy content of the animal can be considered. Due to decreased food availability during winter, the stored reserves might have been depleted in the May cohort. This can actually be seen in the mean dry weight condition (1000*dry weight $\cdot L^{-3}$) of analyzed samples which was lower for the May cohort (1.63 ± 0.25 mg$\cdot$mm^{-3}, mean ± sd) than for the

July cohort (1.87 ± 0.31 mg·mm^{-3}, mean ± sd). The shrimps of the May cohort might have invested less ingested food energy into growth and more into refilling depleted energy stores. Another possibility could be that the overwintering shrimps channel most of their energy into reproduction. A peak in egg-bearing females from latitudes around 54°N in early summer is often reported in literature (Campos & Van der Veer 2008, Siegel et al. 2008). Less energy is needed in the production of spermatophores compared to oocytes, and therefore, mainly females would be expected to be influenced. However, growth rates of both males and females caught in May were much lower when compared to July-caught shrimp. It is therefore most likely that maturation alone is not the reason for the reduced growth rates.

The different growth rates observed in the experiments might also reflect differences in the quality of experimental food. Polychaets and Ulva were provided as food during all the trials and did not differ according to the amount and quality. However the plankton abundance and composition at Helgoland Reede station, which was used as one main food compound, varied considerably between weeks (Figure 5-8). Although the trials with the May and July shrimps overlapped changes in zooplankton composition may have played a role since copepod concentrations were lower in the beginning of the trials in May.

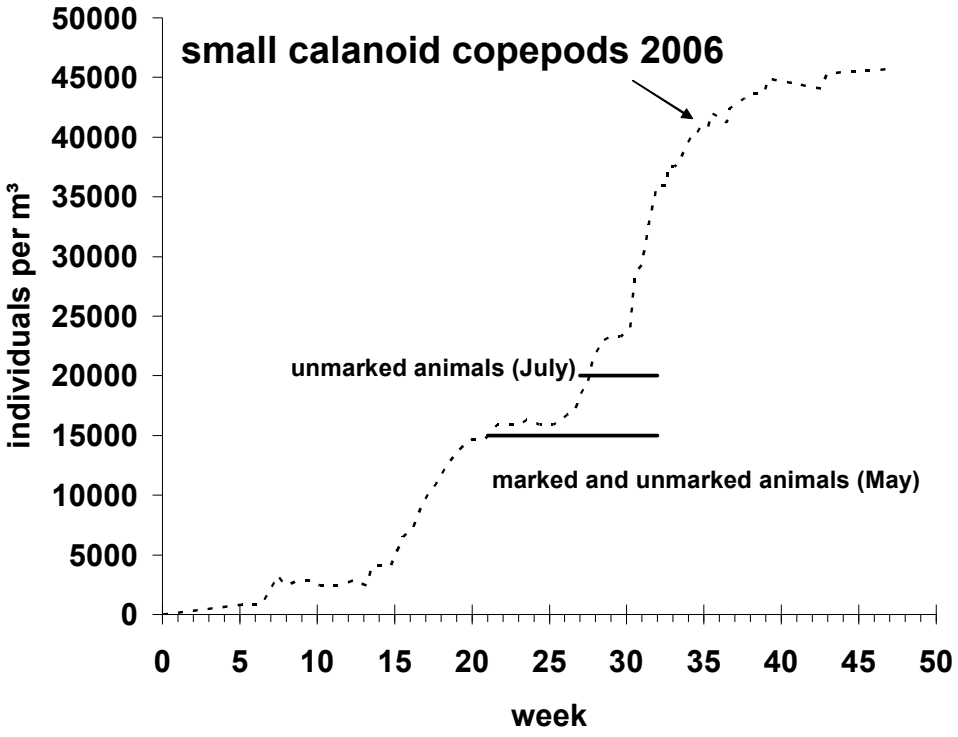

Figure 5-8: Abundance of small calanoid copepods determined at the Helgoland Reede station (MURSEYS, www.bsh.de).

This would underline the importance of life plankton for *C. crangon*. High growth rates were observed in the laboratory when plankton or *Artemia* nauplii were provided as food (Uhlig 2002, Dalley 1980, Meixner 1969) whereas growth rates

were lower when only *C. crangon* (Edwards 1978) or nematodes (Gerlach & Schrage 1969) where used. Uhlig (2002) observed that frozen smelt supported only low growth rates of *C. crangon* (20 mm; 0.25 $mm \cdot d^{-1}$) whereas growth rates of shrimps fed with fresh-caught live plankton doubled (20 mm; 0.56 $mm \cdot d^{-1}$) what is comparable to the results of the preliminary experiments shown in the actual article. Copepods are the main food source and, at least in summer, make up a major part of the stomach content of juvenile *C. crangon* (Boddeke et al. 1986, Plagmann 1939).

Conclusions

Growth rates determined in the actual work strongly depended on the time of catch and were lower for animals caught in May than for animals caught in July

The results suggest that the food source influences growth rates of *C. crangon* and that most probably especially life plankton supports high growth rates

Growth rates determined in this work and by other authors are highly variable

Sources for variability in growth rates

The laboratory trials revealed that growth rates can vary between 0 $mm \cdot d^{-1}$ and 0.5 $mm \cdot d^{-1}$ even in the same setup. However despite of this high scatter the following general patterns can be derived (Figure 5-4):

1. Smaller animals grow faster than larger ones

2. Growth rates increase with temperature for small animals

3. Large shrimps grow fastest at moderate temperatures (10-15°C)

These observations can also be interpreted in the context of von Bertalanffy growth theory (von Bertalanffy 1934). In the actual study the food related term (anabolism) was, following Kuipers & Dapper (1981), expanded with a linear temperature term. The catabolic term was also expanded with an exponential temperature effect. With rising temperatures - especially for large shrimps - the catabolic term increasingly approaches the anabolic term resulting in decelerated growth. The same mechanism could also be realized with exponential temperature terms for both anabolism and catabolism, but with a smaller exponential coefficient on the anabolic term.

As described before the intersection with the x-axis of (5) is an estimate of L_∞ (mean maximum length) that can be reached by the animal at different temperatures. *C. crangon* can therefore reach a maximum size of 57 mm at 25°C whereas at 5°C the maximum size is 80 mm (Figure 5-4). This corresponds to observations in the field where generally animals living in higher latitudes or deeper layers (where water is colder) can reach larger sizes than related species in tropical regions (Angilletta et al. 2004, Gunter 1950). This is also indicated by the L_∞ of *C. crangon* which is (at 10°C, mean North Sea temperature) in this study

comparable to findings of Kuipers and Dapper (1984) who estimated L_∞ = 78 mm and k = 0.95. For *C. crangon* dwelling in warmer water different values for maximum total length are discussed. The data of Labat (1977) suggest a L_∞ of about 50 mm and the largest female Viegas (2007) observed in Portugal was 61 mm long. On the opposite Marques (1982) reports 80 mm and Gelin et al. (2000) found shrimps of 72 mm in the region sampled by Labat (1977).

The temperature dependence which is illustrated by parameter *d* is similar for all scenarios and both genders where growth model (5) was used and varies between 0.0951 and 0.0993. This parameter corresponds to the $Q_{10} = \left(\frac{e^{d \cdot T_1}}{e^{d \cdot T_2}} \right)^{\frac{10}{T_2 - T_1}}$.

C. crangon metabolism thereafter ranges between Q_{10} = 2.59 to 2.70 which is comparable to other ectotherms (Angilletta et al. 2004) and a Q_{10} of 2.85 and 3.01 determined for *Crangon septemspinosa* (Taylor & Peck 2004).

Parameter *b* which mainly determines the intercept of the function with the y-axis is for female shrimps 0.04559 and for male shrimps 0.03180.

Although the data are well described by equation (5) variability within the size and temperature classes is high. This variability might originate from four sources:

1. Methodological effects related to cohort tracking
2. Methodological effects related to laboratory experiments
3. Gender specific growth rates in studies without sex differentiation
4. Natural or genetic variations

1. Cohort tracking

The greatest problem when applying cohort tracking is the limited sample resolution with regard to time and space. *C. crangon* spawns throughout most of the year (Neudecker & Damm 1992, Kuipers & Dapper 1984, Dornheim 1969) and, therefore, slow-growing individuals from early cohorts mix with fast-growing individuals from later cohorts creating cohorts that have mixtures of growth histories.

Concerning the spatial distribution it has to be taken into account that small individuals can be found mainly in shallow water whereas larger animals prefer deeper water and tidal gullies. Gradual disappearance of shrimps larger than 20 mm (Beukma 1992) restricts therefore the analysis of growth from data sampled in shallow water only. Which could have been a problem for example in the study of Oh et al. (1999). Since migration patterns are not only determined by length but also related to age, gender, season and temperature (Spaargaren 2000, Pihl & Rosenberg 1982, Boddeke et al. 1976, Boddeke 1976) obtaining representative length compositions is a difficult task.

In practical applications growth rates of juveniles are most reliable since growth is fast and cohorts are less effected by site dependent emigration into deeper water

(Amara & Paul 2003, del Norte-Campos & Temming 1998). Cohort tracking of larger shrimp will only generate reliable growth estimates if in a local region a single recruitment event dominates and migration is unlikely to occur. Such a situation was analyzed in the French Channel by Tetard (1985).

2. Laboratory artifacts

Laboratory studies avoid many of the problems discussed for cohort tracking but generate others issues relating to prey composition. In nature, *C. crangon* is an opportunistic omnivore (Feller 2006, Boddeke et al. 1986, Pihl & Rosenberg 1984). Our preliminary trials indicated that relatively low growth rates were obtained for shrimp fed either fish, snails, cockles, *Artemia salina* or one of several dry feeds whereas higher growth rates were observed for shrimps fed live copepods (*A. tonsa*).

The increased concentration of animals in the laboratory trials might influence the growth rates by stress, inducing higher energy consumption for activity and hence less energy available for growth. Onnen & Zebe (1982) described that tail flipping, a common escape mechanism of *Crangon cr.,* is very cost intensive. Feeding and walking increases oxygen consumption by 45 % and active swimming by 130% in *Penaeus esculentus* (Laxter & Outhward 1991). Regnault (1970, 1976) observed that *Crangon septemspinosa* growth faster when isolated than in groups or when held on natural substrate. Within his experiments total length of isolated shrimps increased from 8 to 32 mm and of grouped shrimp from 8 to 18 mm. Contrary to Regnaults findings we observed high growth rates with a grouped setup. The reason for the differences might be interactions among individuals. Tiews (1970) reported that animals with missing extremities grew slower over successive moults due to added costs of limb regeneration. If animals do not get optimum food, aggressive interactions (leading to cannibalism) may increase causing more injuries and slower growth rates.

Optimum conditions would therefore be obtained by rearing small groups within tanks having natural substrate using a mixture of prey that includes natural live plankton plus at least one additional food source). The conditions of our experiments, particularly those for the July caught shrimps, were close to this ideal setting.

3. Gender

Growth of female *C. crangon* in our analysis was about 30% higher than that of males. The differences in growth observed between the genders could explain a part of the growth variability reported in studies that did not sex the shrimp. However, even in our study where gender was registered and rearing conditions where close to the perceived optimum for this species, growth variation still existed (e.g. min: 0.4 to max: 0.75 $mm \cdot d^{-1}$ for 20 mm females U20J).

4. Natural or genetic variability

Working with Penaeus monodon, Benzie (1997) reported that the growth rates of 10% of progeny were influenced by genetic factors. A similar study has not been conducted for *C. crangon*. Several authors observed that genetic variability in the field is low and only differs between very remote places (Beaumont & Croucher 2006, Bulnheim & Schwenzer 1993). The most recent study performed by Luttikhuizen et al (2008) who sequenced a 388 bp fragment of the cytochrome-c-oxidase I gene, concluded that genetic subpopulations of *C. crangon* exist on a large spatial scale that are separated by geographic boundaries. Yet at the local scale, variability among individuals can surpass these differences. The Wadden Sea is an open highly turbulent mixed system. It can therefore be assumed that at least the growth studies conducted using animals captured from German and Dutch waters are employing animals from the same stock. Growth rates observed in Northern and Southern Europe are therefore not very different. The growth rates reported by Tetard (1985) and Dalley (1980) from France and the Isle of Man are similar to those found for C. crangon collected in the Wadden Sea.

Conclusions

Laboratory experiments might underestimate growth rates due to feeding effects and/or higher intraspecific interactions leading to injuries and reduced growth due to energy costs associated with regeneration

Cohort tracking might underestimate growth due to insufficient spatio-temporal resolution and due to the mixing of cohorts having different growth histories.

Moult intervals and moult increments

Size- and temperature-specific variability in moult intervals appears lower than the variability observed in growth rates. This indicates that moult intervals are less dependent on the amount and quality of food but mainly dependent on temperature and size. However in starvation trials only those individuals moult which are already in the premoult phase at the start of the experiment (Regnault & Lagardère 1983). Without food the moult is obviously suppressed. Similar results were obtained by Uhlig (2002) who observed a lower moult frequency in starving rather than fed animals. These observations might be explained by the high amount of energy needed for the moulting process. Part of this energy is related to the production of the exhuvia with an energy content of 10.5 $kJ \cdot g^{-1}$ dry weight (Evans 1984) corresponding to up to 17% of the whole dry weight of an animal (Regnault & Luquet 1978). Gerlach & Schrage (1969) observed *C. crangon* eating their exhuvia and, therefore, a portion of the energy might get recycled. The main cost, however is related with the process of moulting. During that process respiratory rates increase up to 2.5 times (Hagerman 1970) and a sharp increase in ammonia production has been observed (Regnault 1979). Tail flipping is most

often employed to shed the old carapace and this behavior has shown to be very energy consuming (Onnen & Zebe 1982).

Furthermore vulnerability towards mortality due to cannibalism (Evans 1984) and other predation events is much higher after moulting. It is therefore surprising that moulting occurs regularly even if the moult increment is zero. It is possible that maturation and reproduction trigger moulting independent of growth rate and recent feeding since female *C. crangon* can only be fertilized after moult when the exosceleton is not hardened (Boddeke et al. 1991). Moulting is required even under bad conditions as a precondition for reproduction. Buchholz et al. (2006) suggested that moulting outside the breeding season might be necessary to minimize the level of parasites in *Meganycthiphanes norvegica*. Although parasites of *C. crangon* have so far only been described for animals from the Mediterranean (Azevedo 2001) other shell diseases like black spots (Porter et al. 2001, Vogan et al. 2001, Dyrynda 1998) or fouling might have lead to the evolution of a regular moulting that is only suppressed during severe starvation events (Hartnoll 2001).

Since the moult interval under feeding conditions is relatively stable the main variance observed in the experiments growth rates must therefore be due to high variability in the moult increments. In fact the moult increments of the animals caught in May were about only half of those caught in July.

Maximum moult increments can be deduced by multiplying the maximum growth rate, (derived from the 0.75 percentile) with the moult interval. This reveals increments of up to 7 mm at a length of 40 mm. The animals caught in May exhibited maximum increments of 5 to 6 mm but mean increments of 2 to 3 mm if zero growth animals were included. The results for the 40 mm sized animals are comparable to those observed by Meixner (1969), whereas the 30 mm animals caught in July showed larger increments either than those reported by Meixner (1969) and the calculated increments. It must be stated that increments of the unmarked animals might be overestimated by unrecognized exhuvia that might have been flushed out of the aquarium or were eaten by the shrimps. However as the moult intervals were comparable to those of the marked animals it is most likely that the higher growth performance is achieved in *C. crangon* by larger increments.

Conclusions

Moult intervals can be described as $mi = 5.7066 \cdot L^{0.7364} \cdot e^{-0.09363 \cdot T}$

From the growth model and the moult interval model the increments for different size classes and temperatures can be calculated

Inter-individual growth rate variability is mainly due to varying moult increments and, to a lesser extent, to moult intervals.

Application of the growth model

The growth trajectories based on mean growth rates and ambient temperatures reveal that 15 mm L shrimp in May and June do not reach 50 mm L prior to October. While these shrimp represent the main recruitment peak that is observed each year in the shallow intertidal areas of the Wadden Sea, the predicted growth rates are insufficient to explain the peak of commercial catches in September. The match with the catch peak is much closer, however if maximum growth (G_{MAX}) from 75 percentile analysis is used for the predictions. This implicates that growth in nature is generally faster than observed in the most studies. This is supported by the results of the only field study where well distinct cohorts could be analyzed (Tetard 1985) and where growth rates comparable to the 75 percentile model were determined. Additionally the mean growth rate of the population and the animals that build up the adult population maximum in October is most probably shifted upwards by selective higher cumulative mortality (and removal from the population) of slower-growing animals. A 5 mm L *C. crangon* in January requires 650 days to reach 80 mm with maximum growth and 892 days with mean growth rates. Assuming a mortality of Z = 4.2 (derived from Temming et al. (1993)), 0.06% of the fast growing and 0.003 % of the mean growing cohort would be left at that size which is 1/20 of the fast-growing fraction.

Another reason for the better fit of the maximum growth model with the observed conditions in the field could be that juvenile C. crangon may experience much higher water temperatures during low tide when these animals reside on the tidal flats and in small pools (Berghahn 1991, Berghahn 1983). Although temperatures from Büsum were measured at the low tide, temperatures on the flats might be higher and, if sufficient food exists, this could increase growth rates of the small juveniles. As only animals < 20 mm dwell in these pools and start migrating into deeper water at larger body sizes (Beukema 1992), a 5°C difference of experienced temperature would only lead to a time shift of 7 to 17 days. For larger shrimps this warmer temperature exposure was assumed and incorporated into the growth model. Temperature might therefore only explain a part of the difference.

The results of the preliminary feeding experiment and the results of the July experiments indicated that maximum growth rates can be observed under ad libitum feeding. This suggests that, at least for the animals that reach marketable size prior to October, there is no food limitation along the German and Dutch coasts.

The survivors of the autumn peak that all started with a length of 5 mm in the actual year are all within the length class 60 to 70 mm in winter. Within this size class nearly all of the female shrimps carry eggs (Siegel et al. 2008, Oh et al. 1999) which subsequently spawn the larvae for the recruitment peak in spring (Temming & Damm 2002b). This indicates that the whole life cycle and the whole population is mainly driven by a yearly cycle: hatch from winter eggs, invasion into the shallow Wadden Sea areas in late spring, maximum growth during summer, marketable size in fall and egg carriage during winter.

This life cycle description also underscores the importance of winter eggs for the North Sea /Wadden Sea population. The whole life cycle is based on the juvenile shrimps that finished their last larval instar between January and June. These juveniles all hatched from winter eggs (Temming & Damm 2002). Reduction of the winter eggs carrying females by increased winter fishery might therefore have implications on the stock size and also on the landings in fall.

Conclusion

High mortality leads to a net increase in growth rates as slow growing animals are reduced by higher cumulative mortality.

Maximum growth can be described as $dL/dt = 0.03218 \cdot T - 0.001227 \cdot e^{0.09777 \cdot T} \cdot L$

The growth model was used to determine length trajectories that explain the life cycle of *C. crangon*

The peak of juveniles in May is made up by shrimps that hatched during winter

The peak in commercial catches in August and September is composed of juveniles observed in the spring

Adults obtain>60 mm L (become mature) prior to winter

Increase in winter fishery might have implications on the stock size and fall landings.

Acknowledgements

Thanks for the support to J.P. Hermann, Prof. Dr. F. Buchholz, Dr. R. Saborowski, C. Rückert and R. Perger. The study was financially supported by the Federal Ministry of Food, Agriculture and Consumer Protection, Germany Project No. 03HS030.

References

Amara R, Paul C (2003) Seasonal pattern in the fish and epibenthic crustaceans community of an intertidal zone with particular reference o the population dynamics of plaice and brown shrimp. Estuarine, Coastal and Shelf Science 56:807-818

Angilletta MJJ, Steury TD, Sears MW (2004) Temperature, Growth Rate, and Body Size in Ectotherms: Fitting Pieces of a Life-History Puzzle. Integrative and Comparative Biology 44:498-509

APEC. APEC Air Shipment of Live and Fresh Fish & Seafood Guidelines. APEC #98-FS-03.1, -157. 1999. First Coastal Corporation.

ASEAN. ASEAN Cooperation in Food, Agriculture and Forestry: Asean good shrimp farm management practice. Fisheries Publication Series No.1, -35. 1998. Walailak University.

Azevedo C (2001) Ultrastructual aspects of a new species, *Vavraia mediterranica* (Microsporidia, Pleistophoridae), Parasite of the French Mediterranean shrimp, *Crangon crangon* (Crustacea, Decapoda). Journal of Invertebrate Pathology 78:200

Beaumont AR, Croucher T (2006) Limited stock structure in UK populations of the brown shrimp, *Crangon crangon*, identified by morphology and genetics. Journal Marine Biology Association UK 86:1107-1112

Benzie, J.A.H. (1997) A review of the effect of genetics and environment on the maturation and larval quality of the giant tiger prawn *Penaeus monodon*. Aquaculture 155:69-85

Berghahn R (1983) Untersuchungen an Plattfischen und Nordseegarnelen (*Crangon crangon*) im Eulitoral des Wattenmeeres nach dem Übergang zum Bodenleben. Helgoländer Meeresuntersuchungen 36:163-181

Berghahn R (1991) Winterfischerei Ja oder Nein? Einige biologische Grundlagen für die Beurteilung dieser Frage sowie erste Untersuchungsergebnisse. Das Fischerblatt 6:173-180

Beukema JJ (1992) Dynamics of juvenile shrimp *Crangon crangon* in a tidal flat-nursery of the Wadden Sea after mild and cold winters. Marine Ecology Progress Series 83:157-165

Boddeke R (1976) The seasonal migration of the brown shrimp *Crangon crangon*. Netherlands Journal of Sea Research 10:103-130

Boddeke R (1982) The occurrence of winter and summer eggs in the brown shrimp (*Crangon crangon*) and the pattern of recruitment. Netherlands Journal of Sea Research 16:151-162

Boddeke R, Bosschieter JR, Goudswaard P (1991) Sex change, mating and sperm transfer in *Crangon crangon*. In: Bauer RT, Martin JW (eds) Crustacean sexual biology. Columbia University press, New York, p 164-182

Boddeke, R., Dijkema, R., and Stemelink, M. E. (1976) The patterned migration of shrimp populations: A complete study of *Crangon crangon* and *Penaeus brasiliensis*. 200, 31-49. 1976. FAO Fisheries report. Symposium on Proceedings in marine Biology in the Carribean Corsas.

Boddeke R (1966) Sexual cycle and growth of Brown Shrimp (*Crangon crangon*). International Council for the exploration of the Sea, Shellfish Committee C.M. 1966 / M:6:

Boddeke R, Driessen G, Doesburg W, Ramaekers G (1986) Food availability and predator presence in a costal nursery area of the brown Shrimp (*Crangon crangon*). Ophelia 26:77-90

Buchholz CM, Buchholz F, Tarling GA (2006) On the timing of moulting processes in reproductively active Northern krill *Meganyctiphanes norvegica*. Marine Biology 149:1443-1452

Bulnheim H-P, Schwenzer DE (1993) Zur Populationsgenetik von *Crangon crangon* und *C. allmanni* (Crustacea, Decapoda) im Bereich der europäischen Küsten. Zoologische Jahrbücher. Abteilung für allgemeine Zoologie und Physiologie der Tiere 97:327-347

Cattrijsse A, Dankwa HR, Mees J (1997) Nursery function of an estuarine tidal marsh for the brown shrimp *Crangon crangon*. Journal of Sea Research 38:109-121

Calado R, Dinis MT (2008) Collection of marine invertebrates for the aquarium trade in European waters: is anyone surveying? Aquatic conservation: Marine and Freshwater Ecosystems 18:335-338

Campos J, van der Veer HW (2008) Autecology of *Crangon crangon* (L.) with an emphasis on latitudinal trends. Oceanography and Marine Biology, Annual Reviews 46:65-104

Caudri LWD (1937) Einfluss der Temperatur und des Salzgehalts auf die Sterblichkeit von Garnelen (*Crangon crangon* L.). Archives Neerlandaises de Zoologie 3:179-196

Cowan JH, Houde ED, Rose KA (1996) Size-dependent vulnerability of marine fish larvae to predation: An individual-based numerical experiment. ICES Journal of Marine Science 53:23-37

Criales MM, Anger K (1986) Experimental studies on the larval development of the shrimps *Crangon crangon* and *C. allmanni*. Helgoländer Meeresuntersuchungen 40:241-265

Cuesta JA, González-Ortegón E, Rodríguez A, Baldó F, Vilas C, Drake P (2006) The decapod crustacean community of the Guadalquivir Estuary (SW Spain): seasonal and inter-year changes in community structure. Hydrobiologia 557:85-95

Dalley R (1980) The survival and developement of the shrimp *Crangon crangon* (L.), reared in the laboratory under non-circadian light-dark cycles. Journal of Experimental Marine Biology and Ecology 47:101-112

del Norte-Campos AGC, Temming A (1998) Population dynamics of the brown shrimp *Crangon crangon* L., in shallow areas of the German Wadden Sea. Fisheries Management and Ecology 5:303-322

Dornheim H (1969) Beiträge zur Biologie der Garnele *Crangon crangon* (L.) in der Kieler Bucht. Berichte der Deutschen Wissenschaftlichen Komission für Meeresforschung 20:179-215

Dyrynda EA (1998) Shell disease in the common shrimp *Crangon crangon*: variations within an enclosed estuarine system. Marine Biology 132:445-452

Edwards RRC (1978) Effects of water-soluble oil fractions on metabolism, growth and carbon budget in the shrimp *Crangon crangon*. Marine Biology 46:259-265

Evans S (1984) Energy budgets and predation impact of dominant epibenthic carnivores on a shallow soft bottom community at the Swedish west coast. Estuarine, Coastal and Shelf Science 18:651-672

Feller RJ (2006) Weak meiofaunal trophic linkages in *Crangon crangon* and *Cracinus maenas*. Journal of Experimental Marine Biology and Ecology 330:274-283

Gamito R, Cabral H (2003) Mortality of brown-shrimp discards from the beam trawl fishery in the Tagus estuary, Portugal. Fisheries Research 63:423-427

Gelin A, Crivelli AJ, Rosecchi E, Kerambrun P (2001) Can salinity changes affect reproductive success in the brown shrimp *Crangon crangon*. Journal of Crustacean Biology 21:905-911

Gerlach SA, Schrage M (1969) Freilebende Nematoden als Nahrung der Sandgarnele *Crangon crangon*. Oecologia 2:362-375

Gillooly JF, Brown JH, West GB, Savage VM, Charnov EL (2001) Effects of size and temperature on metabolic rate. Science 293:2248-2251

Gunter G (1950) Correlation between temperature of water and size of marine fishes on the Atlantic and Gulf coast of the United States. Copeia 4:298-304

Gurney AR (1982) The larval developement of *Crangon crangon* (Fabr. 1795) (Crustacea: Decapoda). Bulletin of the British Museum, Natural History 42:247-262

Hagerman L (1970) The oxygen consumption of *Crangon vulgaris* (Fabricius) (Crustacea, Natantia) in relation to salinity. Ophelia 7:283-292

Hartnoll R (2001) Growth in Crustacea – twenty years on. Hydrobiologia 449:111-122

Havinga B (1930) Der Granat (*C. vulgaris*, Fabr.) in den holländischen Gewässern. Journal du Conseil International pour l'Exploration de la Mer 5:57-87

Henderson PA, Holmes RHA (1987) On the population biology of the common shrimp *Crangon crangon* (L.) (Crustacea: Caridea) in the Severn Estuary and Bristol Channel. Journal of the Marine Biological Association U.K. 67:825-847

Henderson PA, Seaby RMH, Somes JR (2006) A 25-year study of climatic and density-dependent population regulation of common shrimp *Crangon crangon* (Crustacea: Caridea) in the Bristol Channel. Journal of the Marine Biological Association U.K. 86:287-298

Houde ED (1987) Fish Early Life Dynamics and Recruitment Variability. Am.Fish.Soc.Symp. 2:17-29

ICES (2006) Report of the *Crangon* Fisheries and life history. ICES CM 2006/G:XX, 1-32. 2006.

ICES (2007) Report of the Working Group on Crangon Fisheries and Life History. International Council for the exploration of the Sea Living resources comittee:1-33

Janssen GM, Kuipers BR (1980) On tidal migration in the shrimp *Crangon crangon*. Netherlands Journal of Sea Research 14:339-348

Kinne O (1962) Irreversible nongenetic adaptation. Comparative Biochemistry and Physiology 5:265-281

Koenker R, Bassett G (1978) Regression quantiles. Econometrica 46:33-50

Kuipers BR, Dapper R (1981) Production of *Crangon crangon* in the tidal zone of the Dutch Wadden Sea. Netherlands Journal of Sea Research 15:33-53

Kuipers BR, Dapper R (1984) Nursery function of Wadden Sea tidal flats for the brown shrimp *Crangon crangon*. Marine Ecology Progress Series 17:171-181

Labat J-P (1977) Écologie de *Crangon crangon* (L.) (Decapoda, Caridea) dans un étang de la côte languedocienne. Vie Milieu XXVII:273-292

Lagardère JP (1982) Effects of Noise on Growth and Reproduction of *Crangon crangon* in Rearing Tanks. Marine Biology 71:177-185

Lapinska E, Szaniawska A (2006) Environmental Preferences of *Crangon crangon* (Linnaeus, 1758), *Palaemon adspersus* Rathke, 1837, and *Palaemon elegans* Rathke, 1837 in the littoral zone of the gulf of Gdansk. Crustaceana 79:649-662

Laxter JHSB, Outhward AJS (1991) Moulting and growth. Advances in Marine Biology 27:213-250

Lloyd AJ, Yonge CM (1947) The biology of *Crangon vulgaris* L. in the Bristol Channel and Severn Estuary. Journal of Marine Biology UK 26:626-661

Luttikhuizen PC, Campos J, van Bleijswijk J, Peijnenburg KTCA, van der Veer HW (2008) Phylogeography of the common shrimp, *Crangon crangon* (L.) across its distribution range. Molecular Phylogenetics and Evolution 46:1015-1030

Maes J, Taillieu A, Van Damme PA, Cottenie K, Ollevier F (1998) Seasonal Patterns in the Fish and Crustacean Community of a Turbid Temperate Estuary (Zeeschelde Estuary, Belgium). Estuarine Coastal and Shelf Science 47:143-151

Marques, J. C. Estudo ambiental do estuário do Tejo - Decapodes do estuário do Tejo: distribução das espécies e estudo da biologia das populações de *Carcinus maenas*(Decapoda brachyura) *Crangon crangon, Palaemon longirostris* e *Palaemon serratus* (Decapoda, Caridea). 1982. Labóratorio Marítimo da Guia, Portugal.

Meixner R (1969) Wachstum, Häutung und Fortpflanzung von *Crangon crangon* (L.) bei Einzelaufzucht. Berichte der Deutschen Wissenschaftlichen Komission für Meeresforschung 20:93-111

Meyer P-F (1936) Die Nordseekrabbe (Granat, Garnele) *Crangon vulgaris* Fabr. im Jadebusen. Zeitschrift für Fischerei, Neudamm. 34:319-358

NACA. Better Management Practices (BMP) Manual for Black Tiger Shrimp (*Penaeus monodon*) Hatcheries in Viet Nam. Network of Aquaculture Centers in Asia-Pacific. 2006.

Neudecker, T. and Damm, U. (1992) Seasonality of egg-bearing shrimp (*Crangon crangon* L.) in costal waters of the German Bight. K: 28, 1-9. 1992. ICES Shell Fish Committee.

Oh C-W, Hartnoll RG (2000) Effects of food supply on the growth and survival of the common shrimp *Crangon crangon* (Linnaeus, 1758) (Decapoda, Caridea). Crustaceana 73:83-99

Oh C-W, Hartnoll R, Nash RDM (1999) Population dynamics of the common shrimp, *Crangon crangon* (L.), in Port Erin Bay, Isle of Man, Irish Sea. ICES Journal of Marine Science 56:718-733

Onnen T, Zebe E (1982) Energy metabolism in the tail muscles of the shrimp *Crangon crangon* during work and subsequent recovery. Comparative Biochemistry and Physiology 74 A:833-838

Paschke KA, Gebauer P, Buchholz F, Anger K (2004) Seasonal variation in starvation resistance of early larval North Sea shrimp *Crangon crangon* (Decapoda: Crangonidae). Marine Ecology Progress Series 279:183-191

Peterson I, Wroblewski JS (1984) Mortality rate of Fishes in the Pelagic Ecosystems. Canadian Journal Fisheries and Aquatic Science 41:1117-1120

Pihl L, Rosenberg R (1982) Production, abundance, and biomass of mobile epibenthic marine fauna in shallow waters, western Sweden. Journal of Experimental Marine Biology and Ecology 57:273-301

Pihl L, Rosenberg R (1984) Food selection and consumption of the shrimp *Crangon crangon* in some shallow marine areas in western Sweden. Marine Ecology Progress Series 15:159-168

Plagmann J (1939) Ernährungsbiologie der Garnele (*Crangon vulgaris* Fabr.). Helgoländer wissenschaftliche Meeresuntersuchungen 2:113-162

Porter L, Butler M, Reeves RH (2001) Normal bacterial flora of the spiny lobster *Panulirus argus* and its possible role in shell disease. Marine Freshwater Research 52:1405

Regnault M, Luquet P (1978) Variations quantitatives de l'acide desoxyribonucléique (ADN), au cours de cycle de mue, dans les téguments, le muscle et l'hépatopancréas de la crevette *Crangon crangon*. Journal of Physiology Paris 74:21-30

Regnault M (1970) Croissance au laboratoire de *Crangon septemspinosa* Say (Crustacea Decapoda, Natantia), de la métamorphose a la maturité sexuelle. Bulletin du muséum national d'histoire naturelle 2e Série, Tome 42:1108-1126

Regnault M (1976) Influence du substrat sur la mortalité et la croissance de la crevette *Crangon crangon* (L.) en élevage. Cahiers de Biologie Marine Tome XVII:347-357

Regnault M (1979) Ammonia Excretion of the Sand-Shrimp *Crangon crangon* (L.) During the moult cycle. Journal of Comparative Physiology 133:199-204

Regnault M (1981) Respiration and Ammonia Excretion of the Shrimp *Crangon crangon L.:* Metabolic Response to Prolonged Starvation. Journal of Comparative Physiology 141:549-555

Regnault M, Lagardère JP (1983) Effects of ambient noise on the metabolic level of *Crangon crangon* (Decapoda, Natantia). Marine Ecology Progress Series 11:71-78

Revill AS, Holst R (2004) Reducing discards of North Sea brown shrimp (*C. crangon*) by trawl modification. Fisheries Research 68:113-122

Schatte J, Saborowski R (2005) Change of external sexual characteristics during consecutive moults in *Crangon crangon* L. Helgoland Marine Research

Schockaert E (1968) La croissance de la crevette grise (*Crangon crangon* L.) au large de la cote Belge. Annales de la Societe royale Zoologique de Belgie 98:217-231

Siegel V, Damm U, Neudecker T (2008) Sex-ratio, seasonality and long-term variation in maturation and spawning of the brown shrimp *Crangon crangon* (L.) in the German Bight (North Sea). Helgoland Marine Research 62:

Sogbesan AO, Olowosegun T, Ibiyo LMO (2006) Shell fish farming in Nigeria: Aquaculture potential and investment opportunities projections. American-Eurasian Journal of Agriculture & Environmental Science 1:106-112

Spaargaren DH (2000) Seasonal and annual variations in the catches of *Crangon crangon* (L.1758) (Deacapoda, Natantia) near the coast of Texel, the Netherlands. Crustaceana 73:547-563

Taylor DL, Peck MA (2004) Daily energy requirements and trophic positioning of the sand Shrimp *Crangon septemspinosa.* Marine Biology 145:167-177

Temming A, Damm U (2002) Life cycle of *Crangon crangon* in the North sea: A simulation of the timing of recruitment as a function of the seasonal temperature signal. Fisheries Oceanography 11:45-58

Temming, Axel, Damm, Ulrich, and Neudecker, Thomas. Trends in the size composition of commercial catches of brown shrimp (*Crangon crangon* L.) along the German coast. K: 53. 1993. ICES Shell fish Committee.

Tetard A (1985) Elements sur la croissance de la crevette grise, *Crangon crangon* (L.), en manche-est et sud mer du nord. ICES C.M. K:17:16pp.

Tiews K (1970) Synopsis of biological data on the common shrimp *Crangon crangon* (Linaeus, 1758). FAO Fisheries Reports 4:1167-1223

Uhlig, Nadine. Wachstum und Ernährungssituation der Nordseegarnele *Crangon crangon* (L.). -114. 2002. Diploma thesis: Institute for Hydrobiology and fishery science, University of Hamburg.

van Leeuwen F (1975) Verspreidingvan verschillende garnalensoorten in de Zuidelijke Noordzee gedurende verschillende seizoenen. Interne Verslagen Nederlands Instituut voor Onderzoek der Zee, Texel 8:1-23

van Lissa JHL (1977) Aantallen, voedselopname, groei en produktie van de garnaal (*Crangon crangon* L.) in een getijdengebied, alsmede de voedselopname en groei onder laboratoriumomstandigheden. Interne Verslagen Nederlands Instituut voor Onderzoek der Zee, Texel 1977-C:1-100

Viegas I, Martinho F, Neto J, Pardal M (2007) Population dynamics, distribution and secondary production of the brown shrimp *Crangon crangon* (L.) in a southern European estuary. Latitudinal variations. Scientia Marina 71:451-460

Vogan CL, Costa-Ramos C, Rowley AF (2001) A histological study of shell disease syndrome in the edible crab *Cancer pagurus*. Marine Ecology Progress Series 47:209-217

von Bertalanffy L (1934) Untersuchungen über die Gesetzlichkeit des Wachstums I. Teil: Allgemeine Grundlagen der Theorie; Mathematische und Physiologische Gesetzlichkeiten des Wachstums bei Wassertieren. Development Genes and Evolution 131:613-651

World Wide Web

http://www.bsh.de/de/Meeresdaten/Beobachtungen/MURSYS-Umweltreportsystem/Mursys_031/seiten/nozo1_01.jsp

Appendix

Table 5-5: Data of different authors used in the actual work to determine growth rates of *Crangon crangon*. From left to right: reference, method (cohort or laboratory experiments), temperature range of observed growth rates, observed length classes, range of observed growth rates, determined moult intervals, gender specific growth rates, special treatments like influence of starvation or light on growth rates, specific laboratory conditions, used net for cohort tracking and if net selectivity was included or not, ranking if distinct cohorts were observed or not, area where shrimps were caught, time of catch, temporal and spatial sample resolution for cohort tracking.

Reference	Method	Temp. [°C]	Length [mm]	Growth [mm d^{-1}]	moult intervals	gender sep.	special treatment	feeding	laboratory conditions	net selection type of net	distinct cohorts	sample area	year time	sample resolution
(Amara & Paul 2003)	Coh.	15-20	21-31	0.163	-	no	-	-	-	yes according to (Kuipers and Dapper 1981)	only small size classes	La Canche France	Apr-Sept. 2000	good, weekly and LW and HW
(Beukma 1992)	Coh.	8-12	10, 15, 22	0.20, 0.63, 0.54	-	no	-	-	-	-	medium	Balgzand Netherlands	Apr-Sept 1983-1991	high 3 stations 40 cores
(Boddeke 1976)	Coh.	16	60-63	0.11	-	no	-	-	-	-				
(Boddeke 1966)	Coh.	20	50	0.18	-	no	-	-	-	no	no graph	Netherlands	-	-
(Boddeke et al. 1986)	Coh.	ca. 20	23-67	0.22 - 0.48	-	no	-	-	-	no push net + beamtrawl	medium	Netherlands	Jul- Oct 1984 + Mar 1985	high from 4 depth 8 hauls but low in time
(Caudri 1937)	Lab.	6 19	20-32 20-51	0.05-0.52 -0.05-0.2	-	no	differing salinities	*Mytilus* and algae	stationary + sand and shells	-	-	-	Aug-Oct ~1937	-
(Criales & Anger 1986)	Lab.	6 - 18	Larvae	0.01 - 0.14	3 - 18	no	only larvae test of antibiotics	*Artemia*, rotifers, phytopl.	20 cm^3 30-32 psu water change 2 d	-	-	Helgoland	~1986	-
(Dalley 1980)	Lab.	20	7 - 26	0.17 - 0.57	3-4 (Larvae)	no	light differences	Artemia + Mytilus (3d)	2 l + sand, water change 3-6 d	-	-	Isle of man	Jan + Feb	-
(del Norte-Campos & Temming 1998)	Coh.		10-50	0.23	-	no	growth model	-	-	no push net	only small size classes	Germany	Apr. 1991 - Oct 1992	medium: monthly
(Dornheim 1969)	Coh.	2-17	10-74	0.03 - 0.14	-	yes	-	-	-	no push net		Baltic	May-Oct 1965	high: 1-4 per month
(Edwards 1978)	Lab.	10-20	25	0.02	13 - 47	no	oil addition	teleosts + *Crangon*	-	-	-	Scotland	Jul Aug 1976	-
(Evans 1984)	Field	5 + 20	15-45	no	9 - 175	no	-	-	-	no drop net	-	Swedish west coast	1976-1977	-
Fonds (pers. comm.)	Lab.	2-26	8-57	0.01 - 0,50	-	no	-	-	-	-	-	Netherlands	-	-
(Gerlach & Schrage 1969)	Lab.	5-25	25-60	0.02 - 0.14	-	no	-	Panagrellus (5d per week 1-4 portions)	20 psu, water change 30d	push net	-	Germany	Oct 1967Jan Apr Sept1968	-
(Henderson & Holmes 1987)	Coh.	3-23	35-66	0.00 - 0.10	14 - 144	no	-	-	-	capture - recapture power station	medium	Bristol Channel	1980-1982	medium: monthly
(Kuipers & Dapper 1981)	Coh.	7-19	15-30	0.12 - 0.35	-	no	-	-	-	yes (van Lissa)	no	Netherlands	1976-1979	monthly

Reference	Method	Temp. [°C]	Length [mm]	Growth [mm d^{-1}]	moult intervals	gender sep.	special treatment	feeding	laboratory conditions	net selection type of net	distinct cohorts	sample area	year time	sample resolution
(Labat 1977)	Coh.	10-20	8-46	0.01 - 0.38	-	yes	-	-	-	no 5 mm net mesh size	medium	France	Apr. 1974 - Jul 1974	low 11 per 2 a
(Lagardère 1982)	Lab.	15-23	39-52	0.03-0.17	-	yes	noise	Mytilus	flow through 55 l + sand +nylon wool	-	-	France	April 1981	-
(Lloyd & Yonge 1947)	Lab.	12	32-68	-	Increments	-	-	Mytilus	sand, circulation	-	-	Bristol Channel	~1947	-
(Meixner 1969)	Lab.	14	10-50	0.13 - 0.50	11 - 24	yes		Artemia	0.5-5.5 l 18-30 psu	short catches	-	Germany	Mai June 1964	-
(Meyer 1936)	Coh.	5-20	15-38	0.03 - 0.21		no	-	-	-	placed nets (Stellhamen)	no graph	Germany	May 1930-Aug 1931	high >4 per month
(Oh & Hartnoll 2000)	Lab.	15	35-45	0.06 - 0.10	10 - 55	no	feeding every d or every 5 d	*Nephrobs* + prawns	34 psu, compartments in 150 l tanks flow through	-	-	Isle of man	~2000	63 shrimps
(Oh et al. 1999)	Coh.	av. 12	32 - 81	0.01 - 0.11	-	yes	-	-	-	no 1.5 (2) m beam trawl 3 (6) mm	yes	Isle of man	Apr 1995 - Jul 1998	5 transects every 2-4 weeks
(Regnault 1976)	Lab.	15-19	19-40	0.11 - 0.33	-	no	with and without substrate	Carcinus maenas	45-52 l, flow through	-	-	-	Jun-Oct 1975	-
(Regnault 1979)	Lab.	19	25	no	10	no	moult cycle	Carcinus maenas	standin aerated	-	-	France	Jul	
(Regnault 1981)	Lab.	12-14	ca. 40	0.20		no	starv.	Carcinus maenas	-	-	-	France	-	-
(Schatte & Saborowski 2005)	Lab.	5 - 16	26 - 39	no	23 - 46	males	obs. sex change	Mytilus + Crangon	flow through, sand	-	-	Germany	Nov 2003	70 males
(Schockaert 1968)	Coh.	1.1-15.5	45-65	0.00 - 0.17	-	no								
(Tetard 1985)	Coh.	15 - 18	26-61	0.07 - 0.40	-	no	-	-	-	-	yes	France	Jul 1977 Sep 1982	low monthly in summer
(Tiews 1970)		5,10,15	35,73	no	15 - 28	no								
(Uhlig 2002)	Lab.	16 - 17	20-40	0.20 - 0.56	13 - 14	yes	-	planc.,*Nereis*, smelt, *Mytilus*	flow through	-	-	Germany	Jul 2000	-
(van Leeuwen 1975) in (Kuipers & Dapper 1981)	Coh.	14	43-51	0.13	-	no	-	-	-	-	-	-	-	-
(van Lissa 1977)	Lab.	10 - 25	13-44	0.06 - 0.27	-	no	-	Mytilus	30 psu	-	-	Netherlands	~1977	-
(Viegas et al. 2007)	Coh.	15-18 (annual mean)	10-50	-0.11-0.23	-	no	-	-	-	night, high tide,beam trawl 5 mm	no graph	Portugal	2003-2005	monthly

Is *Crangon crangon* (L. 1758, Decapoda, Caridea) food limited in the Wadden Sea?

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Abstract

Dry weight, caloric content and RNA·DNA^{-1} of Brown shrimp *Crangon crangon* were monitored in temperature controlled starvation experiments and field sample. Within the experiments in all length classes (20, 30, 45 and 60 mm) and at all temperatures (9, 12, 17 and 20°C) a significant decrease of dry weight and RNA·DNA^{-1} was observed. Greatest weight loss of -1.9%·d^{-1} occurred at 20°C. Dry weight condition (dry weight at length) and RNA·DNA^{-1} in field samples from the Wadden Sea (2005-2007) peaked in summer and were lowest in winter, while calorific content increased during winter, what might indicate vitellogenesis. Applying the results of the starvation experiments to field conditions suggests that throughout the year at least 25% of the shrimp population is food-limited and therefore not capable of generating maximum growth. Furthermore, maximum food limitation was determined for the period November until April where up to 75% of the population exhibited signs of starvation or food limitation.

Keywords: *Crangon crangon*, brown shrimp, food limitation, bottom up, starvation, RNA·DNA^{-1}, caloric content, weight at length, condition

Introduction

The Wadden Sea covers an area of 9500 km² in the South-Eastern North Sea. Due to its proximity to the coast high concentrations of nutrients are transported by rivers and land runoff into the sea. With respect to biomass production it is therefore one of the most productive areas in the world (Field et al. 1998). Compared to upwelling areas with mean primary production of 300 g $C \cdot m^{-2} \cdot d^{-1}$, inshore waters of up to 182 g $C \cdot m^{-2} \cdot d^{-1}$ and neretic water of up to 365 g $C \cdot m^{-2} \cdot d^{-1}$ (Bunt 1973), peak primary production of over 500 g C m^{-2} a^{-1} can be observed in the Wadden Sea (de Jonge 1997). One of the key species in the Wadden Sea food web is the Brown shrimp *Crangon crangon* (Kuipers & Dapper 1984). Via this species, energy is transferred from the lower trophic levels (Pihl & Rosenberg 1984) to top predators such as cod (*Gadus morhua*), whiting (*Merlangius merlangius*) (Hamerlynck & Hostens 1993) and also humans (ICES 2006). Larger shrimps are mainly situated in deeper water (Janssen & Kuipers 1980),(Siegel et al. 2005, Welleman & Storebeck 2002, Tiews 1954) whereas juveniles use the shallow tidal flats (Evans 1984) (Gunnarsson et al. 2007, Neves et al. 2007)as nurseries (Beukema 1992, Kuipers & Dapper 1984). There densities can reach 80 individuals per m² (Boddeke et al. 1986). Due to the high primary production, total biomass of the shrimp population is assumed to increase by a factor of four in the shallow nursery areas (Kuipers & Dapper 1981). On the other hand density-dependent regulation has been suggested to occur in this species. E.g. in the Bristol Channel variable recruitment that results in relatively stable adult populations was found (Henderson et al. 2006). Kuipers & Dapper discussed that based on a daily consumption of 6.7 g ash-free dry weight (ADW)$\cdot m^{-2}$ (Kuipers & Dapper 1981), food limitation would be expected if mainly macrobenthic production would be considered and competition with other consumers would be assumed.

Food limitation and bottom-up control of the brown shrimp population might therefore be expected even in highly productive areas like the Wadden Sea and might in future become more severe due to increasing temperatures. It was suggested that food intake of *C. crangon* increases by about 6% with each degree water temperature rise (Pihl & Rosenberg 1984). One effect of food limitation and bottom-up control might be a reduction of condition and consequential growth. One major aspect of the actual work is therefore to determine if there are indications for reduced condition and food limitation of *C. Crangon*.

To quantify the condition of aquatic organisms dry weight at length, $RNA \cdot DNA^{-1}$ and calorific content are commonly used and were thus chosen our study. The dry weight at length or dry weight condition index (DWCI) is based on the assumption that a heavier animal of a given length is in better condition than a lighter one (Bolger & Connely 1989). It is a relative index that enumerates the weight deviation of one specific animal of a certain length from a reference sample of similarly-sized individuals of the same species. For example, this index was used to determine density depend variations of *Farfantepenaeus* species (Perez-Castaneda & Defeo 2008) and to describe seasonal variations in condition of *Euphausia superba* (Nicol et al. 2000).

The amount of RNA in a cell is a measure of protein synthesis, and therefore of growth rate, whereas the amount of DNA, as carrier of genetic information, is assumed to be constant per cell. RNA·DNA^{-1} has previously been used as an index of nutritional condition reflecting starvation, feeding and growth (Norkko et al. 2006, Peck et al. 2003, Wagner et al. 1998, Moss 1994b, Buckley 1984).

The caloric content yields information on an organism's nutritional state. Storage lipids have a higher energy per unit dry weight and energy is only stored when a surplus of food is available.

In the present work, the development of dry weight and RNA·DNA^{-1} of *C. crangon* that were experimentally starved under a controlled temperature regime was examined to establish reference values for the interpretation of field data. The aim was to determine whether *C. crangon* is food limited during the year.

Material and Methods

Field samples and collection of the animals for the starvation experiments

Animals were collected at three sites: Büsum (54°07'N, 8°51'E), Meldorf (54°05'N, 8°56'E) and Wilhelmshaven (53°33'N, 8°09'E) (Table 6-1). In Büsum animals were sampled during low tide with a push net in about 1 m depth. The sampled distance was between 400 and 700 m per haul and, depending on the shrimp density, one to five hauls were performed per sampling. The net had a mesh size of 1.8 mm, a width of 144 cm and a height of 23 cm. Sampling time was limited to 10 min to avoid damage of the shrimps (Gamito & Cabral 2003). Live animals were transported to the Institute for Hydrobiology and Fisheries Science in Hamburg in aerated transport boxes. Specimen for the field analysis were frozen at -80°C for RNA·DNA^{-1} and at -20°C for length, dry weight and caloric content analyses.

Meldorf was only sampled in 2005 and at occasionally in 2006. Due to the muddy ground sampling could only be performed during high tide, what lead to a reduced size spectrum of the shrimps caught. In Wilhelmshaven samples were obtained from the cooling water intake of the local power plant between 2005 and 2007. Aquatic organisms and other material was filtered from the cooling water by two consecutively mounted grid screens, the second of which retained *C. crangon* >35-40 mm effectively. The grid was automatically cleaned every 120 min and the retained animals were brushed into metal tubes, collected and stored frozen until further processing.

Table 6-1: Amount of performed samplings and amount of *Crangon crangon* caught in Büsum, Wilhelmshaven, Meldorf and at all sites in 2005, 2006 and 2007.

year	month	Büsum samplings	n *C.cr.*	WHV samplings	n *C.cr.*	Meldorf samplings	n *C. cr.*	all sites sum *C.cr*
2005	4			1	94			94
	5			1	78			78
	6			1	374	1	112	486
	7			3	806	2	405	1211
	8			8	1888	1	134	2022
	9			1	66	1	184	250
	10			2	413	1	307	720
	11			1	177	1	294	471
	12			1	188	1	0	188
2006	1			2	110	1	0	110
	2	1	281	1	24			305
	3			1	41			41
	4	4	1357	1	873	1	0	2230
	5	4	912	1	122	2	0	1034
	6	1	210	3	355	1	402	967
	7			1	77			77
	8	1	716			1	0	716
	9	1	416	1	68			484
	10	1	780	1	171			951
	11	1	172			1	315	487
	12	1	222	1	162			384
2007	1			1	107			107
	2	1	56	1	30			86
	3	1	334	1	258			592
	4	3	604	1	143			747
	5	4	304			2	0	304
	6	3	1056			1	7	1063
	7	1	910					910
	8	1	964					964
	10	1	620					620
	11	1	620					620
	12	1	1100					1100
sum samplings		32	11634	36	6625	18	2160	20419

Length, weight, sex

Length was determined as total length from the tip of the telson to the tip of the scaphocerite to the lower mm with a mm-paper. For comparability of the results with work of other authors additionally the carapace length was determined from a subset of animals. Wet weight was determined after thawing. Dry weight was determined after 24 h freeze-drying (scale: Satorius ± 0.0001 g).

Caloric content

To determine the caloric density, the freeze-dried shrimps were divided into 5 mm size classes (10-14 mm, 15-19 mm …), mashed and pressed to pellets of ~0.1 g.

Caloric density was determined with a microbomb calorimeter (IKA Analysetechnik Heitersheim, IKA-Calorimeter C4000).

Length weight relation and dry weight condition

Wet weight- and dry weight-length relationships were described as:

$$w = a \cdot L^b \qquad (10)$$

with w = dry or wet weight and L = length.

Dry weight condition index was calculated as

$$DWCI = \frac{dw}{a \cdot L^b} \qquad (11)$$

with dw = dry weight in [g] and constants a and b derived from the length weight relation. For the determination of the DWCI of berried females eggs were removed from the setae with a washing bottle and tweezers. The dry weight and DWCI of ovigerous female shrimps was therefore determined without eggs.

Water content, ash-, ash-free-dry weight and protein content

From all samples water content was determined by freeze-drying. From a subset of 122 pooled samples additionally the ash free dry weight (3 replicates per pooled sample) was determined. Samples contained animals of one size class and sampling date. Ash free dry weight was determined after combusting the samples for 2 h at 550°C. Protein content was determined from nine pooled freeze-dried samples (3 replicates per pooled sample) applying the Kjeldahl method. For the calculation of protein from total N the standard factor of 6.25 was used. Protein samples were analyzed from May, July, August, September and October for the size class 51-55 mm and from October additionally for the size classes 41-45, 66-70 and 76-80 mm.

RNA·DNA^{-1} analysis

RNA·DNA^{-1} were determined following the procedure described by Caldarone et al. (2001). Dry weight was determined after one day freeze drying. For analysis the third and fourth segment was cut from the shrimp with a scalpel. The muscle was transferred to 1.5 ml Eppendorf Cap for weight determination after the surrounding chitin was removed. Glass pearls (Ø 1 mm) and 300 µl of 3% Sarcosil Tris-EDTA buffer (N-lauroylsarcosine, 5 mM Tris-HCl, 0.5 mM EDTA) were added and the samples were shaken on a Vortexer for 45 min. Although fluorescence increases with concentration of Tris (Caldarone & Buckley 1991) this concentration was chosen as preliminary experiments with varying concentrations allowed the best sample dissociation and stable results. The solution was treated with an ultrasonic homogenizer for 20 sec in 1 s pulses (20 kHz). To prevent a

temperature increase in the sample during sonification the caps were stored in crushed ice. 1200 µl Tris-EDTA buffer were added and the samples were treated following Calderone et al. (2001). All chemicals and standards used were purchased from Sigma Aldrich, Germany, the microplates from BioLabProducts GmbH, Germany. The fluorescence reader was a Xenius (SAFAS, Monaco). Emission wavelength peaked at 605 nm for an excitation wavelength of 520 nm. From 5 measurements over 0.1 s the mean fluorescence for each well was calculated. For each plate two calibration lines were added, each sample was measured twice and as internal standard a homogenate made of homogenized *C. crangon* muscles was used. Linearity was given for the whole spectrum of concentrations analyzed. Limit of detection was determined according to IUPAC (International Union of Pure and Applied Chemistry) as blind value +3 times the standard deviation of the lowest calibration line value and was 0.67 µg ml^{-1} and 0.54 µg ml^{-1} for RNA, DNA, respectively. This value falls in the range found by Chícharo et al. (1998b), who determined 0.4 and 0.1 $µg \cdot ml^{-1}$ for RNA and DNA, respectively but with only 2 times the standard deviation.

Repeatability was determined by the coefficient of variation between the calibration standards of all measurements as 8, 10% for RNA, DNA respectively over one year of measurement (21 laboratory days, n = 294). The variation between the wells (each sample analyzed two fold) in our samples was 2% before addition of RNase and DNAse and 6% after addition. The slope ratio for intercalibration was $RNA \cdot DNA^{-1}$ 0.85 (Caldarone et al. 2006).

Starvation experiments

Starvation experiments were performed in 2006 and 2007. After one day of acclimatization after the catch the animals were placed in a separate closed water circulation system, containing approximately 1100 l. To clear any potential food items from the system, artificial sea water (Aquamedic, Germany) was produced with tap water a few days before each experiment. The temperatures in the experiments were ± 2°C of the respective field temperatures at catch of the animals. Salinity was adjusted between 27 and 30 PSU. Animals were held separately in 500 ml Kautex bottles to prevent cannibalism. Ten bottles were placed in one aquarium (polypropylene, 40 x 30 x 20 cm, 17 l when filled). Water exchange was guaranteed by little holes (Ø ~ 1mm) in the bottles and an alternating water level in the aquarium induced by a hose sling in the outlet. Water was flowing permanently into the aquarium until the water level was higher then the top of the sling (Figure 6-1). Beyond that point water started to drain of until the lowest point of the sling was reached. By this about half of the water in the aquarium was emptied approximately every 30 min.

After approximately 10, 20, 30, 60 130, 190, 230, 280 and 330 degree days (Temperature[°C]·time[d]) 5 shrimps were removed and frozen at -80°C for later analysis. Three different size classes were analyzed (20, 30, 45, 60 mm) at four different temperatures (9, 12, 17, 21°C). For each experiment 100 animals were

used. The 9°C (30, 45 mm) experiment started on November 29th 2006, the 12°C experiment (20, 60 mm) at October 4th 2007, the 12°C (30, 45 mm) at October 30th 2007, the 17°C (30, 45 mm) experiment at June 27th 2007, the 21°C (30, 45 mm) experiment at August 29th 2007.

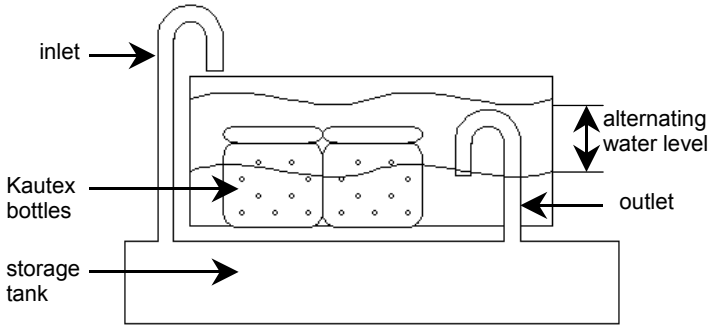

Figure 6-1: Sketch of experimental setup of the starvation experiment showing the Kautex bottles used to hold *Crangon crangon* separately and the hose sling used to guarantee the water exchange in the Kautex bottles.

Starvation-times in the field

Starvation times in the field were estimated from the share of the actual condition (quartile of dry weight, RNA·DNA^{-1} and caloric content), the mean annual condition in the field, the slope determined in the starvation experiments and the temperature.

Daily temperature was derived from a sinus function fitted to the temperatures measured in Büsum

$$T = 11.77 + 8.557 \cdot \sin\left(0.018 \cdot (\mathrm{d} - 120)\right) \qquad (12)$$

with d = day and T = temperature [°C].

In steps of one day the DWCI loss at the given temperature was added to the observed DWCI in the field until a value of 1 was reached. If for example the DWCI on February 10th (day 40) was 0.81 then the temperature of the day before (day 39) was 11.77+8.557·sin(0.018·(39-120)) = 3.2 what corresponds to 3.2 degree days. During 3.2 degree days 3.2·0.000741 of the DWCI is lost (slope of the regression see Table 6-5). Therefore the DWCI at day 39 was 0.81+ 0.00237 = 0,81237.

As for RNA·DNA^{-1} no significant temperature dependence could be determined the RNA·DNA^{-1} loss during starvation was determined without temperature but comparable to the previous described procedure. Here a starting value of RNA·DNA^{-1}= 0.93 was assumed as this represents the mean value of all samples.

Results

Field data

Temperature and Salinity

The seawater temperature in Büsum (Meldorf) varied between -0.6°C in January 2006 and 22°C measured in July 2005, August 2006 and June 2007. The mean salinity was 23.8 pSU.

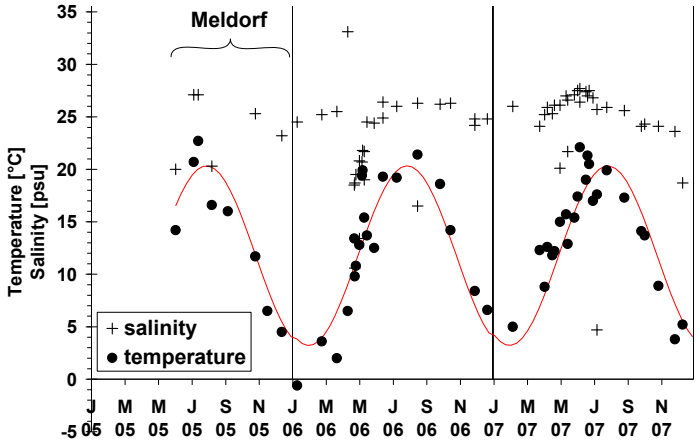

Figure 6-2: Temperature [°C] (dots) and salinity [pSU] (rectangles) measured in Büsum and Meldorf during the samplings in 2005 to 2008.

Length and Abundance

Mean length of *C. crangon* caught in Meldorf was about 10 to 20 mm (Figure 6-3), mean length in Büsum ranged from 30 to 40 mm in August and September 2007 larger (43-45 mm) animals were caught. During the first half of 2006 a constant increase of the mean length was observed from 19.5 mm in February to 40 mm in May. These length distributions were made up by one cohort until in mid May larger animals occurred in the catches and in June the new recruits arrived.

The abundance of shrimps in Büsum/Meldorf was generally lower in winter (0.2-0.6 Ind.·m^{-2}) than in summer (2-2.8 Ind.·m^{-2}, Figure 6-4). In Meldorf, in 2005 the highest abundance was determined in June (6.22 Ind.·m^{-2}). In Büsum, abundance peaked in October 2006 (3.58 Ind.·m^{-2}) after a relatively low density in September (0.67 Ind.·m^{-2}). From early 2007 on abundance constantly increased to values of 2 Ind.·m^{-2} from September on. The abundance in Wilhelmshaven peaked in June 2005 (10 shrimps per m³) and April 2006 (18 shrimps per m³). In 2005 in Meldorf between 1.1 and 6.1 Ind.·m^{-2} but mainly around 3.5 Ind.·m^{-2} were counted.

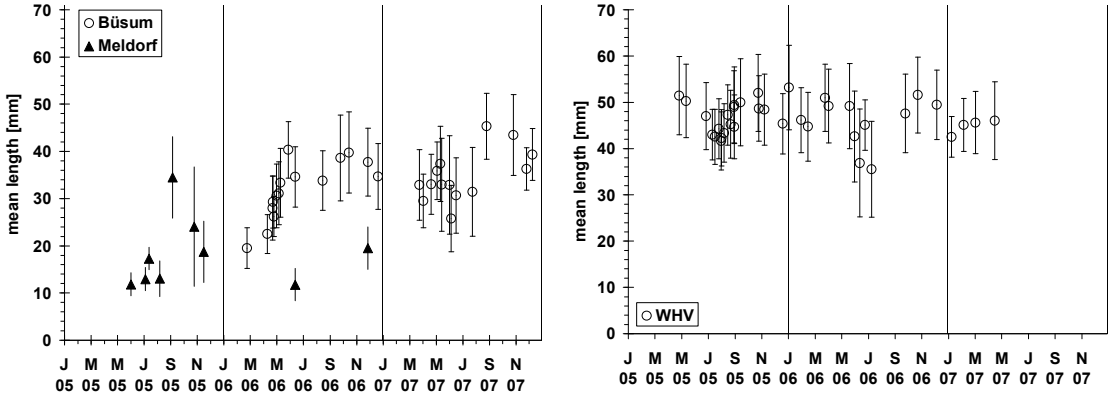

Figure 6-3: Mean length and standard deviation of *Crangon crangon* caught in Büsum/Meldorf (left) and Wilhelmshaven (right).

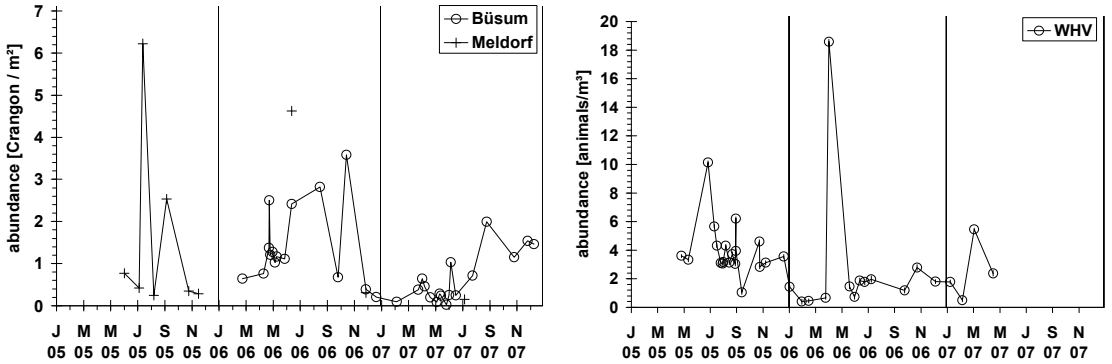

Figure 6-4: Abundance of *Crangon crangon* caught in Büsum/Meldorf (left) and Wilhelmshaven (right). Abundance in pushnet catches (Meldorf and Büsum) are given in $n \cdot m^{-2}$ and in power station samples in $n \cdot m^{-3}$.

Water content, ash free dry weight, protein content

For all samples a mean water content of 74.1 ± 1.8% (standard deviation) was determined with no seasonal variation. The content of ash free dry weight varied only slightly ± 3% (minimum maximum) with no distinct seasonal pattern. The mean ash free dry weight (related to wet weight) was 19.2 ± 1.6% (standard deviation), the mean ash content 6.7 ± 0.8% (std). Within the protein subsamples no seasonal or size-related trend was detectable. The mean percentage of protein (related to dry weight) was 45.5 ± 2.2% (std). The following contents according to wet weight were determined:

water	mineral	protein	remaining organic material
74.2 %	6.5 %	11.9 %	7.4 %

Dry and wet weight, carapace length

Fitting function (10) to the length and weight data yielded the following functions:

wet weight $= 4.0625 \cdot 10^{-6} \cdot L^{3.084}$ $r^2 = 0.985$ (n = 8305)

dry weight $= 1.30054 \cdot 10^{-6} \cdot L^{3.062}$ $r^2 = 0.977$ (n = 8305)

The lower and upper 95% confidence intervals for the wet weight parameters were $4.3772 \cdot 10^{-6}$, $4.8278 \cdot 10^{-6}$ and 3,072, 3.097; those for the dry weight function $1.22176 \cdot 10^{-6}$, $1.37931 \cdot 10^{-6}$ and 3.047, 3.077.

For carapace (CL) and total length (TL) the following relation was determined

$$CL = 0.2317 \cdot TL - 0.7486 \quad (13)$$

The lower and upper 95% confidence intervals for the slope were 0.229, 0.234 and for the intercept -0.831, -0.642 with $r^2 = 0.974$ (n = 929, $p<0.001$).

Condition, caloric content and RNA·DNA^{-1}

Caloric content of different sized shrimps were significant different (10-25 mm and 25-45 mm, p=0.121; 10-25 mm and >45 mm, p:0.089; 25-45 mm and >45 mm, p=0.679). Although high variability was detected, especially in spring, a general seasonal pattern was observed (Figure 6-5, Figure 6-8). Values were always highest in May and June, decreasing to a minimum in September. Over the winter an increase until May was observed. In 2007 calorific contents (Büsum) were slightly lower than in 2005 and 2006. The range of the caloric content values was between 14000 and 17000 J·g^{-1}dry weight in Büsum and from 15000 to 17000 J·g^{-1} dry weight in Wilhelmshaven.

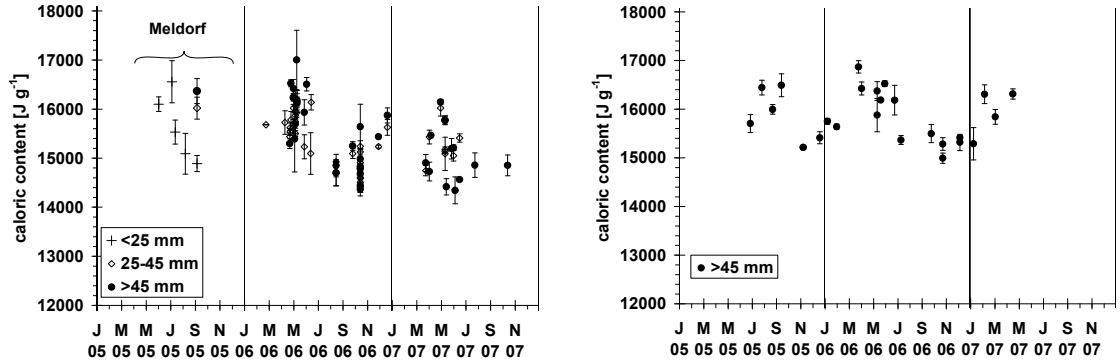

Figure 6-5: Caloric content [J·g^{-1}] of different size classes of *Crangon crangon* caught in Büsum/Meldorf (left) and Wilhelmshaven (right)

The dry weight condition index (DWCI) followed the seasonal temperature pattern. The highest values of 1.2 to 1.3 were measured in Büsum in May, June and in Wilhelmshaven in July, August (Figure 6-6, Figure 6-8). Towards March the mean DWCI decreased, approaching 0.9 in Büsum and to 0.8 in Wilhelmshaven. In Wilhelmshaven shrimps had a significantly lower mean dry weight per unit length in 2005 than in 2006 for the period May-September (t-test, $p < 0.001$). Between the size classes 25-45 mm and >45 mm no significant differences could be detected (t-test, p=0.695).

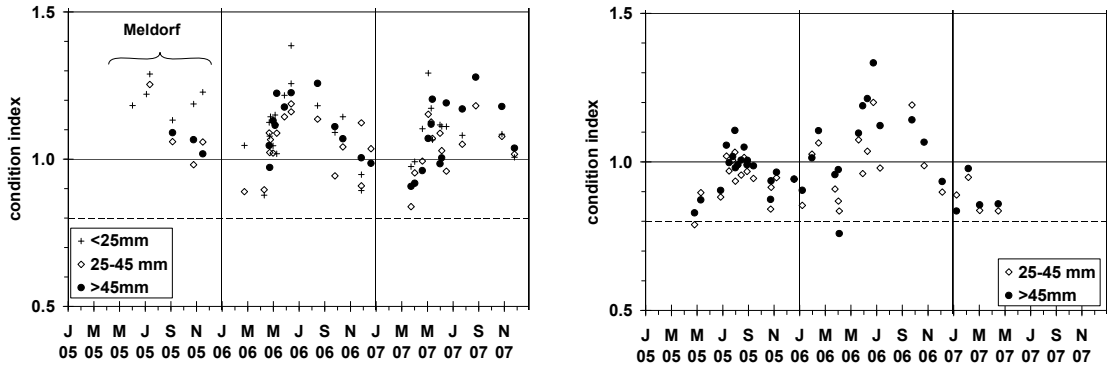

Figure 6-6: Mean dry weight condition of different size classes of *Crangon crangon* caught in Büsum/Meldorf (left) and Wilhelmshaven (right). Dots represent the mean and sd of the DWCI in the referred size class at each sampling.

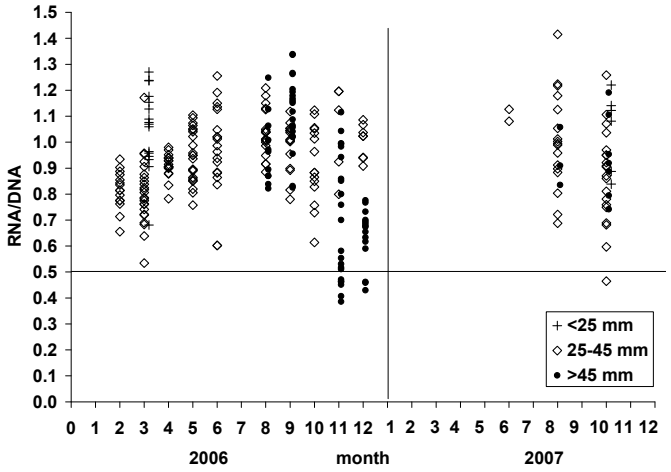

Figure 6-7: RNA·DNA^{-1} of different sized *Crangon crangon* caught in Büsum in 2006 and 2007

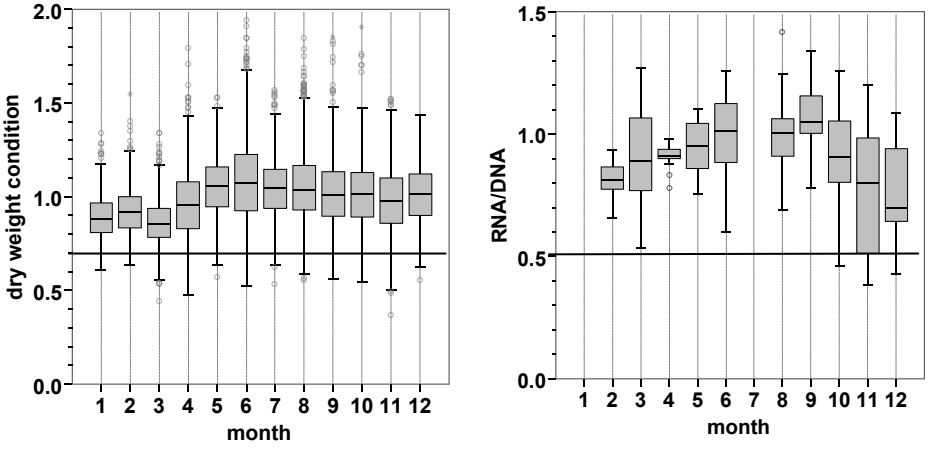

Figure 6-8: Variability of dry weight condition (left) and RNA·DNA^{-1} (right) of all samples per month. Lines indicate the dry weight condition and RNA·DNA^{-1} at the end of the starvation experiments.

Maximum RNA·DNA^{-1} for the 25-45 mm size class were determined in August (1.04) minimum ratios in October and February (0.87, 0.82). For the larger (>45 mm) and lower (<25 mm) size class available samples were too small to represent a complete seasonal pattern, but in 2006 a decrease from a ratio of 1.11 in September to 0.64 in December was apparent.

Starvation experiments

Mortality and growth

In the 9°C experiment 5 animals died on day 15, 19, 28 and two on day 33 the last day of that experiment. At 12°C the run time was 26 days. In the experiment with the 20 and 60 mm size classes one animal died at day 9 and in the experiment with 30 and 45 mm size classes two died (day 16 and 23 of 26). At 17°C three shrimps died on day 2, 7 and 12 of 21 days run time. At 21°C during the whole 18 days of the experiment no shrimp died.

Within the starvation experiments 75 shrimps moulted. Of these animals 4% shrank and more than the half (53%) did not grow. Two shrimps (3%) grew with increments of 4 mm. (Table 6-2)

Table 6-2: Moult increments, number of moultings and rel. number of observed moultings (in comparison to all moultings) with this length increment.

Increment [mm]	number of observed moultings	rel. number [%]
-1	3	4
0	40	53
1	9	12
2	14	19
3	7	9
4	2	3

Dry weight condition index

The DWCI constantly decreased in all starvation experiments performed (r^2 see Figure 6-11). The slope (decrease of the DWCI over the time of starvation) increased with temperature (Table 6-3). In the beginning of each experiment the DWCI was about 1.1 and only those animals of the smallest length class (20 mm, 12°C) had a higher DWCI (intercept = 1.26). At the end of the experiments the DWCI was generally about 0.8 (Figure 6-11). In some cases the decrease seems to be higher during the first days but when fitting an exponential function to the data the r^2 varied only slightly (maximum 0.02) from those of the linear regression (Table 6-3), therefore the linear model was chosen.

DWCI was determined at different days from subgroups of 5 animals. Length in the subgroups did not vary significantly (ANOVA, $p>0.05$), except for day 19 in the 9°C experiment in the different experiments. As the DWCI is calculated as dry

weight per length and length in the experiments was not significantly different the dry weight loss corresponds directly to the DWCI decrease. Therefore the decrease per day corresponds to the variable K (K = 365·slope) in the weight-based von Bertalanffy growth equation and K/3 to k (k_L Table 6-3, Figure 6-9) in the length based Bertalanffy growth equation.

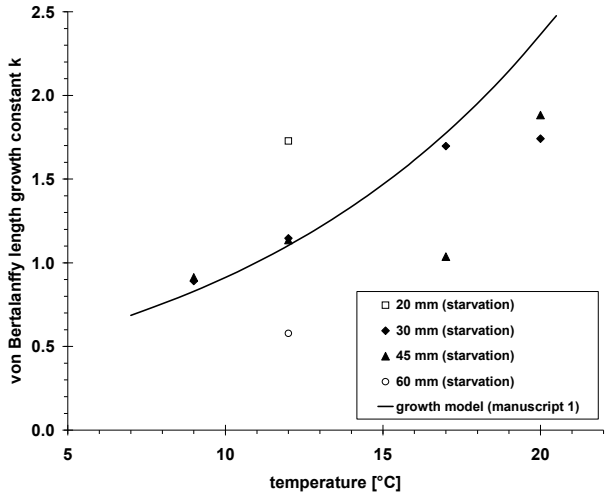

Figure 6-9: Comparison of von Bertalanffy length growth constant k determined in starvation and growth experiments at different temperatures.

Table 6-3: Temperature in the experiment, length class, intercept (with 95% confidence intervals), slope (with 95% confidence intervals) and r² of the decrease of the dry weight condition index of *Crangon crangon* over time of starvation for different length classes and temperatures over time of starvation

T [°C]	L [mm]	intercept	95% conf. interval		slope	95% conf. interval		r²	k_L	p
9	30	1.041	1	1.082	-0.0089	-0.0117	-0.0061	0.488	1.08	<0.001
9	45	1.051	1.004	1.098	-0.0091	-0.0124	-0.0059	0.356	1.11	<0.001
12	20	1.256	1.212	1.3	-0.0173	-0.0208	-0.0138	0.602	2.10	<0.001
12	30	1.057	1.025	1.088	-0.0115	-0.0138	-0.0091	0.609	1.39	<0.001
12	45	1.075	1.033	1.117	-0.0114	-0.0146	-0.0082	0.461	1.38	<0.001
12	60	1.129	1.085	1.174	-0.0058	-0.0091	-0.0025	0.442	0.70	0.001
17	30	1.125	1.085	1.164	-0.0170	-0.0211	-0.0128	0.547	2.07	<0.001
17	45	1.118	1.08	1.156	-0.0104	-0.0137	-0.0070	0.442	1.26	<0.001
20	30	1.072	1.032	1.112	-0.0174	-0.0221	-0.0127	0.479	2.12	<0.001
20	45	1.132	1.105	1.16	-0.0188	-0.0221	-0.0156	0.695	2.29	<0.001

RNA·DNA^{-1}

The RNA·DNA^{-1} declined significantly ($p<0.05$) in every starvation experiment performed (Table 6-4, Figure 6-11) but in most cases not before about five days where the RNA-DNA-ratio remained on a nearly constant level. The slopes (RNA·DNA^{-1} per day) varied between 0.0077 for 45 mm sized *C. crangon* at 12°C (minimum) and 0.0204 for 20 mm sized animals also at 12°C (maximum). The ratios at the beginning of the experiment varied between 0.7 (17°C 45 mm) and 1.36 (9°C 30 mm) and at the end between 0.4 and 0.8 (Figure 6-11). The r² values varied between 0.327 (21°C 30 mm) and 0.911 (9°C 30 mm). The weakest

correlations were determined for 21°C and 30 and 45 mm size animals and for 17°C and 45 mm sized shrimps.

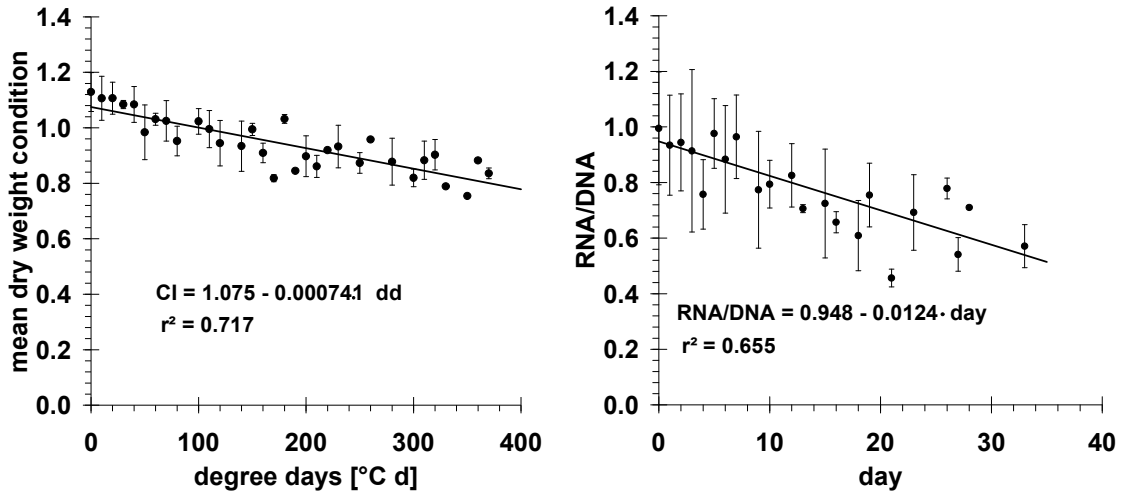

Figure 6-10: Dry weight condition (left) and RNA·DNA^{-1} (right) of *Crangon crangon* in the starvation experiments (all temperatures and size classes) according to the degree day [°C·d]

Table 6-4: Temperature in the experiment, length class, intercept (with 95% confidence intervals), slope (with 95% confidence intervals) and r² of the decrease of RNA·DNA^{-1} of *Crangon crangon* over time of starvation for different length classes and temperatures over time of starvation

T [°C]	L [mm]	intercept	95% conf. interval		slope	95% conf. interval		r²	p
9	30	1.355	1.268	1.442	-0.0251	-0.0313	-0.0188	0.567	<0.001
9	45	1.030	0.961	1.098	-0.0168	-0.0216	-0.0120	0.504	<0.001
12	20	1.352	1.25	1.455	-0.0204	-0.0285	-0.0124	0.286	<0.001
12	30	0.838	0.778	0.897	-0.0105	-0.0158	-0.0052	0.221	<0.001
12	45	0.813	0.768	0.859	-0.0077	-0.0115	-0.0039	0.232	<0.001
12	60	0.994	0.903	1.085	-0.0118	-0.0184	-0.0052	0.200	0.001
17	30	0.753	0.678	0.829	-0.0122	-0.0201	-0.0043	0.140	0.003
17	45	0.703	0.599	0.807	-0.0112	-0.0203	-0.0021	0.098	0.017
20	30	1.045	0.975	1.114	-0.0153	-0.0236	-0.0071	0.181	<0.001
20	45	0.889	0.824	0.954	-0.0098	-0.0176	-0.0021	0.087	0.013

Decline of RNA·DNA^{-1} and dry weight condition index during starvation

Plotting the DWCI and RNA·DNA^{-1} against degree days [°C·d] revealed a linear decline in both cases. Yet the linear regression of RNA·DNA^{-1} with degree days did not increase the explained variability (r²) over the linear regression that only took days and not temperature into account (Table 6-5). As also the confidence intervals of the slopes for different starvation temperatures did overlap (Table 6-3), a temperature dependence of RNA·DNA^{-1} and starvation was neglected. For DWCI an intercept of 1.075 and for RNA·DNA^{-1} of 0.948 was detected.

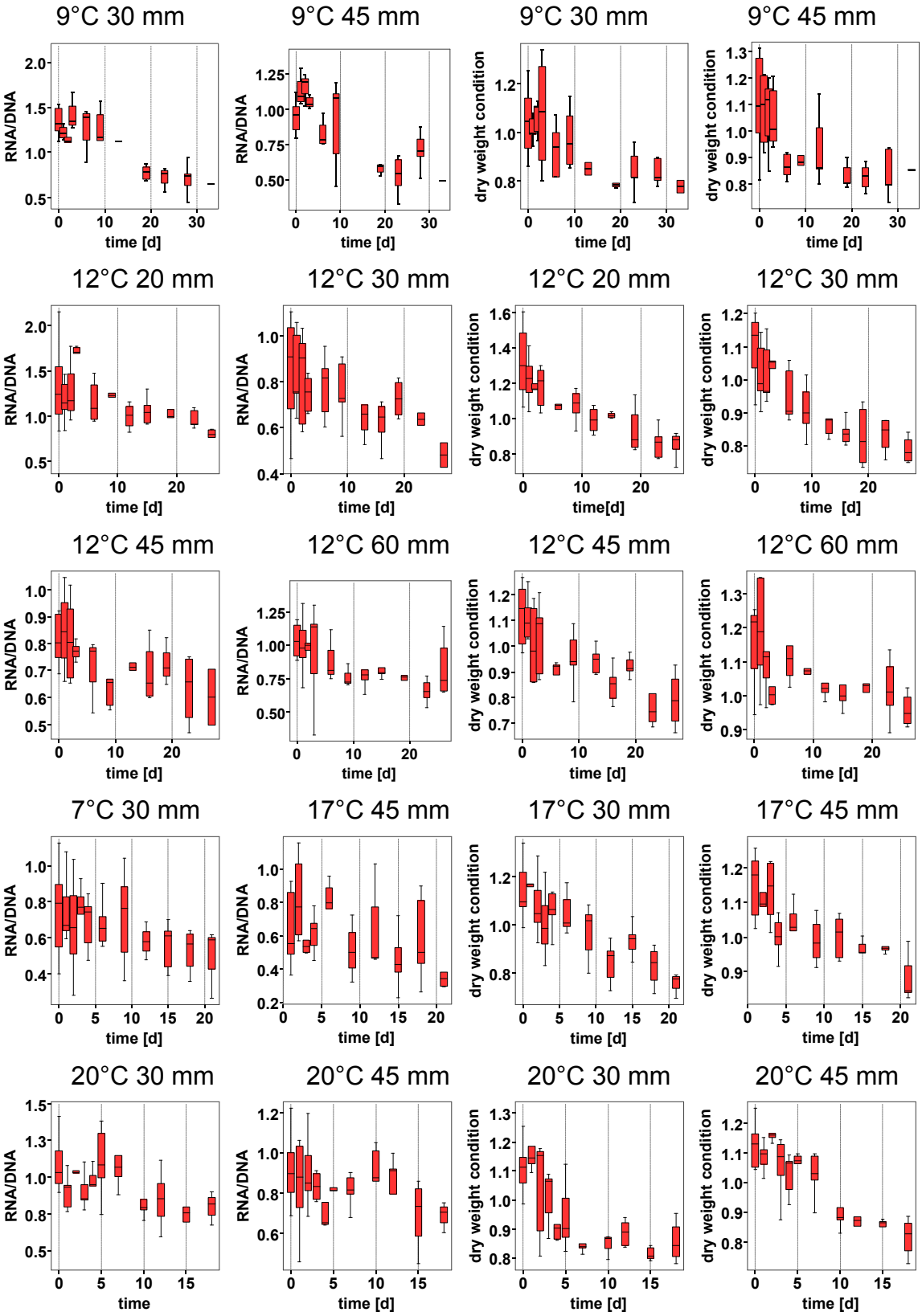

Figure 6-11: 1st and 2nd column: RNA·DNA^{-1} for different sized *Crangon crangon* after starvation at different temperatures over ~330 degree days. 3rd and 4th column: Dry weight condition index for *Crangon crangon* after starvation at different temperatures over different time spans.

Table 6-5: Intercept and slope for linear regression of dry weight condition index and RNA·DNA^{-1} against starvation time in days and degree days.

dependent variable	independent variable	intercept	min 95%	max 95%	slope	min 95%	max 95%	r^2
DWCI	degree days	1.075	1.038	1.111	-0.000741	-0.000914	-0.000567	0.717
RNA·DNA^{-1}	days	0.948	0.878	1.018	-0.012423	-0.016625	-0.008222	0.655
RNA·DNA^{-1}	degree days	0.981	0.905	1.057	-0.001129	-0.001529	-0.000729	0.554

Estimating starvation time

Based on the DWCI the median time of starvation in the field increased from 17 days in November to 48 days in March. There the calculated days of starvation peak and 25% of the population starved up to 17 days, 50% up to 48 days, 75% up to 76 days and animals with the lowest condition up to 134 days (Figure 6-12). In November and December 25% of the population starved more than 21 and 24 days and in summer more than 5 days.

Estimates based on RNA·DNA^{-1} (Figure 6-13) revealed starvation periods for 25% of the population of more than 2-7 days during summer (May - Sept.). In November and December 50% of the population starved more than 7 and 19 days, respectively. In February all measured RNA-DNA-ratios were below 0.93 indicating that the whole population starved at that point of time.

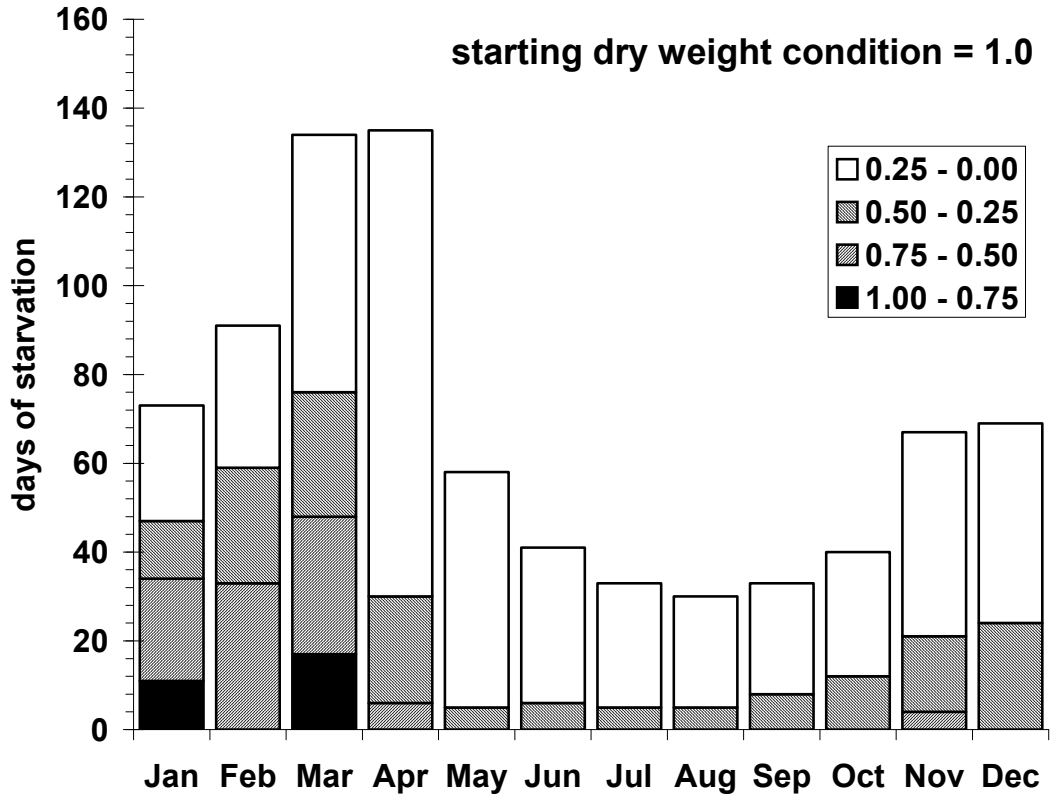

Figure 6-12: Calculated amount of days that *Crangon crangon* has starved in the field based on the dry weight condition index starting with a value of 1.

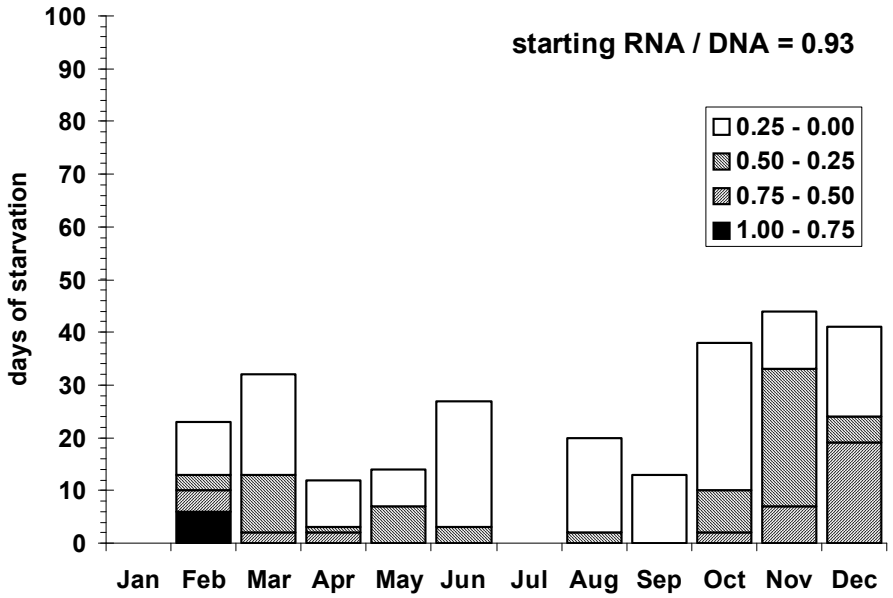

Figure 6-13: Calculated amount of days that *Crangon crangon* has starved in the field based on the RNA·DNA^{-1} starting with a ratio of 0.93.

Discussion

Starvation experiments

Dry weight and therefore also DWCI constantly declined in the starvation experiments. Body mass decreased with increasing temperature and varied between 0.9 and 1.7% per day. This value is lower than that of Carcinus maenas (2.88 %/d) (Dawirs 1983) which inhabits the same habitat, but the determined k values corresponded to values derived from field data. Tiews & Schumacher (1989) determined k of 1.12 and Kuipers & Dapper a k of 0.95. A recent combination of laboratory analysis and literature research on growth of *C. crangon* (Hufnagl & Temming, 2009) delivered $k = 9.659 \cdot 10^{-4} \cdot \exp^{0.09509 \cdot T}$ fitting the observations of the actual starvation experiments of the 30 and 45 mm length classes ($r^2 = 0.775$, $p=0.004$, Figure 6-9). This suggests that starvation experiments might be a suitable method to determine von Bertalanffy growth parameters for species where cohort tracking is complicated.

Parallel to the dry weight decrease a significant decrease of the RNA·DNA^{-1} was observed. Starvation influences the RNA·DNA^{-1} in two ways: before complete cell degradation and therefore a loss of DNA the cell shrinks due to depletion of resources. Thus the amount of DNA per unit dry weight increases. Also, RNA concentration is reduced during starvation as several proteins are not used and therefore RNA per unit dry weight decreases. As a net result RNA·DNA^{-1} decreases over time. A decrease of RNA in the cell is generally an early signal of cell death (del Prete et al. 2003). RNA·DNA^{-1} was in the present study work determined for muscle samples and muscle protein is most probably the major energy source during starvation (see below). Due to the fast response of the RNA·DNA^{-1} to starvation and due to the linear decrease during starvation it can be

concluded that RNA·DNA^{-1} is a good proxy for starvation and condition in *C. crangon*.

During the first 5 days of starvation of *C. Crangon*, Regnault (1981) observed that oxygen consumption remained on a high level and decreased significantly thereafter. This is comparable to the patterns observed in our experiments, where during the first days RNA·DNA^{-1} remained fairly constant and decreased thereafter. Such a response time of RNA·DNA^{-1} to starvation was also observed in sole *Solea solea* (Richard et al. 1991) and summer flounder *Paralichthys dentatus* (Malloy & Targett 1994). Furthermore, a reduction of metabolism under unfavorable conditions is not only described for *C. crangon* but also for other crustaceans (Tarrant et al. 2008).

Field observations

Protein, ash free dry weight, ash and water content

In this work the protein content of *C. crangon* was determined to be 46% of dry weight or 12 % of wet weight. These values are in the same order of magnitude as reported by Regnault (1981), who found a water content of 72-73% and protein content of 42-63% of dry weight. Our value is also comparable to other crustaceans: for *Penaeus esculentus* 1-2% lipids, 13% protein and 71-74 % water content were determined (Barcley et al. 1983) and Childress & Nygaard (1973) found a lipid content of 3-6%, a protein content of 9-12% and a water content of 77-78% in three different Penaeid-species. A low lipid content and the high protein levels suggest a use of protein as a major energy source, what could be demonstrated for *C. crangon* and other crustaceans under food deprivation (Sánchez-Paz et al. 2007, Dall & Smith 1986, Clifford & Brick 1983, Barcley et al. 1983, Regnault 1981, New 1976). The tissue containing the bulk of protein in *C. crangon* is the tail muscle. Due to its large size losses in muscle degradation are visible in dry weight losses and it can be expected that DWCI is a useful measure of *C. crangon* condition.

Abundance and population structure

The highest abundance was observed in Büsum and Wilhelmshaven in late spring and early summer, May and June. The high values are due to the immigration of new recruits entering the shallow areas of the Wadden Sea. The generally low abundance in Büsum in 2007 and the late increase was presumably caused by predation on juvenile *C. crangon* by 0-group whiting (Singh-Renton & Bromley 1999) that invaded the Wadden Sea in large numbers in May (unpublished data) (Hislop et al. 1991).

Shrimp density in Büsum was slightly positively correlated with DWCI ($r^2 = 0.275$), whereas no correlation was found between RNA·DNA^{-1} or the calorific content and shrimp density. In Wilhelmshaven no correlation between the caloric content or DWCI with density could be determined. This indicates that higher densities do not

lead to a decrease of actual condition of the shrimp. On the contrary, the immigration wave seems to be well adjusted in time of maximum production, so that the recruitment wave is not food limited.

Until April the DWCI of the shrimps was low and increased from then on rapidly. These changes might be due to increased feeding conditions (next section) or due to a cohort change. The latter might point to worse condition of the overwintering shrimps in comparison to the recruits of the actual year. In a previous study (Hufnagl & Temming, 2009) shrimps from the cohort caught in May (most probably overwintering shrimps) were found not to be able to generate maximum growth, unlike similarly-sized animals caught in July after the new recruits invaded. This indicates that even under ad libitum feeding growth performance of previously starved animals is low.

Dry weight, $RNA \cdot DNA^{-1}$ and caloric content in field data

DWCI and $RNA \cdot DNA^{-1}$ followed a seasonal pattern: low in winter and increasing towards summer. For other species in temperature-influenced areas the same seasonal patterns were observed (Norkko et al. 2006, Tardif et al. 2005, Rosa & Nunes 2004, Nicol et al. 2000) and in all cases food and temperature were the main determinants. In our study temperature correlated significantly with field temperature ($RNA \cdot DNA^{-1}$: $r^2 = 0.677$, $p=0.000$; DWCI: $r^2 = 0.264$, $p =0.001$), but not with caloric content. Zooplankton abundance for the catch dates were not available, but although *C. crangon* feeds on a variety of food items, zooplankton and especially small crustaceans are preferred (Feller 2006, del Norte-Campos & Temming 1994, Plagmann 1939). The first phytoplankton bloom in the North Sea occurs usually in late April (Reid et al. 1990, Henderson & Holmes 1987) and zooplankton follows thereafter with increasing density and diversity from April on (Beaugrand et al. 2001, Colebrook 1979). This is also the time when $RNA \cdot DNA^{-1}$ and DWCI start to increase. It can thus be assumed that the correlation of temperature and condition is an indirect correlation mainly determined by available food.

Within the starvation experiments a significant decrease of $RNA \cdot DNA^{-1}$ was observed, but the seasonal pattern was not as distinct as the DWCI pattern. The reason for that might be the quick change of RNA/DNA due to refeeding events (discussed below). Food may still be available in winter, but not in amounts sufficient to allow for weight growth, what may cause lower DWCI but increase the $RNA \cdot DNA^{-1}$. Yet *C. crangon* is an opportunistic omnivor and several food sources are utilized, and even the available caloric content of detritus, which can be found in the stomach of *C. crangon,* is within the same order of magnitude as the remaining food (Hanson 1982). The response time of $RNA \cdot DNA^{-1}$ to feeding was determined to be short: Malloy & Targett (1994) determined an increase in previously starved flounder white muscle $RNA \cdot DNA^{-1}$ after one day of refeeding. A rapid $RNA \cdot DNA^{-1}$ increase after refeeding was also observe in the blue crab *Callinectes sapidus* (Wang & Stickle 1986). Furthermore Regnault (1981) revealed that metabolism remains at a high level during the first 5 days of starvation

coinciding with the ratios observed in the present study. If the shrimps feeds during that time span RNA·DNA^{-1} is likely to remain at that level even longer.

This suggests that RNA·DNA^{-1} is a reliable measure of the short time feeding history only. The DWCI on the opposite is a proxy for net weight gain and loss independent from previously ingested small meals. If the ingested amount of food is below a certain threshold no weight gain is possible and DWCI decreases..

The caloric content is high in May and June (16800 J·g^{-1}) and low in October (14800 J·g^{-1}). Over the winter an increase of the caloric content was observed. The DWCI maximum values (>1.2) were observed in June, July. Over the winter a steady decrease until March was observed. The main part of the dry weight of *C. crangon* is made up by the tail muscle and therefore protein. Lipid reserves are mainly concentrated in hepatopancreas and are depleted within a few days (Regnault 1981) during starvation, comparable to other crustaceans (Muhlia-Almazán & García-Carreño 2008, Sánchez-Paz et al. 2007). The DWCI relates to body weight, and therefore mainly to muscle weight. The caloric content on the other hand was determined for pooled samples. Caloric content thus integrates the energetic composition of all tissues in the shrimp. As storage lipids make up only a small fraction of the whole body weight it is likely that the caloric content is determined by other energy rich components such as ripening gonads (not eggs, as these were removed prior to drying). This would also explain the low values determined in September and October as during this time vitellogenesis is suppressed (Klek-Kawinska & Bomirski 1975) and no egg carrying females can be observed in the field (Neudecker & Damm 1992, Kuipers & Dapper 1984). In spring, when the amount of egg bearing shrimps is high, the caloric content is also high. Moreover, the ovary development is linked to hepatopancreas weight and hepatopancreas weight decreases during vitellogenesis (Haefner & Spaargaren 1994, Haefner & Spaargaren 1993). This might indicate that the caloric content is mainly influenced by the developing ovaries of the females (at least during winter).

Is *Crangon crangon* food limited in the Wadden Sea?

The calculated amount of starving days based on the DWCI suggest 50% of the population starved more than 80, 60 and 40 days in March, February and January, respectively. This indicates that there is not enough food available to build up body mass during the whole winter. Furthermore, both indices used to calculate starvation time, dry weight and RNA·DNA^{-1}, predict that food deprivation already starts in October, November. The RNA·DNA^{-1} values determined in October, November and December suggest a proportion of 25% of *C. crangon* that did not feed for more than 10, 33 and 24 days, respectively comparable to DWCI based results of 12, 21 and 24.

From January to March RNA·DNA^{-1} values predict shorter starvation periods than the DWCI, either due to the influence of egg release on dry weight or the short response time of RNA·DNA^{-1} to feeding events.

In Wilhelmshaven in February 5% of all female shrimps >35 mm (mean 2005 - 2007) carried eggs and numbers increased to 36 and 35 % in April and May. Siegel et al (2008) and Boddeke (1982) observed increasing numbers of egg-carrying females from December on. That the percentage of berried females was higher in their catches is due to the lower water depth an therefore smaller shrimps in our catches. The fraction of berried females in the present study with poor DWCI (< 0.8) was 30% (of all animals) and comparable to the fraction of females with good condition (28.9%). This indicates that dry weight loss due to egg release cannot be the sole factor for differences in the calculated starvation time. Densities in our catches were low and the population migrates further offshore during winter (up to 90 km according to Boddeke 1976). Our samples might therefore represent animals that are in bad condition as they stayed close to the shore.

Anyhow even if food limitation is present it might not necessarily lead to a significant reduction of the population. More than 75% of the field-caught animals had a higher DWCI and nearly 100% had higher $RNA \cdot DNA^{-1}$ than those animals analyzed at the end of the starvation experiments. As mortality in the starvation experiments was low and did not rise towards the end of the experiment it can be assumed that values can even decrease further before critical thresholds are reached. The survival of more than 30 days without food and the reduction of the metabolism during starvation (Regnault 1981) exemplifies the high adaptation potential to food limitations.

Influence of food limitation on growth and survival

The results suggest that *C. crangon* is adapted to low food concentration and although growth will be reduced it might still be possible. Regnault & Lagadère (1983) found that only those *C. crangon* moulted that are in the premoult phase. In our starvation experiments 75 of 500 animals moulted and 10% of these grew with increments of 3 and 4 mm but more than the half (53%) did not grow (increments of 0 mm). 60% moulted within the first 5 days and 89% within the first 10 days. Estimated days of starvation suggest that within each month 25% of the animals starved more than 10 days and it can be assumed that these animals do not moult and therefore do not grow. In November and December (based on $RNA \cdot DNA^{-1}$) and from January to February (based on DWCI) 75% starved more than 10 days, indicating that during winter growth is reduced or even stops completely. A growth reduction due to food limitation was already suggested by Amara & Paul (2003) for the French coast of the Eastern Channel. Besides a density control of the abundance (Henderson et al. 2006) and also bottom up control (Boddeke 1996) was suggested earlier.

Survival chances of juvenile shrimps are tightly coupled to growth performance. As in most marine organisms, cumulative mortality will be higher for slow growing shrimps (Cowan et al. 1996, Houde 1987). Likewise, high food concentrations in summer potentially accelerates growth and, in consequence, may result in higher rates of survival to the adult stage. Via this mechanism, food limitation might not

only influence growth but also the density of individuals. Furthermore it underlines the importance of the winter cohort for the population dynamic of *C. crangon*. The juveniles that invade into the Wadden Sea in spring most probably hatched from winter eggs (Temming & Damm 2002). Timing of the invasion of these shrimps fits the timing of zooplankton maximum and during summer food limitation is low. Those animals that hatch from summer eggs only profit from high food concentration for a short time as our results suggest that from October on resources are sparse. Growth performance will therefore be reduced and in conjunction with low temperature these shrimps will not grow for nearly six month. As these shrimps are smaller than those animals hatched from winter eggs the number of survivors of this cohort will be lower.

Conclusions

Dry weight condition reflects long term feeding history and net weight gain

$RNA \cdot DNA^{-1}$ represents short term feeding history

Caloric content most probably reflects ovary ripening and development

Crangon crangon is most probably food limited during winter and the most critical month is, due to increasing temperatures, March

25% of the population is food limited during the whole year.

Growth of the population will mainly occur between April and September what favors the winter egg hatched animals.

Acknowledgements

We wish to thank the employees and support staff at the power plant in Wilhelmshaven for support with the cooling water sampling. The study was financially supported by the Federal Ministry of Food, Agriculture and Consumer Protection, Germany (Project No. 03HS030) and Niedersächsische Wattenmeerstiftung, Germany (Project No. 53-NWS-41/04).

References

Amara R, Paul C (2003) Seasonal pattern in the fish and epibenthic crustaceans community of an intertidal zone with particular reference o the population dynamics of plaice and brown shrimp. Estuarine, Coastal and Shelf Science 56:807-818

Barcley MC, Dall W, Smith DM (1983) Changes in lipid and protein during starvation and the moulting cycle in the tiger prawn *Penaeus esculentus* Haswell. Journal of Experimental Biology and Ecology 68:229-244

Beaugrand G, Ibanez F, Lindley JA (2001) Geographical distribution and seasonal and diel changes in the diversity of calanoid copepods in the North Atlantic and North Sea. Marine Ecology Progress Series 219:189-203

Beukema JJ (1992) Dynamics of juvenile shrimp *Crangon crangon* in a tidal flat-nursery of the Wadden Sea after mild and cold winters. Marine Ecology Progress Series 83:157-165

Boddeke R (1996) Changes in the brown shrimp (*Crangon crangon* L.) population off the Dutch coast in relation to fisheries and phosphate discharge. ICES Journal of Marine Science 53:995-1002

Boddeke R, Driessen G, Doesburg W, Ramaekers G (1986) Food availability and predator presence in a costal nursery area of the brown Shrimp (*Crangon crangon*). Ophelia 26:77-90

Bolger T, Connely PL (1989) The selection of suitable indices for the measurement and analysis of fish condition. Journal of Fish Biology 34:171-182

Buckley LJ (1984) RNA-DNA ratio. an index of larval fish growth in the sea. Marine Biology 80:291-298

Bunt JS (1973) Primary Production: Marine Ecosystems. Human Ecology 1:333-345

Caldarone EM, Buckley LJ (1991) Quantitation of DNA and RNA in crude tissue extracts by Flow Injection Analysis. Analytical Biochemistry 199:137-141

Caldarone EM, Clemmesen CM, Berdalet E, Miller TJ, Folkvord A, Holt GJ, Olivar MP, Suthers IM (2006) Intercalibration of four spectrofluorometric protocols for measuring RNA/DNA ratios in larval and juvenile fish. Limnology and Oceanography: Methods 4:153-163

Caldarone, Elaine M., Wagner, Melissa, St.Onge-Burns, Jeanne, and Buckley, Lawrence J. Protocol and Guide for Estimating Nucleic Acids in Larval Fish Using a Fluorescence Microplate Reader. Northeast Fisheries Science Center Reference Document 01-11, -28. 2001. Woods Hole, Massachusetts, National Oceanic and Atmospheric Administration, National Marine Fisheries Service.

Clifford HC, Brick RW (1983) Nutritional physiology of the freshwater shrimp *Macrobrachium rosenbergii* (De Man) - I. Substrate metabolism in fasting juvenile shrimp. Comparative Biochemistry and Physiology 74A:561-568

Colebrook JM (1979) Continuous Plankton Records: Seasonal cycles of phytoplankton and copepods in the North Atlantic Ocean and the North Sea. Marine Biology 51:23-32

Cowan JH, Houde ED, Rose KA (1996) Size-dependent vulnerability of marine fish larvae to predation: An individual-based numerical experiment. ICES Journal of Marine Science 53:23-37

Dall W, Smith DM (1986) Oxygen consumption and ammonia-N excretion in fed and starved tiger prawns, *Penaeus esculentus* Haswell. Aquaculture 55:23-33

Dawirs RR (1983) Respiration, energy balance and development during growth and starvation of *Carcinus maenas* L. larvae (Decapoda, Portunidae). Journal of Experimental Biology and Ecology 69:105-128

de Jonge VN (1997) High remaining productivity in the Dutch Wadden Sea despite decreasing nutrient inputs from riverine sources. Marine Pollution Bulletin 34:427-436

del Norte-Campos AGC, Temming A (1994) Daily activity, feeding and rations in gobies and brown shrimp in the northern Wadden Sea. Marine Ecology Progress Series 115:41-53

del Prete MJ, Robles AS, Guio A, Martínez-A. C, Izquierdo M, Garcia-Sanz JA (2003) Degradation of cellular mRNA is a general early apoptosis-induced event. The FASEB Journal 16:2003-2005

Evans S (1984) Energy budgets and predation impact of dominant epibenthic carnivores on a shallow soft bottom community at the Swedish west coast. Estuarine, Coastal and Shelf Science 18:651-672

Feller RJ (2006) Weak meiofaunal trophic linkages in *Crangon crangon* and *Cracinus maenas*. Journal of Experimental Marine Biology and Ecology 330:274-283

Field CB, Behrenfeld MJ, Randerson JT, Falkowski P (1998) Primary production of the biosphere: Integrating terrestrial and oceanic components. Science 281:237-240

Gamito R, Cabral H (2003) Mortality of brown-shrimp discards from the beam trawl fishery in the Tagus estuary, Portugal. Fisheries Research 63:423-427

Gunnarsson B, Ásgeirsson þ, Ingólfsson A (2007) The rapid colonization by *Crangon crangon* (Linnaeus, 1758) (Eucarida, Caridea, Crangonidae) of Icelandic coastal waters. Crustaceana 80:747-753

Haefner PA, Spaargaren DH (1993) Interactions of ovary and hepatopancreas during the reproductive cycle of *Crangon crangon* (L.). I. Weight and volume relationships. Journal of Crustacean Biology 13:523-531

Haefner PA, Spaargaren DH (1994) Interactions of ovary and hepatopancreas during the reproductive cycle of *Crangon crangon* (L.). II: Biochemical relationships. Journal of Crustacean Biology 14:6-19

Hamerlynck O, Hostens K (1993) Growth, feeding, production, and consumption in 0-group bib (*Trisopterus luscus* L.) and whiting (*Merlangius merlangus* L.) in a shallow coastal area of the south-west Netherlands. ICES Journal of Marine Science 50:81-91

Hanson RB (1982) Organic Nitrogen and Caloric Content of Detritus. Estuarine, Coastal and Shelf Science 14:325-336

Henderson PA, Holmes RHA (1987) On the population biology of the common shrimp *Crangon crangon* (L.) (Crustacea: Caridea) in the Severn Estuary and Bristol Channel. Journal of the Marine Biological Association U.K. 67:825-847

Henderson PA, Seaby RMH, Somes JR (2006) A 25-year study of climatic and density-dependent population regulation of common shrimp *Crangon crangon* (Crustacea: Caridea) in the Bristol Channel. Journal of the Marine Biological Association U.K. 86:287-298

Hislop JRG, Robb AP, Bell MA, Armstrong DW (1991) The diet and food composition of whiting (*Merlangius merlangus*) in the North Sea. ICES Journal of Marine Science 48:139-156

Houde ED (1987) Fish Early Life Dynamics and Recruitment Variability. Am.Fish.Soc.Symp. 2:17-29

ICES. Report of the *Crangon* Fisheries and life history. ICES CM 2006/G:XX, 1-32. 2006.

Janssen GM, Kuipers BR (1980) On tidal migration in the shrimp *Crangon crangon*. Netherlands Journal of Sea Research 14:339-348

Klek-Kawinska E, Bomirski A (1975) Ovary-inhibiting hormone activity in Shrimp (*Crangon crangon*) eyestalks during the annual reproductive cycle. General and Comparative Endocrinology 25:9-13

Kuipers BR, Dapper R (1981) Production of *Crangon crangon* in the tidal zone of the Dutch Wadden Sea. Netherlands Journal of Sea Research 15:33-53

Kuipers BR, Dapper R (1984) Nursery function of Wadden Sea tidal flats for the brown shrimp *Crangon crangon*. Marine Ecology Progress Series 17:171-181

Malloy KD, Targett TE (1994) The use of RNA:DNA ratios to predict growth limitations of juvenile summer flounder (*Paralichthys dentatus)* from Delaware North Carolina estuaries. Marine Biology 118:367-375

Mathers EM, Houlihan DF, Buren LJ (1994) RNA, DNA and protein concentrations in fed and starved herring *Clupea harengus* larvae. Marine Ecology Progress Series 107:223-231

Moss SM (1994a) Growth rates, nucleic acid concentrations, and RNA/DNA ratios of juvenile white shrimp, *Penaeus vannamei* boone, fed different algal diets. Journal of Experimental Marine Biology and Ecology 182:193-204

Moss SM (1994b) Use of nucleic acids as indicators of growth in juvenile white shrimp, *Penaeus vannamei*. Marine Biology 120:359-367

Muhlia-Almazán A, García-Carreño FL (2008) Influence of moulting and starvation on the synthesis of proteolytic enzymes in the midgut gland of the white shrimp *Penaeus vannamei*. Comparative Biochemistry and Physiology Part B 133:383-394

Neudecker, T. and Damm, U. Seasonality of egg-bearing shrimp (*Crangon crangon* L.) in costal waters of the German Bight. K: 28, 1-9. 1992. ICES Shell Fish Committee.

Neves A, Cabral HN, Gordo LS (2007) Distribution and abundance patterns of decapod crustaceans in the sado estuary, Portugal. Crustaceana 80:97-112

New MB (1976) A review of dietry studies with shrimp and prawns. Aquaculture 9:101-144

Nicol S, Kitchener J, King R, Hosie G, de la Mare WK (2000) Population structure and condition of Antarctic krill (*Euphausia superba*) off East Antarctica (80-150°E) during the Austral summer of 1995/1996. Deep-Sea Research II 47:2489-2517

Norkko J, Thrush SF, Wells RMG (2006) Indicators of short-term growth in bivalves: Detecting environmental change across ecological scale. Journal of Experimental Biology and Ecology 337:38-48

Peck MA, Buckley LJ, Caldarone EM, Bengtson DA (2003) Effects of food consumption and temperature on growth rate and biochemical-based indicators of growth in early juvenile Atlantic cod *Gadus morhua* and haddock *Melanogrammus aeglefinus.* Marine Ecology Progress Series 251:233-243

Pihl L, Rosenberg R (1984) Food selection and consumption of the shrimp *Crangon crangon* in some shallow marine areas in western Sweden. Marine Ecology Progress Series 15:159-168

Plagmann J (1939) Ernährungsbiologie der Garnele (*Crangon vulgaris* Fabr.). Helgoländer wissenschaftliche Meeresuntersuchungen 2:113-162

Regnault M (1981) Respiration and Ammonia Excretion of the Shrimp *Crangon crangon L.:* Metabolic Response to Prolonged Starvation. Journal of Comparative Physiology 141:549-555

Regnault M, Lagardère JP (1983) Effects of ambient noise on the metabolic level of *Crangon crangon* (Decapoda, Natantia). Marine Ecology Progress Series 11:71-78

Reid PC, Lancelot C, Gieskes WWC, Hagmeier E, Weichart G (1990) Phytoplankton of the North Sea and its dynamics. Netherlands Journal of Sea Research 26:295-331

Richard P, Bergeron J-P, Boulhic M, Galois R, Person-Le Ruyet J (1991) Effect of starvation on RNA, DNA and protein content of laboratory-reared larvae and juveniles of *Solea solea*. Marine Ecology Progress Series 72:69-77

Rosa R, Nunes ML (2004) RNA, DNA and protein concentrations and amino acid profiles of deep-sea decapod *Aristeus antennatus*: An indication for seasonal variations of nutrition and growth. Aquatic Living Resources 17:25-30

Sánchez-Paz A, García-Carreño FL, Hernández-López J, Muhlia-Almazán A, Yepiz-Plascencia G (2007) Effect of short-term starvation on hepatopancreas and plasma energy reserves of the Pacific white shrimp (*Litopenaeus vannamei*). Journal of Experimental Biology and Ecology 340:184-193

Siegel V, Gröger J, Neudecker T, Damm U, Jansen J (2005) Long-term variation in the abundance of the brown shrimp *Crangon crangon* (L.) population of the German Bight and possible causes for its interannual variability. Fisheries Oceanography 14:1-16

Singh-Renton S, Bromley PJ (1999) Feeding of small whiting (*Merlangius merlangus*) in the central and southern North Sea. Journal of the Marine Biological Association U.K. 79:957-960

Smith TR (2003) RNA-DNA ratio in scales from juvenile cod provides a nonlethal measure of feeding condition. Transactions of American Fisheries Society 132:9-17

Tardif D, Glémet H, Brodeur P, Mingelbier M (2005) RNA/DNA ratio and total length of yellow perch (*Perca flavescens)* in managed and natural wetlands of a large fluvial lake. Canadian Journal Fisheries and Aquatic Science 62:2211-2218

Tarrant AM, Baumgartner MF, Verslycke T, Johnson CL (2008) Differential gene expression in diapausing and active *Calanus finmarchicus* (Copepoda). Marine Ecology Progress Series 355:193-207

Temming A, Damm U (2002) Life cycle of *Crangon crangon* in the North sea: A simulation of the timing of recruitment as a function of the seasonal temperature signal. Fisheries Oceanography 11:45-58

Tiews K (1954) Die biologischen Grundlagen der Büsumer Garnelenfischerei. Berichte der Deutschen Wissenschaftlichen Kommission für Meeresforschung 13:235-269

Wagner M, Durbin E, Buckley L (1998) RNA:DNA ratios as indicators of nutritional condition of the copepod *Calanus finmarchicus*. Marine Ecology Progress Series 162:173-181

Wang AY, Stickle WB (1986) Changes in nucleic acid concentration with starvation in the blue crab *Callinectes sapidus* Rathbun. Journal of Crustacean Biology 6:49-56

Welleman, H. C. and Storebeck, F. Annex 7: Quantifying the effects of infrastructural works on brown shrimp populations: a habitat modelling approach. CM 2002 IE:05, 45-52. 2002. Report of the Working Group on Marine Habitat Mapping.

Are RNA·DNA^{-1} and dry weight suitable growth proxies for *Crangon crangon* (L., Crustacea) ?

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Abstract

Dry weight-at-length (dry weight condition index, DWC) and RNA-DNA ratio (RD) were tested for the suitability to estimate length growth rates (mm·d-1) of brown shrimp, *Crangon crangon*. Male and female shrimps (20 to 60 mm) were reared at five different temperatures (5 to 25°C) and growth rate, RD and DWC were determined for each individual. Growth rate and DWC were not significantly correlated and a significant correlation between the former and RD was only detected at high temperatures. It was concluded that, while RD explains a portion of the variability in individual growth rates, RD is not a suitable growth proxy for C. crangon. The reason is most probably the effect of short-term variability of RD due to moulting, maturation and feeding intensity in comparison to the relatively long amount of time required to gain robust measures of somatic growth.

Key words: *Crangon crangon*; growth; dry weight; RNA/DNA

Introduction

The brown shrimp *Crangon crangon* is common in coastal areas around Europe (Gunnarsson et al. 2007, Lapinska & Szaniawska 2006, Cuesta et al. 2006, Bulnheim & Schwenzer 1993). The core distribution is the Wadden Sea, a unique area influenced by strong tidal currents and high daily and seasonal variability of temperature and salinity. This species of shrimp feeds on small crustaceans, polychaets, bivalves and zooplankton (Feller 2006, del Norte-Campos & Temming 1994, Plagmann 1939).

It is impossible to accurately determine the age of shrimps since all hard parts are lost during ecdysis (Hartnoll 2001) and, therefore, rates of growth must be determined during laboratory trials or, statistically by means of cohort tracking in the field. Previously determined growth rate estimates are highly variable, and range from 0.0 to 0.1 mm d^{-1} (Oh & Hartnoll 2000, Henderson & Holmes 1987), 0.2-0.54 mm d^{-1} (Beukema 1992) to 0.07-0.4 mm d^{-1} (Tetard 1985) and, at specific body sizes and temperatures, growth rate can range from nearly no growth (Edwards 1978) to 0.54 mm· d^{-1}(Beukema 1992). This high variability produces great uncertainties within age, growth and population analysis. Due to these uncertainties, the life cycle of *C. crangon* is not fully understood. If it would be possible to determine a proxy that facilitated the separation of slow- from fast-growing individuals in the field would significantly improve knowledge of the population dynamics in this species.

$RNA{\cdot}DNA^{-1}$ (RD) is one of the most commonly used indices for estimating growth rates in field individuals and, depending upon the species and life stage, can often be measured on single individuals. The amount of RNA in a cell is supposed to correlate with protein synthesis, and therefore with nutrition and growth, whereas the amount of DNA, as carrier of genetic information, is assumed to be relatively constant in the majority of cell types. RD has been shown to correlate with growth mainly for larval fish (Peck et al. 2003, Buckley et al. 1999, Hovenkamp & Witte 1991, Buckley 1984) but also for corals (Buckley & Szmant 2004), scallops (Lodeiros et al. 1996), anostraca and copepods (Wagner et al. 1998, Dagg & Littlepage 1972) and Crustaceans (Juinio & Cobb 1994, Moss 1994a, Moss 1994b). However in many studies no correlation was determined (Lee et al. 2006, Norkko et al. 2006, Mathers et al. 1994) clearly indicating that a calibration and a suitability determination of RD for each species has to be performed.

Another potential growth index addresses the condition of an animal is dry weight at length. In the past this index has mainly been used for bivalves (Lundebye et al. 1997, Nagasawa & Nagata 1992, Gabbott 1974) and fishes (Kurita et al. 2003, Kosolow et al. 1985, Ehrlich et al. 1976) and in a modified form for crustaceans (Jussila 1999, Dall 1974). Pérez-Castañeda & Defeo (2008) used this index to determine density depend variations of *Farfantepenaeus* species and for *Euphausia superba* the index is used to describe annual condition (Nicol et al. 2000). It is a relative index that shows the weight deviation of one specific animal of a certain length from the mean amount of similar sized individuals. After drying and weighing the animals, the index can easily and accurately be calculated.

Furthermore, it is a suitable index for crustaceans that have a relatively low lipid content (Pearson & Dutson 1997, Barcley et al. 1983) and mainly use protein or carbohydrates as energy sources (Dall & Smith 1986, Clifford & Brick 1983, New 1976). Protein has a higher dry weight than lipids and more protein has to be degraded for the same amount of energy. This results in more or less linear weight gain or loss due to good or bad conditions, respectively.

As previously mentioned, even under ad libitum feeding conditions, growth rates are highly variable and the reasons for this are unclear. Results of preliminary laboratory trials suggested that feeding rates of similar-sized *C. crangon* were different, even under ad libitum feeding conditions which could partly explain the marked growth variability among individuals. If there are differences in feeding history in the course of a growth trial, dry weight and $RNA \cdot DNA^{-1}$ should be lower for slower-growing animals. The aim of this study was, therefore, to determine whether either dry weight condition and/or RD, can be used as a growth proxy for *C. crangon* and whether either of these indices can explain the variability in growth observed for shrimp provided ad libitum prey resources.

Material and Methods

Growth experiments

Growth experiments were performed at 5, 10, 15, 20 and 25°C with shrimps of 20 to 60 mm total length. Conditions, preliminary handling, catch area and feeding were described in detail by Hufnagl & Temming (2009a in press). That study also described temperature- and length-dependent growth rate of *C. crangon*. Two groups of shrimps were examined, one caught in May and the other one caught in July. The latter cohort originated from new recruits whereas the May cohort was mainly composed of overwintering individuals. The group caught in May exhibited significantly lower growth rates than the July group. Therefore growth of these two cohorts were analyzed separately.

Dry weight condition

Dry weight condition index was calculated as

$$CI = \frac{dw}{a \cdot L^b} \qquad (14)$$

with *dw* = dry weight [g] and L = total length [mm]. The constants a = $1.3005 \cdot 10^{-6}$ and b = 3.062 were determined (using SYSTAT 6.0) from the pooled measurements of *C. crangon* caught in Büsum and Wilhelmshaven in 2005, 2006 and 2007 (n = 8305).

RNA·DNA^{-1} analysis

RNA-DNA ratio was determined following largely the procedure described in (Caldarone et al. 2001). Deviations from their method and accuracy of the analysis were described by Hufnagl et al. (2009b, submitted).

Correlation of RNA·DNA^{-1} and dry weight with growth

For all correlation analysis the software SPSS was used and corrected r^2 and p values were calculated. Females that carried eggs and shrimps <20 mm were not included in this analysis.

Data were analyzed separately for homogeneous subgroups (e.g. for each sex and size class and temperature combination growth rate was correlated with either DWC or RD.

Results

Dry weight condition index

No significant correlations were found between RD and GR when all data (all size classes and temperatures) were used in the analysis (Table 7-1, line 1). Reanalysis after dividing the data according to gender did not improve the correlation for female shrimps (r^2 = 0.101) and only slightly improved that (r^2 = 0.068) for male shrimps.

Separating the data according to gender and temperature (but not length) lead to significant correlations (r^2 = 0.564) at high temperatures (20 and 25°C, Table 7-1, lines 8 and 12, 13) but not at low temperatures ($T<15$, $p>0.05$). Further separation according to length delivered slight correlations with $r^2 > 0.15$ for: 40 mm males at 20°C (line 21), 50 mm males at 5 °C and 50 mm males at 10°C (line 23, 24). For female shrimps a correlation of the growth rates with RD could be observed for temperatures > 10°C with exception of 60 mm females at 20°C (line 39). In general comparable results are obtained when analyzing shrimps caught in May and July separately (Table 7-2 and Table 7-3).

RNA·DNA^{-1} ratio

No significant correlations were found between RD and GR when all data (all size classes and temperatures) were used in the analysis (Table 7-1, line 1). Reanalysis after dividing the data according to gender did not improve the correlation for female shrimps (r^2 = 0.101) and only slightly improved that (r^2 = 0.068) for male shrimps.

Table 7-1: Correlation coefficients (corrected r^2) for the regression of *Crangon crangon* growth rates with $RNA \cdot DNA^{-1}$ and the dry weight condition index (DWCI). Data were divided into subsets according to gender, temperature and length and contain data of all trials (marked and unmarked). Bold marked are subclasses showing significant correlation ($p < 0.05$)

	gender	**temperature**	**length**		**RNA/DNA**			**DWCI**		
line	**includes**	**includes**	**includes**	**n**	**r^2**	**F**	**p**	**r^2**	**F**	**p**
1	all	all	all	522	0.076	43.664	0.000	0.067	37.439	0.000
2	male	all	all	163	0.101	19.231	0.000	0.235	50.937	0.000
3	female	all	all	358	0.068	26.932	0.000	0.029	11.631	0.001
4	male	5	all	29	-0.006	0.839	0.368	-0.030	0.159	0.694
5	male	10	all	36	-0.021	0.247	0.623	0.043	2.616	0.115
6	male	15	all	44	0.024	2.102	0.154	-0.010	0.544	0.465
7	male	20	all	34	0.196	9.304	0.004	0.094	4.547	0.041
8	male	25	all	16	0.045	1.751	0.206	0.357	9.888	0.007
9	female	5	all	70	-0.014	0.008	0.930	-0.013	0.102	0.751
10	female	10	all	86	0.045	5.087	0.027	-0.010	0.109	0.742
11	female	15	all	82	0.042	4.639	0.034	-0.008	0.313	0.577
12	female	20	all	79	0.224	23.829	0.000	0.021	2.655	0.107
13	female	25	all	37	0.564	48.907	0.000	0.018	1.671	0.204
14	male	5	30	10	0.024	1.241	0.294	0.171	3.058	0.114
15	male	10	30	3	-0.203	0.493	0.555	0.632	6.144	0.131
16	male	15	30	19	-0.055	0.008	0.930	0.074	2.521	0.130
17	male	20	30	10	0.081	1.971	0.191	0.012	1.135	0.312
18	male	5	40	6	0.088	1.581	0.264	0.054	1.344	0.299
19	male	10	40	20	-0.035	0.315	0.581	0.037	1.772	0.199
20	male	15	40	23	0.088	3.229	0.086	-0.042	0.083	0.776
21	male	20	40	22	0.330	11.840	0.002	0.103	3.526	0.074
22	male	25	40	13	0.065	1.899	0.193	0.334	7.509	0.018
23	male	5	50	11	0.240	4.467	0.061	0.297	5.653	0.039
24	male	10	50	11	0.191	3.598	0.087	-0.049	0.448	0.501
25	male	15	50	11	0.081					
26	female	5	30	18	-0.057	0.022	0.885	-0.050	0.145	0.708
27	female	15	30	8	-0.123	0.126	0.733	-0.082	0.391	0.552
28	female	5	40	13	-0.050	0.383	0.548	0.171	3.676	0.079
29	female	10	40	17	0.021	1.357	0.261	0.020	1.344	0.263
30	female	15	40	50	0.305	22.967	0.000	-0.019	0.071	0.791
31	female	20	40	43	0.150	8.575	0.005	0.111	6.389	0.015
32	female	25	40	24	0.550	30.298	0.000	0.024	1.597	0.219
33	female	10	50	41	0.079	4.505	0.040	-0.140	0.433	0.514
34	female	15	50	11	0.181	3.424	0.094	-0.078	0.202	0.663
35	female	20	50	24	0.171	5.941	0.023	0.061	2.547	0.124
36	female	25	50	11	0.534	13.626	0.004	-0.710	0.268	0.616
37	female	10	60	25	0.041	2.064	0.164	-0.340	0.168	0.686
38	female	15	60	10	0.140	2.629	0.139	0.020	1.205	0.301
39	female	20	60	9	-0.022	0.803	0.396	-0.020	0.824	0.390

Table 7-2: Correlation coefficients (corrected r²) for the regression of *Crangon crangon* growth rates with RNA·DNA^{-1} and the dry weight condition index (DWC). Data were divided into subsets according to gender, temperature and length and contain only data of the trials with the_ animals caught in July. Bold marked are subclasses showing significant correlation ($p < 0.05$)

gender	temperature	length		RNA/DNA			DWCI		
			n	r²	F	p	r²	F	p
all	all	all	202	0.035	8.345	0.004	0.036	8.569	0.004
male	all	all	84	0.112	11.673	0.001	0.185	20.350	0.000
female	all	all	116	0.043	6.240	0.014	0.008	0.051	0.822
male	10	all	13	-0.008	0.884	0.364	0.379	9.534	0.009
male	15	all	36	0.012	1.463	0.234	-0.027	0.033	0.860
male	20	all	29	0.074	3.311	0.080	0.077	3.411	0.075
female	10	all	8	-0.101	0.176	0.686	-0.066	0.443	0.524
female	15	all	44	-0.013	0.411	0.525	0.059	3.797	0.058
female	20	all	46	0.065	4.287	0.044	-0.022	0.007	0.935
female	25	all	12	0.345	7.836	0.016	0.506	14.309	0.003
male	10	30	2	-0.203	0.493	0.555	0.632	6.144	0.131
male	15	30	18	-0.055	0.008	0.930	0.074	2.521	0.130
male	20	30	10	0.081	1.971	0.191	0.012	1.135	0.312
male	10	40	9	0.012	1.122	0.317	-0.109	0.018	0.896
male	15	40	16	0.400	12.346	0.003	0.118	3.282	0.089
male	20	40	16	0.181	4.758	0.044	0.170	3.932	0.065
male	25	40	1	0.993	306.124	0.036	-0.961	0.200	0.911
female	15	30	7	-0.123	0.126	0.733	-0.082	0.391	0.552
female	10	40	6	0.044	1.322	0.294	0.497	7.919	0.031
female	15	40	34	0.069	3.596	0.066	0.027	1.975	0.169
female	20	40	36	0.004	1.145	0.292	-0.016	0.416	0.523
female	25	40	10	0.191	3.596	0.087	0.670	23.290	0.001
female	20	50	7	0.436	5.400	0.053	-0.050	0.617	0.458

Separating the data according to gender and temperature (but not length) lead to significant correlations ($r^2 = 0.564$) at high temperatures (20 and 25°C, Table 7-1, lines 8 and 12, 13) but not at low temperatures ($T<15$, $p>0.05$). Further separation according to length delivered slight correlations with $r^2 > 0.15$ for: 40 mm males at 20°C (line 21), 50 mm males at 5 °C and 50 mm males at 10°C (line 23, 24). For female shrimps a correlation of the growth rates with RD could be observed for temperatures > 10°C with exception of 60 mm females at 20°C (line 39). In general comparable results are obtained when analyzing shrimps caught in May and July separately (Table 7-2 and Table 7-3).

Table 7-3: Correlation coefficients (corrected r²) for the regression of *Crangon crangon* growth rates with RNA·DNA^{-1} and the dry weight condition index (DWC). Data were divided into subsets according to gender, temperature and length and contain only data of the trials with the animals caught in May. Bold marked are subclasses showing significant correlation ($p < 0.05$)

gender	temperature	length		RNA/DNA			DWCI		
with marking			n	r²	F	p	r²	F	p
all	all	all	317	0.021	7.700	0.006	0.053	18.813	0.000
male	all	all	76	0.062	6.052	0.016	0.151	14.684	0.000
female	all	all	239	0.009	3.127	0.078	0.030	8.322	0.004
male	5	all	28	-0.006	0.839	0.368	-0.030	0.159	0.694
male	10	all	20	-0.028	0.420	0.524	-0.030	0.393	0.538
male	15	all	5	0.228	2.768	0.157	0.192	1.188	0.325
male	20	all	3	0.902	37.763	0.009	0.846	23.014	0.017
male	25	all	12	-0.083	0.003	0.955	0.056	1.775	0.207
female	5	all	69	-0.014	0.008	0.930	-0.013	0.102	0.751
female	10	all	75	0.117	11.063	0.001	-0.013	0.023	0.879
female	15	all	35	-0.028	0.011	0.915	-0.008	0.708	0.406
female	20	all	30	0.030	1.960	0.172	0.170	7.331	0.011
female	25	all	22	-0.045	0.004	0.948	0.085	3.150	0.090
male	5	30	9	0.121	1.241	0.294	0.171	3.058	0.114
male	5	40	5	0.088	1.581	0.264	0.054	1.344	0.299
male	10	40	8	-0.080	0.336	0.578	0.107	2.084	0.187
male	15	40	4	-0.144	0.370	0.576	-0.176	0.253	0.642
male	20	40	3	0.902	37.763	0.009	0.846	23.014	0.017
male	25	40	9	-0.110	0.006	0.939	-0.020	0.807	0.392
male	5	50	10	0.240	4.467	0.061	0.297	5.653	0.039
male	10	50	10	0.191	3.598	0.087	-0.049	0.488	0.501
female	5	30	17	-0.057	0.022	0.885	-0.050	0.145	0.708
female	5	40	12	-0.050	0.383	0.548	0.171	3.676	0.079
female	10	40	8	0.127	2.306	0.167	-0.099	0.190	0.675
female	15	40	13	0.038	1.557	0.234	0.032	0.435	0.521
female	20	40	4	0.192	2.185	0.213	0.766	17.408	0.014
female	25	40	11	-0.053	0.401	0.539	-0.081	0.102	0.755
female	10	50	39	0.134	6.057	0.018	-0.023	0.083	0.775
female	15	50	9	-0.091	0.164	0.695	-0.110	0.006	0.938
female	20	50	14	0.304	7.540	0.016	0.000	0.997	0.335
female	25	50	8	-0.111	0.097	0.763	0.156	2.668	0.141
female	10	60	24	0.041	2.064	0.164	-0.034	0.168	0.686
female	15	60	9	0.140	2.629	0.139	0.020	1.205	0.301
female	20	60	8	-0.022	0.803	0.396	-0.020	0.824	0.390

Variability of RNA·DNA^{-1} and dry weight condition for similar sized animals

Within the present experiment, the variability of RD and DWC for animals of the same length, gender and with the same growth rate was high (Figure 7-1). For example, RD was between 0.6 and 1.7 and condition between 1.1 and 1.6 for 31 mm males having identical growth rates (0.39 mm d^{-1}.) at 15°C. The observed coefficients of variation were between 3.6% (male, 35 mm, 20°C) and 16.7%

(female 39 mm 20°C) for DWC and between 14.4% (female 39 mm 20°C) and 35% (male, 31 mm, 15°C) for RD.

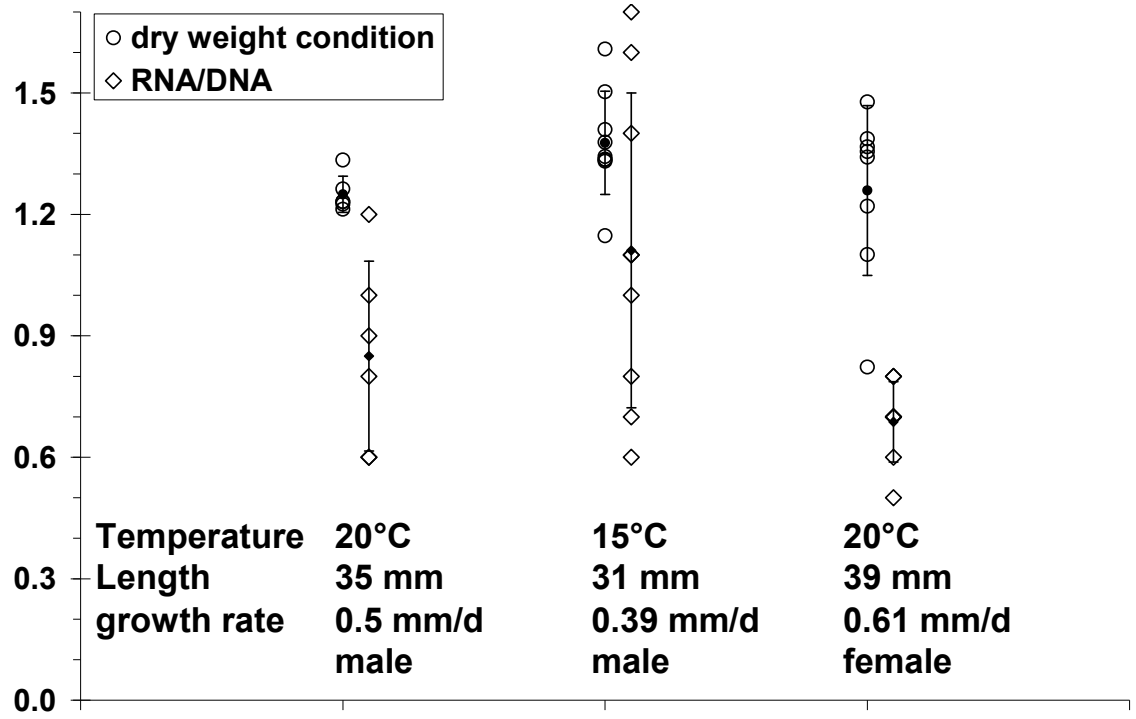

Figure 7-1: Variability of dry weight condition and RNA·DNA^{-1} within similar length, temperature, growth class and same gender.

Discussion

Dry weight condition index

Within this work two possible proxies were tested for their suitability to estimate length growth rates of *C. crangon* in the field. Our results indicate that DWC alone could not be applied as growth proxy, as different values were observed for animals with the same growth rate. The correlation coefficient for growth rate and DWC of the different subgroups (separated according to gender, length and temperature and catch date) was generally low. Since total length and dry weight can be determined with sufficient accuracy it can be assumed that the growth variability must largely result from sources other than measurement error. Other factors contributing to growth variability among individuals include differences in the stage of the moult cycle, the stage of maturation of female shrimp and differences in feeding behavior among individuals.

Moulting

In the present study, moulting was only measured for marked shrimps. This only allowed us to determine the time span since the last moult and does not provide a prediction of the timing of the next moult. For the unmarked shrimps, the moult

interval was determined by counting the exhuvia in a tank containing several animals and therefore only a mean intermoult period can be given. Since crustaceans exhibit continuous weight growth but incremental (discontinuous) length growth as this is hindered by the hard structure of the exosceleton (Laxter & Outhward 1991) dry weight at length can vary according to the moult stage. Growth is therefore performed by steady replacement of water with protein during the moult cycle (Rosa & Nunes 2004). Two similar sized shrimps can therefore have different dry weights only because they are in different stages of the moult cycle. Additionally the whole animal is considered when calculating the DWC and therefore differences in the thickness or calcification of the carapace might influence this proxy. The exhuvia can make up to 17% of the whole dry weight of *C. crangon* (Regnault & Luquet 1978).

Another effect of moulting on DWC could be not only the weight gain at same length but also the weight loss. Before ecdysis, crustaceans stop feeding (Laxter & Outhward 1991). Effects of this premoult starvation and the energy loss during that phase are discussed differently. In most Crustaceans, glycogen, glucose and lipids derived from the hepatopancreas and haemolymphe are used (Sánchez-Paz et al. 2007, Clifford & Brick 1983, Barcley et al. 1983). These substances have high energy but low dry weight and therefore DWC will change very little. However, on the other hand, dry weight is mainly a measure of muscle protein which is expected to decrease when growth is low (negative) which would be captured by the DWC. During the moult, Regnault & Lagadère (1983) assumed that *C. crangon* loses 10% of its protein biomass. Therefore even a growing shrimp fed ad libitum can lose weight.

Maturation

The second factor influencing dry weight is the maturation (ovary development) of the females. At about 55 mm total length, 50% of females can be expected to carry eggs (Henderson & Holmes 1987, Boddeke 1961). Prior to this are costs due to gonadogenesis which can not only influence somatic growth rates (Taylor & Peck 2004) but also body dry weight. According to Haefner & Spaargaren (1993) the weight of the ovary can increase the body weight of *C. crangon* by 5.5%. The exact proportion of shrimps influenced by this factor can not be determined since maturation is a sigmoid function of length. However, according to Henderson & Holmes (1987), the smallest egg-bearing females are about 38 mm. In our experiments, a higher correlation for DWC and growth rate was observed for male shrimps than for female shrimps. This could indicate the influence of maturation on DWC. On the other hand, we did not observe an increase in the growth correlation of DWC with decreasing female size which refutes this theory.

Gut content

Another factor that might slightly influence DWC is the point of time where the animal is sampled from the experimental setup. *C. crangon* mainly feeds at night (Feller 2006) and Pihl & Rosenberg (1984) determined that the stomach content is about 1 to 2% of the body weight.

Conclusion

Dry weight at length is not a suitable proxy to estimate growth rates of *C. crangon*. Most probably moulting, maturation and gut fullness are the major factors influencing scatter within the data. Moulting accounts for about 10% or more, maturation for another 5.5% and gut fullness for approximately 2%.

RNA / DNA

RD correlated significantly with growth rates of male *C. crangon* and with growth rates of female *C. crangon* at high temperatures (20 and 25°C). The correlation coefficient was low when data were not separated according to length or temperature. Therefore, the utility of this growth proxy to field-caught *C. crangon* is limited. The correlation coefficients determined in this work were low in comparison to other species where RD significantly improved growth models (Peck et al. 2003, Smith 2003, Hovenkamp & Witte 1991, Buckley 1984). However, in several species, previous studies have reported that RD was not correlated with growth or that RD can only be used during special life stages or under special conditions. For *Artemia salina* RNA·µg^{-1} depends on the growth phase and is higher in the exponential phase (Dagg & Littlepage 1972). In the scallop *Euvola ziczac* only juvenile growth rates are correlated with RD (Lodeiros et al. 1996). For *Solea solea* RD can only be used to compare larvae of the same stage (Richard et al. 1991). Although RD is generally a good proxy for growth determination of larval fish, for larval dab and sprat (Lee et al. 2006) and larval herring (Mathers et al. 1994) no correlation between recent growth and RD could be determined. Anger & Hirche (1990) detected no correspondence of RD with instantaneous growth rates in carbon and nitrogen of the spider crab, *Hyas araneus*. RD of S*ardina pilchardus* varies only with length and food (Chícharo et al. 1998a) and even within well fed fish larvae a high variability of the RD ratio can be observed (Bergeron & Boulhic 1994). This shows that high RD does not necessarily have to correspond with high growth rates.

There are several other factors that might influence RD but not growth or vice versa. These factors are for *C. crangon* mainly those already discussed for DWC: moulting, maturation and feeding. These parameters do not only influence dry weight but also a variety of biochemical processes and therefore the RNA-DNA ratio. RD can react to feeding events within a very short time span in the order of days or even diurnal (see below).

Moulting

In the actual work RD was determined from the muscle of *C. crangon*. A review about biochemical changes during moult (Chang 1995) gives indication that many biochemical, physiological and behavioral processes are influenced by the moult cycle and that even muscle protein and amino acid incorporation in the muscle can be influenced. This is also indicated by transposition mitochondria during the moult cycle (Miyawaki & Tsuruda 1984). It can therefore be assumed that the RD ratio changes during the moult cycle, what was already shown *Homarus americanus*

post larvae (Juinio et al. 1992), and that this does not necessarily has to parallel changes in growth.

Moulting is a complex procedure and the process is regulated by several hormones (Hartnoll 2001). Different behavior can be observed during the different stages (Boddeke 1976). It was observed that moulting influences the transcription of 16sRNA (Sánchez-Paz et al. 2003), which is the main part of RNA in a cell. Further for *C. crangon* a sharp increase of ammonia excretion during the moult could was observed, indicating a high rate of protein degradation and activity (Regnault 1979) (Regnault & Lagardère 1983). This is quite similar to Nott & Mavin (1986) who described that metabolic activity of *C. crangon* peaks at premoult but decreases during moult. RNA regulates all processes mentioned and therefore changes in the RNA concentration are most likely during moult. Dedicating RNA concentration in a cell only to feeding processes is therefore most probably not possible for C. crangon. Further the already mentioned intermission in feeding during moult might lead to a decrease of RD whereas growth rate is not affected.

Maturation

Comparable to moult, maturation is also a complex procedure that depends on the interaction of several hormones and might therefore also influence RD. Biochemical parameters have been shown to change significantly during vitellogenesis (Haefner & Spaargaren 1994). In our results females of the respective size classes did show varying correlation of growth rate and RD. The 60 mm female growth rates at 20°C did not correlate significantly with RD whereas the 40 mm size classes did. An influence of maturation on RD has been described before for molluscs. In the bivalve *Pecten maximus* RD can be used to determine sexual maturity (Robbins et al. 1990) and in *Euvola ziczac* RD was correlated with growth of the muscle for juveniles but not with maturing scallops. In the actual work correlations were only slightly better for male shrimp than for female shrimp therefore the scatter in the data will most probably be not only an effect of maturation alone, but an influence is possible. Chícharo et al. (2007) observed higher RNA/DW in females than in male *C. crangon* and also concluded that a possible reason for that might be due to maturation or sex specific behavior.

Feeding

Besides moulting and maturation, mainly feeding, short-term starvation during moult and the quick response of RD to feeding might increase scatter in the data. It could be shown for bivalves, fish and decapods that RD reacts on feeding and starving within some days (Norkko et al. 2006, Rosa & Nunes 2004) (Bergeron 2000, Malloy & Targett 1994, Richard et al. 1991) or even within hours as indicated by diurnal differences of larval fish (Lee et al. 2006, Chícharo et al. 1998b, Lough et al. 1996). In comparison to the shortest experimental time span of 30 days necessary to determine growth rates accurately this time intervals are very short. The actual RD ratio can therefore be considered more or less as a snap shot and correlation would only be given if growth and RD remains constantly high or low.

Analyzed tissue

Another factor influencing correlation might have been the analyzed tissue. In the actual work RD ratio of the tail muscle of *C. crangon* was examined. Muscle cells grow either by hyperplasia (new production of muscle fibres) or by hypertrophy (muscle enlargement) (Koumans & Akster 1995). Hypertrophy emerges from incorporation of satellite cells into the muscle fibres and subsequent enlargement. During hypertrophy muscle cells can therefore contain more DNA than muscles growing by hyperplasia. Further incorporated satellite cells might be degraded first during starvation what is indicated by decreasing DNA during atrophy in human cells (Willoughby et al. 2003, Clarke 1990). This indicates that DNA in a cell is not constant as assumed and therefore RD ratio might be even due to changes in DNA or RNA, increasing the variability. The different growth of muscle cells in comparison to other cells that make the analyzed tissue unique. Another aspect is that muscle cells contain a large amount of mitochondria that also contain DNA. Changes in activity increase the numbers of mitochondria and therefore DNA content in a cell. The amount of mitochondrial DNA in comparison to nucleus DNA has not been determined for *C. crangon* yet but in *Drosophila* (whole animal) it is about 1% (Calleja et al. 1993). In muscles it can be assumed to be higher. One animal might therefore show a lower RD ration only because it is more active than another one that is passive and muscles are degraded. Both animals might have fed the same amount of food and also have the same growth rate.

Anyhow even when only muscle tissue was analyzed, like it has been done in the actual study, correlations of growth and RD have been observed (Peck et al. 2003, Smith 2003).

Regeneration

Injuries and subsequent regeneration might influence and reduce growth of C. crangon (Tiews 1970). Regeneration under ad libitum feeding could therefore lead to high RD ratios but low growth rates.

Perspective

RD and dry weight are no suitable proxies to determine growth rates of *C. crangon* in the field. Although it is not possible to directly measure growth with these values it might be possible to determine the nutrition and feeding history. Applying this detour would make it possible to estimate the amount of starving animals in the field and to derive from this fraction the amount of animals not growing due to starvation. Another future focus shall be set to the changes of DWC and RD during the moult cycle.

Acknowledgements

The study was financially supported by the Federal Ministry of Food, Agriculture and Consumer Protection, Germany Project No. 03HS030. Thanks to Prof. Myron Peck for help with the manuscript and the RNA/DNA method.

References

Anger K, Hirche H-J (1990) Nucleic acids and growth of larval and early juvenile spider crab, *Hyas araneaus*. Marine Biology 105:403-411

Barcley MC, Dall W, Smith DM (1983) Changes in lipid and protein during starvation and the moulting cycle in the tiger prawn *Penaeus esculentus* Haswell. Journal of Experimental Biology and Ecology 68:229-244

Bergeron J-P (2000) Effect of strong winds on the nutritional condition of anchovy (*Engraulis encrasicolus* L.) larvae in the Bay of Biscay, Norhtern Atlantic, as inferred from an early field application of DNA/C index. ICES Journal of Marine Science 57:249-255

Bergeron J-P, Boulhic M (1994) Rapport ARN/ADN et évaluation de l'état nutritionelle et de la croissance des larves de poissions marins: un essai de mise au point expérimentale chez la sole (*Solea solea* L.). ICES Journal of Marine Science 51:181-190

Beukema JJ (1992) Dynamics of juvenile shrimp *Crangon crangon* in a tidal flat-nursery of the Wadden Sea after mild and cold winters. Marine Ecology Progress Series 83:157-165

Boddeke R (1961) Sex in the brown shrimp *(Crangon crangon)*. ICES Shellfish Committee C II 1961 Nr.50

Boddeke R (1976) The seasonal migration of the brown shrimp *Crangon crangon*. Netherlands Journal of Sea Research 10:103-130

Buckley BA, Szmant AM (2004) RNA/DNA ratios as indicators of metabolic activity in four species of Caribbean reef- building corals. Marine Ecology Progress Series 282:143-149

Buckley L, Caldarone E, Ong TL (1999) RNA-DNA ratio and other nucleic acid-based indicators for growth and condition of marine fishes. Hydrobiologia 401:265-277

Buckley LJ (1984) RNA-DNA ratio. an index of larval fish growth in the sea. Marine Biology 80:291-298

Bulnheim H-P, Schwenzer DE (1993) Zur Populationsgenetik von *Crangon crangon* und *C. allmanni* (Crustacea, Decapoda) im Bereich der europäischen Küsten. Zoologische Jahrbücher. Abteilung für allgemeine Zoologie und Physiologie der Tiere 97:327-347

Caldarone, Elaine M., Wagner, Melissa, St.Onge-Burns, Jeanne, and Buckley, Lawrence J. Protocol and Guide for Estimating Nucleic Acids in Larval Fish Using a Fluorescence Microplate Reader. Northeast Fisheries Science Center Reference Document 01-11, -28. 2001. Woods Hole, Massachusetts, National Oceanic and Atmospheric Administration, National Marine Fisheries Service.

Calleja M, Peña P, Ugalde C, Ferreiro C, Marco R, Garesse R (1993) Mitochondrial DNA remains intact during *Drosophila* aging, but the levels of mitochondrial transcripts are significantly reduced. The Journal of Biological Chemistry 268:18891-18897

Chang ES (1995) Physiological and biochemical changes during the moult cycle in decapod crustaceans: an overview. Journal of Experimental Marine Biology and Ecology 193:1-14

Chícharo MA, Chícharo L, Valdés L, López-Jamar E, Ré P (1998a) Does the nutritional condition limit survival potential of the sardine Sardina pilchardus (Wlabaum, 1792) larvae off the north coast of Spain? RNA/DNA ratios and their variability. Fisheries Research 39:43-54

Chícharo MA, Chícharo L, Valdés L, López-Jamar E, Ré P (1998b) Estimation of starvation and diel variation of the RNA/DNA ratios in field-caught *Sardina pilchardus* larvae off the north of Spain. Marine Ecology Progress Series 164:273-283

Clarke PGH (1990) Developmental cell death: morphological diversity and multiple mechanisms. Anatomy and Embryology 181:195-213

Clifford HC, Brick RW (1983) Nutritional physiology of the freshwater shrimp *Macrobrachium rosenbergii* (De Man) - I. Substrate metabolism in fasting juvenile shrimp. Comparative Biochemistry and Physiology 74A:561-568

Cuesta JA, González-Ortegón E, Rodríguez A, Baldó F, Vilas C, Drake P (2006) The decapod crustacean community of the Guadalquivir Estuary (SW Spain): seasonal and inter-year changes in community structure. Hydrobiologia 557:85-95

Dagg MJ, Littlepage JL (1972) Relationships between growth rate and RNA, DNA, Protein and dry weight in *Artemia salina* and *Euchaeta elongata.* Marine Biology 17:162-170

Dall W (1974) Indices of nutritional state in the western rock lobster *Panulirus longipes* (Milne Edwards). I. Blood and tissue constituents and water contents. Journal of Experimental Marine Biology and Ecology 16:167-180

Dall W, Smith DM (1986) Oxygen consumption and ammonia-N excretion in fed and starved tiger prawns, *Penaeus esculentus* Haswell. Aquaculture 55:23-33

del Norte-Campos AGC, Temming A (1994) Daily activity, feeding and rations in gobies and brown shrimp in the northern Wadden Sea. Marine Ecology Progress Series 115:41-53

Edwards RRC (1978) Effects of water-soluble oil fractions on metabolism, growth and carbon budget in the shrimp *Crangon crangon*. Marine Biology 46:259-265

Ehrlich KF, Blaxter JHS, Pemberton R (1976) Morphological and histological changes during the growth and starvation of herring and plaice larvae. Marine Biology 35:105-118

Feller RJ (2006) Weak meiofaunal trophic linkages in *Crangon crangon* and *Cracinus maenas*. Journal of Experimental Marine Biology and Ecology 330:274-283

Gabbott PA (1974) A note on the relationship between the dry weight condition index and the glycogen content of adult oysters (*Ostrea edulis* L.) kept in the laboratory. Journal du Conseil Permanent International pour l' Exploration de la Mer 35:359-361

Gunnarsson B, Ásgeirsson þ, Ingólfsson A (2007) The rapid colonization by *Crangon crangon* (Linnaeus, 1758) (Eucarida, Caridea, Crangonidae) of Icelandic coastal waters. Crustaceana 80:747-753

Haefner PA, Spaargaren DH (1993) Interactions of ovary and hepatopancreas during the reproductive cycle of *Crangon crangon* (L.). I. Weight and volume relationships. Journal of Crustacean Biology 13:523-531

Haefner PA, Spaargaren DH (1994) Interactions of ovary and hepatopancreas during the reproductive cycle of *Crangon crangon* (L.). II: Biochemical relationships. Journal of Crustacean Biology 14:6-19

Hartnoll R (2001) Growth in Crustacea – twenty years on. Hydrobiologia 449:111-122

Henderson PA, Holmes RHA (1987) On the population biology of the common shrimp *Crangon crangon* (L.) (Crustacea: Caridea) in the Severn Estuary and Bristol Channel. Journal of the Marine Biological Association U.K. 67:825-847

Hovenkamp F, Witte JIJ (1991) Growth, otolith growth and RNA/DNA ratios of larval plaice Pleuronectes platessa in the North Sea 1987 to 1989. Marine Ecology Progress Series 70:105-116

Juinio MAR, Cobb JS (1994) Estimation of recent growth rates of field-caught postlarval amercian lobsters, Homarus americanus, from RNA:DNA ratios. Canadian Journal Fisheries and Aquatic Science 51:286-294

Juinio MAR, Cobb JS, Bengtson D, Johnson M (1992) Changes in nucleic acids over the moult cycle in relation to food availability and temperature in *Homarus americanus* postlarvae. Marine Biology 114:1-10

Jussila J (1999) Comparison of selected condition indices between intermolt and post-molt marron, *Cherax tenuimanus*, of different feeding status raised under intensive culture conditions. Journal of applied Aquaculture 9:57-65

Kosolow JA, Brault S, Dugas J, Fournier RO, Hughes P (1985) Condition of larval cod (Gadus morhua) of southwest Nova Scotia in 1983 in relation to plankton abundance and temperature. Marine Biology 86:113-121

Koumans JTM, Akster HA (1995) Myogenic cells in development and growth of fish. Comparative Biochemistry and Physiology 110:3-20

Kurita Y, Meier S, Kjesbu OS (2003) Oocyte growth and fecundity regulation by atresia of Atlantic herring (*Clupea haerengus*) in relation to body condition throughout the maturation cycle. Journal of Sea Research 49:203-219

Lapinska E, Szaniawska A (2006) Environmental Preferences of *Crangon crangon* (Linnaeus, 1758), *Palaemon adspersus* Rathke, 1837, and *Palaemon elegans* Rathke, 1837 in the littoral zone of the gulf of Gdansk. Crustaceana 79:649-662

Laxter JHSB, Outhward AJS (1991) Moulting and growth. Advances in Marine Biology 27:213-250

Lee O, Danilowicz BS, Dickey-Collas M (2006) Temporal and spatial variability in growth and condition of dab (*Limanda limanda*) and sprat (*Sprattus sprattus*) larvae in the Irish Sea. Fisheries Oceanography 15:490-507

Lodeiros CJM, Fernández RI, Bonmatí A, Himmelman JH, Chung KS (1996) Relation of RNA/DNA ratios to growth for the scallop *Euvola (Pecten) ziczac* in suspended culture. Marine Biology 126:245-251

Lough RG, Caldarone E, Rotunno TK, Broughton EA, Burns BR, Buckley LJ (1996) Vertical distribution of cod and haddock eggs and larvae, feeding and condition in stratified and mixed waters on southern Georges Bank, May 1992. Deep-Sea Research II 43:1875-1904

Lundebye A-K, Langston WJ, Depledge MH (1997) Stress proteins and condition index as biomarkers of tributyltin exposure and effect in mussels. Ecotoxicology 6:127-136

Malloy KD, Targett TE (1994) The use of RNA:DNA ratios to predict growth limitations of juvenile summer flounder (*Paralichthys dentatus)* from Delawarer North Carolina estuaries. Marine Biology 118:367-375

Mathers EM, Houlihan DF, Buren LJ (1994) RNA, DNA and protein concentrations in fed and starved herring *Clupea haerengus* larvae. Marine Ecology Progress Series 107:223-231

Miyawaki M, Tsuruda T (1984) The transposition of Mitochondria in the hindgut epithelial cells of the crayfish, *Procambarus clarki*, during the moult cycle. Proceedings of the Japan Acadamy Ser.B 60:81-84

Moss SM (1994a) Growth rates, nucleic acid concentrations, and RNA/DNA ratios of juvenile white shrimp, *Penaeus vannamei* boone, fed different algal diets. Journal of Experimental Marine Biology and Ecology 182:193-204

Moss SM (1994b) Use of nucleic acids as indicators of growth in juvenile white shrimp, *Penaeus vannamei*. Marine Biology 120:359-367

Nagasawa K, Nagata M (1992) Effects of *Pectenophilus ornatus* (Copepoda) on the biomass of cultured japanese scallop *Patinopecten yessoensis*. The Journal of Parasitology 78:552

New MB (1976) A review of dietry studies with shrimp and prawns. Aquaculture 9:101-144

Nicol S, Kitchener J, King R, Hosie G, de la Mare WK (2000) Population structure and condition of Antarctic krill (*Euphausia superba*) off East Antarctica (80-150°E) during the Austral summer of 1995/1996. Deep-Sea Research II 47:2489-2517

Norkko J, Thrush SF, Wells RMG (2006) Indicators of short-term growth in bivalves: Detecting environmental change across ecological scale. Journal of Experimental Biology and Ecology 337:38-48

Nott JA, Mavin LJ (1986) Adaptation of a quantitative program for the X-ray analysis of solubilized tissue as microdroplets in the transmission electron microscope: application to the moult cycle of the shrimp *Crangon crangon* (L). Histochemical Journal 18:507-518

Oh C-W, Hartnoll RG (2000) Effects of food supply on the growth and survival of the common shrimp *Crangon crangon* (Linnaeus, 1758) (Decapoda, Caridea). Crustaceana 73:83-99

Pearson AM, Dutson TR (1997) Production and processing of healthy meat, poultry and fish products. Aspen Publishers Inc,

Peck MA, Buckley LJ, Caldarone EM, Bengtson DA (2003) Effects of food consumption and temperature on growth rate and biochemical-based indicators of growth in early juvenile Atlantic cod *Gadus morhua* and haddock *Melanogrammus aeglefinus.* Marine Ecology Progress Series 251:233-243

Pérez-Castañeda R, Defeo O (2002) Morphometric relationships of penaeid shrimps in a coastal lagoon: spatio-temporal variability and management implications. Estuaries 25:282-287

Pihl L, Rosenberg R (1984) Food selection and consumption of the shrimp *Crangon crangon* in some shallow marine areas in western Sweden. Marine Ecology Progress Series 15:159-168

Plagmann J (1939) Ernährungsbiologie der Garnele (*Crangon vulgaris* Fabr.). Helgoländer wissenschaftliche Meeresuntersuchungen 2:113-162

Regnault M, Luquet P (1978) Variations quantitatives de l'acide desoxyribonucléique (ADN), au cours de cycle de mue, dans les téguments, le muscle et l'hépatopancréas de la crevette *Crangon crangon*. Journal Physiolgie Paris 74:21-30

Regnault M (1979) Ammonia Excretion of the Sand-Shrimp *Crangon crangon* (L.) During the moult cycle. Journal of Comparative Physiology 133:199-204

Regnault M, Lagardère JP (1983) Effects of ambient noise on the metabolic level of *Crangon crangon* (Decapoda, Natantia). Marine Ecology Progress Series 11:71-78

Richard P, Bergeron J-P, Boulhic M, Galois R, Person-Le Ruyet J (1991) Effect of starvation on RNA, DNA and protein content of laboratory-reared larvae and juveniles of *Solea solea*. Marine Ecology Progress Series 72:69-77

Robbins I, Lubet P, Besnard J-Y (1990) Seasonal variations in the nucleic acid content and RNA:DNA ratio of the gonad of the scallop *Pecten maximus*. Marine Biology 105:191-195

Rosa R, Nunes ML (2004) RNA, DNA and protein concentrations and amino acid profiles of deep-sea decapod *Aristeus antennatus*: An indication for seasonal variations of nutrition and growth. Aquatic Living Resources 17:25-30

Sánchez-Paz A, García-Carreño FL, Hernández-López J, Muhlia-Almazán A, Yepiz-Plascencia G (2007) Effect of short-term starvation on hepatopancreas and plasma energy reserves of the Pacific white shrimp (*Litopenaeus vannamei*). Journal of Experimental Biology and Ecology 340:184-193

Sánchez-Paz A, García-Carreño FL, Muhlia-Almazán A, Hernández-Saavedra NY, Yepiz-Plascencia G (2003) Differential expression of trypsin mRNA in the white shrimp (*Penaeus vannamei*) midgut gland under starvation conditions. Journal of Experimental Marine Biology and Ecology 292:1-17

Smith TR (2003) RNA-DNA ratio in scales from juvenile cod provides a nonlethal measure of feeding condition. Transactions of American Fisheries Society 132:9-17

Taylor DL, Peck MA (2004) Daily energy requirements and trophic positioning of the sand Shrimp *Crangon septemspinosa.* Marine Biology 145:167-177

Tetard A (1985) Elements sur la croissance de la crevette grise, *Crangon crangon* (L.), en manche-est et sud mer du nord. ICES C.M. K:17:16pp.

Tiews K (1970) Synopsis of biological data on the common shrimp *Crangon crangon* (Linaeus, 1758). FAO Fisheries Reports 4:1167-1223

Wagner M, Durbin E, Buckley L (1998) RNA:DNA ratios as indicators of nutritional condition of the copepod *Calanus finmarchicus*. Marine Ecology Progress Series 162:173-181

Willoughby DS, Sultemeire S, Brown M (2003) Human muscle disuse atrophy after 28 days of immobilization in a lower-limb walking boot: a case study. Journal of Excercise Physiology online 6:88-96

An evaluation of various models for the estimation of total mortality from length frequency distribution data confronted with seasonal variability in growth, recruitment and mortality.

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Abstract

Seven different methods for estimating total mortality (modified Wetherall et al.; Powell; Beverton & Holt; Jones & Zalinge; Hoenig; Ssentongo & Larkin; as well as the seasonal and the non-seasonal Length Converted Catch Curve) were critically evaluated with respect to their sensitivity against violation of basic assumptions such as non-seasonality of growth, mortality and recruitment, fixed mean maximum length L_∞ and constant growth parameter k. All methods were applied to artificially created length frequency distributions with different combinations of known L_∞, Z and k, and the deviation of the resulting θ (Z/k) and L_∞ from the input values was analyzed. In general all methods underestimated the real mortality and seasonality in recruitment particularly biased the results. The level of Z, and to a lesser extent k, considerably influenced the results whereas different L_∞ values only slightly altered the mortality estimates.

In a species-specific approach, length frequency distributions comparable to those of the brown shrimp *Crangon crangon* were created and again θ and L_∞ was estimated for this special case. For all methods, limitations existed and the case study performed for *C. crangon* illustrates how estimates of θ and Z can be improved by selecting suitable methods and correcting observed bias.

Keywords: Total mortality, seasonality, length frequency

Introduction

Management of fish stocks or invertebrate crustaceans requires the quantitative estimation of growth and mortality. In most cases determination of these parameters is coupled to an accurate measurement of age of the studied animal. In short-lived crustaceans and tropical fishes, age determination is not always possible. Crustaceans regularly loose all hard parts during each moult (Jennings & Reynolds 2001) and the lack of a strong seasonality in tropical areas makes the distinction of seasonal- and annual-rings in otoliths problematic (Sparre et al. 1989).

Several methods and models have been developed to determine total mortality (Z) or $\theta = Z/k$ (k = Bertalanffy growth parameter) for assessments of tropical fish stocks. Most methods are based on the von Bertalanffy growth function (von Bertalanffy 1938, von Bertalanffy 1934) and assume an exponential decrease of the abundance of individuals within cohorts over time due to mortality (Wetherall et al. 1987). The methods are restricted to specific conditions such as no seasonality in growth (with exception of the seasonal Length Converted Catch Curve), no seasonality in hatching and mortality and no variability in k and L_∞. However, seasonal recruitment and growth is very common and can be observed in tropical fishes adapted to rainy seasons or seasonal changing currents (Admassu 1996) and is especially important for populations inhabiting higher latitudes (Gonzáles-Gordillo et al. 2003, Marin E. et al. 2003, Wehrtmann 1989). The strong seasonality of recruitment and growth most likely introduces considerable bias in the estimation of total mortality.

The brown shrimp *Crangon crangon* is an ecologically (Kuipers & Dapper 1981) and commercially (ICES 2007) important invertebrate species in the North Sea. The following observations and characteristics violate the general preconditions of length frequency-based methods.

Recruitment occurs in waves and a major pulse can be observed in late spring followed by several smaller events in summer (Temming & Damm 2002, Beukema 1992).

Mortality most likely follows the seasonal abundance of predators, mainly 0 and I group cod (Gadus morhua) and whiting (Merlangius merlangus) in coastal waters with peak abundances in autumn (Jansen 2002, Hamerlynck & Hostens 1993).

The growth pattern exhibits strong seasonal oscillation due to strong seasonal temperature variations.

Individual growth rates can be assumed to be highly variable according to findings of several authors who determined growth rates from 0.0-0.1 mm d^{-1} (Oh & Hartnoll 2000) to 0.07-0.4 mm·d^{-1} (Tetard 1985) to 0.2-0.54 mm·d^{-1} (Beukema 1992).

The aim of this work was therefore twofold 1.): to evaluate methods for the determination of θ from length frequency distributions using simulated data with known characteristics representing a wide range of species and conditions. 2.): to determine the bias of the methods when applied to simulated data representing

brown shrimp in the North Sea. These two case studies were conducted to demonstrate how biases that occur when applying mortality estimation methods to seasonally influenced data can potentially be corrected. This leads to a more accurate interpretation of the estimated mortalities.

For the general and the brown shrimp-specific case studies, artificial length frequency distributions were generated with known mortality (Z), growth (k) and mean maximum length (L_∞). Seasonal variable, mortality, growth and recruitment were applied in the simulations. Furthermore, in specific runs, L_∞ and k were variable between cohorts with coefficients of variations of 10, 20 and 30% around a given mean value. The different length frequency-based methods were applied to the simulated length frequency distributions and the mortality and L_∞ estimates were compared with the known input values.

Material and Methods

Description of the model

For the generation of the length frequency distributions with the known input values, a PASCAL routine was written. Each day, 100 cohorts start, each one with a different k and L_∞ that were generated from a normally distributed random process with fixed mean and standard deviation. In scenarios where k and L_∞ were kept constant, only one cohort is simulated each day. The initial size of each cohort was determined by an assumed level of recruitment and a fraction of the cohort died each day according to the mortality assumed in the specific scenario (see below). Furthermore, the length of the animals in the cohort increased each day.

To assure that the model was in a steady state when the length frequency distribution was created, the runtime of the model depended upon growth constants and the mortality and was adjusted so that the runtime equals the life span of the first cohort plus one year in each simulation.

The mean length frequency distribution for a whole year was based on the length frequencies calculated each day of the last (equilibrium) model year.

Recruitment

Recruitment is realized in the model by different sizes of the cohorts that start each day. Cohorts starting the same day have the same size but the starting size (N_0) on different days, was varied by a sinusoidal function (suggesting a recruitment maximum in spring).

$$N_0 = N_0 \cdot 0.5 + 0.5 \cdot \sin\left(\frac{2 \cdot \pi}{365} \cdot t\right) \qquad (15)$$

Growth

The simulated length of each cohort at a given day was calculated using the non-seasonal (16) or the seasonal version (17) of the von Bertalanffy growth function according to

$$L_t = L_\infty \cdot \left(1 - e^{-k\cdot(t-t_0)}\right) \quad (16)$$

$$L_t = L_\infty \cdot \left(1 - e^{-k\cdot(t-t_0)-\left(\frac{C\cdot k}{2\cdot\pi}\right)\cdot[\sin(2\cdot\pi\cdot(t-t_s))-\sin(2\cdot\pi\cdot(t_0-t_s))]}\right) \quad (17)$$

where L_t is the length a time t (in years), L_∞ is the mean maximum length which an individual can approach, k is the growth constant per year and t_o is the theoretical time when the animal has a length of zero. C and t_s in (17) determine the amplitude and the time of the minimum of the yearly growth oscillation. If C equals zero, there is no oscillation, if C equals 1 then the growth oscillation is strongest. Seasonally varying length growth was simulated with C=1 and t_s=0.5 which resulted in minimum growth in winter (day 365).

Bertalanffy parameters k and L_∞ were randomly varied so that each cohort starting at one day had slightly different growth parameters. To accomplish this, a random number was multiplied to the parameters based on a normal distribution calculated by transformation of two equal distributed numbers using the polar method. For L_∞ the borders of the maximum and minimum variation within the normal distribution was set to

$$L_{\infty\max} = L_\infty + 3\cdot\sigma L_\infty$$
$$L_{\infty\min} = L_\infty - 3\cdot\sigma L_\infty \quad (18)$$

where σL_∞ is the used coefficient of variation of L_∞.

Mortality

Total mortality in the model was calculated according to

$$N_t = N_0 \cdot e^{-Z\cdot t} \quad (19)$$

where N_t is the amount of animals left from the starting amount N_0 after t years. Alternatively a seasonally varying Z was applied which oscillates around a given mean value according to the cosine function

$$Z = Z + \Delta Z \cdot -\cos(2\pi\cdot t) \quad (20)$$

where ΔZ is the amplitude (percentage of Z) of the variation of Z. This function simulates maximum mortality in summer.

The number of surviving animals from a starting cohort size N_0 at a given day t is then

$$N_t = N_0 \cdot e^{-\frac{1}{365}\cdot\sum_{t=1}^{n} Z+\Delta Z\cdot Z\cdot\left(-\cos\left(2\cdot\pi\cdot\frac{t}{365}\right)\right)} \quad (21)$$

As the calculation of the sum in (21) is very computer time consuming, the result of (21) was determined for different t and a cosine function was fit to the results only dependent on time t (r^2=1,). Depending on the time where the maximum mortality should occur four calculations are possible:

maximum mortality in summer:

$$-\frac{1}{365}\cdot\sum_{t=1}^{n} Z+\Delta Z\cdot Z\left(-\cos\left(2\cdot\frac{t}{365}\right)\right)=-\frac{Z\cdot t}{365}-\frac{\Delta Z}{365}\cdot\left(-\sin\left(\frac{t}{365}\cdot 2\cdot\pi\right)\cdot\frac{365}{2\cdot\pi}\right) \quad (22)$$

maximum mortality in autumn:

$$-\frac{1}{365}\cdot\sum_{t=1}^{n} Z+\Delta Z\cdot Z\left(-\sin\left(2\cdot\frac{t}{365}\right)\right)=-\frac{Z\cdot t}{365}-\frac{\Delta Z}{365}\cdot\left(-\cos\left(\frac{t}{365}\cdot 2\cdot\pi\right)\cdot\frac{365}{2\cdot\pi}-\frac{365}{2\cdot\pi}\right) \quad (23)$$

maximum mortality in spring:

$$-\frac{1}{365}\cdot\sum_{t=1}^{n} Z+\Delta Z\cdot Z\left(\sin\left(2\cdot\frac{t}{365}\right)\right)=-\frac{Z\cdot t}{365}-\frac{\Delta Z}{365}\cdot\left(-\cos\left(\frac{t}{365}\cdot 2\cdot\pi\right)\cdot\frac{365}{2\cdot\pi}+\frac{365}{2\cdot\pi}\right) \quad (24)$$

maximum mortality in winter:

$$-\frac{1}{365}\cdot\sum_{t=1}^{n} Z+\Delta Z\cdot Z\left(\sin\left(2\cdot\frac{t}{365}\right)\right)=-\frac{Z\cdot t}{365}-\frac{\Delta Z}{365}\cdot\left(\sin\left(\frac{t}{365}\cdot 2\cdot\pi\right)\cdot\frac{365}{2\cdot\pi}\right) \quad (25)$$

Assuming a variable hatch day t_0 and including e.g. (22) in (21) leads to (shown here only for maximum mortality in summer)

$$N_t = N_0\cdot e^{-\frac{Z\cdot t}{365}-\frac{\Delta Z}{365}\cdot\left[\left(-\sin\left(\frac{t}{365}\cdot 2\cdot\pi\right)\cdot\frac{365}{2\cdot\pi}\right)-\left(-\sin\left(\frac{t_0}{365}\cdot 2\cdot\pi\right)\cdot\frac{365}{2\cdot\pi}\right)\right]} \quad (26)$$

Estimating θ and L_∞

For estimating θ=Z/k and L_∞ different methods were available which are briefly outlined below:

1. The classical method developed by Beverton and Holt (1956)

$$\theta_{BH}=\frac{L_\infty-\overline{L}}{\overline{L}-L_c} \quad (27)$$

where $\overline{L}$ is here and in the following text the mean length of all fish above L_c, which is the size at first capture meaning the first length class fully vulnerable to the fishing gear.

2. Jones and Zalinge (1981), where θ_{JZ} is estimated by linear regression:

$$\ln\left(C\left(L,L_\infty\right)\right)=a+\theta_{JZ}\ln\left(L_\infty-L\right) \quad (28)$$

In this equation $C(L,L_\infty)$ is the amount of fish caught between L and L_∞.

3. Hoenig (1983), which is based on the median length of all fish above L_c:

$$\theta_H=\frac{\ln(2)}{-\ln\left(1-\frac{L_{median}}{L_\infty}\right)+\ln\left(1-\frac{L_c}{L_\infty}\right)} \quad (29)$$

where L_{median} is the median length of all fish above length of first capture L_c.

4. Ssentongo & Larkin (1973), which is based on the logarithm of the mean length and length at first capture:

$$\bar{y} = \ln\left(1 - \frac{\bar{L}}{L_\infty}\right) \quad (30)$$

$$y_c = \ln\left(1 - \frac{L_c}{L_\infty}\right) \quad (31)$$

$$\theta_{SL} = \frac{n}{n+1} \cdot \frac{1}{\left(\bar{y} - y_c\right)} \quad (32)$$

where n = total number of animals greater than L_c

5. Powell (1979), which includes the variance of the length distribution and also allows to give an estimate of L_∞.

$$L_{\infty P} = \bar{L} + \frac{2 \cdot S_l^2\left(\bar{L} - L_c\right)}{\left(\bar{L} - L_c\right)^2 - S_l^2} \quad (33)$$

$$\theta_P = \frac{2 \cdot S_l^2}{\left(\bar{L} - L_c\right)^2 - S_l^2} \quad (34)$$

where $\bar{L}$ is the mean length of all animals above L_c and S_i^2 is the variance of all animals above L_c.

6. The Wetherall et al. (1987) method modified by Sparre et al. (1989) is a regression method with the slope b, the intercept a and L' > L_c:

$$\bar{L} - L' = a \cdot L' + b \quad (35)$$

$$L_{\infty W} = -\frac{b}{a} \quad (36)$$

$$\theta_W = -\frac{\left(1+a\right)}{a} \quad (37)$$

7. Miranda (2002) is quite similar to the modified Wetherall et al. (1987) method

$$\bar{L} = L_\infty + \frac{Z}{Z+k}\left(L_\infty - L_i\right) \quad (38)$$

$$\theta_M = -\frac{a}{1-a} \quad (39)$$

8. The non-seasonal Length Converted Catch Curve (Pauly 1983), which requires an estimate of L_∞ and of the growth constant k as initial input.

$$\ln\left(\frac{C(L_1, L_2)}{\frac{1}{k} \cdot \ln\left(\frac{L_\infty - L_2}{L_\infty - L_1}\right)}\right) = a - \theta_{sLCCC} \cdot \ln\left(1 - \frac{L_1 + L_2}{2 \cdot L_\infty}\right) \quad (40)$$

$C(L_1,L_2)$ is the amount of animals caught within the length classes L_1 and L_2.

9. The seasonal Length Converted Catch Curve as presented by de Graaf & Dekker (2006). As in this calculation the seasonal von Bertalanffy growth equation (17) is used an estimate of t_0 is needed. In this work t_0 is always set to 0 and t_s was chosen as 0.5 to simulate maximum growth in summer.

Parameter range for the general evaluation

Although k, L_∞ and Z are most probably interlinked, they have been varied separately in this study. To approximate the possible range of the realized combinations of the parameters, a literature review was conducted to extract estimates for L_∞, k and Z of different marine species (Table 8-2, Figure 8-1).

According to these data the total mortality Z range in the model was chosen from 0.1 to 8, the growth coefficient k from 0.2 to 4 and L_∞ from 30 to 220. L_∞ in the model is unitless as the estimate from the Bertalanffy growth function is only determined by k and not by the unit of L_∞, which only changes the scaling. L_∞, k and Z were varied in 20 steps with step sizes of 10, 0.2 and 0.4, respectively. According to these variations, each simulation generated 20x20x20=8000 estimates for each method. The basic assumptions for the different scenario runs are shown in Table 8-1.

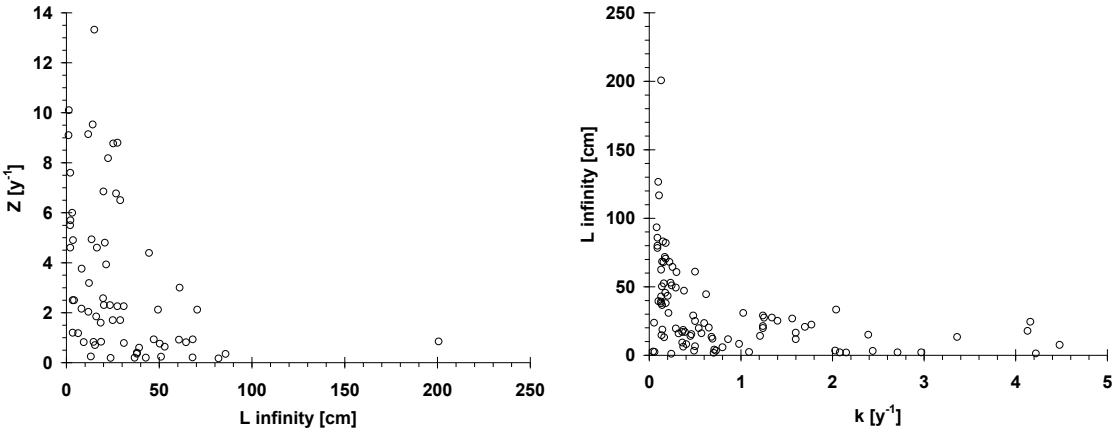

Figure 8-1: Plot of total mortality Z against mean maximum length L∞ (left) and L∞ against growth constant k (right) for different animals. Values and references are given in Table 8-1

Table 8-1: Assumptions for the different scenario runs used to create the artificial length frequency distributions. Column 1: scenario number. Column 2: Use of seasonal or nonseasonal von Bertalanffy growth. Column 3 to 6: Maximum of the sinus function used to simulate seasonal recruitment. Column 7 to 10: Maximum of the total mortality if assumed seasonal and deviation of the maximum from the mean yearly mortality. column 11, 12: Variability of k and L_∞ in the model between the different cohorts.

run	Bertalanffy	Recruitment maximum				Mortality				std k	std L_∞
		spring	summer	fall	winter	spring	summer	fall	winter	[%]	[%]
1	non seasonal	-	-	-	-	-	-	-	-	-	-
2	non seasonal	X	-	-	-	-	-	-	-	-	-
3	non seasonal	-	-	-	-	-	+20%	-	-20%	-	-
4	non seasonal	X	-	-	-	-	+20%	-	-20%	-	-
5	Seasonal	-	-	-	-	-	-	-	-	-	-
6	Seasonal	X	-	-	-	-	-	-	-	-	-
7	Seasonal	-	-	-	-	-	+20%	-	-20%	-	-
8	Seasonal	X	-	-	-	-	+20%	-	-20%	-	-
9	Seasonal	-	X	-	-	-	-	-	-	-	-
10	Seasonal	-	-	X	-	-	-	-	-	-	-
11	Seasonal	-	-	-	X	-	-	-	-	-	-
12	Seasonal	-	-	-	-	+50%	-	-50%	-	-	-
13	Seasonal	-	-	-	-	-	+50%	-	-50%	-	-
14	Seasonal	-	-	-	-	-50%	-	+50%	-	-	-
15	Seasonal	-	-	-	-	-	-50%	-	+50%	-	-
16	Seasonal	X	-	-	-	+50%	-	-50%	-	-	-
17	Seasonal	-	X	-	-	+50%	-	-50%	-	-	-
18	Seasonal	-	-	X	-	+50%	-	-50%	-	-	-
19	Seasonal	-	-	-	X	+50%	-	-50%	-	-	-
20	Seasonal	X	-	-	-	-	+50%	-	-50%	-	-
21	Seasonal	-	X	-	-	-	+50%	-	-50%	-	-
22	Seasonal	-	-	X	-	-	+50%	-	-50%	-	-
23	Seasonal	-	-	-	X	-	+50%	-	-50%	-	-
24	Seasonal	X	-	-	-	-50%	-	+50%	-	-	-
25	Seasonal	-	X	-	-	-50%	-	+50%	-	-	-
26	Seasonal	-	-	X	-	-50%	-	+50%	-	-	-
27	Seasonal	-	-	-	X	-50%	-	+50%	-	-	-
28	Seasonal	X	-	-	-	-	-50%	-	+50%	-	-
29	Seasonal	-	X	-	-	-	-50%	-	+50%	-	-
30	Seasonal	-	-	X	-	-	-50%	-	+50%	-	-
31	Seasonal	-	-	-	X	-	-50%	-	+50%	-	-
32	Seasonal	-	-	-	-	-	-	-	-	10	-
33	Seasonal	X	-	-	-	-	-	-	-	10	-
34	Seasonal	-	-	-	-	-	+50%	-	-50%	10	-
35	Seasonal	X	-	-	-	-	+50%	-	-50%	10	-
36	Seasonal	-	-	-	-	-	-	-	-	-	10
37	Seasonal	X	-	-	-	-	-	-	-	-	10
38	Seasonal	-	-	-	-	-	+50%	-	-50%	-	10
39	Seasonal	X	-	-	-	-	+50%	-	-50%	-	10
40	Seasonal	-	-	-	-	-	-	-	-	10	10
41	Seasonal	X	-	-	-	-	-	-	-	10	10
42	Seasonal	-	-	-	-	-	+50%	-	-50%	10	10
43	Seasonal	X	-	-	-	-	+50%	-	-50%	10	10
44	non seasonal	-	-	-	-	-	+50%	-	-50%	-	-
45	non seasonal	X	-	-	-	-	+50%	-	-50%	-	-
46	Seasonal	-	-	-	-	-	+50%	-	-50%	-	-
47	Seasonal	X	-	-	-	-	+50%	-	-50%	-	-
48	Seasonal	X	-	-	-	-	+50%	-	-50%	20	20
49	Seasonal	X	-	-	-	-	+50%	-	-50%	30	30

Table 8-2: Bertalanffy growth constant k, mean maximum length L_∞, total mortality Z and references for different marine species used for estimating parameter range (see also Figure 8-1).

	name	k	L_∞	Z	reference		name	k	L_∞	Z	reference
1	*Lutjanus griseus*	0.17	71.70		Burton	28	*Penaeus duorarum*	0.13	14.61	0.831	Campos
		0.13	62.50					0.14	18.70	0.838	Campos
							Macrobrachium				
2	*Zeus faber*	0.30	60.71	0.92	Akyol	29	*vollenhovenii*	1.24	21.36	3.93	Nwosu
3	*Zygochlamys patagonica*		0.00					1.24	19.90	6.85	Nwosu
4	*Salmo trutta*	0.24	51.00	0.24	Brown	30	*Uca cumulanta*	4.22	1.31	10.1	Koch
		0.18	82.00	0.17				2.4	1.11	9.1	Koch
		0.16	68.00	0.21		31	*Uca maracoani*	2.03	3.52	4.9	Koch
					Amezcua (Madrid						
5	*Lutjanus guttatus*	0.09	80.01		Vera)			2.44	3.10	6	Koch
		0.10	39.50		Amezcua (Maupome)	32	*Uca rapax*	2.08	2.05	4.6	Koch
		0.14	68.44		Amezcua (Rojas)			2.15	2.00	5.5	Koch
		0.15	83.00		Amezcua (Siefke)	33	*Uca vocator*	2.71	2.16	5.7	Koch
		0.09	78.30		Amezcua			2.97	2.06	7.6	Koch
		0.08	93.42		Amezcua	34	*Nephrops norvegicus*	0.14	50.31	0.76	Ulmestrand
		0.09	85.82	0.35	Amezcua			0.16	52.43		Ulmestrand *(ICES)*
							Helicolenus				
6	*Haliporoides sibogae*	0.37	18.50	1.6	Baelde	35	*dactylopterus*	0.13	39.20	0.6	Ragonese
		0.49	3.35	1.2	Baelde	36	*Merluccius merluccius*	0.18	70.50	2.12	Ragonese
	Macrobrachium										
7	*macrobrachion*	1.21	14.14	9.53	Nwosu			0.29	49.40	2.12	Ragonese
		1.60	11.76	9.14	Nwosu	37	*Mullus barbatus*	0.60	23.50	2.3	Ragonese
8	*Callinectes sapidus*	1.09	2.35		Bunnell			0.65	20.20	2.3	Ragonese
9	*Palaemon gravieri*	0.80	5.80		Sunghan	38	*Mullus surmuletus*	0.48	29.00	1.7	Ragonese
		0.70	1.86		Sunghan			0.50	25.00	1.7	Ragonese
10	*Octopus maya*	1.40	25.20	8.77	Arreguin-Sanchez	39	*Pagellus erythrinus*	0.18	38.00	0.4	Ragonese
							Peristedion				
11	*Trypaea australiensis*	0.37	6.28	1.18	Rotherham	40	*cataphractum*	0.20	43.40		Ragonese
		0.98	8.25	3.76	Rotherham			0.18	45.60		Ragonese
					Pauly (Clark and						
12	*Penaeus plebejus*	4.16	24.50		Kirkwood)	41	*Phycis blennoides*	0.22	68.10	0.93	Ragonese
13	*Penaeus duorarum*	2.04	33.37		Pauly (Garcia)			0.38	47.10	0.93	Ragonese
									126.5		
		3.36	13.32		Pauly (Garcia)	42	*Raja clavata*	0.10	0		Ragonese
									116.7		
14	*Renaeus indicus*	4.13	17.80		Pauly (Marcille)			0.11	0		Ragonese
							Aristaeomorpha				
		4.48	7.62		Pauly (Marcille)	43	*foliacea*	0.50	6.58		Ragonese
15	*Pandalus borealis*	0.46	15.44	0.7	Clark (Terceiro)	44	*Nephrops norvegicus*	0.14	36.72	0.2	Ragonese
	Pandalus borealis	0.29	19.54		Clark (Apollonio)			0.13	42.86	0.2	Ragonese
							Parapenaeus				
	Pandalus borealis	0.36	16.98		Clark (Clark)	45	*longirostris*	0.71	4.09	2.5	Ragonese
	Pandalus borealis	0.39	16.98		Clark (Clark)			0.73	3.43	2.5	Ragonese
	Pandalus borealis	0.32	15.92		Clark (Terceiro)	46	*Epinephelus striatus*	0.12		0.55	Sadovy
									200.6		
	Pandalus borealis	0.45	14.09		Clark (Fournier)	47	*Epinephelus itajara*	0.13	0	0.85	Sadovy
16	*Penaeus semisulcatus*	1.77	22.40	8.18	Mehanna	48	*spec.*			1.4	Guénette
		1.56	26.80	6.77	Mehanna	49	*spec.*			2.44	Guénette
17	*Merlicertus Kerathurus*	1.03		2.26	Conides	50	*spec.*			0.54	Guénette
		1.03	30.91	2.26	Conides	51	*spec.*			2.24	Guénette
		1.25		2.26	Conides	52	*spec.*			2.54	Guénette
		1.25	27.42	2.26	Conides	53	*spec.*			0.68	Guénette
18	*Pandalus jordani*	0.04	2.69		Gotshall	54	*spec.*			0.6	Guénette
		0.06	2.57		Gotshall	55	*spec.*			0.4	Guénette
19	*Farfantepenaeus brasiliensis*	1.24	29.00	6.5	Leite	56	*Homarus gammarus*			0.15	Sheehy
20	*Farfantepenaeus paulensis*	1.34	27.50	8.8	Leite					0.08	Sheehy
21	*Egeria radiata*	0.36	9.30	0.82	Mose	57	*Oreochromis niloticus*	0.50	61.00	3	Kolding
22	*Hemisynodontis membranaceus*	0.62	44.50	4.39	Ofori-Dnason	58	*Penaeus semisulcatus*	1.60	16.43	4.6	Mohammed
				0.81							
23	*Oreochromis niloticus*	0.25	64.50	8	Getabu			1.70	20.75	4.8	Mohammed
							Monacanthus				
24	*Astacus leptodactylus*	0.41	8.08	2.16	Deval	59	*tomentosus*	0.86	11.79	2.033	Peristiwady
25	*Haliotis iris*	0.16	13.19	0.25	Sainsbury K.J.	60	*Pterogymnus laniarius*	0.13	37.94	0.36	Booth
26	*Lithognathus mormyrus*	0.21	30.94	0.79	Kallianiotis	61	*Acipenser medirostris*	0.05	23.80	0.19	Beamesderfer
27	*Cancer polyodon*	0.57	16.05	1.85	Wolff	62	*Scylla paramamosian*	2.39	15.00	13.32	Vay
		0.54	19.80	2.58	Wolff	63	*Cynoscion guatucupa*	0.23	53.00	0.64	Villwock
						64	*Procambarus clarkii*	0.68	13.52	4.94	Scalici
								0.69	12.14	3.19	Scalici

Parameter settings for Crangon crangon case study

Growth

For *C. crangon* L_∞ estimates of 78 mm (Kuipers & Dapper 1984) and 85 mm (Tiews 1989) have been reported. Thus L_∞ was varied in the simulations between 70 and 100 and a coefficient of variation was 5 %. The assumption of a 5% CV was based on several lines of reasoning: one is that if the variability chosen is too high, very large animals occur in the simulation which are not observed in the field. The largest animals caught within the German Demersal Young Fish survey during the last 10 years were mainly between 80 and 90 mm with only one exception in 2005 where an animal of 109 mm was caught (pers. comm. Neudecker). Taking 100 mm as L_∞ in the simulation and the three-fold standard deviation as maximum length criterion leads to simulated maximum size of 115 mm comparable to the field value. The second reason for choosing 5 % is the definition of Taylor (1985) who calculated L_∞ as maximum length in the catch divided by 0.95.

The parameter k was chosen between 1 and 2.5 and the variability of k was chosen to be 30% (Hufnagl & Temming submitted).

Total mortality Z and seasonality of Z

Temming et al (1993) determined total mortalities (Z) for *C. crangon* ranging from2.78 to 6.08 but even high values have been reported including 8.7 (2003/4) and 10.9 (2004/5) (Viegas et al. 2007). Values of Z between 2 and 11 were chosen for the simulations.

For the simulation of the artificial *C. crangon* length frequency distributions mortality levels were adjusted seasonally to match the abundance of the predators of large (>45 mm) *C. crangon* which are mainly 0 and I-group cod and whiting (Hislop et al. 1991, Daan 1989, Daan 1973). Therefore, a higher mortality was assumed in fall (October +80%) and lower in spring (April -80%) following (Hamerlynck & Hostens 1993). The focus is set only on the large animals as the applied methods to determine total mortality are dependent on L_c. In the surveys that monitor the brown shrimp L_c is ~45 mm. In the actual study no length dependent decrease in mortality was assumed which might be present as predators for small shrimps are more numerous than for adult shrimps.

Recruitment

The recruitment pattern was calculated as described in Temming & Damm (2002). First, an egg index representing the amount of eggs present in the field was calculated based on the amount of egg bearing females, the amount of eggs carried per female, the temperature-dependent moult interval, and an index shrimp density. Temperature-dependent egg development was then calculated based on the function presented by Redant (1978).

Sampling date

There are three, main surveys that target *C. crangon*: the Demersal Fish Survey (DFS) - Netherlands, the Demersal Young Fish Survey (DYFS) - Germany, and the Bycatch Series (BS) - Germany. Although the BS, is based upon samples taken during the entire year from commercial catches (Meyer-Waarden & Tiews 1965),the DFS and DYFS are mainly conducted in the autumn. Due to the differences in these surveys, he influence of catch date on the mortality estimation from such season-specific data was tested. In the first scenario, a mean length frequency distribution of the whole year was calculated and in the second approach only the length frequency distribution calculated for autumn (day 280) was used to estimate Z and L_∞.

Results

Due to the stepwise changes of L_∞, k and Z to cover the range of these parameter values, each run resulted in 8000 θ and L_∞ estimates. Not all of these results can be shown here and thus only boxplots illustrating the deviation of the estimated values (L_∞, θ) with the input values assumed in the model are shown. The deviation is shown as percentage and a value of 100 denotes that the method determines exactly the true value assumed in the simulation. Outliers and extreme values are taken into account in the analysis but are not shown in the graphs for clearness. The boxes show the 0.25 and 0.75 quartile, the line represents the median and the bars (whiskers) indicate the largest and smallest values which are not outliers or extreme values. Outliers are all values between 1.5 and 3 times the box-length and extreme values are those with more than 3 box-length difference.

More detailed contour plots for each method and run are available as supplementary material and is available at *ICESJMS* online. These graphs are d are cited as Figure S1, S2 etc. In these graphs green colour relates to 100% conformity, blue to values which are 200% ore more below and red to those 200% or more above the known input values of L_∞ and θ.

Evaluation of L_∞ estimation

Only two of the tested methods can also be applied to estimate L_∞: the methods of Powell and the modified Wetherall et al. method. In nearly all model runs the Wetherall method slightly underestimated the given L_∞, but generally the deviation from the real L_∞ is lower than 20% (Figure 8-2). Only in run 48 and 49 where L_∞ and k were varied (20, 30%) the input L_∞ is clearly overestimated by about 30 and 50%. Accuracy of the L_∞ estimation is only slightly influenced by the level of L_∞. Only for $L_\infty < 60$ and $\theta > 20$ ($Z > 4$ and $k < 0.5$) was L_∞ underestimated by up to 80%.

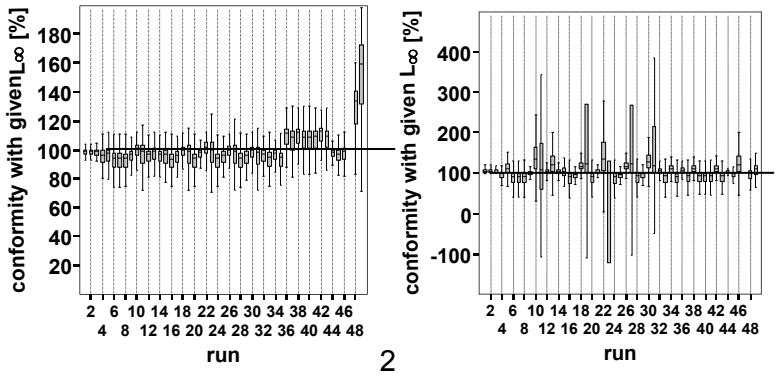

Figure 8-2: Boxplot of the conformity of the estimates with the given values in %. 1: modified Wetherall et al. method, 2: Powell method. On the x-axis the number of the scenario run is plotted (detailed information in Table 8-1).

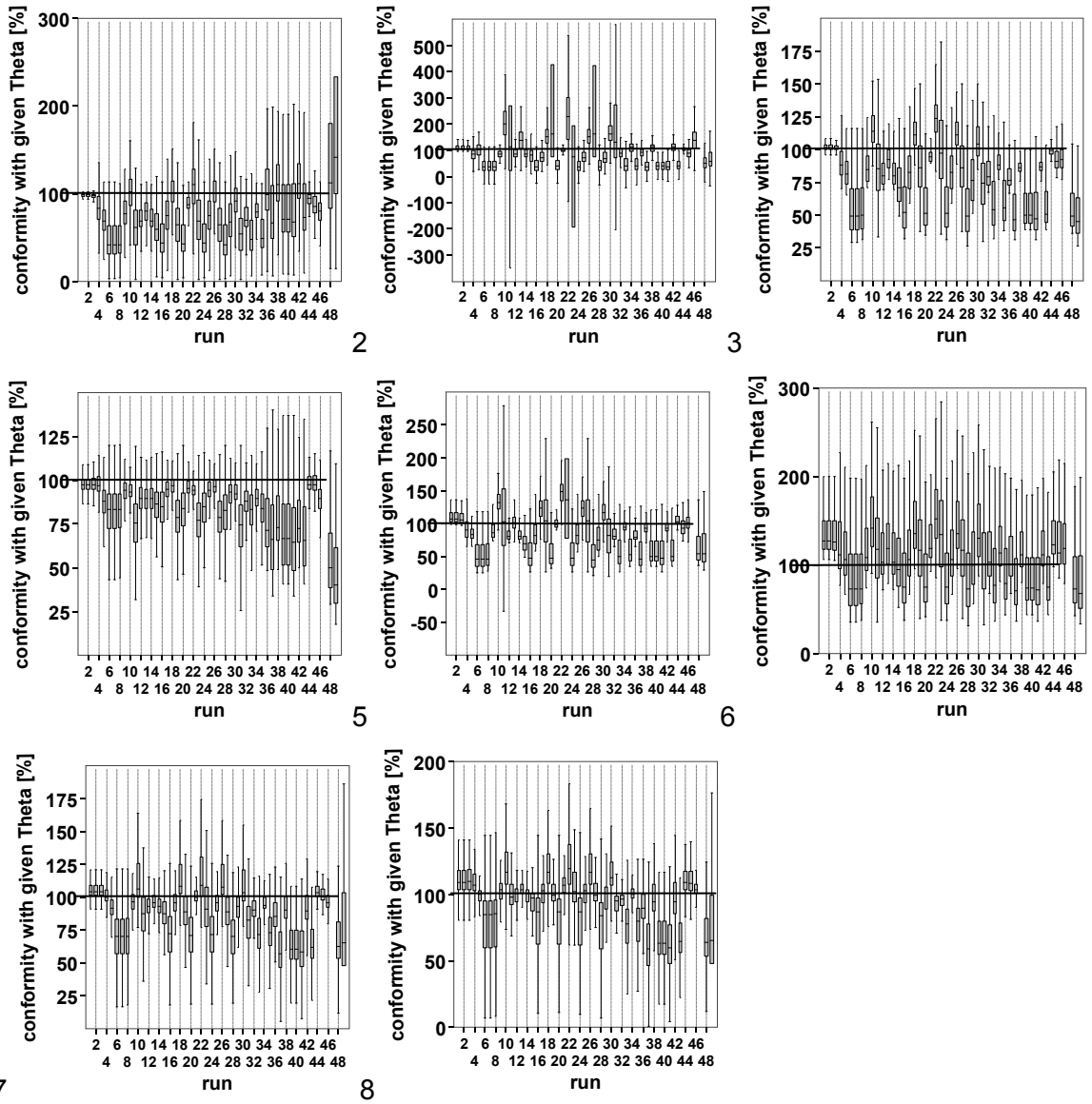

Figure 8-3: Boxplots showing the conformity of the estimates with the given values in % according to the different methods (1: modified Wetherall et al., 2: Powell, 3: Beverton & Holt, 4: Jones & Zalinge, 5: Hoenig, 6: Ssentongo & Larkin, 7: Length Converted Catch Curve, 8 seasonal Length Converted Catch Curve). On the x-axis the number of the scenario run is plotted (detailed information in Table 8-1).

For all runs, the Powell method predicted median values close to 100%, varying mainly between 80 and 120% (Figure 8-2). However, in the scenarios based on winter recruitment (11, 19, 23, 27, 31, Table 8-1), deviations often exceeded 300%. In those runs with no seasonal recruitment L_∞ is slightly overestimated by 10 to 30% whereas in the remaining runs L_∞ is underestimated by about 10 to 20%. If recruitment peaks are simulated in spring or summer, accurate estimates can only be calculated if $\theta<10$. The parameter range where results deviate less than 5% from the input values is $Z < 4$ and $k > 2$.

Evaluation of θ estimation

The modified Wetherall method exhibited similar deviation patterns when θ or L_∞ is determined. The same was observed for the Powell method. The variability of the estimates is higher for the Powell method while the modified Wetherall function consistently underestimated the real θ (Figure 8-3). The highest deviations for θ estimated with the Wetherall method were determined within run 48 and 49 (with 20 and 30% variation of k and L_∞) and for the Powell method in those runs where seasonal recruitment peaks in winter (deviations of more than 200%). The accuracy of the Wetherall method did not depend on the level of L_∞ but was sensitive to the actual value of θ (k as well as Z). Although low values of θ were accurately estimated, values were progressively overestimated with increasing values of θ. For example, for $\theta \sim 20$, the estimated value was about 40-80% lower than the assumed values. The influence of k and Z levels on the estimation of θ increased with increasing values of the standard deviation of k and L_∞. Therefore, with variability in growth parameters, accurate estimates are only possible under specific combinations ($Z > 2.5$ and $1 < k < 2.5$, supplement Figures). The Powell method underestimated the real θ in nearly every run. This method was extremely sensitive to the level of k and Z and the combination of theses two parameters. If recruitment peaks in spring or summer, input values of $k > 2$ and $Z < 3$ lead to an underestimation of θ by 10 to 40%. The smaller the input k and the higher the input Z the higher the deviation of the estimated θ (deviation of more than 80%).

The Beverton & Holt method exhibited the highest deviations in estimates in runs where the recruitment maximum was located in the spring. In contrast, accurate estimates were available from runs with winter recruitment while overestimates of θ were apparent in runs where the recruitment maximum was in the autumn. In all cases, best estimates were achieved with the Beverton & Holt method for $Z < 3$. If recruitment takes place in winter or spring, the deviation of the estimated θ from the real θ is about -40 to -60% for $Z>2$. L_∞ has only a low influence on the accuracy of the estimate.

In most cases, the Jones & Zalinge method underestimated θ with median estimates that were about 20% lower than the actual value. Variability in L_∞ lead to estimates of θ of that were about half of the given value. The Jones & Zalinge method underestimated the true θ values when winter and spring recruitment was assumed whereas summer and fall recruitment led to accurate estimates. With

increasing k and Z in the simulations, this method becomes less accurate and θ is underestimated by about 20% for Z > 6 and k > 2. Variations of k with a standard deviation of 10% exhibited only limited influence but variations of L_∞ (10%) lead to an underestimation of θ by 20%.

Similar to the other methods, The Hoenig method exhibited a conformity of nearly 100% when non-seasonal growth was assumed and was (in this special case) not dependent on seasonal recruitment or mortality. Within those runs where growth was assumed to be seasonal, highly fluctuating estimates for the different k, Z and L_∞ combinations were determined when recruitment took place in winter. Spring recruitment lead to an underestimation of the known input θ by 50%. Recruitment in summer and fall resulted in accurate estimates. For small animals (input L_∞ < 100) θ values deviate from the true θ by more then 100% if the simulated θ was above 15. When recruitment was in summer or fall, the best results were achieved for Z > 3 but are still overestimated by 20 to 40%.

The Ssentongo & Larkin method generally exhibited greater deviations from the known input θ values compared to the other methods and 100% overestimation of θ was observed in nearly every run. Like most of the other methods, spring recruitment lead to underestimates and fall recruitment to overestimates of θ by (median) 30%. Even when non-seasonal growth was assumed, the median of the calculated θ was about 25% higher than the known value. Seasonal mortality has only little influence on the accuracy of the method. The best results were achieved when 2 < Z < 6 and 1 < k < 3 (supplement Figures) where estimates deviate ± 20% from the true value. The exception was for fall recruitment where estimates improve with increasing Z and decreasing k. Then estimates are exactly the true value for k < 2 and Z > 4.

The results of the Miranda method were very similar to those of the modified Wetherall method and, therefore, these results will not be discussed and shown explicitly.

The results of the seasonal and the non-seasonal Length Converted Catch Curve were very similar to one another. In both methods, spring recruitment lead to underestimates of θ by 30% (median) and to a greater variability of predicted values whereas fall recruitment lead to a slight overestimation of θ (5 to 10%). In scenarios where recruitment was in the autumn or winter, θ was strongly overestimated by more than 100% if Z was smaller than 2. In nearly all runs, if k < 2 and Z > 2 the estimated θ deviates less then 10% from the true θ.

Evaluation of the model when applied for *Crangon crangon*

Evaluation of L_∞ estimation

L_∞ is estimated accurately with the modified Wetherall et al. and Powell methods no matter if samples were only collected in the autumn (Figure 8-4, Figure 8-5) or during the whole year (Figure 8-7, Figure 8-8). For small k, L_∞ was slightly underestimated (5%) with the modified Wetherall method, whereas it was

overestimated by 10% with the Powell method. Plotting the calculated L_∞ over the known L_∞ indicated that estimates of the Powell method highly fluctuate when L_∞ > 90 (Figure 8-5). It was possible to determine L_∞ values of up to 110 if the true value was 70. Additionally, if samples were only available from the autumn L_∞ was overestimated for Z = 5 by 20% with the Powell method. Plotting the calculated L_∞ from the modified Wetherall method against the true input values indicated that calculated values below 70 mm underestimate the true value by about 15%.

Evaluation of θ estimation

Generally, estimates of θ based on the modified Wetherall and Powell methods had the same patterns of variability as estimates of L_∞. Variability in the results and deviations from the assumed values was higher (Figure 8-4, Figure 8-7). If samples were only available from the autumn, θ was overestimated by 30% for Z = 3, slightly overestimated (up to 10%) for 3 < Z < 5 and underestimated (up to50%) with the modified Wetherall method. In comparison, the Powell method was less sensitive to Z but more sensitive to k. When applying Powell's method, underestimates θ increased (up to 50%) with increasing values of K.

The median of the Beverton & Holt model applications underestimated the input θ by 30 % based on only autumn samples (Figure 8-4, Figure 8-6) whereas the estimation improved if samples were available during the entire year (Figure 8-7). Overestimates occurred using the Beverton & Holt model when θ was > 6 (Figure 8-9).

Estimating θ with the Jones & Zalinge method leads to consistent underestimation of the input θ by about 40%. Different levels of Z, k and L_∞ do not influence this bias of the estimates (Figure 8-4, Figure 8-7).

The Hoenig, Ssentongo & Larkin and the nonseasonal Length Converted Catch Curve only slightly underestimated θ with increasing Z. The underestimation was more pronounced for the seasonal Length Converted Catch Curve and increased with increasing θ (Figure 8-4, Figure 8-7).

The seasonal Length Converted Catch Curve generally underestimated the input θ. This was especially the case when samples were only available from the autumn and if k is smaller than 1.5 and Z is larger than 5 (Figure 8-4).

From these correlations between predicted and "real θ" (Figure 8-5, 6, 8 and 9), correction functions are available for estimating θ from field length frequency distributions of *C. crangon*. The θ and the L_∞ range as well as the parameters of the correlations are shown in Table 8-4.

Autumn sampling

Calculated θ values from fall data were between 2 to 30% lower (Table 8-3) than θ estimates based on whole year data (Calculated as deviation = $(\theta_{fall} / \theta_{year}) \cdot 100 - 100$). The method less biased by the time of sampling is the Jones & Zalinge (-2%)

and the Length converted catch curve (-7%). Highest deviation and also the highest variability within the results were observed for the Powell and the Hoenig methods with -28 ± 27% and -20 ± 45%, respectively.

Table 8-3: Deviation = (θ_{fall} / θ_{year}) ·100 -100 of calculated θ values based on fall data in comparison to θ calculated with whole year data.

	mean deviation	**standard dev.**
L_∞ mod. Wetherall	-1.67	1.83
L_∞ Powell	-1.06	58.43
seasonal length converted catch curve	1.85	7.73
length converted catch curve	1.86	8.64
Ssentongo & Larkin	-0.52	10.17
Beverton & Holt	-0.73	12.43
mod. Wetherall	-4.37	11.90
Jones & Zalinge	6.50	10.32

Table 8-4: Range of application (min, max derived from Fig. 5,6,8 and 9) and parameters (a=slope, b=intercept) of the linear correction functions. The determined functions correct estimated θ for the bias that occurs when seasonal growth, recruitment and mortality as observed for *Crangon crangon* alter the results of length frequency based mortality methods (Figure 8-5 and Figure 8-6). Regressions were all significant (p<0.001)

	samples available over the year					**samples available from fall**				
	min	**max**	**a**	**b**	**r²**	**min**	**max**	**a**	**b**	**r²**
L_∞ Wetherall	70	105	0.92	4.30	0.98	65	105	0.82	13.49	0.96
L_∞ Powell	70	95	1.05	-7.56	0.94	70	90	1.24	-18.52	0.97
θ Wetherall	1	6	1.38	-0.59	0.99	1	6	0.78	1.59	0.90
θ Powell	1	6	0.85	1.11	0.96	1	8	0.89	1.00	0.89
θ Beverton & Holt	1	7	1.34	0.21	1.00	1	8	1.10	0.83	0.99
θ Jones Zalinge	1	7	1.41	0.74	0.97	1	7	1.09	1.23	0.97
θ Hoenig	2	10	1.04	0.63	0.93	1	6	0.95	0.92	0.93
θ Ssentongo & Larkin	1	8	1.33	-0.39	1.00	1	9	1.08	0.36	0.99
θ Length Converted Catch Curve	1	6	1.60	-0.63	1.00	1	8	1.24	0.24	0.99
θ seasonal Length Converted Catch Curve	1	6	1.90	-1.08	1.00	1	6	1.56	-0.30	0.98

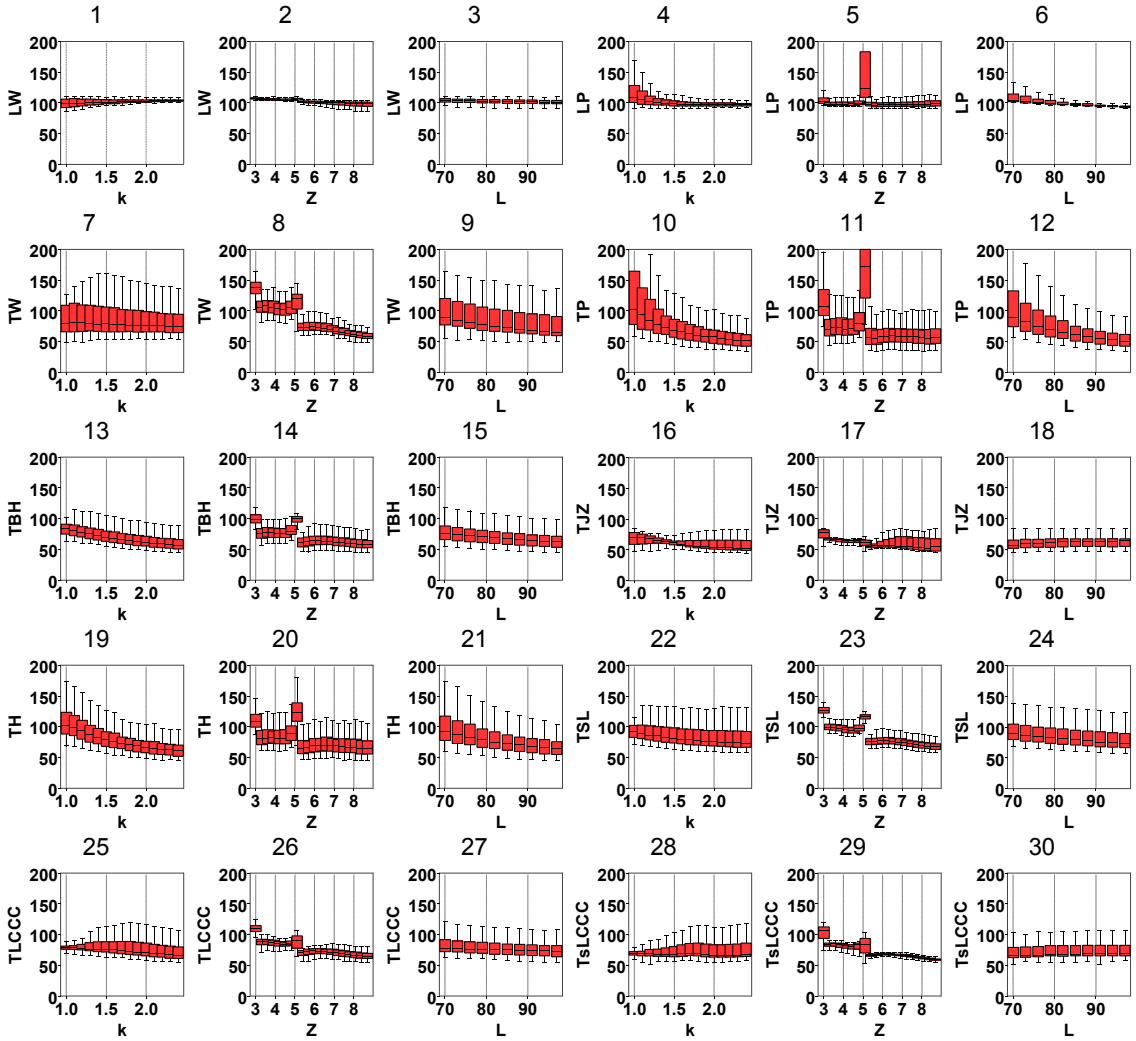

Figure 8-4: Mean deviation of the calculated θ from the assumed θ in dependency of growth rate k, total mortality Z and mean maximum length L_∞. 100 is a 100% accordance of the calculated value with the assumed value. (1-3) L_∞ modified Wetherall et al. (LW); (4-6) L_∞ Powell (LW); (7-9) θ modified Wetherall (TW); (10-12) θ Powell (TP); (13-15) θ Beverton & Holt (TBH; (16-18) Jones & Zalinge (TJZ; (19-21) Hoenig (TH); (22-24) Ssentongo & Larkin (TSL); (25-27) the nonseasonal (TLCCC); and (28-30) seasonal Length Converted Catch Curve (TsLCCC) were applied on artificial length frequency distributions simulating *Crangon crangon* and samples that are only available from fall.

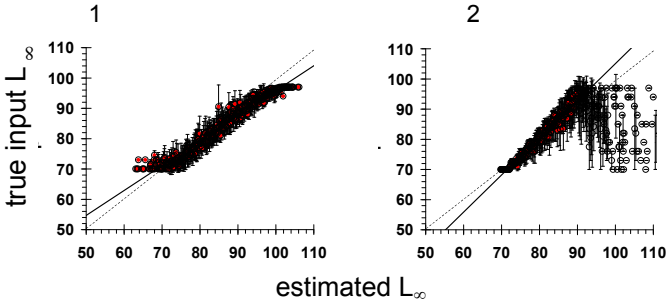

Figure 8-5: Deviation of the estimated L_∞ from the true input L_∞ med for generating the length frequency distribution based on *Crangon crangon* growth, mortality and recruitment parameters. Applied methods: (1) modified Wetherall et al., (2) Powell. Samples were assumed to be only available from fall.

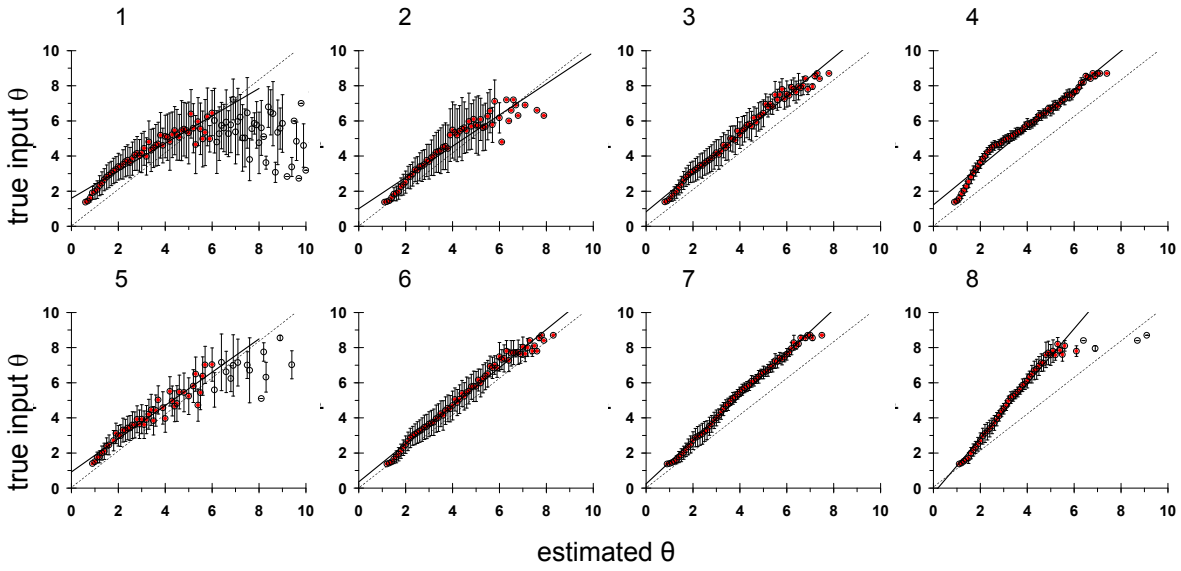

Figure 8-6: Deviation of the estimated θ from the true input θ used for generating the length frequency distribution based on *Crangon crangon* growth, mortality and recruitment parameters. Applied methods: (1) modified Wetherall et al., (2) Powell, (3) Beverton & Holt, (4) Jones & Zalinge, (5) Hoenig, (6) Ssentongo & Larkin, (7) nonseasonal and (8) seasonal Length Converted Catch Curve. Samples were assumed to be only available from fall.

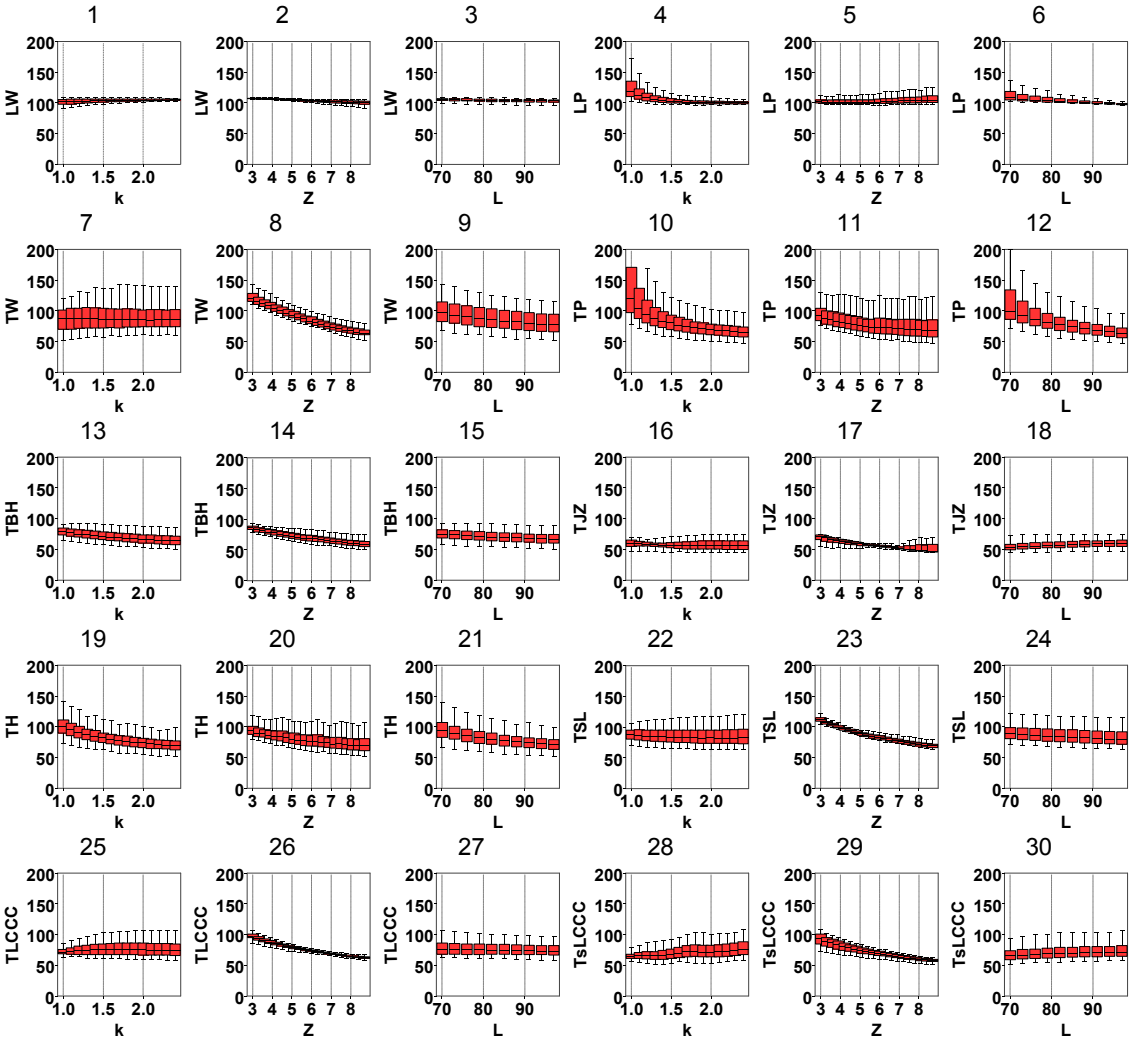

Figure 8-7: Mean deviation of the calculated θ from the assumed θ in dependency of growth rate k, total mortality Z and mean maximum length L_∞. 100 is a 100% accordance of the calculated value with the assumed value. (1-3) L_∞ modified Wetherall et al. (LW); (4-6) L_∞ Powell (LW); (7-9) θ modified Wetherall (TW); (10-12) θ Powell (TP); (13-15) θ Beverton & Holt (TBH; (16-18) Jones & Zalinge (TJZ; (19-21) Hoenig (TH); (22-24) Ssentongo & Larkin (TSL); (25-27) the nonseasonal (TLCCC); and (28-30) seasonal Length Converted Catch Curve (TsLCCC) were applied on artificial length frequency distributions simulating *Crangon crangon* and samples that are available from the whole year.

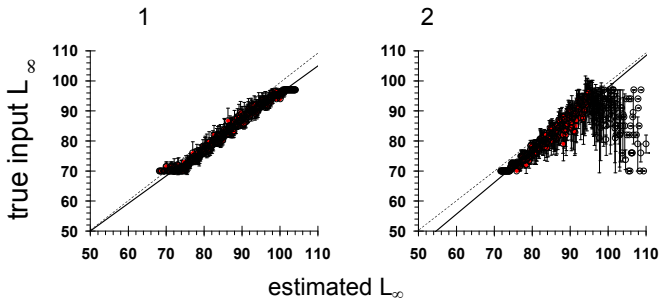

Figure 8-8: Deviation of the estimated θ from the true input θ used for generating the length frequency distribution based on *Crangon crangon* growth, mortality and recruitment parameters. Applied methods: (1) modified Wetherall et al., (2) Powell, (3) Beverton & Holt, (4) Jones & Zalinge, (5) Hoenig, (6) Ssentongo & Larkin, (7) nonseasonal and (8) seasonal Length Converted Catch Curve. Samples were assumed to be available from the whole year.

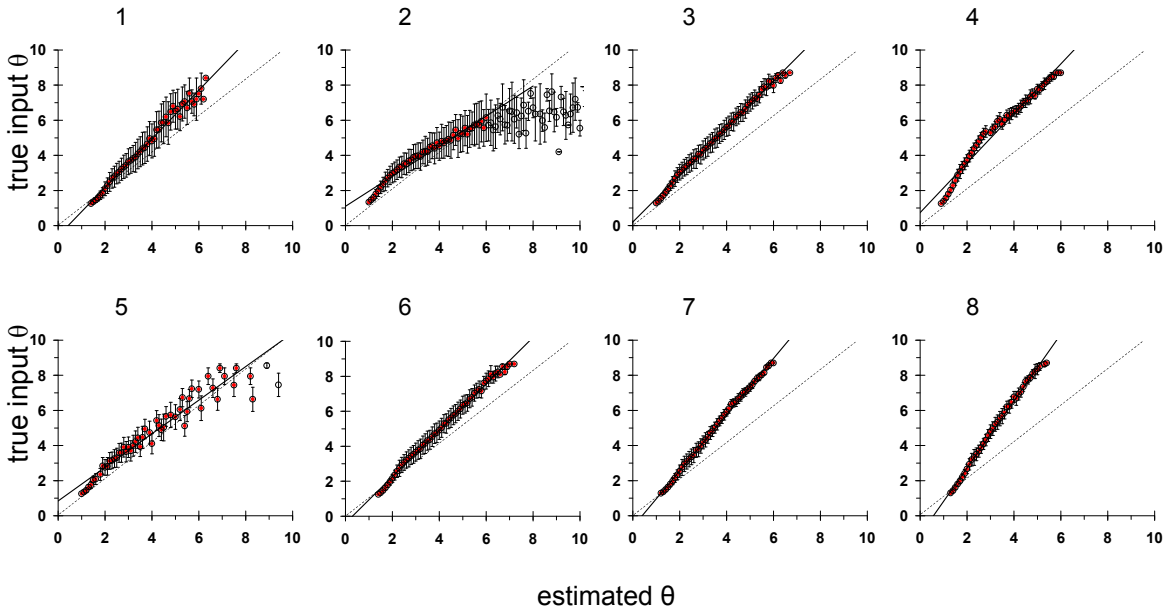

Figure 8-9: Deviation of the calculated θ from the θ assumed for generating the length frequency distribution based on *Crangon crangon* growth, mortality and recruitment parameters. Applied methods: (1) modified Wetherall et al., (2) Powell, (3) Beverton & Holt, (4) Jones & Zalinge, (5) Hoenig, (6) Ssentongo & Larkin, (7) nonseasonal, (8) and seasonal length converted catch curve. Samples were assumed to be available from the whole year.

Discussion

General Problems of all methods

Ralston (1989) stated that the Beverton & Holt estimator is stable over a broad range of simulated conditions according to seasonal recruitment. This can only partially be confirmed in this work as our results indicated a general underestimation of θ when applying the Beverton & Holt method on length frequency distributions for populations having recruitment maximum in spring. Erhardt & Ault (1992) demonstrated that the Beverton & Holt estimator was highly biased when the maximum age in the catch was less than the maximum age in the field. Although this has not been tested in this work,it can be compared with the effect of seasonality in recruitment, as both factors skew the length frequency distribution towards a lower mean length (discussed in the next section).

All seven methods were particularly sensitive to seasonal growth and seasonal recruitment and, to a lesser extent, seasonal mortality. Seasonal growth generally leads to an underestimation of θ, and this effect is increased if recruitment is maximal in spring. On the other hand, autumn recruitment generally leads to an overestimation of θ. For a better understanding of these results one must consider that all methods are based on the mean or median length of the catch and how that value is related to L_∞. If recruitment takes place in spring, the juveniles immediately experience high temperatures and growth performance is high. Consequently the mean length distribution over the whole year shifts towards L_∞. On the other hand, if recruitment takes place in the autumn or winter, animals

remain small throughout the winter and, therefore, the annual mean length is shifted towards L_c. In both cases, the same mortality was assumed but the resulting mean length will be different and therefore θ will be either over- or underestimated, respectively.

There is a general trend in all methods to underestimate θ and this is especially problematic in assessments of stock exploitation. However they have been used extensively to calculate θ and with it Z of marine animals (e.g. see Table 8-2) especially with the FISAT and ELEFAN routines (Gayanilo et al. 1996). These programs include among other the Wetherall et al and the Beverton & Holt method. The results shown here might help to improve mortality and L_∞ estimates. For future studies of populations with seasonally varying growth and recruitment, it is recommended that simulations are performed similar to the one performed here on C. crangon, in order to quantify the case-specific bias and find the best method for estimating the target population.

Advantages and disadvantages of single methods

L_∞ estimates

For estimating L_∞, the modified Wetherall et al. method is the better choice over the Powell method. It generated robust estimates and is insensitive to seasonality in recruitment, mortality and growth. Increased bias might arise when intra-cohort variation of L_∞ is high since large animals occurring in catches would alter the regression in the Wetherall method. These outliers strongly influence the left term of (35) for large L' and thus the slope of the regression and subsequently L_∞ increases.

From field samples, the CV of L_∞ can be estimated from length at age data but, in general, such data do not exist for most invertebrates. Taylor (1958) suggested that L_∞ be calculated using 0.95 times the maximum length observed in all catches of a target species. A standard deviation of L_∞ of 5% might therefore be a good estimate of the CV of L_∞. Seabream length at age data are presented in (Domínguez-Seoane et al. 2006) and for the older age classes a CV of 1.4 to 3.3 % can be calculated. For demersal cod and juvenile salmonids a coefficient of variation L_∞ of about 5% was determined (Gurney et al. 2007). In stocks with a 5% cov of L_∞ and lower our results suggest that the Wetherall et al method can be applied and estimates will only be slightly biased.

Higher cov of L_∞ are imaginable, for example in food limited or density-regulated stocks since the share of "winners" and "losers" will be increased in these stocks. Furthermore, sex-specific growth rates and sex-dependent maximum sizes exist in the field (Sainte-Marie et al. 2006) and if these are not included in analyses high variability in L_∞ can be expected, depending on the actual mixture of the two genders. In this cases, Wetherall et al method will most likely underestimate L_∞ by 40% or more.

The Powell method is mainly influenced by seasonal recruitment with maximum deviation of the estimated θ from the true θ value if winter recruitment is simulated. Our simulations suggested that, under such conditions, L_∞ estimations can deviate from the true value of by 300% or more. This is mainly caused by the use of the variance of the length in the catch in the Powell method. If, for example, Z is high and k is small, a large variance in the simulated length frequency distribution is generated as mainly the young cohorts influence the length frequency distributions. Similarly, the mean length of the length frequency distribution is low in comparison to L_∞ as mortality is high. In combination, this leads to small nominator values in (33) and (34) and, consequently, to a large value added to mean length $\overline{L}$. This will increase the estimated L_∞ and θ in comparison the real value. The Powell method should not be applied when a high θ is expected that might exist, for example,, in slow-growing, highly-exploited species.

θ estimates

Under non-seasonal conditions, only the Ssentongo & Larkin method are likely to overestimate θ. In most cases, θ was underestimated, particularly when the recruitment maximum is in spring or summer and seasonal growth is present. The restrictions and problems discussed in the previous section are comparable when applying the Powell or Wetherall method to estimate θ.

For all other methods except the Jones & Zalinge method, the problems discussed in the general section represent the main limitations. Recruitment maximum in spring leads to an underestimation, and recruitment in fall and winter to an overestimation of θ. Different combinations of k, L_∞ and Z produced the highest variability in the Ssentongo & Larkin methods. If the method generates good estimates for a species with given k, L_∞ and Z values, it might be significantly biased for another species with only slightly different k, L_∞ and Z values. This implies that the Ssentongo & Larkin method, similar to the Powell method, should be carefully evaluated for each target species prior to application.

Mortalities are generally underestimated by 25% using the Jones & Zalinge method (Figure 8-2). This negative bias increases further with increasing variation (cov) of k and L_∞, for similar reasons as discussed for the Wetherall method.

Case study *Crangon crangon*

As expected from the results of run 25 and 43, which are closest to the conditions of *C. crangon*, the Wetherall and Jones & Zalinge method underestimate θ (cov k), the Beverton & Holt method and the non-seasonal Length Converted Catch Curve produce accurate θ estimates, and the seasonal LCCC underestimates θ. For the Powell and Ssentongo & Larkin methods, the bias depends upon the parameter values chosen for k and Z.

The Ssentongo & Larkin method turned out to be the most accurate method in the case study where samples were collected throughout the year. Problems with that

method were only observed for increasing Z values > ~ 6 resulting in up to 30% bias.

The same response to increasing values of Z can also be observed for the other methods whereas k values in the range of 1 to 2.5 and L_∞ values, in the range of 70-100, did not influence the estimates, with exception of the Powell and Hoenig method. Similar results were obtained if length frequency data were only available from the autumn. In this special case, differences mainly occurred for the Powell and the modified Wetherall methods where estimates of θ were more accurate if they were based on one sampling in autumn.

Possible correction of occurring bias

As all of the methods either under- or overestimate θ, simple methods were designed to correct for model bias (Figure 8-9).

The Ssentongo & Larkin method, the Beverton & Holt and the LCCC (seasonal and nonseasonal) exhibited a constant and linear deviation from the true input θ. This result can be used to correct the bias to achieve a more accurate estimate of θ.

The remaining methods: Powell, Hoenig and the modified Wetherall et al methods exhibited no linear deviation over the whole parameter range. For these methods, an estimated θ of 8 can be a true θ of 3 to 7 if samples are taken in fall and a θ of > 10 if samples are taken during the whole year. Moreover L_∞ should not be estimated with the Powell method if values larger than 90 are determined whereas, on the other hand, the results of modified Wetherall et al. method should be neglected if values < 60 are estimated.

The seasonal Length Converted Catch Curve deviates more strongly from the real values than the non-seasonal version. The reason is that the seasonal LCCC can only account for seasonal growth for one main cohort. If several cohorts start at different dates in the year, the main cohort is estimated correctly, but estimates for all cohorts that start earlier or later are biased. *C. crangon* spawns over an extended period and, therefore, no one single recruitment event can be observed (Temming & Damm 2002, Neudecker & Damm 1992, Kuipers & Dapper 1984). This leads to an underestimation of the real θ when the seasonal LCCC is used.

Acknowledgements

The study was financially supported by the Federal Ministry of Food, Agriculture and Consumer Protection, Germany Project No. 03HS030.

References

Admassu D (1996) The breeding season of tilapia, *Oreochromis niloticus* L. in Lake Awassa (Ethiopian rift valley). Hydrobiologia 337:77-83

Akyol O (2001) Some biological properties and stock estimates of *Zeus faber* L., 1758 (Pisces, Zeidae) in the Aegean Coasts of Turkey. E.U.Journal of Fisheries & Aquatic Science 18:39-46

Amezcua F, Soto-Avila C, Green-Ruiz Y (2005) Age, growth, and mortality of the spotted rose snapper *Lutjanus guttatus* from the southeastern Gulf of California. Fisheries Research 77:293-300

Arreguín-Sanchéz F, Solís-Ramírez MJ, González de la Rosa M (2000) Population dynamics and stock assessment for *Octopus maya* (Cephalopoda: Octopodidae) fishery in the Campeche Bank, Gulf of Mexico. Revista de Biologia Tropical 48:323-331

Baelde P (1994) Growth, mortality and yield-per-recruit of deep-water royal red prawns (*Haliporoides sibogae*) of eastern Australia, using the length-based MULTIFAN method. Marine Biology 118:617-625

Beamesderfer RCP, Simpson ML, Kopp GJ (2007) Use of life history information in a population model for Sacramento green sturgeon. Environmental Biology of Fishes 79:315-337

Beukema JJ (1992) Dynamics of juvenile shrimp *Crangon crangon* in a tidal flat-nursery of the Wadden Sea after mild and cold winters. Marine Ecology Progress Series 83:157-165

Beverton RJH, Holt SJ (1956) A review of methods for estimating mortality rates in fish populations, with special reference to sources of bias in catch sampling. Rapport et Procès-verbaux des Réunions, Conseil international pour L'Exploration de la Mer 140:67-83

Booth AJ, Buxton CD (1997) The biology of the panga, Pterogymnus laniarius (Teleostei: Sparidae). Environmental Biology of Fishes 49:207-226

Burton ML (2001) Age, growth, and mortality of gray snapper, *Lutjanus griseus*, from the east coast of Florida. Fisheries Bulletin 99:254-265

Campos, W. L. and Berkeley, S. A. Impact of the commercial fishery on the population of Bait Shrimp (Penaeus spp.) in Biscayne Bay 1986. NOAA Technical Memorandum NOS NCCOS CCMA 165, 1-34. 2003. NOAA/University of Miamay Joint Publication, University of Miami RSMAS TR 2003-02.

Clark SH, Cadrin SX, Schick DF, Diodati PJ, Armstrong MP, McCarron D (2000) The Gulf of Maine Northern Shrimp (*Pandalus borealis*) fishery: a review of the record. Journal of Northwest Atlantic Fishery Science 27:193-226

Conides A, Glamuzina B, Jug-Dujakovic J, Papaconstantinou C, Kapiris K (2006) Age, growth, and mortality of the karamote shrimp *Melicertus kerathurus* (Forskal, 1775), in the East Ionian Sea (western Greece). Crustaceana 79:33-52

Daan N (1973) A quantitative analysis of the food intake of north sea cod, *Gaduas morhua*. Netherlands Journal of Sea Research 6:479-517

Daan, N. ed. Data base report of the stomach sampling project 1981. Cooperative research report 164, 144. 1989.

Domínguez-Seoane R, Pajuelo JG, Lorenzo JM, Ramos AG (2006) Age and growth of the sharpsnout seabream *Diplodus puntazzo* (Cetti, 1777) inhabiting the Canarian archipelago, estimated by reading otoliths and by backcalculation. Fisheries Research 81:142-148

Erhardt NM, Ault JS (1992) Analysis of Two length-based mortality models applied to bounded catch length frequencies. Transactions of American Fisheries Society 121:115-122

Gayanilo FC, Sparre P, Pauly D (1996) The FAO-ICLARM stock assessment tool (FiSAT) user's guide. FAO computerized information series / Fisheries 8:-126

Gonzáles-Gordillo JI, Arias AM, Rodriguez A, Drake P (2003) Recruitment patterns of decapod crustacean megalopae in a shallow inlet (SW Spain) related to life history strategies. Estuarine Coastal and Shelf Science 56:593-607

Gotshall DW (1972) Population Size, Mortality Rates, and Growth Rates of Northern Californi Ocean Shrimp, *Pandalus jordani*, 1965 through 1968. Californian Fish Bulletin 155 :1-47

Guénette, Sylvie and Morato, Telmo. The Azores,Archipelago, 1997. Guénette, S., Christensen, V., and Pauly, D. Fisheries Impact on North Atlantic Ecosystems: Models and Analysis, 241-270. 2001. Fisheries Centre, University of British Columbia, Vancouver, BC, Canada Vol. 9 (4) 344 pp.

Gurney WSC, Tyldesley G, Wood SN, Bacon PJ, Heath MR, Youngson A, Ibbotson A (2007) Modelling length-at-age variability under irreversible growth. Canadian Journal Fisheries and Aquatic Science 64:638-653

Hamerlynck O, Hostens K (1993) Growth, feeding, production, and consumption in 0-group bib (*Trisopterus luscus* L.) and whiting (*Merlangius merlangus* L.) in a shallow coastal area of the south-west Netherlands. ICES Journal of Marine Science 50:81-91

Hewitt DA, Lambert DM, Hoenig JM, Lipicius RN (2007) Direct and indirect estimates of natural mortality for Chesapeake Bay blue crab. Transactions of American Fisheries Society 136:1030-1040

Hislop JRG, Robb AP, Bell MA, Armstrong DW (1991) The diet and food composition of whiting (*Merlangius merlangus*) in the North Sea. ICES Journal of Marine Science 48:139-156

Hoenig JM, Lawing WD, Hoenig NA (1983) Using mean age, mean length and median length data to estimate the total mortality rate. International Council for the exploration of the Sea ICES-CM-1983:

ICES (2007) Report of the Working Group on Crangon Fisheries and Life History. International Council for the exploration of the Sea Living resources comittee:1-33

Jansen, Soenke. Das Räuber-Beutesystem juveniler Gadiden, Grundeln und Garnelen im Wattenmeer nördlich von Sylt. 1-155. 2002. Thesis, Institute for Hydrobiology and Fisheries Science, University Hamburg.

Jennings S, Reynolds JD (2001) Marine Fisheries Ecology. Blackwell Publishing, London

Jones R, van Zalinge NP (1981) Estimates of mortality rate and population size for shrimp in Kuwait waters. Kuwait Bulletin of Marine Science 2:273-288

Kallianiotis A, Torre M, Argyri A (2005) Age, growth, mortality, reproduction, and feeding habits of the striped seabream, Lithognathus mormyrus (Psices: Sparidae), in the coastal waters of the Thracian Sea, Greece. Scientia Marina 69:391-404

Kim S (2004) Population structure, growth, mortality, and size at sexual maturity of *Palaemon gravieri* (Decapoda: Caridea: Palaemonidae). Journal of Crustacean Biology 25:226-232

Koch V, Wolff M, Diele K (2005) Comparative population dynamics of four fiddler crabs (Ocypodidae, genus *Uca*) from a North Brazilian mangrove ecosystem. Marine Ecology Progress Series 291:177-188

Kolding J (1993) Population dynamics and life-history styles of Nile tilapia, *Orechromis niloticus*, in Ferguson´s Gulf, Lake Turkana, Kenya. Environmental Biology of Fishes 37:25-46

Kuipers BR, Dapper R (1981) Production of *Crangon crangon* in the tidal zone of the Dutch Wadden Sea. Netherlands Journal of Sea Research 15:33-53

Kuipers BR, Dapper R (1984) Nursery function of Wadden Sea tidal flats for the brown shrimp *Crangon crangon*. Marine Ecology Progress Series 17:171-181

Le Vay L, Ut VN, Walton M (2007) Population ecology of the mud crab *Scylla paramamosain* (Esampador) in an estuarine mangrove system; a mark-recapture study. Marine Biology 151:1127-1135

Leaman BM (1991) Reproductive styles and life history variables relative to exploitation and management of *Sebastes* stocks. Environmental Biology of Fishes 30:253-271

Leigh GM, Hearn WS, Pollock KH (2006) Time-dependent instantaneous mortality rates from multiple tagging experiments with extract times of release and recovery. Environmental and Ecology Statistics 13:89-108

Leite Jr NO, Petrere Jr M (2006) Growth and mortalities of the pink-shrimp *Farfantepenaeus brasiliensis* Latreille, 1970 and *F. paulensis* Pérez-farfante 1967 in Southeast Brazil. Brazilian Journal of Biology 66:523-536

Marin E. BJ, Quintero A, Bussiere D, Dodson JJ (2003) Reproduction and recruitment of white mullet to a tropical lagoon as revealed by otolith microstructure - *Mugil curema* - Margarita Island, Venezuela. Fishery Bulletin 101:809-821

Mehanna SF (2000) Population dynamics of *Penaeus semisulcatus* in the Gulf of Suez, Egypt. Asian Fisheries Science 13:127-137

Meyer-Waarden PF, Tiews K (1965) Grundlagenmaterial zu "Die Zusammensetzung der Fänge der deutschen Garnelenfischerei in den Jahren 1954 - 1960". Veröffentlichungen des Instituts für Küsten- und Binnenfischerei1-29

Mohammed HMA, Bishop JM, Xu X (1996) Population characteristics of green tiger prawns, *Penaeus semisculatus*, in Kuwait waters prior to the Gulf War. Hydrobiologia 337:37-47

Moses BS (1990) Growth, biomass, mortality, production and potential yield of the West African clam, *Egeria radiata* (Lamarck) (Lamellibranchia, Donacidae) in the Cross River system, Nigeria. Hydrobiologia 196:1-15

Neudecker, T. and Damm, U. Seasonality of egg-bearing shrimp (*Crangon crangon* L.) in costal waters of the German Bight. K: 28, 1-9. 1992. ICES Shell Fish Committee.

Nwosu FM, Holzlöhner Sieghard, Enin UI (2007) The exploited population of the brackish river prawn (*Macrobrachium macrobrchion* Herklots 1851) in the Cross River estuary, Nigeria. Scientia Marina 71:115-121

Nwosu FM, Wolfi M (2006) Population Dynamics of the giant African river prawn *Macrobrachium vollenhovenii* Herklots 1857 (Crustacea, Palaemonidae) in the Cross River Estuary, Nigeria. West Africa Journal of Applied Ecology 9:1-14

Oh C-W, Hartnoll RG (2000) Effects of food supply on the growth and survival of the common shrimp *Crangon crangon* (Linnaeus, 1758) (Decapoda, Caridea). Crustaceana 73:83-99

Pauly D (1982) A method to estimate the stock-recruitment relationship of shrimps. Transactions of American Fisheries Society 111:13-20

Pauly D (1983) Length converted catch curves: A powerful tool for fisheries research in the tropics (part I). Fish Bytes 1:9-13

Peristiwady T, Geistdoerfer P (1991) Biological aspects of *Monacanthus tomentosus* (Monacanthidae) in the seagrass bed of Kotania Bay, West Seram, Moluccas, Indonesia. Marine Biology 109:135-139

Powell DG (1979) Estimation of mortality and growth parameters from the length frequency of a catch. Rapport et Procès-verbaux des Réunions, Conseil international pour L'Exploration de la Mer 175:167-169

Ragonese, S., Andreoli, M. G., Bono, G., Giusto, G. B., Rizzo, P., and Sinacori, G. Overview of the available biological information on demersal resources of the Strait of Sicliy. FAO-FI--GCP/RER/010/ITA/MSM-TD-02, 67-73. 2004. Gzira (Malta),10-12 Dec 2002 / FAO, Mazara del Vallo (Italy). MedSudMed Project, 2004, 102 p.

Ralston S (1989) Effect of seasonal recruitment on bias of the Beverton-Holt length-based mortality estimator. American Fisheries Society Symposium 6:190-197

Rotherham, Douglas. Relative growth and preliminary estimates of absolute growth parameters, mortalities and exploitation ratios of *Trypaea australiensis* in south-eastern Australia. 1-25. 2004. Thesis, University of Wollongong.

Sadovy, Yvonne and Eklund, Anne-Marie. Synopsis of Biological Data on the Nassau Grouper, *Epinephelus striatus* (Bloch, 1792), and the Jewfish, E. itajara (Lichtenstein,1822). Fishery Bulletin. NOAA Technical Reports NMFS 146, 1-65. 1999.

Sainte-Marie B, Bérubé I, Brillo S, Hazel F (2006) Observations on the growth of the sculptured shrimp *Sclerocrangon boreas* (Decapoda: Caridea). Journal of Crustacean Biology 26:55-62

Sheehy MRJ, Bannister RCA, Wickins JF, Shelton PMJ (1999) New perspectives on the growth and longevity of the European lobster (*Homarus gammarus*). Canadian Journal Fisheries and Aquatic Science 56:1904-1915

Sparre P, Ursin E, Venema SC (1989) Introduction to tropical fish stock assessment. Food and Agriculture organization of the united nations, Rome

Ssentongo GW, Larkin PA (1973) Some simple methods of estimating mortality rates of exploited fish populations. Journal of the Fisheries Research Board Canada 24:2355-2453

Taylor CC (1958) Cod growth and temperature. Journal du Conseil Permanent International pour l' Exploration de la Mer 23:365-370

Temming A, Damm U (2002) Life cycle of *Crangon crangon* in the North sea: A simulation of the timing of recruitment as a function of the seasonal temperature signal. Fisheries Oceanography 11:45-58

Temming, Axel, Damm, Ulrich, and Neudecker, Thomas. Trends in the size composition of commercial catches of brown shrimp (*Crangon crangon* L.) along the German coast. K: 53. 1993. ICES Shell fish Committee.

Tetard A (1985) Elements sur la croissance de la crevette grise, *Crangon crangon* (L.), en manche-est et sud mer du nord. ICES C.M. K:17:16pp.

Ulmenstrand M, Eggert H (2001) Growth of Norway lobster, *Nephrops norvegicus* (Linnaeus 1758), in the Skagerrak, estimated from tagging experiments and length frequency data. ICES Journal of Marine Science 58:1326-1334

Viegas I, Martinho F, Neto J, Pardal M (2007) Population dynamics, distribution and secondary production of the brown shrimp *Crangon crangon* (L.) in a southern European estuary. Latitudinal variations. Scientia Marina 71:451-460

Villwock de Miranda L, Haimovici M (2007) Changes in the population structure, growth and mortality of striped weakfish Cynoscion guatucupa (Sciaenidae, Teleostei) of Southern Brazil between 1976 and 2002. Hydrobiologia 589:69-78

von Bertalanffy L (1934) Untersuchungen über die Gesetzlichkeit des Wachstums I. Teil: Allgemeine Grundlagen der Theorie; Mathematische und Physiologische Gesetzlichkeiten des Wachstums bei Wassertieren. Development Genes and Evolution 131:613-651

von Bertalanffy L (1938) A quantitative theory of organic growth (inquiries on growth laws. II). Human Biology a record of research 10:181-213

Wehrtmann IS (1989) Seasonal occurrence and abundance of caridean shrimp larvae at Helgoland, German Bight. Helgoländer Meeresuntersuchungen 43:87-112

Wetherall JA, Polovina JJ, Ralston S (1987) Estimating growth and mortality in steady-state fish stocks from length frequency data. In: D.Pauly and G.R.Morgan, Editors, Length-based Methods in Fishery Research53-74

Estimating total mortality Z and maximum length L_∞ of *Crangon crangon* (Crustacea L. 1758) between 1955 and 2006

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Abstract

Total mortality of Southern North Sea brown shrimp (*Crangon crangon*) was determined by estimating θ, a parameter that is the quotient of total mortality (Z) and the von Bertalanffy growth constant (k). Estimates were based on length frequency distributions obtained from three, long-term data series: German Demersal Young Fish Survey, Dutch Demersal Fish Survey and German (Büsum, East Frisia) bycatch series). The surveys covered the period from 1955 until 2006. Different methods to estimate θ and L_∞ were applied and evaluated according to their applicability to the survey and brown shrimp conditions.

For the time period 1955 to 2006, median estimates of Z ranged from 5.74 (Ssentongo & Larkin), 5.65 (Beverton & Holt), 5.64 (Jones & Zalinge) and 5.35 (Length-converted Catch Curve) from all survey data. Highest mortality of Z = 8 was observed during the early 1990s lowest of Z = 4 during the 1960s. Over the whole period an increase in θ and a decrease of the fraction of animals larger >60 mm within the catches was observed, whereas maximum length L_∞ remained constant, L_∞ = 79.3 mm (total length). This is most likely due to an increase of Z and not to a decrease of k.

Key words: Mortality, maximum length, *Crangon crangon*

Introduction

The brown shrimp *Crangon crangon* is one of the most common species in European coastal waters (Gunnarsson et al. 2007, Neves et al. 2007, Pihl & Rosenberg 1982, Kuipers & Dapper 1981). In the southern part of the North Sea, mainly in Germany, the Netherlands and Denmark, it supports the most lucrative regional fishery. Total commercial landings from the North Sea constantly increased since the 1970s with for the present highest landings in 2005 of about 37000 t (ICES 2006, Neudecker & Damm 2006).

Until now uncertainties exist about the life cycle and maximum age of *C. crangon* which is assumed to be between two and five years (Oh et al. 1999, Tiews 1954, Lloyd & Yonge 1944, Havinga 1930, Havinga & Willer 1929). Additionally many different estimations of the total mortality exist that range from $Z = 2$ (Henderson et al. 2006, Henderson & Holmes 1987) over $Z = 4$ (Temming et al. 1993, Knijn & Boddeke 1991) and $Z = 11$ (Viegas et al. 2007) to $Z = 22$ (del Norte-Campos & Temming 1998).

These different findings are mainly due to difficulties in ageing shrimps but also to problems with length based methods like cohort tracking. Ageing is nearly impossible in short lived fast growing Crustaceans as all hard parts are lost after ecdysis (Hartnoll 2001). Additionally, and in particular for the brown shrimp, cohort tracking is difficult mainly because of its extended spawning, length and seasonal dependent migration patterns (Boddeke 1976, van der Baan 1975, Hartsuyker 1966) as well as sexual dimorphism (Labat 1977)(Meixner 1969).

Methods of choice for estimating mortality are therefore based on length frequency distributions and estimate θ, the quotient of total mortality Z and von Bertalanffy growth parameter k (de Graaf & Dekker 2006, Miranda 2002, Sparre et al. 1989, Wetherall et al. 1987, Pauly 1983, Hoenig et al. 1983, Jones & van Zalinge 1981, Powell 1979, Ssentongo & Larkin 1973, Beverton & Holt 1956). These methods are all based on the assumption that growth, mortality and recruitment does not vary seasonally. In a previous approach of the authors it has been shown that violating the assumption of no seasonality can lead to considerable bias in total mortality and maximum length estimates (Hufnagl & Temming, manuscript 1). The results further indicated that up to now mortalities have generally been underestimated. Under conditions comparable to those of the brown shrimp population, L_∞ can be estimated with high accuracy when the Wetherall and the Powell method are applied in combination. θ was estimated most accurate applying the Ssentongo & Larkin, Beverton & Holt and Jones & Zalinge method or the Length Converted Catch Curve, but in all cases estimates were negatively biased. To correct for the bias correction function were defined in the preceding study.

The aim of the actual work is to further investigate the accuracy of the length frequency methods based on field data and to apply both, the results of the theoretical and the field based evaluation, to improve the accuracy of the estimate of *C. crangon* total mortality. Another aspect of the actual study will be to determine if long term changes and regional differences of Z exist. Until now no

long time data series have been used to examine variations and trends of Z in annual resolution. In this work data are available for half a century from 1955 to 2006 along the whole coast of Germany and the Netherlands, the core distribution area of *C. crangon*.

Material and Methods

Sampling Surveys

Four different data sets from three long time series from Germany and one from the Netherlands were used. The German bycatch data span the period from 1955 to 1996 (Büsum) and 1958 to 1993 (East Friesian) and were taken from weekly, monthly commercial catches (Meyer-Waarden & Tiews 1965). The resolution of the length data is 5 mm (total length) and data are available for the whole year. Animals were taken from the unsieved and unsorted catch. For this survey no water depth and haul duration is known.

The third data set spans the period 1997 to 2007 along the German coast and were gained in a scientific survey (Demersal Young Fish Survey, DYFS) performed by the former “Bundesforschungsanstalt für Fischerei”, Germany every autumn. The Demersal Young fish survey is performed regularly since 1974 (Neudecker 2001). The survey covers mainly shallow water areas and a 3 m beam trawl with 20 mm mesh size (stretched mesh) and without tickler chain is used. Fifteen-minute standard tows are carried out with the prevailing tidal current at a towing speed of 2–4 knots over ground with a mean distance covered of approximately 0.75 nautical miles (Neudecker et al. 1998)

The Dutch Demersal Fish Survey (DFS) covers the coastal zone from the border between the Netherlands and Belgium up to Esbjerg (DK), the Dutch Wadden Sea, Ems-Dollard estuary, Schelde estuary (Wester-/Oosterschelde) and is conducted each autumn since 1969. A 6 m and a 3 m beam trawl with tickler chain and 20 mm mesh opening (cod end) is applied (van Keeken et al. 2008).

All data were used without correction for net selectivity functions as only animals > 45 mm were included in our analysis. 45 mm was chosen as from 40-45 mm on numbers of animals per length decreased which is according to Pauly (1990) the first point to choose in the regression.

Methods used to estimate θ and L_∞

Estimating $\theta=Z/k$ and L_∞ was performed according to the following methods (briefly reviewed in (Hufnagl & Temming, manuscript 1).

Powell method (Powell 1979)

Modified Wetherall et al. method (Wetherall et al. 1987) (Sparre et al. 1989)

Beverton and Holt (Beverton & Holt 1956)

Jones and Zalinge (Jones & van Zalinge 1981)

Hoenig (Hoenig et al. 1983)

Ssentongo & Larkin (Ssentongo & Larkin 1973)

nonseasonal Length Converted Catch Curve (Pauly 1983)

seasonal Length Converted Catch Curve based on a spreadsheet method presented by (de Graaf & Dekker 2006)

Estimating the sensitivity of the estimates according to uncertainties in the data

Influence of seasonality

The scientific cruises (DYFS and DFS) are mainly performed in autumn whereas the bycatch data are available for the whole year. To determine the differences that occur if only autumn data are used for the mortality estimations the Büsum bycatch data were used. The mortality was estimated 1st based on all the whole year data of the Bycatch series and 2nd based only on autumn data of the same series. The received results were then compared with each other.

Influence of depth

Density and size spectra of *C. crangon* are dependent on the depth were the animals are caught (Siegel et al. 2005, Welleman & Storebeck 2002, Janssen & Kuipers 1980, Tiews 1954). Larger animals can mainly be found in deeper water, whereas smaller animals are caught on the tidal flats and sometimes in mud puddles during ebb tide (Boddeke et al. 1986, Berghahn 1983). The influence of the depth on the estimations was tested in the actual work by splitting the DYFS data (as here the catch depth is known) into subsets including all catches < 5 m, 5-10 m and 10 - 25 m. Mortality was estimated for each depth separately.

Influence of interlink of k and L_∞

The growth constant k and the mean maximum length L_∞ are correlated (Summerfelt & Hall 1987) and several combinations of k and L_∞ can be used to describe the growth of the same population without changes in explained variance. In the introduction it was discussed that age determination of *C. crangon* is not possible what further implied that also growth and an accurate estimation of k is difficult. On the opposite L_∞ can be estimated accurate with the Wetherall and Powell method from length frequency distributions. Would it be possible to determine k based on L_∞ could increase the accuracy of the Z ($\theta = Z/k$) estimate.

For this purpose a growth model derived from growth experiments and literature data as well as a sinus temperature function (Hufnagl & Temming, submitted) was used to calculate a length trajectory of *C. crangon*. The seasonal von Bertalanffy

growth equation was fitted to that trajectory. L_∞ was in this regression set to a constant value (70. 75, 80 ... 95) and k was estimated. The parameters of the seasonal von Bertalanffy growth function determining seasonality were kept constant: C = 1 (maximum seasonal oscillation), t_s = 0.5 (minimum of oscillation during winter). The relation of k and L_∞ and the resulting growth trajectories are plotted in Figure 9-1. Further L_∞ and k determined by (Kuipers & Dapper 1984, Tiews & Schumacher 1982, Tiews 1954) were used to establish the correlation (Figure 9-1).

$$k = 10430 \cdot L_\infty^{-2.0824} \qquad (41)$$

The influence of k on the estimated Z was determined 1st with a static k and 2nd with a k depending on L_∞ as descried in equ. (10). As static value k = 1.25 was used as this value was derived from the growth model (Hufnagl & Temming, submitted) for a mean North Sea temperature of 10°C.

Influence of the region

C. crangon dwells over a wide geographical and latitudinal range. Factors that might influence mortality are for example food quality, pollution or hydrographical conditions like temperature and salinity. Additionally the fishing effort is variable not only over time but also according to the different nations and within different areas (ICES 2007). Further occurrence of predators might vary regional. Whiting for example seems in winter to be bound to the sea surface temperature (Zheng et al. 2002). Summarizing regional differences in mortality are possible.

DYFS and DFS data show an overlap in time and also in space. It was possible for several years to calculate mortality for different regions along the coast of the Netherlands, Germany and Denmark. Several stations within one area were aggregated and L_∞ and Z were calculated for these regions (Figure 9-3). The data were aggregated into five-year classes, as not all stations were sampled all years.

Influence of seasonal mortality, growth and recruitment on the used methods

The prerequisite of the methods used to determine L_∞ and θ are no seasonality in mortality, recruitment and growth. Further it is assumed that there is no intracohort variability of L_∞ and k. This is not the case for the brown shrimp population. Therefore prior to the application the methods were evaluated according to the applicability for conditions presumable prevalent for *C. crangon* in the field (Hufnagl & Temming, manuscript 1). The results of that prior study suggested that the Powell, Wetherall and Hoenig method as well as the seasonal Length Converted Catch Curve are not suitable for estimating mortality of *C. crangon*. For completeness and comparison with other species in the actual work these four methods were used to determine Z and the results are shown in the appendix. However, in the final mortality estimation of *C. crangon* they are not included.

Table 9-1: Range of applicability, slope and intercept of the correction functions for the different methods.

	samples available over the year				samples available from autumn			
	min	**max**	**slope**	**intercept**	**min**	**max**	**slope**	**intercept**
L_∞ Wetherall	70.0	105.0	0.92	4.30	65.0	105.0	0.82	13.49
L_∞ Powell	70.0	95.0	1.05	-7.56	70.0	90.0	1.24	-18.52
θ Beverton & Holt	1.0	7.0	1.34	0.21	1.0	8.0	1.10	0.83
θ Jones Zalinge	1.0	7.0	1.41	0.74	1.0	7.0	1.09	1.23
θ Ssentongo & Larkin	1.0	8.0	1.33	-0.39	1.0	9.0	1.08	0.36
θ LCCC	1.0	6.0	1.60	-0.63	1.0	8.0	1.24	0.24

Another result of the preceding evaluation study was that not all methods were suitable for the whole range of θ and bias is bound to the level of θ. Supplementary all methods are susceptible to singular combination of k, Z, L_∞ and seasonality, especially if only autumn samples are available. Basing the mortality on more than one method is therefore recommended (see discussion). The final mortality estimate included the following methods: Beverton Holt, Jones & Zalinge, Ssentongo & Larking, LCCC. For each year L_∞ and θ was estimated from the annual or autumn length frequency distribution with each method. If the determined θ or L_∞ was inside the application range (minimum and maximum θ and L_∞, Table 9-1) it was corrected with the linear function (slope and intercept,) otherwise it was rejected. From all remaining estimates $Z=\theta \cdot k$ was determined. The Bertalanffy growth parameter k was derived from the estimated L_∞ according to equ. (10).

Regression of determined annual mortalities with different biotic and abiotic factors

Annual total mortality was correlated with different parameters using the Jones & Zalinge method and including all surveys. This was done to determine whether annual variations in mortality are due to biotic or abiotic variables. The Jones & Zalinge method was chosen, as it showed in the preceding evaluation study (Hufnagl & Temming, manuscript 1) the lowest sensitivity to changes of seasonal recruitment and to different combinations of k and Z. As explaining variables NAOI (winter, whole year), bottom temperatures, *C. crangon* landings (Netherlands, Germany form ICES WGCRAN), *C. crangon* density (DFS: $n \cdot haul^{-1}$), whiting densities (DYFS, $n \cdot 100m^{-2}$) were examined. At the moment no long time North Sea bottom temperature data series exists. Although the water body is well mixed in the Wadden Sea area a preliminary study revealed that temperatures can vary between bottom and surface. Therefore bottom temperatures were taken from the „Hamburg Shelf Ocean Model“ (HAMSOM) (Schrum et al. 2002, Schrum & Backhaus 1999). Four positions representing German and Netherlands shallow and coastal area were chosen: NL1: N52.98, E4.67, NL2: N52.98, E4.67, G1: N54.18 E8.67, G2: N54.08 E8.17.

Results

Influence of seasonality on the estimation of θ

L_∞ estimates did vary only slightly when based on autumn data in comparison to whole year length frequency distributions (Wetherall -2 ± 4%, Powell 1 ± 6%) for the Büsum bycatch data (Table 9-2).

θ estimates of the Wetherall, Ssentongo & Larkin, Beverton & Holt, Jones & Zalinge and the LCCC are 1 to 8% lower when based on autumn data in comparison to annual estimates. The Powell, Hoenig and the seasonal LCCC overestimated θ (autumn in comparison to whole year) by 3 to 5%. When calculating θ the highest year to year differences were observed for the Hoenig (±44%) and Powell (±25%) method and the seasonal LCCC (21%) whereas lower variability was observed for the Jones & Zalinge (±14%), the Wetherall (±16%), the LCCC (±16%), the Ssentongo & Larkin (±17%) and the Beverton & Holt (±18%) methods.

Table 9-2: Deviation of θ and L_∞ determined with Büsum bycatch annual data for different depth ranges (<5 m, 5-10 m, 10-25 m) in comparison to θ and L_∞ determined for length frequency distributions of all depth. Values represent the mean deviation and standard deviation based on all analyzed years (n=42). The last row shows the mean deviation of θ and L_∞ estimation based on autumn length frequency distributions related to θ and L_∞ derived from annual data. (W = Wetherall et al. P = Powell, LCCC = Length Converted Catch Curve, SL = Ssentongo & Larkin, JZ = Jones & Zalinge, BH = Beverton& Holt, sLCCC = seasonal Length Converted Catch Curve, H = Hoenig)

	<5 m		**5-10 m**		**10-25 m**		**autumn**	
	mean	**std**	**mean**	**std**	**mean**	**std**	**Mean**	**std**
L_∞ W	-2	2	-1	3	-2	3	-2	4
L_∞ P	-1	2	0	5	-4	7	1	6
θ JZ	-8	5	-10	8	-12	19	-4	14
θ SL	-9	4	-11	10	-13	21	-4	17
θ BH	-10	4	-11	9	-17	17	-2	18
θ LCCC	-9	4	-10	9	-15	16	-1	16
θ W	-14	7	-14	11	-12	14	-8	16
θ P	-7	5	-8	14	-16	23	5	25
θ sLCCC	-10	7	-14	9	-24	10	3	21
θ H	-16	11	-16	13	-22	16	3	44

Influence of depth

The length frequency distribution of the DYFS data set was used to determine L_∞, θ for different water depth. The estimates are based on a total of 138708, 82118 and 26939 animals for a water depth of < 5 m, 5-10 m and 10-25 m, respectively. Mean depth of the DFS sampling stations was greater (8.4 - 10.5 m) than mean depth of the DYFS stations (5 - 6.6 m, Table 9-3)

Highest deviations of the depth specific θ were determined for the 10 - 25 m subset while θ was determined for all animals of one year (all depth strata) depth.

Mean deviations were calculated ranging from -12% (Wetherall) to -24% (seasonal LCCC).

All depth segregated subsets underestimate θ in comparison to the integrated θ including all data (Table 9-2). This is especially true for the Hoenig (-16 to -22%), the seasonal LCCC (-10 to -24%) and the Beverton & Holt (-10 to -17%) methods. The standard deviation of θ also increases with depth and is highest for the 10-25 m subset (±10 to 23%).

The deviation for the depth determined L_∞ varies only slightly (2 - 4%) for the Wetherall and the Powell method.

Table 9-3: Mean and standard deviation of the water depth at the sampling point of the DFS and the DYFS

DYFS year	mean depth [m]	std
1998	6.2	6.2
1999	5.4	5.9
2000	5.5	6.3
2001	5.6	6.1
2002	5	5.9
2003	6.2	7
2004	5.4	4.8
2005	6.6	7.5
2006	6.6	6.7

DFS year	mean depth [m]	std
1970	8.8	5.1
1971	9.5	5.3
1972	9.3	4.7
1973	9.0	4.4
1974	9.8	4.7
1975	9.3	4.8
1976	8.4	4.1
1977	10.5	5.6
1978	10.5	5.0
1979	10.4	4.8
1980	8.8	5.1
1981	9.8	4.8
1982	9.4	4.9
1983	8.8	4.5
1984	9.2	4.8
1985	8.9	4.6
1986	9.0	4.6
1987	8.9	4.9
1988	8.9	4.2

DFS Year	mean depth [m]	std
1989	9.4	5.1
1990	9.7	5.0
1991	9.4	4.7
1992	9.3	5.5
1993	9.1	4.6
1994	9.5	5.5
1995	8.8	4.8
1996	9.7	5.1
1997	9.9	5.6
1998	9.2	5.5
1999	10.3	5.9
2000	10.1	4.8
2001	11.0	5.8
2002	9.8	5.5
2003	9.0	4.3
2004	9.5	4.6
2005	9.5	5.5
2006	10.4	4.9

Influence of a variable k

Calculating $Z = \theta \cdot k$ requires accurate knowledge of the growth rate k and as described before different combinations of k and L_∞ are potentially suitable to describe a population. To estimate the influence of this relationship fact Z was once calculated with a static k and once based on the cubic relation shown in Figure 9-1 and equ. (10). This was done for all data sets: DYFS, DFS, Büsum and East Frisian bycatch data.

The mortality estimates based on the DFS and the East Frisian are 11 and 12 % higher if a variable k instead of a constant k of 1.25 is applied. Based on DYFS and Büsum data the deviation is 5% and 1%, respectively (Table 9-4). The standard deviation is between 7% for the East Frisian Bycatch series and 9% for the Büsum Bycatch data.

Table 9-4: Deviation of Z calculated with a static growth constant k for each year in relation to Z calculated with an annual varying k according to the function shown in Figure 9-1.

	mean dev [%]	std	n (years)
DYFS	5	8	10
DFS	11	8	37
Büsum	1	9	42
East Frisia	12	7	33

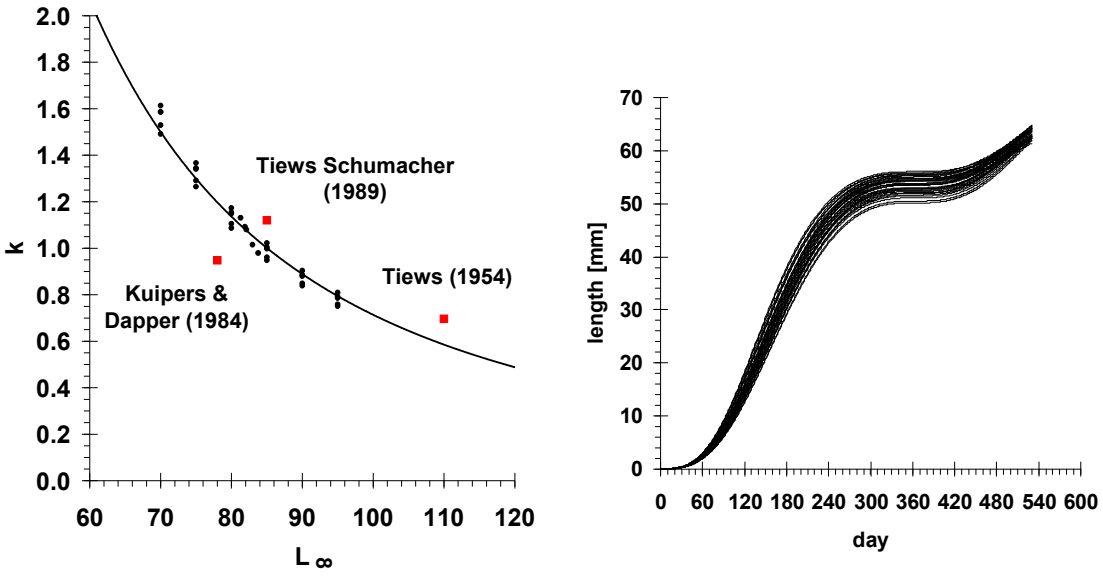

Figure 9-1: Left: Correlation of mean maximum length and growth constant k as estimated by Tiews (1954), Kuipers & Dapper (1984) and Tiews & Schumacher (1989) and based on the growth function published in Hufnagl & Temming (submitted). Right: Length trajectories calculated with the seasonal Bertalanffy growth function and the different parameter combinations of L_∞ and k.

Influence of region

L_∞ is about 75 to 85 mm in all areas and for all analyzed periods (Figure 9-3). Only for area NL1 and NL2, the most western survey locations the determined L_∞ are slightly higher L_∞ = 86 -92 mm.

Total mortalities calculated for the various areas vary between Z = 4 and Z = 6 (Figure 9-2). The different regions show no significant difference when grouping the three time periods (ANOVA, p>0.01).

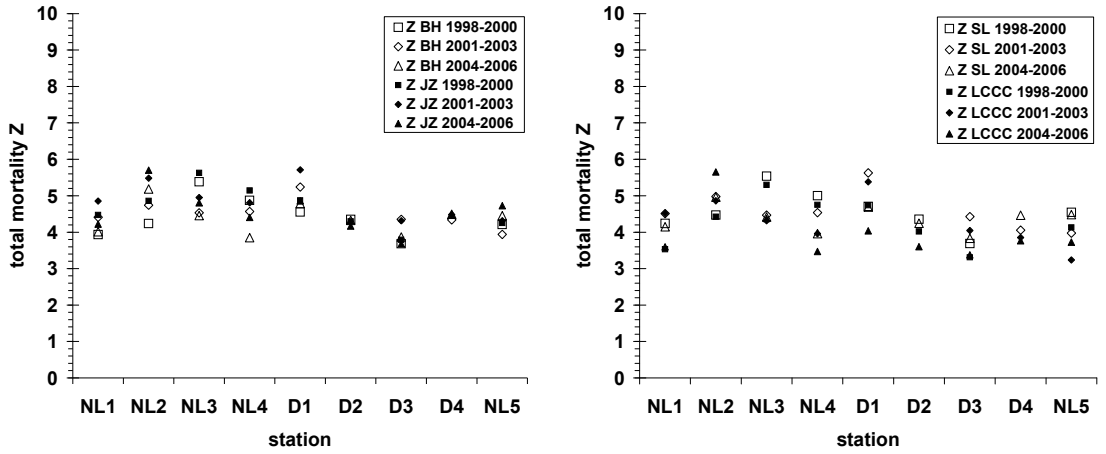

Figure 9-2: Mortality calculated with the Beverton & Holt (Z BH), Jones & Zalinge (Z JZ), Ssentongo & Larkin (Z SL) and the Length Converted Catch Curve (Z LCCC) for different regions derived from the German Demersal Young Fish Survey (D1-D4) and the Dutch Demersal Fish Survey (NL1-NL5) for three time periods.

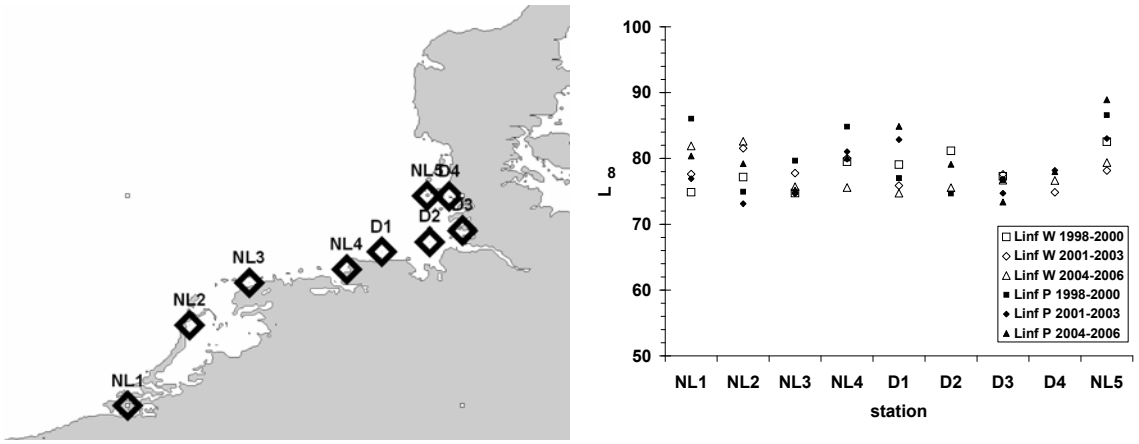

Figure 9-3: Left: mean latitude and longitude of the created subareas for testing the influence of position on mortality and L_∞. Points NL1 to NL5 are derived from the Dutch DFS data, points D1 to D4 are derived from the German DYFS data. Right: L_∞ calculated for the different positions with the Wetherall et al. (W) and the Powell (P) function for the different periods.

Annual total mortalities based on the DYFS, DFS, Büsum and East Frisian bycatch data

Results of the previous section suggested that total mortality of *C. crangon* is not specific to sampling site. Therefore, in this section we analyzed pooled length frequency distributions of all sampling stations. The Z values shown in this section were derived from mean θ determined with the Wetherall, Beverton & Holt, Jones & Zalinge, Ssentongo & Larkin methods and the LCCC. All θ and also L_∞ were corrected using the functions given in Table 8-4 while k was determined by equ. (10).

L_∞ values varied only slightly over the whole period from 1955 to 2006 (Figure 9-4, left). For all years a mean of 79.3 mm was determined with lower L_∞ during 1970 - 1990 (exception: DFS) and higher L_∞ since 1995. According to the cubic relation between k and L_∞ the trend in the determined k is mainly reverse to that of L_∞. For all years a mean k of 1.17 was determined (Figure 9-4, right).

A steady increase of total mortality with time was observed for the data of the Büsum bycatch series (Figure 9-5, left). Between 1955 to 1980 lowest mortality values were about 4, generally estimates of the LCCC. Highest values were generally determined by the Ssentongo & Larkin method; these reached values around 6 with peaks of 7.0 and 7.3 in 1973 and 1975 respectively. In the beginning of the 1990s maximum values were calculated ranging from 8.5 (1995) to 9.1 (1993), while minimum values did not exceed Z= 5-7.

Based on the East Frisian bycatch data series again highest mortalities were calculated by the Ssentongo & Larkin method in most years and lowest mortalities were obtained from the LCCC (Figure 9-5, right). Lowest mortalities vary between Z = 4 and 5 until 1983 and increased to Z = 8.2 in 1990.

The share between the mortalities, estimated with the different methods, is lower when the methods are applied on the DFS data then when applied to the bycatch data. In this case highest values were mainly determined by the Jones & Zalinge method whereas the mortalities determined by the LCCC are again mainly the lower estimates. The general pattern over the whole period is comparable to the previously described surveys. Highest mortalities of Z=7.7 were calculated for 1993 (Figure 9-6, left).

The calculation of an annual mean mortality was based on all surveys but separated according to the methods. The result suggests an increase of maximum total mortality from 1955 toward the period 1988 to 1996 (Figure 9-6, right). The observed range of the determined mortalities is mainly between Z = 4 and 6 for the period 1955 to 1985. Since 1985 towards 1993 lowest estimates increased from Z = 4.9 to Z = 6.6 and maximum estimate from Z = 6.8 to Z =9.1. Afterwards a decrease can be observed until 2006, when the estimated mortality range was between Z = 3.7 and Z = 7.7.

The median was determined with the different methods for the whole period and all surveys; results are given as

Beverton & Holt:	5.65
Jones & Zalinge:	5.64
Ssentongo & Larkin:	5.74
Length Converted Catch Curve:	5.35

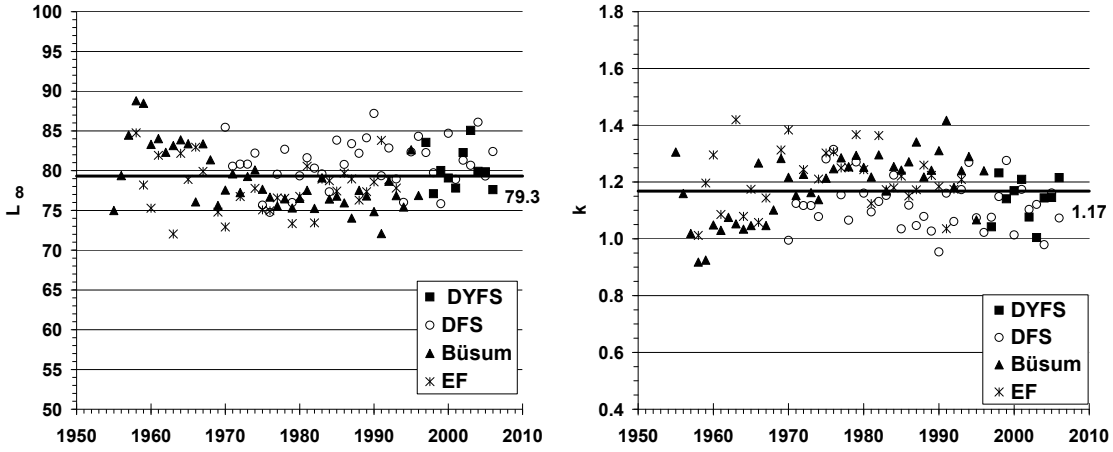

Figure 9-4: Left: L_∞ calculated as mean of the Wetherall et al. and Powell method for the DYFS (Germany), DFS (Netherlands), Büsum (Germany) and East Frisia (Germany) bycatch data. Right: Calculated growth constant k based on the determined L_∞.

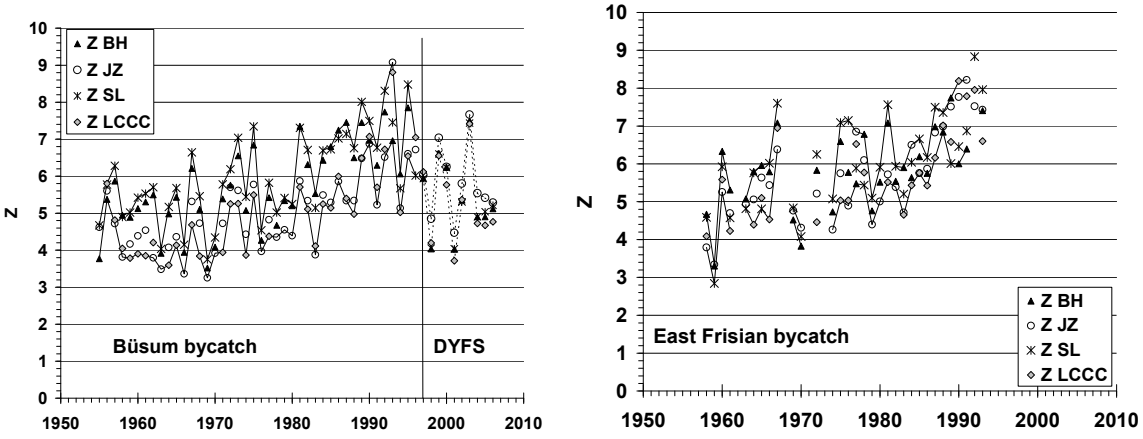

Figure 9-5: Mortalities determined with the Beverton & Holt (triangles), Ssentongo & Larking (stars), Jones & Zalinge (circles) methods and the Length Converted Catch Curve (diamonds). Left: Calculations based on Büsum bycatch (1955 – 1996) and DYFS (1997-2006) data. Right: Total mortality estimations based on the DFS data.

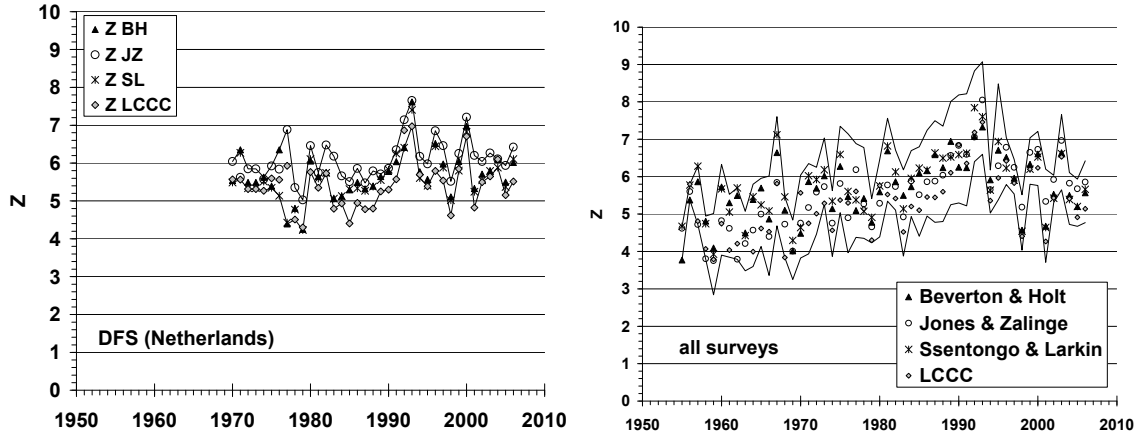

Figure 9-6: Mortalities determined with the Beverton & Holt (triangles), Ssentongo & Larking (stars), Jones & Zalinge (circles) methods and the Length Converted Catch Curve (diamonds). Left: Calculations based on East Frisian bycatch (1955 – 1996) data. Right: Total mortality estimations calculated as the mean result of all surveys per year. Lines indicate the maximum and minimum values determined within each year.

Fraction of large animals in the catches

The calculations of the fraction of animals > 70 mm and > 60 mm in comparison to all animals > 45 mm show a constant decline from 1959 (mean 15% and 36%) towards 1990s (<1% and ~10%). Since then values remained at a constant low level. During the last two decades the fraction of *C. crangon* > 70 mm in the catches was below 2 % and of animals > 60 mm, but mainly below 20%.

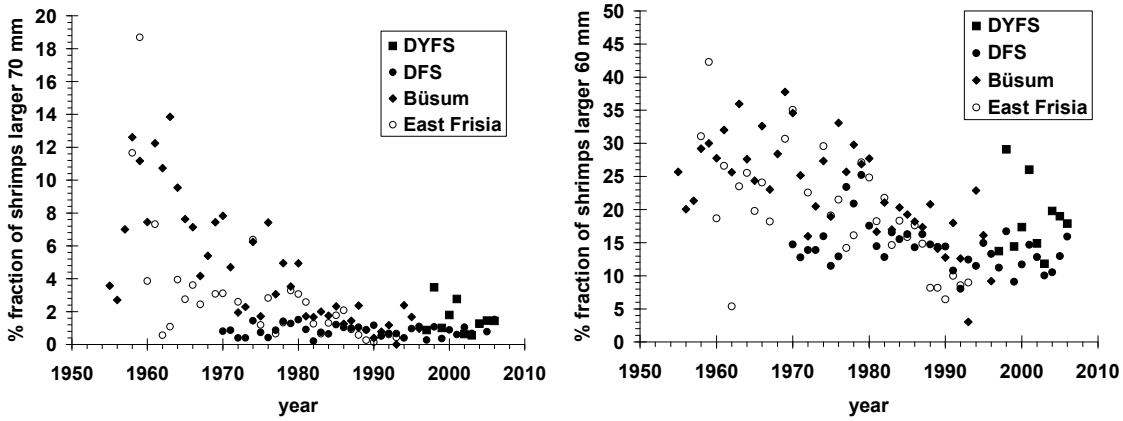

Figure 9-7: Fraction of shrimps larger than 70 mm (left) and 60 mm (right) in relation to 45 mm shrimps in the catches of the Demersal Young Fish Survey (Germany), Demersal Fish Survey (Netherlands), the Büsum and the East Frisia Bycatch data.

Regression of determined annual mortalities with different biotic and abiotic factors

A significant correlation ($p<0.005$) was observed between total mortality (Jones & Zalinge) and the winter North Atlantic Oscillation index (NAOI Dec-Mar, Z, $r^2 = 0.356$, $p = 0.000$). Another correlation was found between total mortality and bottom temperature. The latter correlation was present for each position in March, April, May and June ($r^2 = 0.199$-0.323, $p<0.002$) and the mean temperature of these months ($r^2 = 0.300$, $p = 0.000$). If we skip the outlying values for the years 1993 and 1996 the correlation coefficient of the latter regression would increase ($r^2 = 0.506$, $p = 0.000$). Further T (dependent) and NAOI were positively correlated ($r^2 = 0.338$, $p = 0.000$)

Discussion

Accuracy of the estimates

Evaluation of the different length frequency based methods carried out in a study prior to this work (Hufnagl & Temming, manuscript 1) indicated that among the evaluated methods none was capable to determine θ (Z/k) with 100% accuracy if a population is influenced by seasonality. The Powell, Wetherall and Hoenig method and the seasonal Length Converted Catch Curve were not suitable to determine θ

of *C. crangon*, although the Wetherall and Powell method accurately calculated L_∞. The remaining four evaluated methods were: Ssentongo & Larkin, Jones & Zalinge, Beverton & Holt and the nonseasonal Length Converted Catch Curve. These showed different advantages and disadvantages. Under simulation conditions the Ssentongo & Larkin method showed the lowest mean deviation of the calculated θ from the θ used to generate the artificial length frequency distribution. However, the estimates showed a high variability according to specific combinations of specific k and Z. Since both values are not known with high accuracy, the estimates of the Ssentongo & Larkin method might be biased. The Jones & Zalinge method showed only low variability and lowest sensitivity to seasonal recruitment, but consequently underestimated θ and was sensitive to inner-cohort variability of L_∞ and k. The Beverton & Holt method was sensitive to the timing of recruitment and estimates based on the assumption of spring recruitment (maximum in March) were underestimated by 50%, whereas summer recruitment (maximum in June) was underestimated by only 25%. According to Beukema (1992) timing of recruitment of *C. crangon* is variable and depends on the winter temperature, which might influence the Beverton & Holt estimates. Besides this bias reliable estimates with only low variability were calculated by that method in the brown shrimp specific simulations (PAPER 1). The Length Converted Catch Curve was not influenced by inner-cohort variability of L_∞ and k. Furthermore, influence of seasonal recruitment was lower than for the Beverton & Holt method, but the higher θ the more it is underestimated with the Length Converted Catch Curve (under conditions comparable to the brown shrimp).

Despite the previously mentioned biases it was possible to derive correction functions for each method that should minimize the bias, but due to the high variability in the field uncertainties still exist. Therefore, in the present study θ was estimated using all four methods and Z is presented as a range for each year.

Comparing the findings of the present work with the simulation study reveals some similarities. Highest variability of estimates derived from autumn and whole year data were obtained for the Hoenig, Powell and Wetherall method and the seasonal LCCC. Additionally, these methods were also more sensitive when applied to length frequency distribution of different depth strata.

When considering only fall data or data from sampling point with unknown water depth, Both factors most probably lead to an underestimation of θ. The reason for that is that a shift of the mean length in the catch towards L_∞ implies a low mortality independent of the method applied. Mean length in autumn length frequency distributions is higher compared to whole year distributions as abundance of adult (>50 mm) *C. crangon* peaks in autumn (Siegel et al. 2005, Spaargaren 2000) whereas new recruits are numerous in early summer (Beukema 1992). Additionally, length dependent migration into deeper water of larger animals is possible (Beukema 1992, Kuipers & Dapper 1981, Janssen & Kuipers 1980). Both conditions would increase the mean length in the catch in comparison to considering all available data and therefore θ would be underestimated.

The Büsum and East Frisian bycatch samples were taken from commercial catches and no depth information were available. If water depth of the preferred fishing grounds varies between the years the estimated Z might be biased. This uncertainty is at top -13 ± 21 % (Ssentongo & Larkin 10 - 25 m). For the DFS and DYFS depth is known and sampling depths in the German survey (DYFS) are lower than in the Dutch survey (DFS) as shown in Table 9-3. Therefore, mortality determined from the DFS data might underestimate the real Z more than those based on the DYFS data.

Application of a static k in comparison to a variable k mainly altered the results of the East Frisian bycatch and DFS (Netherlands) estimates. The growth constant k might have varied over the observed period from 1955 to 2006 and as these differences cannot be determined the estimated maximum deviation of 12 ± 7% has to be taken as an additional uncertainty range of the estimate.

Present and previously determined total mortalities

The median mortality for all surveys and years determined in this work was between Z = 5.35 and 5.74, the mean L_∞ was 79.3 mm, and the resulting k = 1.17. These values are within the range of previous findings. Temming et al. (1993) estimated total mortality applying the Wetherall method and the LCCC on the Büsum bycatch data (five year means) from 1955 to 1988. The estimated Z varied between 2.78 and 6.08. These values are comparable to the actual results without correction.

According to Allen (1971) the Production to Biomass ratio (P/B) equals the total mortality Z. For the Dutch Wadden Sea a P/B ratio of 7.7 to 9.3 was calculated for the late 1970s (Kuipers & Dapper 1981). For *C. crangon* inhabiting the coasts of Portugal a P/B ratio of 8.7 (2003/4) and 10.9 (2004/5) was calculated (Viegas et al. 2007). These values are higher than our estimates for these years but estimating P/B ratios is generally complicated and as faulty as using length frequency distributions. This is mainly based on the size based migration of *C. crangon* (Beukema 1992, Kuipers & Dapper 1981, Janssen & Kuipers 1980) which makes sampling of the whole stock complicated. Additionally, estimation of biomass is based on an accurate knowledge of the catchability and vulnerability of the target species to the sampling gear. Kuipers & Dapper corrected their catches which were taken by a 5 mm mesh size net according to a function given by van Lissa (1977). This might have led to an overestimation of small shrimps (<30 mm) what might have falsified the results.

Furthermore, it has also to be taken into account that in our work total mortality was estimated only for large animals as the L_c was chosen as 45 mm (full catchability of the net). It can be assumed, that mortality of larger shrimps is lower than for smaller ones what is indicated by the following observations: For the German Wadden Sea P/B ratios of 22 were determined for small shrimps (del Norte-Campos & Temming 1998). The same restrictions and errors that have been discussed before have to be mentioned here, but as this work focused mainly on

small shrimps and a mesh with a small mesh size was applied, it might indicate that Z for smaller size classes is higher. In a more generalized approach this was also suggested by Peterson & Wroblewski (1984).

For *C. crangon* the great number of potential predators for small shrimps is most probably the main reason for a higher mortality. It is assumed that 60-80% of juvenile *C. crangon* are eaten by predators (Evans 1984).

For *C. crangon* caught in the Bristol Channel mortality was calculated as 2.92 (1981-2004) (Henderson et al. 2006) and 1.92-2.28 (Henderson & Holmes 1987), respectively. These estimates are based on the decline of the abundance after the first recruitment peak of the year. Again, migration was not included in these values as sampling was performed by power station entrainment catches and therefore at a fixed station. Recruitment waves immigrating after the main recruitment wave in August could therefore increase the abundance and could have led to an underestimation of Z.

For berried females a Z of 3.9 (Nov-Apr) and 1.7 (May-Aug) was determined for the period 1985-1990 by Knijn & Boddeke (1991). This is lower than our estimates during that period. These estimates are based on the amount of ripe and unripe eggs and are dependent on migration and temperature.

Is decline of large shrimp based on growth or mortality changes?

Estimating total mortality based on length frequency methods always depends on an accurate knowledge of k as all methods only determine $\theta = Z/k$. In this work k was calculated based on L_∞ which over the whole period from 1955 to 2006 was nearly constant. Despite the constant L_∞ the fraction of large shrimps constantly declined since 1955. This indicates that either the growth rate must have decreased or the mortality must have increase during that time.

For the years prior to the 1990s an increase in θ can be observed (not shown). Assuming that Z is constant and the decline in the fraction of large shrimp would be due to changes in k a bisection of the growth rate would be necessary to explain the increase. This is unlikely as the growth rate is mainly dependent on the temperature which even increased since the 1970s. Especially during the winters 1989-1994 very mild temperatures were measured (Becker & Pauly 1996). Therefore it is most likely that the observed increase in θ is an increase in mortality and that the decline of the fraction of large shrimp is a subsequent response of this.

Reasons for year to year variation of Z

The data analysis revealed strong year-to-year variation of mortality. Some of these variations might be due to methodical errors as discussed before and indicated by the range of mortality determined each year. However, some parallel trends can even be seen between overlapping surveys and different methods. A

significant correlation was determined for the bottom temperature (March to June) and total mortality (Jones & Zalinge method) as well as for the winter NAOI and total mortality.

A negative correlation of the winter NAOI with the abundance and recruitment of *C. crangon* has been reported by Siegel et al. (2005) and Henderson et al. (2006). A high index indicates strong westerlies and a high transport of bottom and surface water along the coasts towards the Skagerrak whereas a low index represents weak westerlies and low surface and bottom currents. One possible reason for the influence might therefore be seen in the studies mentioned earlier on the transport of hatched winter larvae to favorable or unfavorable areas. But this does not explain the influence of the NAOI on adult shrimp mortality as observed in our study. Zooplankton and phytoplankton abundance has been reported to depend positive on previous year NAO (Piontkovsi et al. 2006, Beaugrand et al. 2002) and zooplankton especially copepods are one of the major food sources of *C. crangon* (Boddeke et al. 1986, Plagmann 1939). Changes of food composition and abundance influenced by NAO might therefore force recruitment, abundance and mortality of *C. crangon* by bottom up control.

Another possibility could be that correlation between NAOI and mortality is a auto correlation and that mainly temperature influences Z. This is also indicated by the high r^2 of 0.5 that occurs if the years 1993 and 1996 are left out of the regression. Metabolic costs increase with temperature and also with size (van Donk & de Wilde 1981, Ivleva 1980, Hagerman 1976). Since food concentrations are low early in the year and energy reserves of the large shrimp are depleted from the winter an increase in temperature might lead to higher mortality rates.

The harvesting of the fishery as source for a higher mortality in the form of landings did not correlate with shrimp mortality. However, no significant correlation does not mean that there is no influence of the commercial shrimping as total abundance has to be taken into account (discussion below).

Another factor that influences mortality of *C. crangon* is the abundance of potential predators. For large individuals, that were only taken into account in this study, this is mainly cod and whiting (Singh-Renton & Bromley 1999a, Daan 1973) and in deeper water grey gurnard (de Gee & Kikkert 1993). Again no correlation between the landings of the predators was found with shrimp mortality and also no correlation with whiting abundances from DYFS survey data. Even a high abundance of age 1 whiting along the coast of Denmark in autumn 1999 (ICES WGNSSK 2007) did not increase the mortality of *C. crangon* compared to the years 2004 to 2006 where nearly no whiting were caught close to the German or Danish coasts. The reason therefore is that consumption and biomass of the predators and *C. crangon* has to be taken into account (discussion below).

Henderson & Holmes (1987) assumed that mortality of *C. crangon* is density dependent. This could not be substantiated in this study as no correlation between *C. crangon* landings or abundance were observed. On the other hand the possibility cannot be excluded, because the density estimate used in the correlation is based on swept area. This method is dependent on the vulnerability

of the shrimp to the sampling gear and furthermore includes only autumn catches of animals >45 mm.

As discussed before there was no correlation between the determined mortality and: catch, biomass of *C. crangon*, biomass of predators, and landings of predators. The reason might be that natural mortality M and fishing mortality F is always related to biomass:

$$F = \frac{C}{\overline{B}} \text{ and } M = \frac{D}{\overline{B}}$$

with *C* = catch, $\overline{B}$ = biomass and *D* the amount of animals consumed by predators.

$$Z = F + M = \frac{C + D}{\overline{B}} \text{ or } Z = \frac{P}{\overline{B}}$$

with P = total production. Further D is the sum of the product of predator biomass and predator consumption rate.

A correlation of Z with the catch may only be observed if the biomass of *C. crangon* and the predators are constant. For a correlation of Z with predator landings firstly the amount of predators and secondly the amount of *C. crangon* actually consumed by these predators has to be constant. As there is no predator that solely feeds on *C. crangon* constant consumption will not be observed in the field.

Long term trends

Besides the year-to-year variability there is a long-term trend indicating an increase in total mortality that was maximum between 1990 and 1993 and a slight declined afterwards. A reason for the high values might be a salinity anomaly (Heath et al. 1991) and a temperature anomaly that led to mild winters between 1989 and 1994 (Becker & Pauly 1996). Salinity might influence mortality directly by greater metabolic needs with higher temperature (Spaargaren & Haefner Jr. 1987) (Hagerman 1970) but indirect factors influencing mortality are more likely.

In the 1990s a regime shift of several planktonic animals was reported (Beaugrand 2003, Beaugrand et al. 2000) which might have changed the feeding conditions of *C. crangon* (Beaugrand et al. 2002). Parallel to that plankton regime shift a fish regime shift was observed. Litzow et al. (2006) compared fish species composition in boreal oceans and observed that lipid-rich (pelagic) species are favored after a temperature decrease and lipid poor (demersal) species benefit after a temperature increase. Most of the latter species are shrimp predators. About 22% (Hislop et al. 1991) to 40% (Singh-Renton & Bromley 1999b) of the food of whiting is made up by *C. crangon* and for *Pleuronectes platessa* it is about 10% (Basimi & Grove 1985) (Aarnio et al. 1996). About 20% of the food of cod is made up by crustaceans (Daan 1989) and the fraction of *C crangon* is between 31% (10-29 cm size class) and 2% (>70 cm) (Daan 1973). Juvenile grey gurnard nearly solely feed on small *C. crangon* and in stomachs of turbot and dab 30 and 20% (Braber

& de Groot 1973) *C. crangon* was observed, respectively. An increase of several benthivorous species might therefore have led to an increase in observed *C. crangon* mortality. An increase of benthivore fishes has also been described by Heath (2005), who reported maximum abundances of turbot, sole and grey gurnard between 1985 and 1992.

In 1990 a strong year class of whiting was observed in the Dutch Groundfish survey (Knijn & Boddeke 1991) which was assumed to have a lasting effect on the shrimp stock and the fisheries for the following years (Berghahn 1996).

The high total mortality determined in this study during the early 1990s is therefore most probably based on three factors: changes in the food composition, high abundances of potential predators and eventually habitat changes.

Maximum age

Based on the mortality level determined in the actual work it is possible to roughly estimate the age of the population in the field. Assuming a total mortality of 5.7 as determined by us would have the consequence that for only three shrimp out of 1.000 survive until year 1, one out of 100.000 would survive until year 2, and four of 100.000.000 until year 3. In the 1990s this fraction was even lower and only 0.9 of 1.000 and 0.08 of 100.000 survived (Z = 7) until year 1 and 2, respectively. It can therefore be assumed that the majority of the animals observed in the field belong to age class 1, whereas before 1970 the fraction of older animals might have been higher.

Acknowledgements

The study was financially supported by the Federal Ministry of Food, Agriculture and Consumer Protection, Germany Project No. 03HS030.

References

Aarnio K, Bonsdorff E, Rosenback N (1996) Food and feeding habits of juvenile flounder *Platichthys flesus* (L.), and turbot Scophthalmus maximus L. in the Åland archipelago Northern Baltic Sea. Journal of Sea Research 36:311-320

Allen RK (1971) Relation between production and biomass. Fisheries Research Board of Canada 28:1573-1581

Basimi RA, Grove DJ (1985) Estimates of daily food intake by an inshore population of *Pleuronectes platessa* L. off eastern Anglesey, North Wales. Journal of Fish Biology 27:505-520

Beaugrand G (2003) Long-term changes in copepod abundances and diversity in the north-east Atlantic in relation to fluctuations in the hydroclimatic environment. Fisheries Oceanography 12:270-283

Beaugrand G, Reid PC, Ibanez F, Lindley JA, Edwards M (2002) Reorganization of North Atlantic marine copepod biodiversity and climate. Science 296:1692-1694

Beaugrand G, Reid PC, Ibanez F, Planque B (2000) Biodiversity of North Atlantic and North Sea calanoid copepods. Marine Ecology Progress Series 204:299-303

Becker GA, Pauly M (1996) Sea surface temperature changes in the North Sea and their causes. ICES Journal of Marine Science 53:887-898

Berghahn R (1983) Untersuchungen an Plattfischen und Nordseegarnelen (*Crangon crangon*) im Eulitoral des Wattenmeeres nach dem Übergang zum Bodenleben. Helgoländer Meeresuntersuchungen 36:163-181

Berghahn R (1996) Episodic mass invasions of juvenile gadoids into the Wadden Sea and their consequences for the population dynamics of brown shrimp (*Crangon crangon*). Marine Ecology 17:251-260

Beukema JJ (1992) Dynamics of juvenile shrimp *Crangon crangon* in a tidal flat-nursery of the Wadden Sea after mild and cold winters. Marine Ecology Progress Series 83:157-165

Beverton RJH, Holt SJ (1956) A review of methods for estimating mortality rates in fish populations, with special reference to sources of bias in catch sampling. Rapport et Procès-verbaux des Réunions, Conseil international pour L'Exploration de la Mer 140:67-83

Boddeke R (1976) The seasonal migration of the brown shrimp *Crangon crangon*. Netherlands Journal of Sea Research 10:103-130

Boddeke R, Driessen G, Doesburg W, Ramaekers G (1986) Food availability and predator presence in a costal nursery area of the brown Shrimp (*Crangon crangon*). Ophelia 26:77-90

Braber L, de Groot SJ (1973) The food of five flatfish species (Pleuronectiformes) in the Southern North Sea. Netherlands Journal of Sea Research 6:163-172

Daan N (1973) A quantitative analysis of the food intake of north sea cod, *Gaduas morhua*. Netherlands Journal of Sea Research 6:479-517

Daan, N. ed. Data base report of the stomach sampling project 1981. Cooperative research report 164, 144. 1989.

de Gee A, Kikkert AH (1993) Analysis of the grey gurnard (*Eutrigla gurnardus*) samples collected during the 1991 International Stomach Project. International Council for the exploration of the Sea C.M. 1993:1-25

de Graaf G, Dekker P (2006) A simple spreadsheet model to incorporate seasonal growth into length-based stock assessment methods. NAGA World Fish Center Quarterly 29:46-54

del Norte-Campos AGC, Temming A (1998) Population dynamics of the brown shrimp *Crangon crangon* L., in shallow areas of the German Wadden Sea. Fisheries Management and Ecology 5:303-322

Evans S (1984) Energy budgets and predation impact of dominant epibenthic carnivores on a shallow soft bottom community at the Swedish west coast. Estuarine, Coastal and Shelf Science 18:651-672

Gunnarsson B, Ásgeirsson þ, Ingólfsson A (2007) The rapid colonization by *Crangon crangon* (Linnaeus, 1758) (Eucarida, Caridea, Crangonidae) of Icelandic coastal waters. Crustaceana 80:747-753

Hagerman L (1970) The oxygen consumption of *Crangon vulgaris* (Fabricius) (Crustacea, Natantia) in relation to salinity. Ophelia 7:283-292

Hagerman L (1976) The respiration during the molt cycle of Crangon vulgaris (Fabr),(Crustacea, Natantia). Ophelia 15:15-21

Hartnoll R (2001) Growth in Crustacea – twenty years on. Hydrobiologia 449:111-122

Hartsuyker L (1966) Daily tidal migrations of the shrimp *Crangon crangon* L. Netherlands Journal of Sea Research 3:52-67

Havinga B (1930) Der Granat (*C. vulgaris*, Fabr.) in den holländischen Gewässern. Journal du Conseil International pour l'Exploration de la Mer 5:57-87

Havinga BA, Willer (1929) Krebse und Weichtiere. Schweitzerbart'sche Verlagsbuchhandlung, Stuttgart

Heath MR, Henderson EW, Slesser G, Woodward EMS (1991) High salinity in the North Sea. Nature 352:116

Heath MR (2005) Changes in the structure and function of the North Sea fish foodweb, 1973 - 2000 and the impacts of fishing and climate. ICES Journal of Marine Science 62:847-868

Henderson PA, Holmes RHA (1987) On the population biology of the common shrimp *Crangon crangon* (L.) (Crustacea: Caridea) in the Severn Estuary and Bristol Channel. Journal of the Marine Biological Association U.K. 67:825-847

Henderson PA, Seaby RMH, Somes JR (2006) A 25-year study of climatic and density-dependent population regulation of common shrimp *Crangon crangon* (Crustacea: Caridea) in the Bristol Channel. Journal of the Marine Biological Association U.K. 86:287-298

Hislop JRG, Robb AP, Bell MA, Armstrong DW (1991) The diet and food composition of whiting (*Merlangius merlangus*) in the North Sea. ICES Journal of Marine Science 48:139-156

Hoenig JM, Lawing WD, Hoenig NA (1983) Using mean age, mean length and median length data to estimate the total mortality rate. International Council for the exploration of the Sea ICES-CM-1983:

ICES. Report of the *Crangon* Fisheries and life history. ICES CM 2006/G:XX, 1-32. 2006.

ICES (2007) Report of the Working Group on Crangon Fisheries and Life History. International Council for the exploration of the Sea Living resources comittee:1-33

ICES WGNSSK (2007) Working Group on the Assessment of Demersal Stocks in the North Sea and Skagerrak. ICES CM 2007 / ACFM:18 and 30:

Ivleva IV (1980) The Dependence of Crustacean respiration rate on body mass and habitat temperature. Internationale Revue der gesamten Hydrobiologie 65:1-47

Janssen GM, Kuipers BR (1980) On tidal migration in the shrimp *Crangon crangon*. Netherlands Journal of Sea Research 14:339-348

Jones R, van Zalinge NP (1981) Estimates of mortality rate and population size for shrimp in Kuwait waters. Kuwait Bulletin of Marine Science 2:273-288

Knijn RJ, Boddeke R (1991) Estimates of total mortality of berried females of the brown shrimp *Crangon crangon* (L.). International Council for the exploration of the Sea, Shellfish Committee C.M. 1991 / K:25:1-11

Kuipers BR, Dapper R (1981) Production of *Crangon crangon* in the tidal zone of the Dutch Wadden Sea. Netherlands Journal of Sea Research 15:33-53

Kuipers BR, Dapper R (1984) Nursery function of Wadden Sea tidal flats for the brown shrimp *Crangon crangon*. Marine Ecology Progress Series 17:171-181

Litzow MA, Bailey KM, Prahl FG, Heintz R (2006) Climate regime shifts and reorganization of fish communities: the essential fatty acid limitation hypothesis. Marine Ecology Progress Series 315:1-11

Lloyd AJ, Yonge CM (1944) The biology of *Crangon vulgaris* L. in the Bristol channel and severn estuary. Marine Biological Association of the UK 26:626-661

Meyer-Waarden PF, Tiews K (1965) Grundlagenmaterial zu "Die Zusammensetzung der Fänge der deutschen Garnelenfischerei in den Jahren 1954 - 1960". Veröffentlichungen des Instituts für Küsten- und Binnenfischerei1-29

Miranda LE (2002) Establishing size-based mortality caps. North American Journal of Fisheries Management 22:433-440

Neudecker T (2001) Der Demersal Young Fish Survey (DYFS) in Schleswig-Holstein, Entwicklung und derzeitiger Stand. In: Landesamt für den Nationalpark Schleswig-Holsteinisches Wattenmeer (Hrsg.): Wattenmeermonitoring 2000.Schriftenreihe Nationalpark Schleswig-Holsteinisches Wattenmeer Sonderheft:24-30

Neudecker T, Damm U (2006) 2005: Rekord-Anlandungen an Nordseegarnelen (*Crangon crangon* L.) in Europa. Informationen aus der Fischereiforschung 53:80-81

Neudecker T, Fischer J, Damm U (1998) Influence of Tidal Currents on Fishing Performance in the Wadden Sea. International Council for the exploration of the Sea C.M. 1998/BB:6:1-11

Neves A, Cabral HN, Gordo LS (2007) Distribution and abundance patterns of decapod crustaceans in the sado estuary, Portugal. Crustaceana 80:97-112

Oh C-W, Hartnoll R, Nash RDM (1999) Population dynamics of the common shrimp, *Crangon crangon* (L.), in Port Erin Bay, Isle of Man, Irish Sea. ICES Journal of Marine Science 56:718-733

Pauly D (1990) Length-converted catch curves and the seasonal growth of fishes. Fish Bytes 8:33-38

Pauly D (1983) Length converted catch curves: A powerful tool for fisheries research in the tropics (part I). Fish Bytes 1:9-13

Peterson I, Wroblewski JS (1984) Mortality rate of Fishes in the Pelagic Ecosystems. Canadian Journal Fisheries and Aquatic Science 41:1117-1120

Pihl L, Rosenberg R (1982) Production, abundance, and biomass of mobile epibenthic marine fauna in shallow waters, western Sweden. Journal of Experimental Marine Biology and Ecology 57:273-301

Piontkovsi SA, O'Brien TD, Umani SF, Krupa EG, Stuge TS, Balymbetov KS, Grishaeva OV, Kasymov AG (2006) Zooplankton and the North Atlantic Oscillation: a basin-scale analysis. Journal of Plankton Research 28:1039-1046

Plagmann J (1939) Ernährungsbiologie der Garnele (*Crangon vulgaris* Fabr.). Helgoländer wissenschaftliche Meeresuntersuchungen 2:113-162

Powell DG (1979) Estimation of mortality and growth parameters from the length frequency of a catch. Rapport et Procès-verbaux des Réunions, Conseil international pour L'Exploration de la Mer 175:167-169

Schrum C, Backhaus JO (1999) Sensitivity of atmosphere-ocean heat exchange and heat content in North Sea and Baltic Sea. A comparative assessment. Tellus 51:526-549

Schrum C, Siegismund M, St.John M (2002) Decadal variations in the stratification and circulation patterns of the North Sea - are the 90's unusual? ICES Symposium on Hydrobiological Variability in the ICES Area 1990-1999

Siegel V, Gröger J, Neudecker T, Damm U, Jansen J (2005) Long-term variation in the abundance of the brown shrimp *Crangon crangon* (L.) population of the German Bight and possible causes for its interannual variability. Fisheries Oceanography 14:1-16

Singh-Renton S, Bromley PJ (1999a) Feeding of small whiting (*Merlangius merlangus*) in the central and southern North Sea. Journal of the Marine Biological Association U.K. 79:957-960

Singh-Renton S, Bromley PJ (1999b) Feeding of small whiting (*Merlangius merlangus*) in the central and southern North Sea. Journal of the Marine Biological Association U.K. 79:957-960

Spaargaren DH (2000) Seasonal and annual variations in the catches of *Crangon crangon* (L.1758) (Deacapoda, Natantia) near the coast of Texel, the Netherlands. Crustaceana 73:547-563

Spaargaren DH, Haefner Jr. PA (1987) The effect of environmental osmotic conditions on blood and tissue glucose levels in the brown shrimp *Crangon crangon* (L.). Comparative Biochemistry and Physiology 87A:1045-1050

Sparre P, Ursin E, Venema SC (1989) Introduction to tropical fish stock assessment. Food and Agriculture organization of the united nations, Rome

Ssentongo GW, Larkin PA (1973) Some simple methods of estimating mortality rates of exploited fish populations. Journal of the Fisheries Research Board Canada 24:2355-2453

Summerfelt RC, Hall GE (1987) Age and Growth of Fish. Iowa State University Press,

Temming, Axel, Damm, Ulrich, and Neudecker, Thomas. Trends in the size composition of commercial catches of brown shrimp (*Crangon crangon* L.) along the German coast. K: 53. 1993. ICES Shell fish Committee.

Tiews K, Schumacher A (1982) Assessment of brown shrimp stocks (*Crangon crangon* L.) off the German coast for the period 1965-1978. Archiv für Fischereiwissenschaft 32:1-11

Tiews K (1954) Die biologischen Grundlagen der Büsumer Garnelenfischerei. Berichte der Deutschen Wissenschaftlichen Kommission für Meeresforschung 13:235-269

van der Baan SM (1975) Migrations of *Crangon crangon* in surface waters near the "Texel" Lightship. Netherlands Journal of Sea Research 9:287-296

van Donk E, de Wilde PAWJ (1981) Oxygen consumption and motile activity of the brown shrimp *Crangon crangon* related to temperature and size. Netherlands Journal of Sea Research 15:54-64

van Keeken, O. A., Bolle, L. J., and Verver, S. Quality manual part I: National data collection and raising procedures. Report Number: C041/05. 2008. RIVO-Netherlands Institute for Fisheries Research.

van Lissa JHL (1977) Aantallen, voedselopname, groei en produktie van de garnaal (*Crangon crangon* L.) in een getijdengebied, alsmede de voedselopname en groei onder laboratoriumomstandigheden. Interne Verslagen Nederlands Instituut voor Onderzoek der Zee, Texel 1977-C:1-100

Viegas I, Martinho F, Neto J, Pardal M (2007) Population dynamics, distribution and secondary production of the brown shrimp *Crangon crangon* (L.) in a southern European estuary. Latitudinal variations. Scientia Marina 71:451-460

Welleman, H. C. and Storebeck, F. Annex 7: Quantifying the effects of infrastructural works on brown shrimp populations: a habitat modelling approach. CM 2002 IE:05, 45-52. 2002. Report of the Working Group on Marine Habitat Mapping.

Wetherall JA, Polovina JJ, Ralston S (1987) Estimating growth and mortality in steady-state fish stocks from length frequency data. In: D.Pauly and G.R.Morgan, Editors, Length-based Methods in Fishery Research53-74

Zheng X, Pierce GJ, Reid DG, Jolliffe IT (2002) Does the North Atlantic current affect spatial distribution of whiting? Testing environmental hypotheses using statistical and GIS techniques. ICES Journal of Marine Science 59:239-253

Appendix

Table 9-5: θ = Z/k calculated with different methods. Used length frequency distributions originating from Demersal Fish Survey data from the Netherlands, Demersal Young Fish Survey data, Germany and the Bycatch Series, East Friesian Wadden Sea.

year	L_∞	k	Linf Wetherall	Linf Powell	Z/k Wetherall	Z/k Powell	Z/k Beverton& Holt	Z/k Jones&Zalinge	Z/k Hoenig	Z/k Ssentongo&Larkin	Z/k LCCC	Z/k sLCCC (Lookup)
Demersal Fish Survey (DFS) Netherlands												
1970	85.4	0.99	79.5	91.4	4.6	4.8	5.6	6.1	5.5	5.5	5.6	8.2
1971	80.6	1.12	78.3	82.9	4.5	3.9	5.6	5.0	6.0	5.6	4.9	6.8
1972	80.8	1.11	74.6	87.1	3.9	4.5	4.9	5.2	5.0	4.8	4.8	7.5
1973	80.8	1.11	74.6	87.1	3.9	4.5	4.9	5.2	5.0	4.8	4.8	7.5
1974	82.2	1.07	80.9	83.5	4.6	3.9	5.2	5.2	6.3	5.1	4.9	6.6
1975	75.7	1.28	72.1	79.2	3.6	3.8	4.2	4.6	4.4	4.2	4.4	5.9
1976	74.7	1.31	76.1	73.4	4.3	3.1	4.8	4.4	4.9	3.9	4.2	6.0
1977	79.5	1.15	78.4	80.7	5.5	4.2	3.8	6.0	3.6	3.8	5.1	5.8
1978	82.7	1.06	78.1	87.3	3.9	4.0	4.5	5.0	4.3	4.5	4.2	7.1
1979	76.0	1.26	77.2	74.7	3.6	2.7	3.3	4.0	3.3	3.3	3.4	4.6
1980	79.3	1.16	79.3		4.2	4.8	5.2	5.6	5.4	5.3	5.0	5.8
1981	81.6	1.09	77.2	86.0	4.2	4.3	5.2	5.3	5.8	5.1	4.9	5.8
1982	80.3	1.13	75.0	85.6	4.1	4.5	5.1	5.7	5.2	5.1	5.1	6.0
1983	79.6	1.15	79.6	79.6	4.5	3.5	4.4	5.4	4.5	4.4	4.2	6.0
1984	77.3	1.22	74.3	80.3	3.7	3.7	4.2	4.6	4.2	4.2	4.0	5.6
1985	83.8	1.03	78.0	89.6	4.1	4.4	5.1	5.3	5.2	5.1	4.3	5.8
1986	80.8	1.11	76.9	84.7	4.1	4.2	4.9	5.2	4.9	4.8	4.4	6.1
1987	83.4	1.04	77.8	89.0	4.1	4.4	5.1	5.2	5.2	5.0	4.6	7.5
1988	82.2	1.07	78.0	86.4	4.3	4.3	5.0	5.4	5.1	5.0	4.5	5.4
1989	84.1	1.02	77.3	91.0	4.2	4.8	5.5	5.6	5.2	5.4	5.1	6.2
1990	87.2	0.95	81.6	92.8	4.9	5.0	6.1	6.1	5.6	6.1	5.6	7.7
1991	79.4	1.16	77.8	80.9	4.7	4.1	5.2	5.5	4.8	5.4	4.8	6.6
1992	82.8	1.06	86.1	79.6		4.2	6.0	6.7	5.7	6.0	6.5	8.4
1993	78.9	1.17	78.9		4.7	5.7	6.5	6.5		6.3	5.9	8.3
1994	76.0	1.26	76.4	75.6	4.4	3.5	4.5	4.9	4.6	4.4	4.5	6.1
1995	82.4	1.07	81.9	82.8	4.9	4.0	5.2	5.6	4.6	5.0	5.0	6.1
1996	84.3	1.02	84.3		5.3	5.2	6.4	6.7	6.1	6.3	5.7	8.4
1997	82.3	1.07	75.1	89.5	4.2	5.0	5.6	6.0	6.1	5.5	5.1	6.5
1998	79.7	1.14	77.0	82.5	4.0	3.8	4.4	4.8	4.3	4.4	4.0	5.4
1999	75.8	1.27	75.6	76.1	4.4	3.7	4.7	4.9	4.5	4.5	4.6	6.0
2000	84.7	1.01	84.7		5.6	5.5	6.9	7.1		6.7	6.6	9.0
2001	78.9	1.17	79.7	78.1	4.5	3.5	4.5	5.3	4.2	4.5	4.1	7.1
2002	81.3	1.10	79.4	83.2	4.6	4.1	5.2	5.5	5.1	5.0	5.0	6.5
2003	80.7	1.12	79.2	82.1	4.7	4.2	5.2	5.6	5.1	5.2	5.0	7.6
2004	86.1	0.97	83.4	88.8	5.3	4.8	6.2	6.2	5.7	6.0	6.2	8.7
2005	79.3	1.16	78.1	80.5	4.3	3.8	4.7	5.1	4.2	4.6	4.4	6.0
2006	82.4	1.07	82.4		4.7	4.9	5.6	6.0	5.9	5.7	5.1	7.6

year	L_∞	k	Linf Wetherall	Linf Powell	Z/k Wetherall	Z/k Powell	Z/k Beverton& Holt	Z/k Jones&Zalinge	Z/k Hoenig	Z/k Ssentongo&Larkin	Z/k LCCC	Z/k sLCCC (Lookup)
Demersal Young Fish Survey (DYFS), Germany												
1997	83.6	1.04	81.7	85.4	5.0	4.3	5.7	5.8	6.2	5.7	5.9	7.6
1998	83.1	1.05	79.3	86.8	3.6	3.4	3.8	4.6	3.9	3.8	4.0	5.3
1999	79.1	1.16	79.1		4.5	5.0	5.7	6.0	5.8	5.7	5.6	6.2
2000	79.0	1.17	79.0		4.1	4.6	5.3	5.3	5.8	5.3	4.9	6.8
2001	78.3	1.19	75.2	81.5	3.2	3.1	3.4	3.8	3.4	3.4	3.1	3.9
2002	81.9	1.08	74.6	89.3	3.8	4.6	4.9	5.3	4.7	4.9	4.9	5.7
2003	76.2	1.26	76.2		4.3	5.3	5.9	6.1	6.0	5.9	5.9	6.4
2004	83.4	1.04	79.1	87.6	4.2	4.1	4.7	5.3	4.8	4.7	4.5	5.5
2005	83.0	1.05	78.2	87.7	4.0	4.1	4.6	5.1	4.8	4.8	4.4	5.4
2006	80.1	1.13	77.8	82.5	4.0	3.7	4.5	4.7	4.5	4.6	4.2	5.4
East Frisian Bycatch Series (EF) Germany												
1958	84.8	1.01	84.5	85.0	2.5	4.1	4.6	3.8	4.1	4.5	4.0	6.2
1959	78.2	1.19	82.0	74.4	1.5	2.6	2.8	2.8		2.4		2.9
1960	75.3	1.29	78.8	71.8	2.4	5.1	4.9	4.1	7.6	4.6	4.3	7.1
1961	81.9	1.08	88.2	75.6	3.6	3.5	4.9	4.3	3.9	4.2	3.9	6.4
1962	78.7	1.18	80.4	77.0	6.4		6.8	7.3	7.3	7.3	8.0	
1963	72.0	1.41	76.4	67.6	2.4	3.1	3.6	3.5	3.1	3.4	2.8	6.2
1964	82.2	1.07	83.4	81.0	3.5	4.5	5.4	4.7	7.9	5.3	4.1	7.9
1965	78.9	1.17	81.5	76.3	3.9	4.4	5.1	4.8	7.4	4.1	4.3	8.4
1966	83.0	1.05	82.1	83.8	3.4	4.8	5.5	5.1	8.0	5.7	4.3	8.2
1967	79.9	1.14	80.6	79.3	3.5	5.2	6.2	5.6	5.9	6.6	6.1	7.9
1969	74.8	1.31	80.5	69.1	2.8	2.8	3.4	3.6		3.7		4.5
1970	72.9	1.38	78.8	67.1	2.2	2.4	2.8	3.1		2.9		4.6
1972	76.7	1.24	80.1	73.4	3.1	3.8	4.7	4.2	5.4	5.0	3.6	6.1
1974	77.8	1.20	78.9	76.6	2.2	3.6	3.9	3.5	2.9	4.2	3.1	4.1
1975	75.1	1.30	78.4	71.7	3.3	4.1	5.1	4.4	3.3	5.4	3.9	6.3
1976	75.0	1.30	80.9	69.1	3.4	3.3	4.4	3.7	5.2	5.5	3.9	5.7
1977	76.0	1.26	82.8	69.3	4.7	4.3	4.3	5.4	5.4	4.6	5.1	7.9
1978	76.5	1.25	83.0	70.0	4.5	4.1	5.4	4.9	5.4	4.3	4.6	7.7
1979	73.3	1.36	79.0	67.7	2.7	2.8	3.5	3.2	4.9	3.7	2.8	4.9
1980	76.8	1.24	80.4	73.1	3.0	3.6	4.4	4.0	5.4	4.8	3.3	6.7
1981	80.6	1.12	82.7	78.4	3.9	5.0	6.3	5.1	6.1	6.7	4.9	8.1
1982	73.4	1.36	80.4	66.5	3.5	3.0	4.1	3.9	4.9	4.4	3.2	6.3
1983	78.9	1.17	78.9		3.8	3.6	5.0	4.0	4.8	4.4	4.0	6.6
1984	78.7	1.17	86.7	70.7	5.1	4.0	4.8	5.5	5.8	5.1	4.6	6.5
1985	77.4	1.22	78.8	76.1	3.4	5.0	5.1	4.7	5.5	5.5	4.7	6.4
1986	79.7	1.15	82.0	77.3	3.8	4.9	5.0	5.1	5.9	5.4	4.7	7.9
1987	79.0	1.17	82.2	75.7	4.6	5.3	6.0	5.8	5.8	6.4	5.3	9.9
1988	76.3	1.25	81.3	71.3	5.4	5.6	5.4	5.5	6.2	5.8	5.6	8.5
1989	75.4	1.28	81.9	68.9	5.6	5.8	6.0	5.8	6.1	4.7	5.1	8.9
1990	74.6	1.31	77.6	71.6	4.4		4.6	5.9	5.9	4.9	6.2	9.7
1991	83.8	1.03	86.2	81.4	6.4		6.2	7.9	7.4	6.6	7.5	8.9
1992	78.9	1.17	86.4	71.4	6.7	4.9		6.4	5.8	7.5	6.8	8.2
1993	77.8	1.20	82.7	73.0	5.1	5.4	6.1	6.1	5.6	6.6	5.5	5.5

Table 9-6: θ = Z/k calculated with different methods. Used length frequency distributions originating from the Büsum Bycatch Series.

year	L_∞	k	Linf Wetherall	Linf Powell	Z/k Wetherall	Z/k Powell	Z/k Beverton& Holt	Z/k Jones&Zalinge	Z/k Hoenig	Z/k Ssentongo&Larkin	Z/k LCCC	Z/k sLCCC (Lookup)
Büsum Bycatch Series (EF) Germany												
1955	75.0	1.30	77.4	72.6	1.8	3.8	2.9	3.5	2.9	3.6		4.9
1956	79.4	1.15	79.9	78.9	3.5		4.6	4.8	3.3	5.0	5.0	6.9
1957	84.5	1.01	86.1	82.8	3.8	4.8	5.8	4.6	8.3	6.2	4.7	8.6
1958	88.8	0.91	89.2	88.4	3.1	4.6	5.4	4.2	8.2	5.3	4.4	6.4
1959	88.5	0.92	90.1	86.9	3.4	4.4	5.3	4.5	8.1	5.4	4.1	7.4
1960	83.3	1.04	83.4	83.2	2.9	4.3	4.9	4.2	7.3	5.2	3.7	6.6
1961	84.0	1.03	84.0		2.5	4.9	5.1	4.4	8.1	5.4	3.7	7.0
1962	82.3	1.07	85.8	78.8	3.1	3.8	5.1	3.5	7.2	5.3	3.9	6.0
1963	83.2	1.05	83.3	83.0	2.0	3.5	3.7	3.3	3.6	3.8	2.6	5.0
1964	83.9	1.03	85.4	82.3	2.9	4.1	4.8	3.9	7.4	5.0	3.5	6.5
1965	83.4	1.04	85.1	81.7	3.2	4.4	5.2	4.2	7.3	5.4	3.9	6.4
1966	76.1	1.26	81.6	70.5	2.2	2.7	3.1	2.7	3.0	3.3		4.9
1967	83.4	1.04	83.0	83.7	3.5	5.1	5.9	5.1	7.3	6.3	4.5	8.2
1969	81.4	1.10	82.2	80.5	2.8	4.1	4.6	4.3	7.0	5.0	3.5	7.7
1970	75.6	1.28	80.0	71.2	1.9	2.6	2.7	2.5	3.0	2.9		4.5
1971	77.6	1.21	77.9	77.2	1.6	3.3	3.4	3.2	2.8	3.6	2.7	4.7
1972	79.6	1.15	80.2	79.0	2.7	4.1	4.7	4.1	6.7	5.0	3.4	7.0
1973	77.3	1.22	80.1	74.4	3.6	4.6	4.7	4.6	6.3	5.0	4.3	7.6
1974	79.3	1.16	79.1	79.4	3.1	4.9	5.6	4.8	5.8	6.1	4.5	8.1
1975	80.1	1.13	79.9	80.3	2.4	4.1	4.5	3.9	6.0	4.8	3.4	7.3
1976	77.7	1.21	79.0	76.3	3.3	4.7	5.6	4.8	5.6	6.1	4.5	7.7
1977	76.7	1.24	79.1	74.2	2.0	3.1	3.4	3.2		3.6	2.6	5.1
1978	75.6	1.28	78.5	72.6	2.7	3.5	4.2	3.8	5.2	4.5	3.4	6.2
1979	76.5	1.25	78.4	74.5	2.2	3.4	3.7	3.5	5.4	4.0	2.9	5.6
1980	75.3	1.29	78.3	72.3	2.5	3.4	4.1	3.5	5.2	4.2	3.2	5.8
1981	76.5	1.25	79.3	73.8	2.5	3.4	4.2	3.5	5.4	4.2	3.1	5.6
1982	77.5	1.21	83.8	71.2	4.5	4.1	6.0	4.8	5.6	6.0	4.7	7.1
1983	75.3	1.29	77.4	73.1	2.8	4.1	4.9	4.1	5.2	5.2	3.9	7.3
1984	79.1	1.16	79.1		3.3	3.0	4.7	3.3	4.8	4.4	3.5	5.5
1985	76.4	1.25	80.1	72.8	3.4	4.0	5.1	4.4	5.4	5.3	4.2	7.5
1986	76.8	1.24	80.8	72.8	3.4	4.0	5.5	4.3	5.5	5.4	4.1	6.9
1987	76.0	1.27	82.7	69.2	4.2	3.7	5.7	4.6	5.4	5.5	4.7	7.2
1988	74.0	1.33	78.8	69.3	3.4	3.8	5.6	4.0	5.0	5.3	4.0	6.4
1989	77.5	1.21	79.8	75.2	3.2	4.3	5.3	4.4	5.6	5.5	4.1	7.5
1990	76.8	1.24	78.6	75.0	3.8	5.4	6.0	5.2	5.4	6.5	5.2	9.1
1991	74.9	1.30	80.3	69.4	4.6	4.6	5.3	5.2	5.2	5.7	5.4	5.5
1992	72.1	1.41	76.8	67.3	3.1	3.5	4.4	3.7	4.7	4.8	4.0	6.4
1993	78.7	1.18	81.5	75.9	4.4	5.6	6.5	5.5	5.8	7.0	5.7	9.3
1994	76.9	1.23	82.2	71.5	7.1	6.2	5.6	7.3	6.3	6.0	7.1	9.6
1995	75.4	1.28	82.8	68.1	3.6	3.0	4.7	4.0	5.3	4.4	3.9	5.7
1996	82.6	1.06	86.6	78.6	5.2	5.5	7.4	6.2	6.4	7.9	6.1	9.6
1997	76.9	1.23	86.2	67.6	6.1	4.5		5.4	5.5	4.9	5.7	8.5

The brown shrimp a protandric hermaphrodite? Relevance of this sexual system for its population dynamics.

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Abstract

Brown shrimp, *Crangon crangon* were obtained from intertidal habitats along the German Wadden Sea coast near Büsum (2000, 2006 and 2007) and Wilhelmshaven (2005−2007). The calculation of length-based and seasonal sex ratios revealed a bias towards females in all size classes during most of the year. This observation, plus the regular occurrence of large males in the catches, indicates that *C. crangon* is, if ever, a facultative hermaphrodite. Variation of the sex ratio for specific length classes (30-40 and 40-50 mm) between the seasons is explained by migration while the decline of the male fraction with increasing length is related to sex-specific growth rates. Including previously determined growth and moulting rates at different temperatures, as well as numbers of length-specific eggs per female, into a mathematical model, the probable amount of eggs released by primary and secondary females can be estimated. The potential maximum egg contribution by secondary females to the population was estimated to be about 4% but is most probably < 1% of the total eggs produced by one cohort of male and female *C. crangon*.

Key words: Sex change, sex ratio, *Crangon crangon*, hermaphroditism, growth

Introduction

The brown shrimp *Crangon crangon* (Crustacea, Decapoda) is common in European coastal waters and of great commercial importance (Pihl & Rosenberg 1984, Henderson & Holmes 1987, Bulnheim & Schwenzer 1993, Oh et al. 1999, Cuesta et al. 2006, Gunnarson et al. 2007). Adult shrimps > 50 mm are fished intensively in German, Dutch and Danish waters and to a smaller extend also in France, Spain, Portugal and Great Britain. Total annual landings from the North Sea exceed 30000 t (ICES 2007) with an estimated value of 100 million €.

Adult and especially egg-bearing shrimps inhabit sublittoral areas where newly hatched larvae are often encountered. After completion of planktonic life phases, consisting of 5 zoeal stages (Criales & Anger 1986), at a total length of 5–7 mm (Boddeke 1966, Boddeke et al. 1991) the juvenile shrimps invade shallow, tidally influenced areas in late spring (Berghahn 1983, Martin 1995). The occurrence of adult shrimps in commercial landings peaks in late fall (Siegel et al. 2005). There is strong evidence that the peak of adult shrimps in fall is generated by the juveniles in spring (Hufnagl & Temming 2009) that were hatched from winter eggs (Temming & Damm 2002).

In the past, *C. crangon* has been considered to be an obligate protandric hermaphrodite, with all male shrimps > 37 mm changing sex between March and July depending on whether they participated in mating in winter or spring. These conclusions were mainly based on research on the structure of gonads (Boddeke 1966). However, sex change was not observed within former laboratory experiments (Meixner 1969b, Tiews 1970) or in ovary studies of field-caught animals (Klek-Kawinska & Bomirski 1975, Oh & Hartnoll 2004).

Histological field investigations found developing oocytes within 2.5–3.0% of males sampled in August and 9.2% of males sampled in September, possibly resulting from sex reversal (Martens and Redant 1986). In that study, only animals actually displaying developing oocytes were described. Completely reversed, secondary females were not detected using their methods; thus it might be possible that the fraction of secondary females was actually higher than the observed 9.2%.

In a recent laboratory trial, 70 mature males between 26-39 mm, total length were observed over several moult cycles (Schatte & Saborowski 2005). During eight months of observations only one of the reared shrimps changed its external sex characteristics from male to female. A factor potentially biasing the results of Schatte & Saborowski (2005) was, that shrimps were kept in isolation in their laboratory study, thus excluding any social and environmental influences that could be important stimuli to induce sex reversal.

The work of Martens & Redant (1986) and Schatte & Saborowski (2005) indicated that *C. crangon* has the potential to display developing oocytes and change external sexual characteristics, such as reducing the appendix masculina, but the relevance of this for the population dynamics of *C. crangon* and the individual benefit remains unclear.

Not only the proportion in a population of shrimps changing sex, but also the exact time span when the reversal occurs remains unknown to date. Boddeke (1966) stated that male shrimps that took part in the mating in October to February change their sex in March, whereas those that mated in March to June change sex in April to July. In contrast to these findings, Martens & Redant (1986) observed a large fraction of males with developing oocytes in September.

However, for a small number of males, it might be profitable to change gender, but only if this change occurs at the right point in time. For example, if sex change occurs mainly in spring only a few large males from the last season would be included in the process. On the other hand, if sex reversal takes place mainly in the fall, a larger number of males would significantly increase the spawning stock of fertile females as secondary female in winter. These winter eggs are the source of the large amount of new recruits in spring (Temming & Damm 2002).

A modelling approach accounting for the actual mortality levels and the seasonal timing of recruitment and maturity is essential to advance the quantitative understanding of sex reversal in brown shrimp populations. However, as a precondition, reliable data are needed on the seasonally varying share of primary females in the central areas of the brown shrimp population.

In recent studies (Martens & Redant 1986, Henderson 7& Holmes 1987, Siegel et al. 2008) declining amounts of males with increasing length were observed. For *Crangon franciscorum* such observations led to the hypothesis that sex change is present (Gavio et al. 2006). The negative relationship between length and the proportion of males (Martens & Redant 1986, Siegel et al. 2008) could also originate from sex-specific growth rate differences.

The objectives of this work were therefore

to determine the share of primary females in the nursery areas and tidal flats in the core distribution area of *C. crangon,*

to develop a simple population model to test if sex specific growth rates can lead to the observed decline of male shrimps towards larger length classes,

to apply the population model to calculate the fractions of eggs of secondary females contributed to the whole egg production of the population.

Material and Methods

Brown shrimp *C. crangon* were caught once a month at Büsum (54°07’N, 8°51’E) from June to October 2000 and from January 2006 to December 2007 with a pushnet (width 1.44 m, height 0.23 m, mesh size 1.8 mm) in a water depth of approximately 1 m during low tide. When enough animals were available, 250 animals from each haul were sexed, sized and weighed.

Samples from the East Frisian Wadden Sea areas (53°34’,N8°09’E)were obtained from the cooling water intake of the power plant in Wilhelmshaven between 2005 and 2007. Aquatic organisms and other material was filtered from the cooling

water by two consecutively mounted grid screens, the second of which retained *C. crangon* >35-40 mm effectively. The grid was automatically cleaned approximately every 120 min (depending on the pressure loss in the cooling cycle) and the retained animals were brushed into metal tubes, collected and stored frozen (-20°C) until further processing.

Total length was measured to the mm below from the telson to the tip of the scaphocerite. The sex of the animals was determined according to Tiews (1970) by the length of the endopodite of the first pleopod and the presence of an appendix masculina. A reliable classification of the sex by using these criteria was possible for all animals > 25 mm. A total of 27000 animals were analyzed.

The sex ratio was calculated as

$$SR = \frac{male}{male + female} \quad (42)$$

The adult sex ratio (ASR) includes only males larger 40 mm and ripe females (see below) therefore it was estimated according to

$$ASR = \frac{males\ (>40mm)}{males\ (>40mm) + egg\ carrying\ \ females} \quad (43)$$

The number of fecund females was calculated according to the sigmoid function for egg bearing females with parameters determined in (Oh & Hartnoll 2004) (L = total length)

$$egg\ carrying\ \ females = \frac{1}{1 + e^{10.35 - 0.83 \cdot \frac{(L+0.816)}{4.456}}} \quad (44)$$

Sex ratio and sex specific growth rates

Artificial length frequency distributions with different growth rates for male and female *C. crangon* were created with a PASCAL routine. In this model several assumptions were made according to the life cycle of the shrimp.

Mean maximum length L_∞ and θ=Z/k

The ratio of total mortality Z and growth k (von Bertalanffy): θ, as well as the mean maximum length L_∞ for male and female shrimps were estimated with the modified Wetherall et al. method (Wetherall et al. 1987, Sparre et al. 1989) from the length frequency distributions gained from the samples taken in Büsum and Wilhelmshaven.

Table 10-1: Parameters b, c and d of growth equation (5) and Bertalanffy growth constant k and maximum length L∞ determined for 10°C water temperature.

values	b	c	d	r^2	k (mean T 10°C)	L_∞ (mean T 10°C)
male	0.03238	0.00187	0.0951	0.51	1.76	67.0
female	0.03946	0.00177	0.0951	0.92	1.67	86.1

Growth

Growth was described as the first derivative of the von Bertalanffy growth curve describing growth as the net effect of anabolism (left term) minus metabolism (right term):

$$\frac{dW}{dt} = H \cdot W^{\frac{2}{3}} - K \cdot W \qquad (45)$$

with W = weight, K = catabolic constant, equivalent to % weight loss per time in starving individuals and H = anabolic constant, related to food intake and synthesis of body mass. Transformation of (2) to length growth ($W \sim L^3$) gives

$$\frac{dL}{dt} = E - k \cdot L \qquad (46)$$

with L = length, k = K/3 and E representing anabolism. This equation was used with temperature included into the constant E to calculate daily length growth of shrimp as a function of length and temperature (Kuipers & Dapper 1981).

$$\frac{dL}{dt} = a + b \cdot T - k \cdot L \qquad (47)$$

where *a* and *b* are constants and *T* is Temperature.

Metabolism is not only length-dependent but also increases exponentially with increasing temperature (Gillooly et al. 2001). An exponential temperature term was therefore included into (4) with $k = c \cdot e^{d \cdot T}$:

$$\frac{dL}{dt} = a + b \cdot T - c \cdot e^{d \cdot T} \cdot L \qquad (48)$$

where *a*, *b, c* and *d* are constants.

In a literature study combined with laboratory data for male and female *C. crangon* parameter *d* was determined. Parameter *a* was not significantly different from zero and therefore omitted. Parameter d, which describes the temperature dependence of *C. crangon* metabolism, was found to be very similar for male and female shrimps and for both sexes combined (Hufnagl and Temming, 2009) (Table 10-1).

The growth model determined for male and female shrimps by Hufnagl and Temming (2009) was based on literature data and data determined by the authors but did include only few data points for large animals. To reduce uncertainties and to parameterize the growth model to the conditions observed in Wilhelmshaven and Büsum, equation (5) was refitted to the data including the L_∞ determined with

the length frequency data. Therefore the parameter *b* in (5) was expressed in dependency of parameter *c* as

$$b = \frac{L_\infty \cdot \left(-c \cdot e^{d \cdot T}\right)}{T} \qquad (49)$$

with T = mean water temperature (10°C), *d* = 0.0951 as determined for the whole stock (male and female shrimps combined along Dutch and German coast) and L_∞ estimated with the Wetherall method, leaving *c* as the only unknown parameter in eq. (5) to determine male and female specific growth rates. This parameter was then estimated with the previously mentioned data set of laboratory and literature data described in (Hufnagl and Temming, 2009).

Total mortality Z

The mortality was calculated from the estimated growth rate k (Table 10-1) and θ (Figure 10-6) determined with the modified Wetherall et al. function as $Z = \theta \cdot k$.

For both male and female shrimps a Z of 7.2 was estimated based on the field data from Wilhelmshaven and Büsum.

For the calculation of the artificial length frequency distributions the mortality of the animals was chosen variable over the year. Based on observations on seasonal predator densities in the Belgian (bib and whiting) and German Wadden Sea (whiting and cod) (Hamerlynck & Hostens 1993, Temming et al. 2000, Jansen 2002) the mortality in the model was increased in fall and lowered in spring by 80%.

Recruitment

The recruitment pattern was calculated as described in Temming & Damm (2002). First an egg index representing the amount of eggs present in the field was calculated based on the amount of egg bearing females, the amount of eggs carried per females and the moult interval. Egg development according to temperature was then calculated based on the function presented by Redant (1978).

Simulating length frequency distributions

To obtain the length frequency distributions for each day of the year, a simulation for a cohort of male and a cohort of female shrimps was run with relative cohort size determined by the recruitment pattern. Equilibrium within the model was assumed to be reached when only 0.01% of the first cohort was left. From that point on, the model was run for another year and the mean length frequency distribution of that last year was calculated.

Estimation of the fraction of eggs contributed to the stock by secondary females

Moult interval and egg development

A function for calculating the moult interval was parameterized based on literature and laboratory data (Hufnagl and Temming, 2009):

$$mi = 5.7066 \cdot L^{0.7364} \cdot e^{-0.09363 \cdot T} \quad (50)$$

The intermoult period was calculated based on the temperature experienced during each day by each cohort in the model. This was achieved by giving the cohort the value zero in the beginning of the intermoult period and adding the reciprocal of equation (9) each day which equals the fraction of the intermoult period contributed by one day at the given temperature. When the value 1 was reached, the moult interval was completed and the value was reset to zero.

The egg development time of *C. crangon* was coupled to the moult cycle of the female shrimps (Havinga 1930) and corresponds to the moult interval described by equ. (9)(Tiews 1954, Meixner 1969a, Wear 1974, Kuipers & Dapper 1984). Therefore equ. (9) was also used to calculate the egg development.

At 55 mm length 50% of the female shrimps in the field carry eggs (Henderson & Holmes 1987, Oh et al. 1999, Oh & Hartnoll 2004). In the calculation this length was therefore chosen as length when 100% of all females are capable of carrying eggs. The total number of eggs carried per primary and secondary female was calculated, according to Redant (1978), as

$$n\,egg = 0.01878 \cdot L^3 \quad (51)$$

Sex change

According to Martens & Redant (1986) most of the male shrimps with developing oocytes had a length of 45–47 mm; therefore 45 mm was chosen in the calculation as length of sex reversal.

Three scenarios were calculated which differed with regard to the percentage of the population that changed sex:

All males changed sex

9% of males changed sex (Martens & Redant 1986)

1.4% of males changed sex (Schatte & Saborowski 2005)

Calculating the fraction of eggs contributed by secondary females

Within the simulation, at the 1st of each month, one cohort of male and one cohort of female shrimps was started at a length of 5 mm, roughly equivalent to postlarval length (Criales & Anger 1986). A fraction of animals in this cohort died each day according to the total mortality Z previously determined (Z =7.2).

A length of 55 mm at maturity was chosen as previously described. Each day after reaching a length of 55 mm, the moult interval according to equ. (9) was used to determine egg development time and equ. (51) was used to calculate the number of eggs released. The number of animals in the cohort remaining at the end of the moult interval when the eggs were released, was multiplied with the calculated number of eggs. The sum of all eggs released by the female cohort during one year (Σ $eggs_{pri.fem.}$) was calculated to provide a relative measure of productivity.

The simulation for the male cohort was started at the same day as for the female cohort. At a length of 45 mm all, 9% or 1.4% of this male cohort were assumed to change sex. After one further moult it was assumed that these males are functional females and are able to carry eggs and the carrying time as well as the amount of eggs was then calculated as described for the female shrimps. For the whole male cohort the reproductive output (Σ $eggs_{se.fem}$) was then calculated and the fraction that secondary females contribute to the whole amount of eggs was determined as

$$eggs_{se.fem.}[\%] = \frac{\sum eggs_{se.fem}}{\sum eggs_{se.fem.} + \sum eggs_{pri.fem.}} \quad (52)$$

Results

Field data

The largest male observed had a length of 70 mm, the largest female a length of 82 mm. In all size classes more females than males were observed. In Wilhelmshaven the ratio of male to female shrimps was higher than in Büsum. Additionally, the shrimps caught in Wilhelmshaven (Kolmogorov-Smirnov: $p>5.54$) were generally larger (t-test: $p<0.001$) than those caught in Büsum (Kolmogorov-Smirnov: $p>4.26$), for females (Kolmogorov-Smirnov: $p=5.516,5.170$ t-test: $p<0.001$) as well as for males (Kolmogorov-Smirnov: $p=4.827,2.651$, t-test: $p<0.001$). The fraction of male shrimps in the catches (quotient male/female) constantly and significantly (p-values see Table 10-2) declined from about 45% to 0% from 25 mm to 70 mm total length (Figure 10-2).

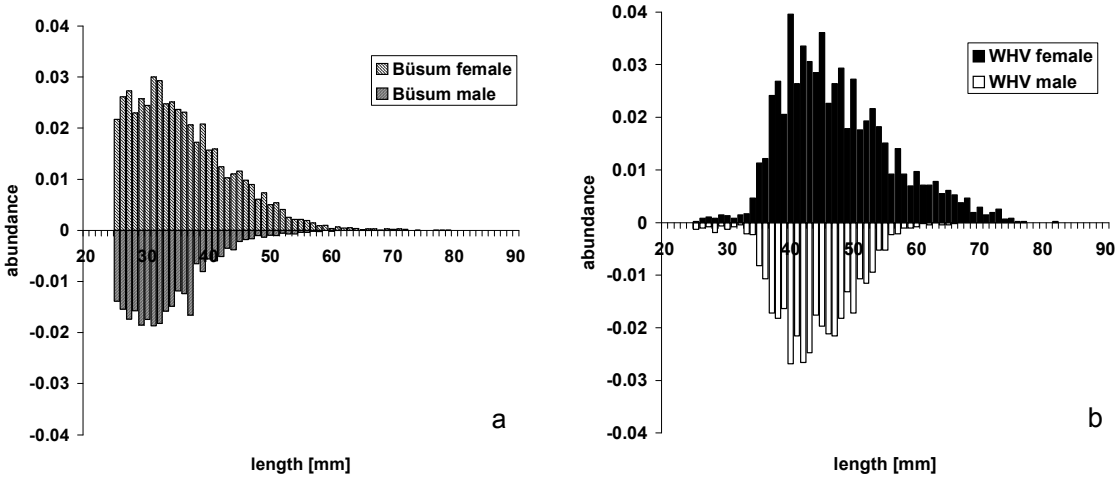

Figure 10-1: Abundance of male, female and juvenile shrimps caught in Büsum in 2000 and 2005-2007 (a) and Wilhelmshaven 2005-2007 (b). All months are combined.

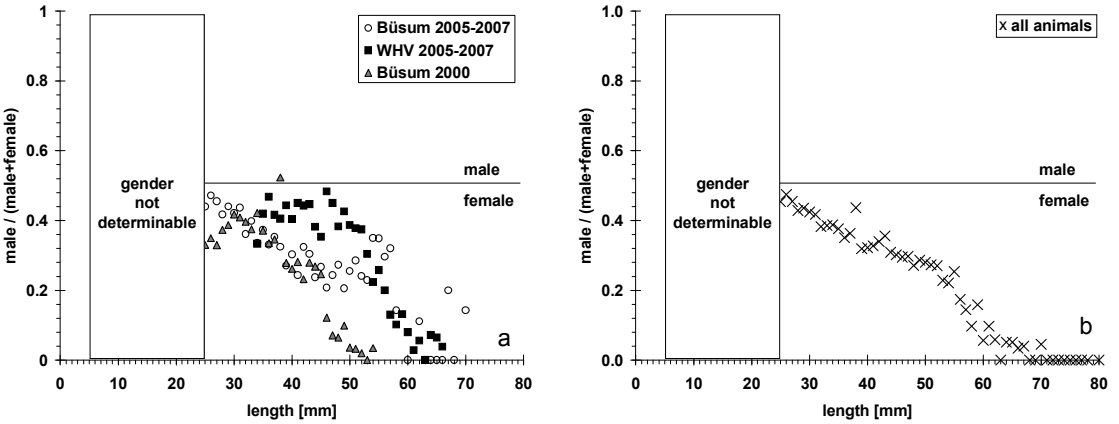

Figure 10-2: Ratios of male to female *Crangon crangon* by length. a: Büsum 2005-2007, Büsum 2000 and Wilhelmshaven 2005-2007. b: Mean of all male-female ratios plotted by length [mm]. Animals below 25 mm were left out of the calculation due to high uncertainties when the sex of small individuals is determined.

Table 10-2: Slope, correlation coefficient and p for decrease of the sex ratio (male/female) vs. length.

	slope	**r^2**	**p**
Büsum 2000	-0.0156	0.765	<0.001
Büsum 2005-2007	-0.009	0.732	<0.001
Wilhelmshaven	-0.0129	0.701	<0.001
all data		0.944	<0.001

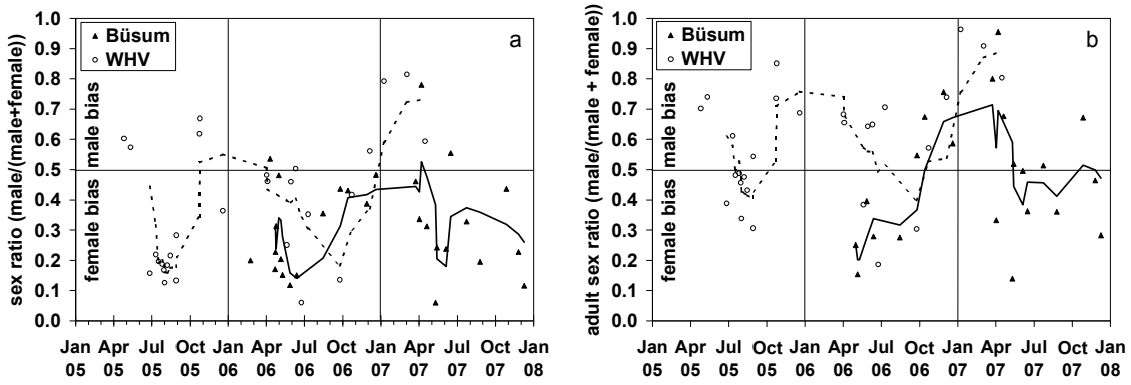

Figure 10-3: Sex (males/ (males+females)) (a) and Adult Sex Ratios (ripe males/(ripe males+ripe females)) (b) for Büsum (triangles) and Wilhelmshaven (dots) and the years 2005-2007. Solid and dashed lines are the according running means (three neighboring points)

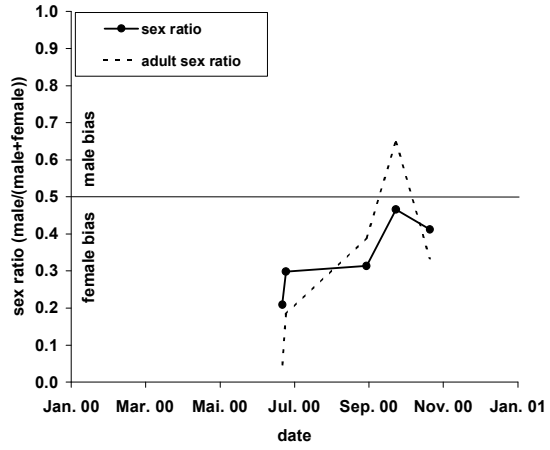

Figure 10-4: Sex ratio and adult sex ratio for Büsum 2000.

From October to April more males than females were caught, while during summer the ratio declined and females clearly dominated the samples (Mann-Whitney-U-test, $p<0.001$),(Figure 10-3, Figure 10-4). Catches were generally below 0.5 Ind·m^{-2} when males dominated (spring and winter), whereas abundance was higher in summer and fall (3–4 m^{-2}). The adult sex ratio exhibited showed the same seasonal pattern as the sex ratio, except that the adult sex ratio was male biased during winter Mann-Whitney-U-test, $p<0.001$).

The values (grouped per 3 month and all years) were normally distributed (Kolmogorov-Smirnoff smallest $p=0.357$). For the size class 30–40 mm (Figure 10-5) the sex ratio was significantly different from 50% (t-test $p<0.01$) and female-biased from Apr-Jun. The rest of the year mean ratios of 42% males in the catches were observed. Near the coast (Wilhelmshaven) 60 and 55% males were observed in the catches between Jan-Mar and Apr-Jun. Between July and September the catches were significantly different from a 50% ratio (t-test, $p<0.01$) and females made up 68% of the catch. For the size class 40–50 mm caught in

Wilhelmshaven this pattern remained with the exception that between Jan-Mar the catches were significantly male biased. The fraction of male shrimps on the tidal flats however increased in the course of the year (r^2=0.491, p=0.017) from 0% in late winter/ early spring to 30% at the end of the year and catches were throughout the year significantly female biased.

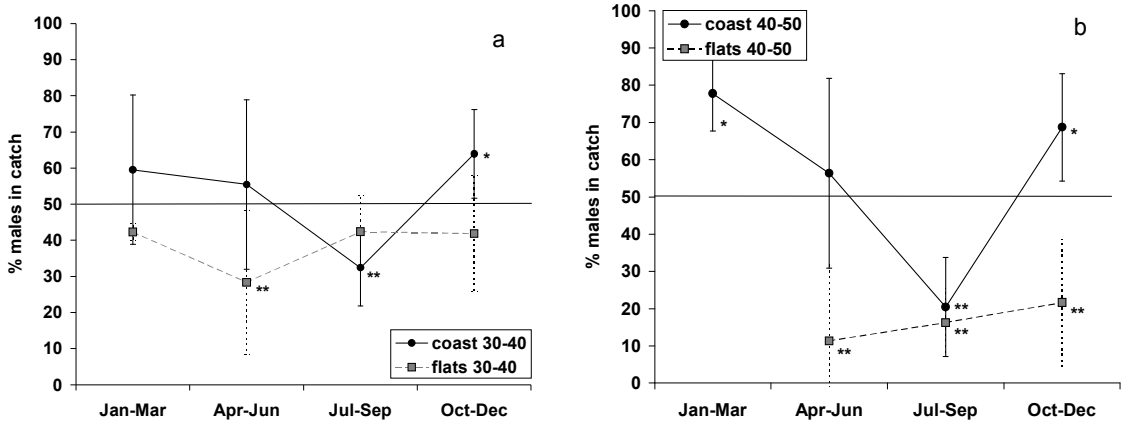

Figure 10-5: Seasonal development of the proportion of male *Crangon crangon* in the catches of Wilhelmshaven (coast) and Büsum (flats). a: 30 to 40 mm length class, b: 40 to 50 mm size class.

Based on the length frequency distributions L_∞ and θ were calculated by the decline of the difference of mean length and length over length using the modified Wetherall method (Wetherall et al. 1987, Sparre et al. 1989). The determined values, derived from Figure 10-6, were L_∞ = 67 and 86 mm and θ = 4.1 and 4.3 for male and female shrimps, respectively.

Calculation of the sex ratio depending on sex specific growth rates

Previously made assumptions included seasonally varying recruitment, a total mortality of Z= 7.2 (θ·k) for male and female shrimp, a k and L_∞ of 1.76 and 67 mm for male shrimps and 1.67 and 86.1 mm for female shrimps, respectively. The simulation generated length frequency distributions as shown in Figure 10-7.

A gradual decline of the theoretical as well as the observed sex ratio with length became evident, reaching zero for the 70 mm length class (Figure 10-8). Significant linear correlations were determined between calculated and observed mean sex ratios (r^2=0.947, p<0.005) calculated and observed male (r^2=0.951, p<0.005) female (r^2=0.884, p<0.005) and total abundances (r^2=0.949, p<0.005).

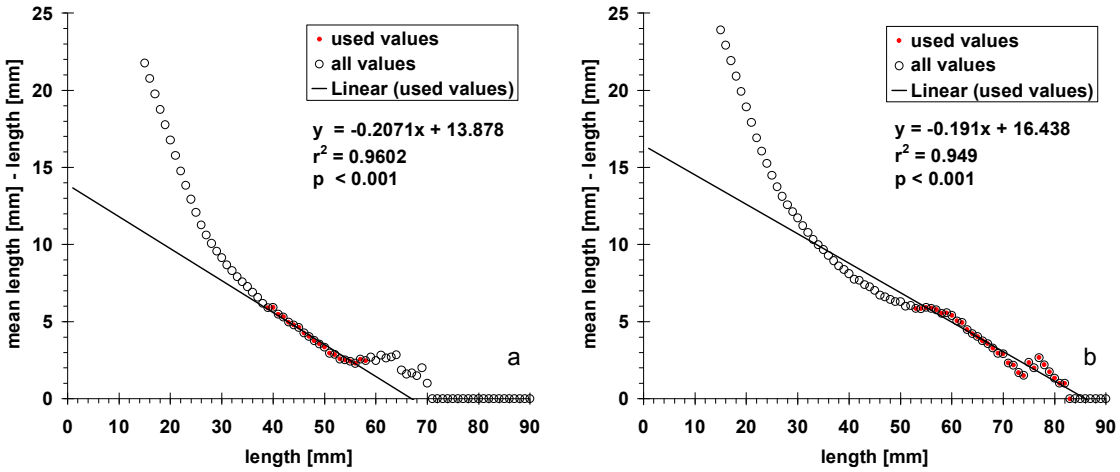

Figure 10-6: Modified Wetherall et al. method to determine L∞ and θ of male (a) and female (b) *Crangon crangon*. Data include all animals caught in Büsum and Wilhelmshaven in 2000 and 2005 to 2007

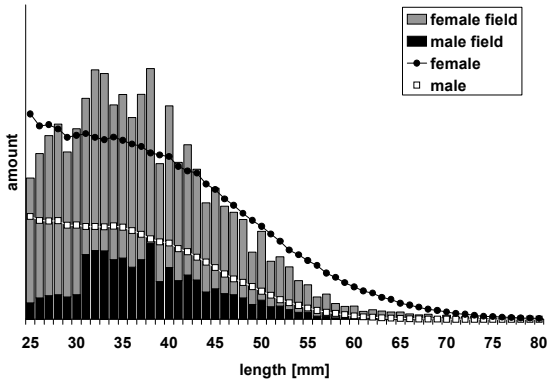

Figure 10-7: Calculated length frequency distributions for male and female *Crangon crangon* and field data (all samples taken in Büsum and Wilhelmshaven). Within the simulation Z = 7.2 (in fall + 80% in spring - 80%), seasonal Bertalanffy growth (Table 10-1) and seasonal recruitment (Temming & Damm 2002) was included.

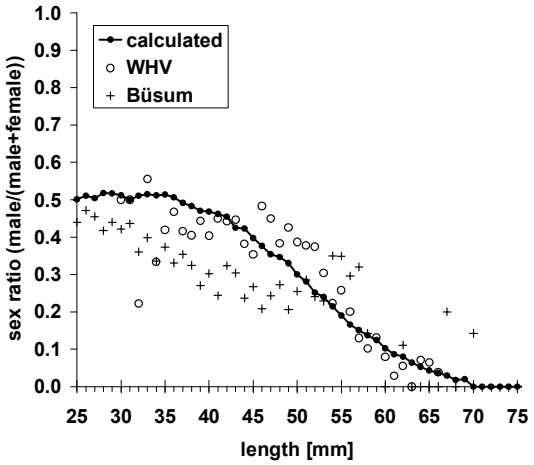

Figure 10-8: Calculated and observed (average of Büsum and Wilhelmshaven) sex ratio of *Crangon crangon*. Assumed parameters for the calculation male shrimps: L∞=67, k=1.76, Z=7.2; female shrimps: L∞: 86, k=1.67, Z=7.2.

Fraction of eggs contributed by secondary females

Based on the total life time production and assuming that all males >45 mm undergo sex reversal the fraction of eggs produced by secondary females would range from 15 to 30% (Table 10-3). If only 9% of the males developed functioning oocytes, the egg production share of secondary females would range from 1 to 3%. If the rate of individuals changing sex was only 1.4%, the amount of eggs of secondary females would be reduced to $\leq$ 0.62 %. The fraction contributed by secondary females was dependent on the season and was lowest in the simulations for cohorts with 5 mm in June and July and highest for cohorts starting in September and October.

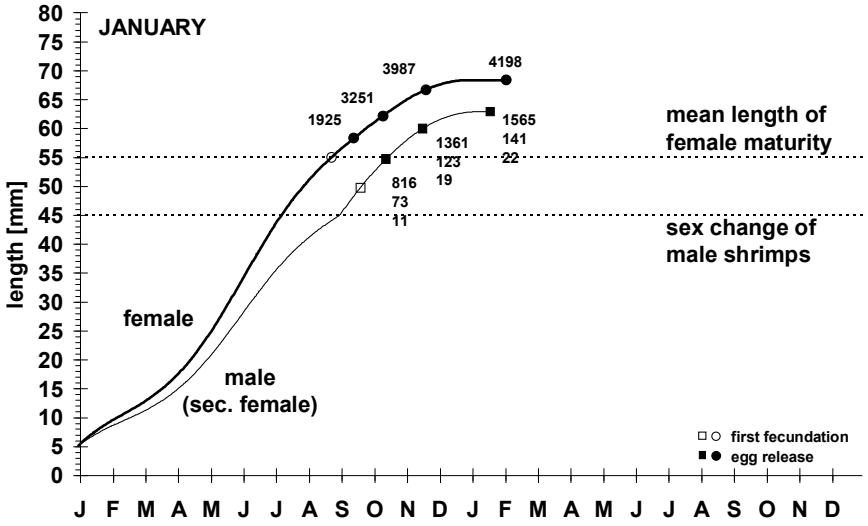

Figure 10-9: Length of female and male shrimps starting in the simulation with 5 mm total length on January 1st. Open circle: first fecundation of female shrimps, open rectangle: first fecundation of secondary females. Full symbols: days when eggs are released. Numbers: total amount (sum) of eggs released from the cohort. Numbers for secondary females refer to scenarios where all, 9% or 1.4% of males change sex.

Female shrimp cohorts starting in the simulation with 5 mm in January start carrying eggs not until September/October (Figure 10-9). Until January the following year the female cohort produces over the whole simulation time an amount of 4188 eggs, whereas total egg production of the former male cohort amounts to 1565 eggs in the corresponding period, when all males change sex. If only 1.4% change sex, 22 eggs are produced (Figure 10-9). In summer the productivity of a cohort is higher and female cohorts starting in the simulation with 5 mm in June produce 13107 eggs within one year. The formerly male cohort produces 2775, 250 and 39 eggs (Figure 10-10). For animals starting in the simulation with 5 mm in September first fertilization takes place in the summer the following year and until September 2525 eggs are released by the female cohort (Figure 10-11).

The decrease of the amount of eggs shown in Figure 10-9 to 11 for secondary females is linear. Assuming that 50% of males change sex therefore leads to a total amount of 783 (Jan.), 1388 (Jun.) and 566 (Sep.) eggs contributed by sec. females. This corresponds to a relative amount of 15.7, 9.6 and 18.3%.

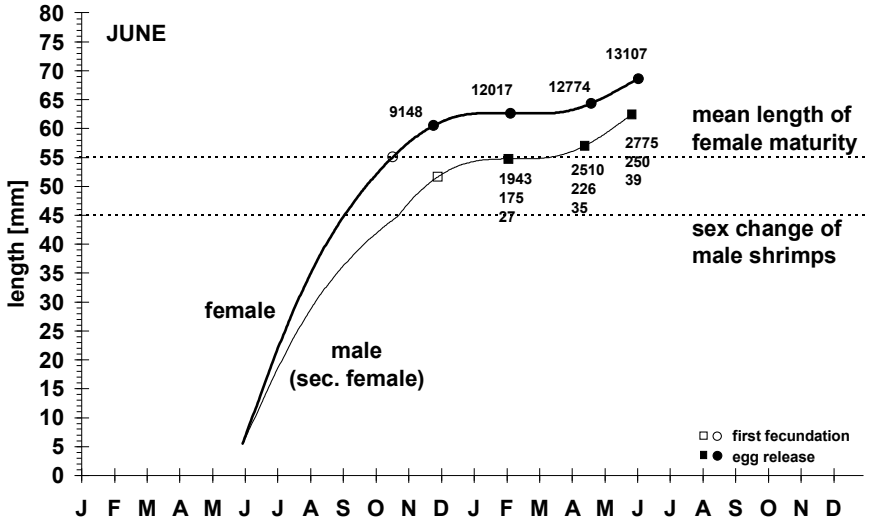

Figure 10-10: Length of female and male shrimps starting in the simulation with 5 mm total length on June 1st. Open circle: first fecundation of female shrimps, open rectangle: first fecundation of secondary females. Full symbols: days when eggs are released. Numbers: total amount (sum) of eggs released from the cohort. Numbers for secondary females refer to scenarios where all, 9% or 1.4% of males change sex.

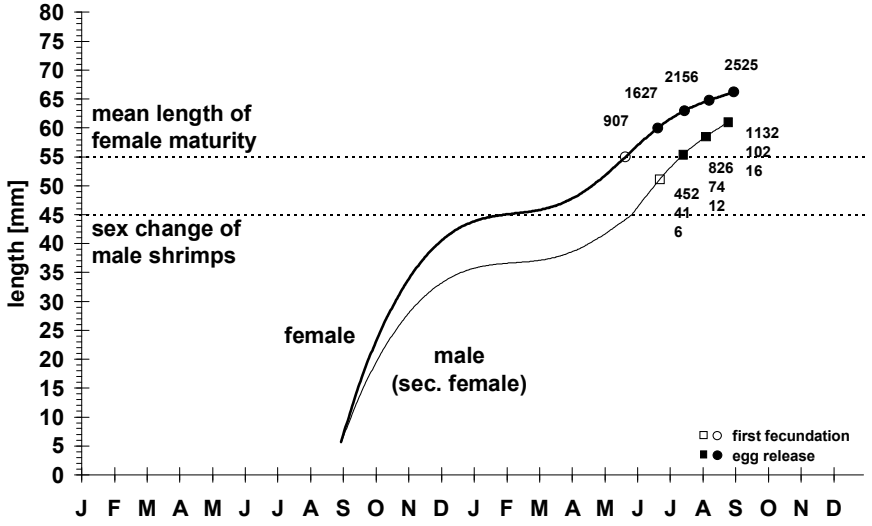

Figure 10-11: Length of female and male shrimps starting in the simulation with 5 mm total length on September 1st. Open circle: first fecundation of female shrimps, open rectangle: first fecundation of secondary females. Full symbols: days when eggs are released. Numbers: total amount (sum) of eggs released from the cohort. Numbers for secondary females refer to scenarios where all, 9% or 1.4% of males change sex.

Table 10-3: Relative amounts of eggs (in percentage) contributed by secondary females to the total amount of eggs produced by primary and secondary females of *Crangon crangon* starting at the same date. 1st column date when male and female cohort starts with 5 mm length in the simulation. Second to fourth column fraction of eggs according to the assumption that all, 9% or 1.4% of the male shrimps change sex.

start (5 mm)	all	9%	1.4%
Jan 1st	27.16	3.25	0.52
Feb 1st	26.22	3.10	0.50
Mar 1st	24.03	2.77	0.44
Apr 1st	23.58	2.70	0.43
May 1st	21.46	2.40	0.38
June 1st	17.47	1.87	0.30
July 1st	17.54	1.88	0.30
Aug 1st	30.67	3.83	0.62
Sep 1st	30.95	3.88	0.62
Oct 1st	30.95	3.88	0.62
Nov 1st	29.78	3.68	0.59
Dec 1st	29.34	3.60	0.58

Discussion

The population is female-biased from the smallest size class on

The fact that the population is female-biased, even in small length classes, indicates that the number of primary females equals that (or is even higher than) the number of males. This is not the case in (obligate) protandric hermaphroditic species such as Pandalus (Bergström 1997), *Athanas indicus* (Gherardi & Calonu 1993), *Hippolyte inermis* (Zupo 2001). In these species a large fraction of small males can be observed that all undergo sex reversal after mating (Correa & Thiel 2003, Allsop & West 2004).

The observed decline in sex ratio with increasing length is in agreement with Martens & Redant (1986), but the fraction of small males was lower in our study. In the Belgian coastal area, the fraction of males was nearly 100% in October which was observed in Wilhelmshaven only during the winter and spring and was not observed at any time at the collection site in Büsum.

As sex specific migration is most likely temperature-dependent (see below), the difference between our data and those of Martens and Redant (1986) might be explained by small males staying longer close to the shore, while female shrimp depart to deeper water earlier when water temperature is decreasing. In fall 1979 during the study of Martens & Redant the temperature was about 3°C lower in Belgium than in 2006 in Germany (sea surface temperature, www.bsh.de).

Large males (70 mm) were observed in the samples

Not all males change their sex as indicated by large males being caught in the field (Figure 10-6). In obligate hermaphrodites all males undergo sex reversal after

reproduction as male (Correa & Thiel 2003), e. g. in *Pandalus* (Carlisle 1959, Hoffman 1972, Clark et al. 2000). Based on the parameters presented in Table 10-1, males starting in the simulation in winter with 5 mm length did not obtain 70 mm until the following summer. When hatched in summer, animals are unable to obtain 70 mm prior to the fall of the following year. In both cases, the time span is long enough that animals would have had the chance to reproduce at least once prior to changing sex. In obligate hermaphrodites therefore no males of that size should be detectable in the field.

The sex ratio constantly declines over all length classes

The decline in sex ratio with increasing total length suggests a facultative or a partial protandric system. In protandric systems a change in the slope of the sex ratio over length can be observed from the length on where most males change sex. According to Boddeke (1966) and Martens & Redant (1986), this size class should be 35 to 45 mm. Such a change in slope was not observed in the present study in the given size range. A slight increase of the decline of the male fraction can be seen in Figure 10-2 (mean of all data) from approximately 55 mm on whereas the separated data set from Büsum (2000) and Wilhelmshaven constantly decline over length. The Büsum data from 2006 and 2007 show no clear trend in the largest size classes as these are influenced by small amounts of animals. During 2006 and 2007 only 97 animals larger than 60 mm were caught in Büsum.

However, the change in sex ratio decline observed in shrimp caught in Wilhelmshaven and the mean sex ratio of all sites at 55 mm is 10 mm above the length range assumed for sex reversal. Moreover, the fraction of males reaching this size is small (see below). Due to the slower growth a higher cumulative mortality occurs which has also been described for larval fish (Houde 1987). Since most of them die before they can reproduce as females, the benefit of sex change is low and therefore also the contribution of these males to the population reproduction.

In Büsum, the samples were taken with a push net in shallow waters on the tidal flats which are regarded as juvenile nursery areas (Cattrijsse 1997). Starting at a length of 30 mm, *C. crangon* migrates to deeper water (Beukema 1992) and, therefore, earlier migration from maturing males can be expected and would explain the differences between the sex ratios of Büsum and Wilhelmshaven.

Explanation of the observed sex ratio decline by sex specific growth rates

Differences in growth rates and size between sexes are common in shrimps (Pauly 1982, Baelde 1994, Mehanna 2000, Campos & Berkeley 2003, Kim 2004, Ragonese et al. 2004, Sainte-Marie et al. 2006) and were also observed for *C. crangon* (Meixner 1969b, Labat 1977, Lagardère 1982, Oh et al. 1999, Uhlig 2002).

Our simulation model based on the assumptions of different growth rates for female and male *C. crangon* with slower growth rates for the latter generated mean sex ratios and abundances over the year which significant correspond to field values. Growth rate differences alone are therefore sufficient to generate the observed decline in the sex ratio. The remaining parameters such as recruitment and mortality were equal for both sexes. The latter assumption was based on the estimates from the length frequency distributions and, is not surprising, as it can be assumed that mortality is not sex specific, but mainly length specific, due to fishery and predation.

The observed sex ratios from Wilhelmshaven are more male-biased, whereas the sex ratios obtained from Büsum are more female-biased than the sex ratios calculated with the model. This is most probably an effect of the more pronounced winter migration of female shrimps (see below). Male shrimps tend to stay closer to the coast in winter, whereas female shrimps seem to migrate further offshore.

Explanation of the observed seasonal pattern

Migration patterns have repeatedly been described to differ seasonally for male and female shrimps (Boddeke 1976) and also for different size classes (Tiews 1970, Boddeke et al. 1976, Henderson & Holmes 1987, Beukema 1992, Reiss & Kröncke 2005). The patterns observed in the field (see Figure 10-3,4 and 5) result from the sex ratio on tidal flats being slightly female-biased in 30–40 mm size class due to the lower growth rate of males (Figure 10-8). Our results suggest that, the male fraction increases until July - September. Beukema (1992) reported that C. crangon begin migrating to deeper waters at >20 mm so the increase in the male fraction most likely resulted from faster-growing females migrating towards deeper water (Beukema 1992). Consequently, the sex ratio becomes female-biased in the deeper water from July through September as females that were hatched in winter and spring begin growing through 30 mm. The slower-growing males reach the size class of 30 mm somewhat later (Oct-Dec) which becomes evident through the constant increase of the male fraction (30-40 mm) on the flats from Apr.-Jun. until Oct.-Dec. and by the strong increase of the proportion of males in deeper water in Oct.-Dec. This increase is only possible if females migrate towards even deeper waters (further offshore) in the winter, which is not only suggested by our data but also indicated by the sex ratio determined by Siegel et al. (2008). The small fraction of females over the winter (Oct.-Dec.) might indicate that males stay closer to the shore than females. The assumptions made fit in large parts the migration patterns described by Henderson & Holmes (1987).

The fraction of eggs contributed by secondary females to the spawning stock

Assuming that all males change sex after reaching 45 mm, the fraction of offspring contributed by secondary females would amount to 15–30%. Our field samples

have shown that large males exist and that the decline in the sex ratio with length can fully be described by sex specific growth rates. This indicates that not all males change sex and that the fraction of males changing sex in the field is low, as suggested by Martens & Redant (1986) and Schatte & Saborowski (2005).

Pandalus spec. a species related to *C. Crangon* secondary females need several moults to be able to develop oocytes after sex reversal (Clark et al. 2000). Likewise, in *C. crangon* it takes several moults until the endopodite of the first pleopod (where the eggs are attached) grows to the size of similarly sized primary females (Schatte & Saborowski 2005). Therefore, it is more probable that more than one moult is needed until secondary females can carry eggs or are fully fertile, comparable to primary females of the same size. If two moults are needed (and all males change sex) the fraction of eggs contributed by secondary females reaches a maximum of 20%, while if three moults are needed the contribution of eggs decreases to 10%. If both the low rate of sex changer and the additional time required for several moults by those animals, are considered, one can conclude that reproductive success of sex changers is very low under the actual conditions in the field.

From the calculated length trajectories and the calculated amount of eggs it can also be derived that winter-spawned shrimp are more relevant for the population than the summer spawned shrimp. Animals that start with 5 mm length from January to June can reach size of maturity earlier and develop eggs during winter (Figure 10-9 to 11). Due to the high temperatures growth is fast and therefore cumulative mortality is low. Therefore a considerable share of females (2−3% of starting cohort size) reaches size of first egg release. Animals that hatched in summer and start with 5 mm in September in our simulation (Figure 10-11) do not carry eggs before the next summer. Until then at top 0.3% of the starting cohort size is left what is about a tenth if the winter cohort.

Advantages of hermaphroditism and why it still exists in *Crangon crangon*

The reasons for hermaphroditism and why sex change in Carideans most often occurs from male to female are well discussed by Bauer (2006). For females fecundity is generally correlated with size which is not true for males. Therefore large males change sex to gain a higher reproductive output. Beside this most probably and important reason, there are several other factors favoring hermaphroditism in contrast to gonochronism.

The disadvantage of being a large male in comparison to a small male is assumed to be higher agility of small males and therefore higher success when searching for ripe females. Smaller males need less energy for the basic metabolism and can therefore put more energy into searching activity. Energy costs increase further if water currents have to be overcome, which can lead to energy deficits and higher mortality of searching males (Ward 1983). As small males are more active, they can fertilize more females per unit time than larger ones.

Guarding females against competing males, and therefore selection of bigger males, might be suppressed not only by the high abundance of co-occuring males but also by high number of potential predators. For example, *C. crangon* must leave the shelter of being buried in the sand during copulation which selects for short interactions and therefore favors more agile small males and not slow (and evt. guarding) large males. Zhang (2005) observed that smaller male of *Lysmata wurdemanni* could mate with larger females but large males sometimes failed to mate with smaller females. This would select for smaller male size as small males can mate with a greater fraction of fecund females.

Applying a general model Morbey & Abrams (2004) suggested that late arriving males invest more in competitive ability than in survival because mating opportunities will only be present for a short period of time (late year). Large males would therefore gain an advantage if they would become female.

If the reproductive input from secondary females is low, as shown in the actual study, but advantages are multiple, the following questions arise: why has the ability to change sex developed, and why does it still exist?. One reason might be that environmental conditions in the past were different from the conditions at present. Conditions may have favored hermaphroditism in the past, whereas the current situation is either neutral for gonochronism (all males stay males) and hermaphroditism for *C. crangon* or favors gonochronism.

Longevity is one factor favoring hermaphroditism (Morbey & Abrams 2004) which might have changed over time. The advantages of protandric hermaphroditism are lost if the secondary female dies before reproduction. This effect is increased if the reproductive output is lower or nonexistent as appears to be the case fort secondary females prior to successive moulting and conclusion of sex change). In combination with a short lifetime and the energy necessary for sex change, the advantage of hermaphroditism is lost (Policansky 1982). This applies to *C. crangon* and longevity might have even have decreased most recently due to intensive fishery. Temming et al. (1993) observed a decrease of the maximum length in commercial catches from the 1950s to the 1980s and Henderson & Holmes (1987) refer to studies where brown shrimps in the 1930s have been much larger. A reduction of maximum length and therefore longevity is present and might have lead to changes in sexual behavior and reproduction of *C. crangon*. Such a response to increasing levels of fishing effort or changes in mortality have been reported for several species (Fukuwaka & Morita 2008, Watanabe & Yatsu 2006, Moiseenko 2002).

Although the North Sea is relatively old, the core distribution area of *C. Crangon,* the Wadden Sea, is geologically young and was formed after the last ice age about 11000 years ago. It can be assumed that *C. crangon* in former times lived in deeper water similar to its congener (*Crangon allmanni)* (Blahudka & Türkay 2002), or at lower concentrations on sandy beaches (Beyst et al. 2001). Luttikhuizen et al (2008) concluded from sequencing a 388 bp fragment of the cytochrome-c-oxidase I gene, that the oldest and most variable population of *C. crangon* inhabit the western Mediterranean. Under these conditions

hermaphroditism might have developed. So far, hermaphroditism and sex ratios have not been examined for *Crangon allmanni* but are well known for another North Sea deep water decapod: *Pandalus borealis* (Hoffman 1972).

Another factor favoring hermaphroditism is population density. Female shrimps can carry eggs only for limited time in the year, whereas males can mate with several females. Hence, a male has the opportunity to achieve more reproductive output, if the population density is high. If density is low, female shrimp have an advantage. An indicator that population densities were lower in the past might be the possible existence of female pheromones used to attract males (Boddeke et al. 1991), which is uncommon in Carideans (Correa & Thiel 2003). Production of pheromones is energetically expensive (Byers 2005, Rantala et al. 2003) and would only be selected for, if the success of attracting males outweighs the disadvantage of energy loss. At densities of 4–5 individuals per $m^{2,}$ as observed in our samples, the encounter rate between males and females can be expected to be high and it could thus be expected that the use of pheromones is not necessary at the present conditions.

Concomitant with the invasion into the highly productive shallow areas of the Wadden Sea, gonochronism might have been the superior mating system. Due to higher predation pressure mortality was greater in shallow water, but due to higher food concentrations, density could increase and with it the advantages of large males. Large males usually produce more high quality sperm (Ceballos- Vázquez et al. 2003), can fertilize more females (Adams et al. 1989, Nakashima 1995) and have an advantage when internal fertilization, as assumed for *C. crangon* (Boddeke et al. 1991), occurs (Urbani et al. 1998). The latter assumption is based on two observations. First, sperm stratification where older sperm are displaced so that younger sperm is in a better position. Second, sperm removal, where older sperm is flushed out and therefore displaced by younger sperm. In both cases larger males are favored as these can produce more sperm. Another reproductive advantage of larger males is that larger males are more able to break the resistance of a female (Jormalainen & Merilaita 1995) and search more efficient for mating partners as they have higher locomotory capacity (Spaargaren 1999). In contrast to the formerly discussed point of higher agility of smaller males this might be important in turbulent water, like the tidal influenced Wadden Sea. Additionally, large males have longer antennae and might be able to detect females more rapidly (Bertin & Cezilly 2003). In summary, not changing sex and growing to a larger size might be favorable under specific conditions. The sum of these effects might have selected against sex changing males and therefore shifted the previously hermaphroditic sexual system of *C. crangon* towards the gonochronistic system observed in the majority of the North Sea population today.

Acknowledgements

We wish to thank the employees and support staff at the power plant in Wilhelmshaven for support with the cooling water sampling. The study was financially supported by the Federal Ministry of Food, Agriculture and Consumer

Protection, Germany (Project No. 03HS030) and Niedersächsische Wattenmeerstiftung, Germany (Project No. 53-NWS-41/04)

References

Adams J, Watt PJ, Naylor CJ, Greenwood PJ (1989) Loading constraints, body size and mating preference in *Gammarus* species. Hydrobiologia 183:157-164

Allsop DJ, West SA (2004) Sex-ratio evolution in sex changing animals. Evolution 58:1019-1027

Baelde P (1994) Growth, mortality and yield-per-recruit of deep-water royal red prawns (*Haliporoides sibogae*) of eastern Australia, using the length-based MULTIFAN method. Marine Biology 118:617-625

Bauer RT (2006) Same sexual system but variable sociobiology: evolution of protandric simultaneous hermaphroditism in *Lysmata* shrimps. Integrative and Comparative Biology 46:430-438

Berghahn R (1983) Untersuchungen an Plattfischen und Nordseegarnelen (*Crangon crangon*) im Eulitoral des Wattenmeeres nach dem Übergang zum Bodenleben. Helgoländer Meeresuntersuchungen 36:163-181

Bergström BI (1997) Do protandric pandalid shrimp have environmental sex determination? Marine Biology 128:397-407

Bertin A, Cezilly F (2003) Sexual selection, antennae length and the mating advantage of large males in *Asellus aquaticus*. Journal of Evolutionary Biology 16:698-707

Beukema JJ (1992) Dynamics of juvenile shrimp *Crangon crangon* in a tidal flat-nursery of the Wadden Sea after mild and cold winters. Marine Ecology Progress Series 83:157-165

Beyst B, Hostens K, Mees J (2001) Factors influencing fish and macrocrustacean communities in the surf zone of sandy beaches in Belgium: temporal variation. Journal of Sea Research 46:281-294

Blahudka S, Türkay M (2002) A population study of the shrimp *Crangon allmanni* in the German Bight. Helogland Marine Research 56:190-197

Boddeke R (1976) The seasonal migration of the brown shrimp *Crangon crangon*. Netherlands Journal of Sea Research 10:103-130

Boddeke R, Bosschieter JR, Goudswaard P (1991) Sex change, mating and sperm transfer in *Crangon crangon*. In: Bauer RT, Martin JW (eds) Crustacean sexual biology. Columbia University press, New York, p 164-182

Boddeke R, Dijkema R, and Stemelink ME The patterned migration of shrimp populations: A complete study of *Crangon crangon* and *Penaeus brasiliensis*. 200, 31-49. 1976. FAO Fisheries report. Symposium on Proceedings in marine Biology in the Carribean Corsas.

Boddeke R (1966) Sexual cycle and growth of Brown Shrimp (*Crangon crangon*). International Council for the exploration of the Sea, Shellfish Committee C.M. 1966 / M:6.

Byers JA (2005) A cost of alarm pheromone production in cotton aphids, *Aphis gossypii*. Naturwissenschaften 92:69-72

Bulnheim H-P, Schwenzer DE (1993) Zur Populationsgenetik von *Crangon crangon* und *C. allmanni* (Crustacea, Decapoda) im Bereich der europäischen Küsten. Zoologische Jahrbücher. Abteilung für allgemeine Zoologie und Physiologie der Tiere 97:327-347

Cattrijsse A, Dankwa HR, Mees J (1997) Nursery function of an estuarine tidal marsh for the brown shrimp *Crangon crangon*. Journal of Sea Research 38:109-121

Carlisle BDB (1959) On the sexual biology of *Pandalus borealis* (Crustacea Decapoda). Journal of the Marine Biological Association U.K. 38:481-491

Ceballos- Vázquez BP, Rosas C, Racotta IS (2003) Sperm quality in relation to age and weight of white shrimp *Litopenaeus vannamei*. Aquaculture 228:141-151

Clark SH, Cadrin SX, Schick DF, Diodati PJ, Armstrong MP, McCarron D (2000) The Gulf of Maine Northern Shrimp (*Pandalus borealis*) fishery: a review of the record. Journal of Northwest Atlantic Fishery Science 27:193-226

Correa C, Thiel M (2003) Mating systems in caridean shrimp (Decapoda: Caridea) and their evolutionary consequences for sexual dimorphism and reproductive biology. Revista Chilena de Historia Natural 76:187-203

Criales MM, Anger K (1986) Experimental studies on the larval development of the shrimps *Crangon crangon* and *C. allmanni*. Helgoländer Meeresuntersuchungen 40:241-265

Cuesta JA, González-Ortegón E, Rodríguez A, Baldó F, Vilas C, Drake P (2006) The decapod crustacean community of the Guadalquivir Estuary (SW Spain): seasonal and inter-year changes in community structure. Hydrobiologia 557:85-95

Fukuwaka M, Morita K (2008) Increase in maturation size after the closure of a high seas gillnet fishery on hatchery-reared chum salmon *Oncorhynchus keta*. Evolutionary Applications 1:376-387

Gavio MA, Orensanz JML, Armstrong D (2006) Evaluation of alternative life history hypothesis for the sand shrimp *Crangon franciscorum* (Decapoda: Caridea). Journal of Crustacean Biology 26:295-307

Gherardi F, Calloni C (1993) Protandrous hermaphroditism in the tropical shrimp *Athanas indicus* (Decapoda, Caridea), a symbiont of sea urchins. Journal of Crustacean Biology 13:675-689

Gillooly JF, Brown JH, West GB, Savage VM, Charnov EL (2001) Effects of size and temperature on metabolic rate. Science 293:2248-2251

Gunnarsson B, Ásgeirsson þ, Ingólfsson A (2007) The rapid colonization by *Crangon crangon* (Linnaeus, 1758) (Eucarida, Caridea, Crangonidae) of Icelandic coastal waters. Crustaceana 80:747-753

Hamerlynck O, Hostens K (1993) Growth, feeding, production, and consumption in 0-group bib (*Trisopterus luscus* L.) and whiting (*Merlangius merlangus* L.) in a shallow coastal area of the south-west Netherlands. ICES Journal of Marine Science 50:81-91

Havinga B (1930) Der Granat (*C. vulgaris*, Fabr.) in den holländischen Gewässern. Journal du Conseil International pour l'Exploration de la Mer 5:57-87

Henderson PA, Holmes RHA (1987) On the population biology of the common shrimp *Crangon crangon* (L.) (Crustacea: Caridea) in the Severn Estuary and Bristol Channel. Journal of the Marine Biological Association U.K. 67:825-847

Hoffman DL (1972) The development of the ovotestis and copulatory organs in a population of protandric shrimp, *Pandalus platyceros* Brandt from Lopez Sound, Washington. Biological Bulletin 142:251-270

Houde ED (1987) Fish Early Life Dynamics and Recruitment Variability. Am.Fish.Soc.Symp. 2:17-29

Hufnagl M, Temming A (2009) Determining growth rates of the brown shrimp *Crangon crangon* (L.): review and laboratory approach. Marine Ecology Progress Series.

ICES (2007) Report of the Working Group on Crangon Fisheries and Life History. International Council for the exploration of the Sea Living resources comittee:1-33

Jansen, S. (2002) Das Räuber-Beutesystem juveniler Gadiden, Grundeln und Garnelen im Wattenmeer nördlich von Sylt. 155pp. Universität Hamburg.

Jormalainen V, Merilaita S (1995) Female resistance and duration of mate-guarding in three aquatic peracarids (Crustacea). Behavioral Ecology and Sociobiology 36:43-48

Klek-Kawinska E, Bomirski A (1975) Ovary-inhibiting hormone activity in Shrimp (*Crangon crangon*) eyestalks during the annual reproductive cycle. General and Comparative Endocrinology 25:9-13

Kuipers B, Dapper R (1984) Nursery function of the Wadden Sea tidal flats for the brown shrimp *Crangon crangon*. Mar.Ecol.Prog.Ser. 17:171-181

Kuipers BR, Dapper R (1981) Production of *Crangon crangon* in the tidal zone of the Dutch Wadden Sea. Netherlands Journal of Sea Research 15:33-53

Lagardère JP (1982) Effects of Noise on Growth and Reproduction of *Crangon crangon* in Rearing Tanks. Marine Biology 71:177-185

Luttikhuizen PC, Campos J, van Bleijswijk J, Peijnenburg KTCA, van der Veer HW (2008) Phylogeography of the common shrimp, *Crangon crangon* (L.) across its distribution range. Molecular Phylogenetics and Evolution 46:1015-1030

Martens E, Redant F (1986) Protandric hermaphroditism in the brown shrimp *Crangon crangon* (L.), and its effects on recruitment and reproductive potential. International Council for the exploration of the Sea Shellfish Committee:1-15

Martin J (1995) Période d'eclosion et abondance des larves de crevette grise Crangon crangon Linnaeus, 1758 en manche et au sud de la Mer Du Nord. K: 53, 2-15. ICES. C.M.

Mehanna SF (2000) Population dynamics of *Penaeus semisulcatus* in the Gulf of Suez, Egypt. Asian Fisheries Science 13:127-137

Meixner R (1969) Wachstum, Häutung, und Fortpflanzung von *Crangon crangon* (L.) bei Einzelaufzucht. Berichte der deutschen wissenschaftlichen Kommission für Meeresforschung 20:93-111

Moiseenko TI (2002) Change in the Life Cycle Strategy of Fish under the Effect of Chronic Water Pollution. Russian Journal of Ecology 33:45-55

Morbey YE, Abrams PA (2004) The interaction between reproductive lifespan and protandry in seasonal breeders. Journal of Evolutionary Biology 17:768-778

Nakashima Y (1995) Can small shrimps achieve copulation in the presence of larger ones? Journal of Ethology 13:9-16

Oh C-W, Hartnoll RG (2004) Reproductive biology of the common shrimp *Crangon crangon* (Decapoda: Crangonidae) in the central Irish Sea. Marine Biology 144:303-316

Oh C-W, Hartnoll R, Nash RDM (1999) Population dynamics of the common shrimp, *Crangon crangon* (L.), in Port Erin Bay, Isle of Man, Irish Sea. ICES Journal of Marine Science 56:718-733

Pihl L, Rosenberg R (1984) Food selection and consumption of the shrimp *Crangon crangon* in some shallow marine areas in western Sweden. Marine Ecology Progress Series 15:159-168

Policansky D (1982) Sex change in plants and animals. Annual Reviews of Ecology and Systematics 13:471-495

Rantala MJ, Kortet R, Kotiaho JS, Vainikka A, Suhonen J (2003) Condition dependence of pheromones and immune function in the grain beetle *Tenebrio molitor*. Functional Ecology 17:534-540

Redant, F (1978) Konsumptie en pruoduktie van post-larval *Crangon crangon* (L.) (Crustacea, Decapoda) in de Belgische kustwateren, DEEL I. TEKST. Vrije Universiteit Brussel, Fakulteit Wetenschappen.

Sainte-Marie B, Bérubé I, Brillo S, Hazel F (2006) Observations on the growth of the sculptured shrimp *Sclerocrangon boreas* (Decapoda: Caridea). Journal of Crustacean Biology 26:55-62

Schatte J, Saborowski R (2005) Change of external sexual characteristics during consecutive moults in *Crangon crangon* L. Helgoland Marine Research

Siegel V, Damm U, Neudecker T (2008) Sex-ratio, seasonality and long-term variation in maturation and spawning of the brown shrimp *Crangon crangon* (L.) in the German Bight (North Sea). Helgoland Marine Research 62:

Siegel V, Gröger J, Neudecker T, Damm U, Jansen J (2005) Long-term variation in the abundance of the brown shrimp *Crangon crangon* (L.) population of the German Bight and possible causes for its interannual variability. Fisheries Oceanography 14:1-16

Spaargaren DH (1999) Shape and Hydrodynamic Properties in relation to size in marine macro crustacea. Crustaceana 72:203-214

Sparre P, Ursin E, Venema SC (1989) Introduction to tropical fish stock assessment. Food and Agriculture organization of the united nations, Rome

Temming, A, Jansen, S, Welleman, HC, and Daan, N (2000) Interrelationships of the brown shrimp population and the multinational brown shrimp fishery with emphasis on recent changes in the population structure. Study in support of the common fisheries policy 96-007. Final Report.

Temming A, Damm U (2002) Life cycle of *Crangon crangon* in the North sea: A simulation of the timing of recruitment as a function of the seasonal temperature signal. Fisheries Oceanography 11:45-58

Tiews K (1970) Synopsis of biological data on the common shrimp *Crangon crangon* (Linaeus, 1758). FAO Fisheries Reports 4:1167-1223

Tiews K (1954) Die biologischen Grundlagen der Büsumer Garnelenfischerei. Berichte der deutschen wissenschaftlichen Kommission für Meeresforschung 13:235-269

Urbani N, Sainte-Marie B, Sévigny J-M, Zadworny D, Kuhnlein U (1998) Sperm competition and paternity assurance during the first breeding period of female snow crab (*Chionoecetes opilio*) (Brachyura: Majidae). Canadian Journal Fisheries and Aquatic Science 55:1104-1113

Ward PI (1983) Advantages and a disadvantages of large size for male *Gammarus pulex* (Crustacea: Amphipoda). Behavioral Ecology and Sociobiology 14:69-76

Watanabe C, Yatsu A (2006) Long-term changes in maturity at age of chub mackerel (*Scomber japonicus*) in relation to population declinesin the waters off northeastern Japan. Fisheries Research 78:323-332

Wear RG (1974) Incubation in British decapod crustacea and the effects of temperature on the rate and success of embryonic development. J.Mar.Biol.Assoc.UK. 54:745-762

Wetherall JA, Polovina JJ, Ralston S (1987) Estimating growth and mortality in steady-state fish stocks from length frequency data. In: D.Pauly and G.R.Morgan, Editors, Length-based Methods in Fishery Research53-74

Zhang D, Lin J (2005) Comparative mating success of smaller male-phase and larger male-role euhermaphrodite-phase shrimp *Lysmata wurdemanni* (Caridea: Hippolytidae). Marine Biology 147:1387-1392

Zupo V (2001) Influence of diet on sex differentiation of *Hippolyte inermis* Leach (Decapoda: Natantia) in the field. Hydrobiologia 449:131-140

Discussion

Growth rates of *C. crangon*

In the present thesis, a growth model was developed based on new laboratory data and existing literature data. This is the first growth model that is able to link the sizes of individuals within the peak of invading juvenile shrimps in spring with the sizes of individuals within the peak of abundance of adults captured fall. These results were only possible when maximum growth rates were assumed, indicating that the fast-growing shrimps mainly determine the population dynamics in North Sea. The slower growth of the remaining population is likely due to food-limitation as indicated by the results in manuscript 2. The slow growing shrimps will suffer high cumulative mortality (Houde, 1987) and a smaller fraction of them finally reaches the age / size of maturity. Comparing the amount of animals left from a cohort that starts in July with 5 mm and experiences a constant Z of 5, the number of fast growing shrimps that reaches 55 mm will be 10 times higher than the number of shrimps growing at mean rates. Assuming a decreasing mortality with length from Z=20 at 5 mm to 5 at 45 mm the factor will even increases to 25.

Analyzing field data taken in Büsum and Wilhelmshaven in comparison to starvation trials implied that mainly during winter (from November to March) potential food sources are absent and animals loose weight, despite the opportunistic omnivorous feeding of the shrimp. As growth at low temperatures is already low (even under ad libitum conditions, manuscript 1) it can therefore be assumed that during winter shrimps do not grow. Moreover, the poor growth performance of these overwintering shrimps in the laboratory even under ad libitum conditions (May shrimps in manuscript 1) is likely directly related to those overwintering conditions experienced prior to the laboratory growth trials. Reasons might be non-genetic irreversible adaptation (Kinne 1962) or genetic differences. The shrimps might either be fast growing summer hatched shrimps that experienced low food concentrations during winter and are now adapted to survive by reducing their catabolism as described by Regnault (1981), or they are slow growing shrimps from winter eggs of the previous year that had slow growth rates (and thus perhaps lower food requirements) during their entire life due to genetic effects. In both cases, a separation of the “slow” shrimps from faster growing conspecifics might not be possible by solely regarding $RNA \cdot DNA^{-1}$ as described in manuscript 2. If a shrimp is adapted to low food conditions it might feed at its maximum and has the same $RNA \cdot DNA^{-1}$ ratio as a shrimp that is adapted to higher feeding conditions which feeds more and therefore growth faster. This shall be explained by regarding exemplary the amount of digestion enzymes. An animal adapted to high food concentrations will have a high concentration of enzymes present and therefore has a high turnover of ingested material and subsequently a high growth performance is possible. This is possible with only a low amount of RNA as no new enzymes have to be produced. An animal adapted to low food concentrations will have to produce additional enzymes to increase growth and therefore its RNA concentration has to increase. Although $RNA \cdot DNA^{-1}$ and dry

weight at length were suitable parameters to describe the condition and feeding history of *C. crangon* the previous explanation might be one reason why it was not possible to correlate these parameters directly to the growth rates observed in the laboratory. Another reason might be food quality. If, for example, a “bad” food source such as sprat was ingested, $RNA \cdot DNA^{-1}$ might be as high as, or even higher than when a “good” food source was ingested. For example the fatty acid composition of the “good” food source will fit better to the shrimp or essential components are lacking in the “bad” food source, growth will be different but $RNA \cdot DNA^{-1}$ might be comparable. Other reasons have been discussed in manuscript 2 and are most probably moulting and maturation of the shrimps. Based on the findings it can be concluded, that $RNA \cdot DNA^{-1}$ and also dry weight at length are not suitable to determine growth of *C. crangon* unless the effect of moulting, maturation and feeding has been examined systematically. Until now it is therefore not possible to separate slow- and fast-growing individuals within field samples.

Beside the growth model presented in manuscript 1 several methods were applied to determine von Bertalanffy growth parameters k and L_∞ to allow a direct calculation of length. In manuscript 1, k and L_∞ were determined from the first derivative of the von Bertalanffy growth equation fitted to different growth rate data sets. Based on mean growth rates k = 1.33 and L_∞ = 76.2 mm and based on 0.75 percentile of the growth rates k = 1.19 and L_∞ = 98.66 mm were determined. These results are comparable to findings of Kuipers & Dapper (1984) who estimated L_∞ = 78 mm and k = 0.95 based on field data. A *C. crangon* specific relation between k and L_∞ was determined in manuscript 5 where further L_∞ was estimated with the Wetherall et al. and the Powell method. Based on data from 1955 - 2006 sampled along the German and Dutch coast a mean L_∞ of 79.3 mm was determined again comparable to findings of Kuipers & Dapper (1984), Temming et al. (1993) and the results of manuscript 1. The growth parameter k was further determined by the rate of weight loss of *C. crangon* measured during starvation experiments at different temperatures. A comparison of the findings with the growth model was presented in manuscript 3. The results are comparable mainly for the 30 and 45 mm class, whereas k determined in the starvation experiments with 20 mm sized shrimps was higher and with 60 mm sized shrimps lower than the value determined in the growth experiment. At 20°C values based on the growth model are slightly higher for the 30 and 45 mm in comparison to the values determined in the starvation experiments. The reasons for the differences are multiple, as both methods, the determination in growth and starvation experiments are difficult as discussed en detail in the manuscrips.

Total mortality

In manuscript 4 methods applicable to determine total mortality from length frequency distributions were evaluated. Suitable methods were then applied to field data of the Dutch and German North Sea. Within the period 1955 to 2006 a mean Z (total mortality) of 5.4 to 5.7 was determined, depending on the chosen

method. Interannual variability was high and a period of high Z in the early 1990s was observed. As abundance and occurrence of predators as well as fishing effort did not match with high mortality rates, bottom up effects and the discussed (manuscript 5) regime shift in the North Sea during that time span might be an important factor. Additionally a correlation between spring temperature and total mortality was observed. Both, the effect of spring temperature and the indication for bottom up effects can be explained by taking the results of manuscript 3 (starvation and food limitation) into account. In late winter, early spring condition of shrimps caught in Büsum and Wilhelmshaven was low and the most critical month seems to be March. Further the data suggested that during winter food is sparse. An increase in temperature at the end of a starvation period would increase metabolic costs and speed up weight and therefore condition loss. This will most probably lead to a loss of a part of the population. As this mainly influences the overwintering cohort and therefore larger juveniles and adults this increase in mortality can be determined with the methods applied in manuscript 5 and explains the correlation with spring temperature.

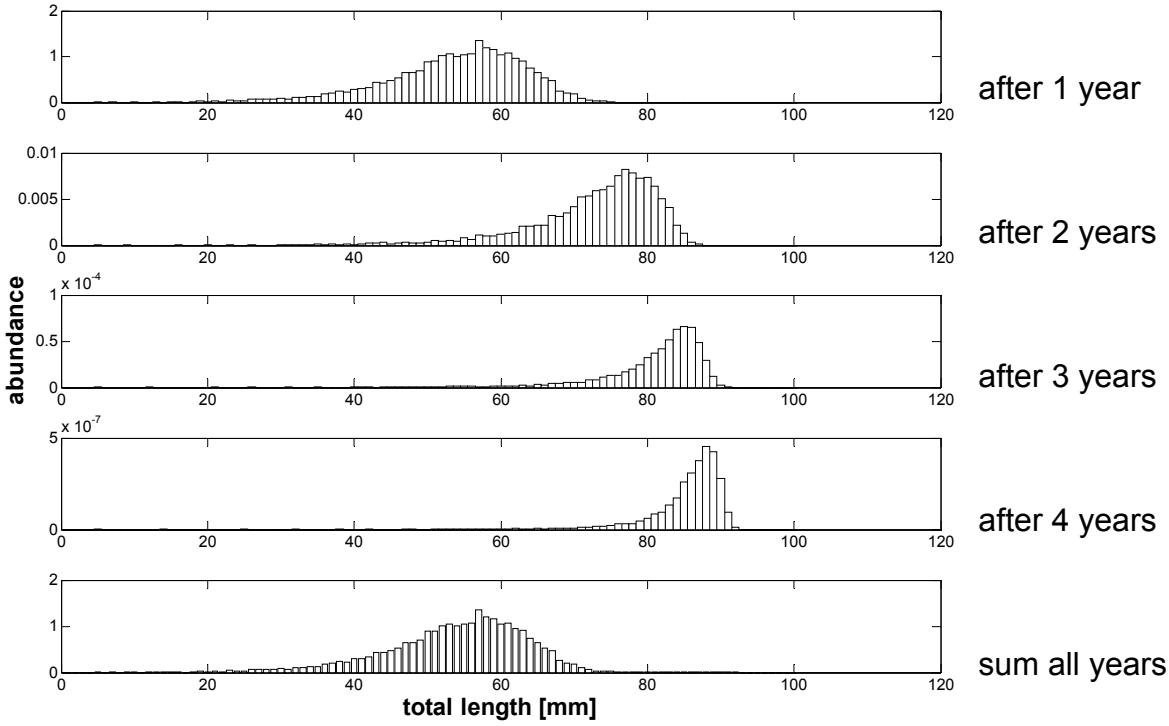

Figure 11-1: Calculated length frequency distributions of *Crangon crangon* based on the growth model determined in manuscript 1 and a total mortality of Z = 5.5 (manuscript 5). Virtual shrimps start at day 120 (May 1st) with 5 mm total length. Growth within a cohort is normally distributed between ±15% of the determined mean growth rate (mean growth rate model manuscript 1).

Combining the determined growth and mortality rates allows to estimate the abundance change of an artificial *C. crangon* cohort with time (Figure 11-1). Mean

length of the cohorts, starting at May 1st with 5 mm total length, increases from 57 mm in year 1 to 89 mm in year 4 whereas relative maximum abundance decreases from 1 to $4 \cdot 10^{-7}$. In this simulation growth rates and temperatures are the same used in manuscript 1 and total mortality was based on manuscript 5 (Z = 5.3). The major part of the population structure is build up by one-year-old shrimps. In relation thereto the abundance of 2, 3 and 4 year old shrimps is low and decreases to 0.003%, $1.6 \cdot 10^{-5}$ and $9.2 \cdot 10^{-8}$% of the start cohort size, respectively.

Sex change

A similar model approach to that one used to create Figure 11-1 was applied in manuscript 6 to determine the influence of male *C. crangon* changing sex on the population. The output of the model was compared with the sex ratios determined from the samples taken in Büsum and Wilhelmshaven. The results suggested that sex specific growth rates alone lead to a decline of the sex ratio from small to large individuals. Nevertheless the results of Schatte & Saborowski (2005), Martens & Redant (1986) and Boddeke indicate that *C. crangon* has developed the ability to change sex, which is an evolutionary difficult step (Charnov, 1982) and which would not be present if it would not have been evolutionary selected against. It can therefore be concluded that sex change was more profitable in the past than today. Under the actual habitat conditions male shrimps do not profit any more from a sex change.

The results of manuscript 5 suggested that the total mortality in the 1950s was about half of that in the 1990s. Furthermore maximum length observed in the catches decreased and the fraction of animals >70 mm making up 15-20% of the catches in the past declined to about 2%. These changes might have shifted the advantages and disadvantages of hermaphroditism. For several marine species changes have been observed regarding maturation and behavior as response to increasing levels of fishing effort or changes in mortality (Fukuwaka and Morita, 2008;Moiseenko, 2002;Watanabe and Yatsu, 2006). The observed changes of the maximum size of *C. crangon* in scientific and commercial catches as well as the observed increase in total mortality might therefore have also lead to changes in the population dynamics of *C. crangon*. As shown sex change from male to female is only profitable if the animals lives long enough to benefit from the change (Policansky, 1982). A higher mortality and therefore a reduction of the maximum age and longevity reduces this benefit.

Based on the data of the present work and the available literature it is not possible to determine if and when changes of the sexual system have occurred. It might therefore have happened due to the increased fishery or due to geographical and habitat changes or be a result of both. Luttikhuizen (2008) assumed that the origin of *C. crangon* is in the Mediterranean. In the present population density observed in the Mediterranean and on sandy shores are generally low (Beyst et al., 2002) in comparison to the Wadden Sea. Changes might therefore also have occurred when *C. crangon* colonized the new habitat Wadden Sea. The Wadden Sea is a

geologically young habitat that developed during the last 11000 years. *C. crangon* got adapted more and more to the life in the shallow highly productive areas. Growth rates and density increased but parallel the amount of predators increased as well. As shown before the high mortality necessarily leads to a reduction of maximum age and therefore the evolutionary selection preferring sex changing males was reduced. The advantages of hermaphroditism got further reduced with increasing shrimping. The fishery added an artificial selection pressure against large animals and decreased maximum age further. Based on the simulations presented in the actual work the contribution of secondary female eggs is therefore very low (<1%).

Life cycle

Plotting the determined L_∞ against k as well the total mortality against L_∞ in comparison to other marine species (values taken from manuscript 5) shows that *C. crangon* has a short maximum length and high growth performance (Figure 11-2). Growth parameters can be compared best (within the listed species) to *Macrobrachium macrobrachion*, an important decapod of the ecology of rivers and estuaries along the west coast of Africa, from Senegal (latitude 20°N) to Angola (latitude 16°S) (Holthuis, 1980). This shrimp inhabits a comparable habitat and holds a comparable trophic and fishery position as *C. crangon* (Nwosu et al., 2007). Parameter estimates for *C. crangon* are not as extreme as though observed for *M. macrobrachion* which is not surprising regarding the multiple habitats inhabited by *C. crangon*.

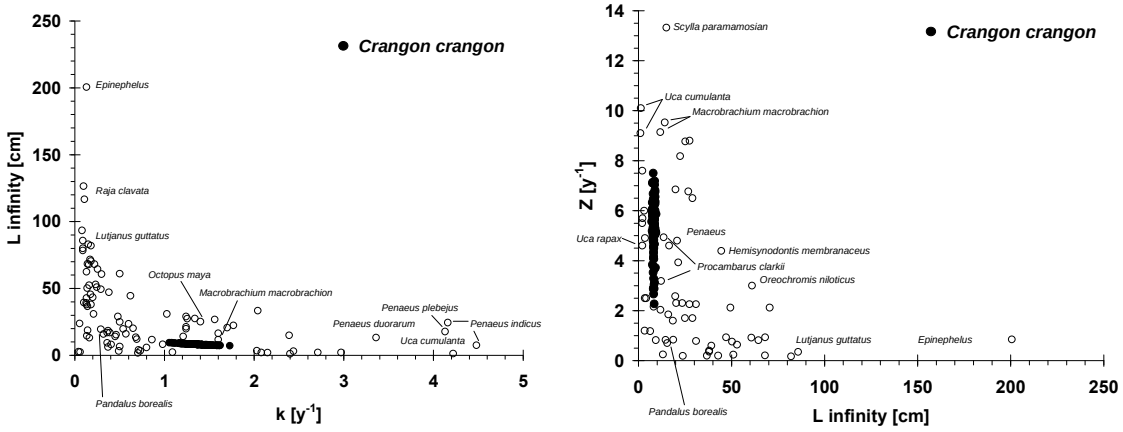

Figure 11-2: von Bertalanffy growth parameters L_∞, k and total mortality Z of *Crangon crangon* related to different marine species (references and single values are listed in manuscript 4).

Extremes such as the high k determined for several penaeid species are, in general, an adaptation to specific niches or results from stable conditions with low variability. Given the high seasonal changes in temperature, salinity, food resources, drift and predation pressure such highly-adapted species would not be able to survive in the Wadden Sea. The observed and described variability of

brown shrimp growth is therefore most likely a necessary adaptation to the environmental conditions based on match mismatch scenarios considering prey availability, prey requirements or preferred prey sizes of predators. Further reasons for the observed variability could be a permanent mixing of the stock over a large latitudinal distribution ares. Taking the anticlockwise flow fields of the North Sea into account it can be suggested that the Northern stocks get “supplied” by southern shrimps and by migration of the large shrimps towards deeper water in winter and offshore spawning the Northern stocks also supply the Southern stocks with new larvae. There are indications that life cycle and breeding season differ with latitude (Kuipers & Dapper 1984). Habitat conditions in colder regions might therefore also select for slower growing shrimps whereas in warmer regions faster one might be preferred.

The actual work improved the knowledge about the life cycle, growth and mortality of *C. crangon* in several ways. Based on the existing literature and the new results the population dynamics will be described in the following section.

Highest numbers of small individuals (Juveniles) are observed in the intertidal areas in late spring and early summer (April-June) (Beukema, 1992; Boddeke, 1976; Hartsuyker, 1966) in more northern areas somewhat later (Pihl and Rosenberg, 1982). These small juveniles and postlarvae can mainly be observed in shallow areas and mud puddles. There they are able to get the highest benefit from the high temperatures and the lowest predation pressure.

In manuscript 1 it could be demonstrated that all juveniles starting between December and June with 5 mm total length reached the size of invading juveniles in May and July and marketable size by October. The latter fits with high landings and large amounts of shrimps >50 mm observed in fall (Maes et al., 1998). These shrimps do not carry eggs before December as in fall (September, October, November) the amount of egg bearing shrimps is low (Neudecker and Damm, 1992) most probably due to suppression by hormones which are produced in the eye stalks (Klek-Kawinska and Bomirski, 1975). From December on the amount of egg bearing shrimps increases (Siegel et al., 2008) and since then berried females can be observed throughout the whole year (Kuipers and Dapper, 1984). The adult animals, mainly females as shown in manuscript 1, avoid the colder water temperatures in winter and migrate up to 90 km offshore (Boddeke 1975,1976). With increasing temperature they return to more shallow areas in spring (Pihl and Rosenberg, 1982).

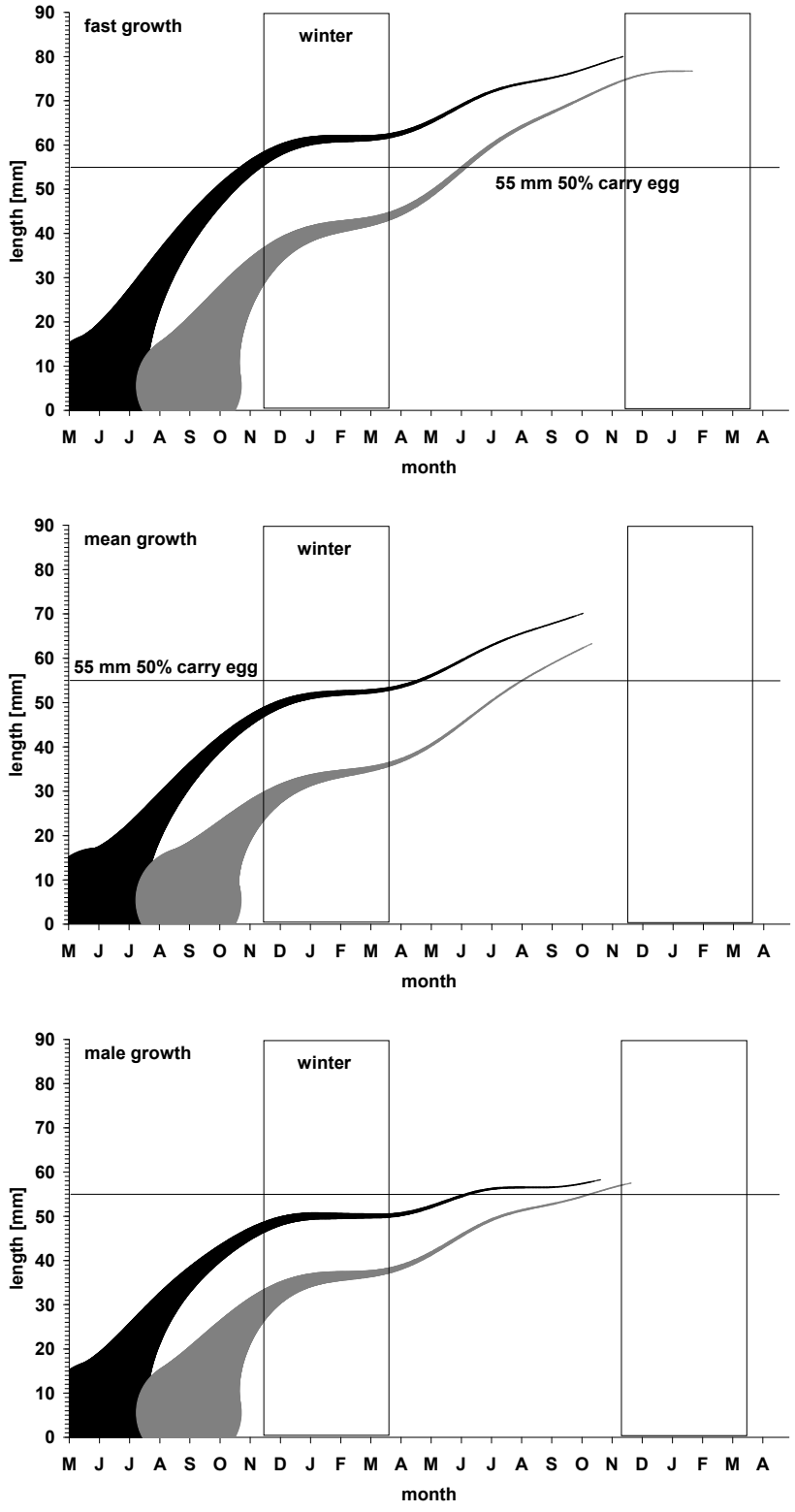

Figure 11-3: Length trajectories and abundance of animals growing at fast (top) or mean (middle) rates and length trajectories calculated for male shrimps (bottom). Tracks end if less than 0.01% of the cohort is left. Growth rates were taken from manuscript 1 and Z decreased with size from Z=20 at 5 mm to Z=5.3 at 45 mm. Animals start either in June (black) or September (grey) with 5 mm total length.

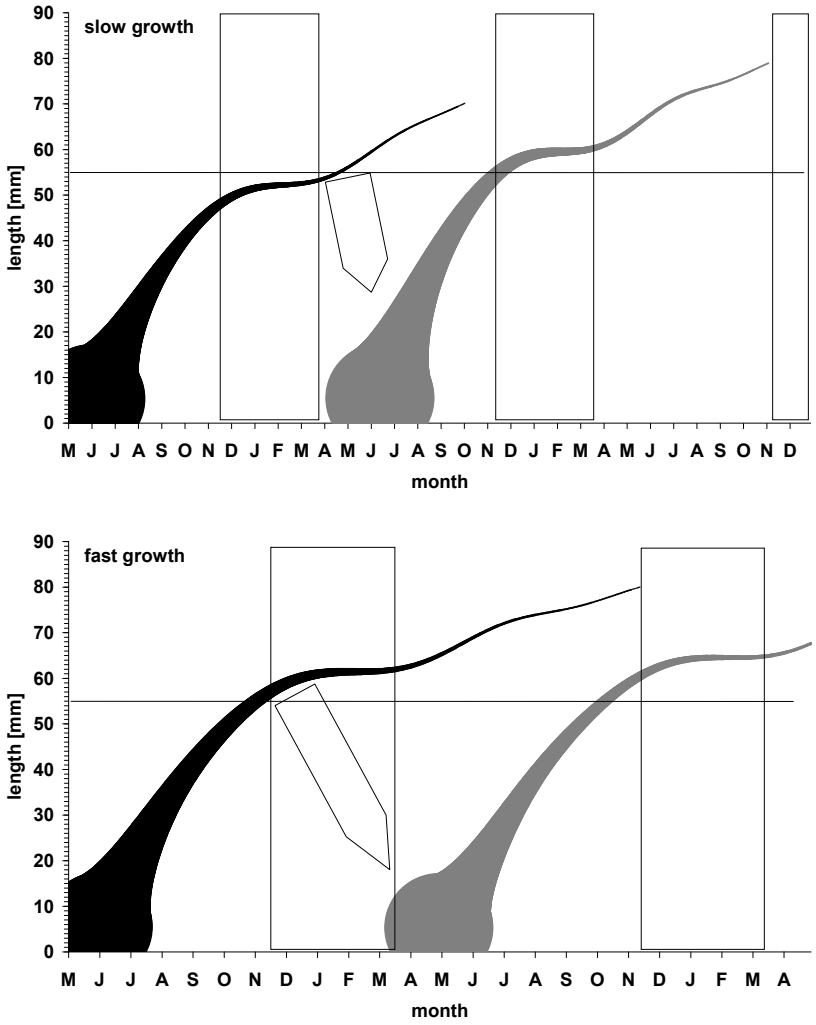

Figure 11-4: Length trajectories and abundance of animals growing at mean (top) or fast (bottom) rates. Conditions as described in Figure 11-3 but animals only start in June. Grey indicated is the cohort that follows from the first eggs released by the first cohort.

Mortality for larvae and juveniles, mainly due to predation, but also according to foodniche overlaps, differs throughout the year and is therefore higher in summer than in winter (Basimi and Grove, 1985;Evans, 1983;Maes et al., 1998;Pohle, 2004). The resulting higher mortality of summer larvae is offset to some extent by a higher amount of eggs carried per female in summer (Oh and Hartnoll, 2004) and a shorter moult interval (manuscript 1) resulting in more eggs released in a comparable time interval. Larvae that hatched during summer most probably do not reach maturity until winter and stay comparable small over a long time period (Figure 11-3). Due to the higher cumulative mortality a smaller fraction of the summer-hatched shrimps survives (in comparison to winter hatched ones) and therefore only contribute to a lower extend to the stock of adult females. The winter hatched larvae on the opposite reach maturity in fall, carry eggs during winter and therefore close the cycle as invasion of juveniles in spring is mainly driven by winter hatched larvae (Temming and Damm, 2002).

Although adult females from summer hatched larvae are less numerous, due to higher temperatures and higher prey amounts these animals can reach maximum length of nearly 80 mm within one year (Figure 11-3). This is comparable to the maximum length observed in catches (manuscript 5). Yet larger females might contribute substantially to the total egg production, as they carry up to 10 times more eggs than smaller females (Redant, 1978). Immature females that survived the winter and the critical warming phase in early spring are able to channel the increasing amount of available prey first into growth and then into the production of summer eggs. In manuscript 6 it was calculated that these summer hatched females (starting with 5 mm in September) first carry eggs in May of the following year, comparable to the slow growing cohort in Figure 11-4. Larvae that hatch from these eggs are able to use the warm temperatures and high amount of available prey. The fast growing shrimps will again reach maturity until winter like the larvae hatched from winter eggs. In short fast growing shrimps from winter eggs again produce winter eggs and later as larger females summer eggs. Slow growing females from winter eggs reach maturity in spring of the following year and produce spring/summer eggs. From these eggs the fast one might be able to produce winter eggs, but the juveniles suffer higher predation pressure in summer. The fraction of females that do not reach maturity until winter will be higher, in comparison to the winter hatched shrimps, due to the late start and therefore a higher fraction will suffer a higher cumulative mortality due to no growth in winter. Summer egg production is therefore not wasteful although most probably only a lower fraction survives.

Less than 1% of the males that hatched from winter eggs, reproduced and survived the winter reach the size window of potential sex change. Due to the long intermoult periods during winter these secondary females will carry eggs not before spring as shown in manuscript 6. Already numerously reduced and smaller than the primary females they will support the stock of summer eggs only to a fraction that is lower than 1%.

Conclusions

The results of the presented work imply that growth rates of *C. crangon* are highly variable. This high variability is most probably not only a laboratory artefact but also actually present in the field. There it might be a successful strategy for survival in a highly variable habitat or an effect of a bottom up (food) and top down (predation and shrimping) controlled population. Based on the high mortality levels it could be shown that the majority of the population is not older than one year, that in the Southern North Sea mainly the fast growing shrimps determine the population and additionally that large males do not profit from changing sex. The whole life cycle of the shrimp can be closed within on year. Anyhow older and larger animals can be observed in the field, especially in areas where shrimping pressure is low (Denmark, ICES 2007). In the southern North Sea catches the observed maximum length decreased during the last 50 years whereas landings continuously increased. In protected areas or natural (not human influenced)

conditions large animals most probably contribute significantly to the population as they carry more eggs. Additionally predation pressure on the larger shrimps is lower. Under such conditions also sex change might be more advantageous what should be further investigated. Another factor that should be disentagled in future work should be the determination of factors that influence RNA/DNA. This might help to approve that the growth rates determined in the actual work are really present in the field.

Although until now there are no signs of overfishing *C. crangon*, problems might arise with increasing winter fishery or increasing predator populations (cod and whiting). The increased winter fishery directly reduces the spawning stock biomass and in combination with low food concentrations in spring or high predation pressure this might have serious implications on the population. Whiting and cod populations are down at the moment but are under management and there are recent signs of recovery. If protection measures are successful and the predator stock increases than not only the winter but also the traditional fishery might influence the brown shrimp population.

References

Basimi RA, D J Grove, 1985, Estimates of daily food intake by an inshore population of *Pleuronectes platessa* L. off eastern Anglesey, North Wales: Journal of Fish Biology, v. **27**, p. 505-520.

Berghahn R, 1996, Episodic mass invasions of juvenile gadoids into the Wadden Sea and their consequences for the population dynamics of brown shrimp (*Crangon crangon*): Marine Ecology, v. **17**, p. 251-260.

Beukema JJ, 1992, Dynamics of juvenile shrimp *Crangon crangon* in a tidal flat-nursery of the Wadden Sea after mild and cold winters: Marine Ecology Progress Series, v. 83, p. 157-165.

Beyst B, J Vanaverbeke, M Vincx, J Mees, 2002, Tidal and diurnal periodicy in macrocrustaceans and demersal fish of an exposed sandy beach, with special emphasis on juvenile plaice *Pleuronectes platessa*: Marine Ecology Progress Series, v. 225, p. 263-274.

Boddeke, R, 1976, The seasonal migration of the brown shrimp *Crangon crangon*: Netherlands Journal of Sea Research, v. 10, p. 103-130.

Charnov EL, 1982, The Theory of Sex Allocation, Princeton,New Jersey Princeton University Press.

del Norte-Campos AGC, A Temming, 1998, Population dynamics of the brown shrimp *Crangon crangon* L., in shallow areas of the German Wadden Sea: Fisheries Management and Ecology, v. **5**, p. 303-322.

Evans S, 1983, Production, predation and food niche segregation in a marine shallow soft-bottom community: Marine Ecology Progress Series, v. 10, p. 147-157.

Fukuwaka M, K Morita, 2008, Increase in maturation size after the closure of a high seas gillnet fishery on hatchery-reared chum salmon *Oncorhynchus keta*: Evolutionary Applications, v. 1, p. 376-387.

Hartsuyker L, 1966, Daily tidal migrations of the shrimp *Crangon crangon* L.: Netherlands Journal of Sea Research, v. **3**, p. 52-67.

Holthuis LB. FAO Species catalogues. Vol. 1. Shrimps and prawns of the world. An annotated catalogue of species of interest to fisheries. FAO Fish.Synop.(125), 271. 1980.

Houde ED, 1987, Fish early life dynamics and recruitment variability: American Fisheries Society Symposium, v. 2, p. 17-29.

Klek-Kawinska E, A Bomirski, 1975, Ovary-inhibiting hormone activity in Shrimp (*Crangon crangon*) eyestalks during the annual reproductive cycle: General and Comparative Endocrinology, v. 25, p. 9-13.

Kuipers BR, R Dapper, 1984, Nursery function of Wadden Sea tidal flats for the brown shrimp *Crangon crangon*: Marine Ecology Progress Series, v. **17**, p. 171-181.

Maes J, A Taillieu, P A Van Damme, K Cottenie, F Ollevier, 1998, Seasonal Patterns in the Fish and Crustacean Community of a Turbid Temperate Estuary (Zeeschelde Estuary, Belgium).: Estuarine Coastal and Shelf Science, v. 47, p. 143-151.

Moiseenko, TI, 2002, Change in the Life Cycle Strategy of Fish under the Effect of Chronic Water Pollution: Russian Journal of Ecology, v. **33**, p. 45-55.

Neudecker T, U Damm. Seasonality of egg-bearing shrimp (*Crangon crangon* L.) in costal waters of the German Bight. K: 28, 1-9. 1992. ICES Shell Fish Committee.

Nwosu FM, Holzlöhner Sieghard, U I Enin, 2007, The exploited population of the brackish river prawn (*Macrobrachium macrobrchion* Herklots 1851) in the Cross River estuary, Nigeria: Scientia Marina, v. 71, p. 115-121.

Oh C-W, R G Hartnoll, 2004, Reproductive biology of the common shrimp *Crangon crangon* (Decapoda: Crangonidae) in the central Irish Sea.: Marine Biology, v. 144, p. 303-316.

Peterson I, J S Wroblewski, 1984, Mortality rate of Fishes in the Pelagic Ecosystems: Canadian Journal Fisheries and Aquatic Science, v. 41, p. 1117-1120.

Pihl L, R Rosenberg, 1982, Production, abundance, and biomass of mobile epibenthic marine fauna in shallow waters, western Sweden: Journal of Experimental Marine Biology and Ecology, v. 57, p. 273-301.

Pohle J. Prädationsdruck auf die Nordseegarnele *Crangon crangon* L. durch Fische im Sublitoral der Sylt-Rømø-Bucht und im vorgelagerten Seegebiet. -72. 2004. Wattenmeerstation Sylt, Alfred-Wegener Institut für Polar- und Meeresforschung.

Policansky D, 1982, Sex change in plants and animals: Annual Reviews of Ecology and Systematics, v. 13, p. 471-495.

Redant F. Konsumptie en pruoduktie van post-larval *Crangon crangon* (L.) (Crustacea, Decapoda) in de Belgische kustwateren, DEEL I. TEKST. 1978. Vrije Universiteit Brussel, Fakulteit Wetenschappen. Thesis/Dissertation

Siegel V, U Damm, T Neudecker, 2008, Sex-ratio, seasonality and long-term variation in maturation and spawning of the brown shrimp *Crangon crangon* (L.) in the German Bight (North Sea): Helgoland Marine Research, v. 62.

Temming A, U Damm, 2002, Life cycle of *Crangon crangon* in the North sea: A simulation of the timing of recruitment as a function of the seasonal temperature signal: Fisheries Oceanography, v. 11, p. 45-58.

Temming A, U Damm, T Neudecker. Trends in the size composition of commercial catches of brown shrimp (*Crangon crangon* L.) along the German coast. K: 53. 1993. ICES Shell fish Committee.

Watanabe C, A Yatsu, 2006, Long-term changes in maturity at age of chub mackerel (*Scomber japonicus*) in relation to population declines in the waters off northeastern Japan: Fisheries Research, v. 78, p. 323-332.

Acknowledgements

I am grateful for all the support in all kinds of different ways, that I have received whilst researching and writing my dissertation.

Foremost I want to express my special gratitude to my supervisor Prof. Axel Temming for introducing me into the interesting world of the brown shrimp, for giving me tons of inspirations and for many, not only work related, chats and discussions that in some ways changed my view of the world.

Furthermore I would like to thank Prof Myron Peck for many good ideas, correcting my manuscripts, improving my English, travelling to interesting meetings and especially for widening my understanding of marine sciences and to always remind me on the “big picture”.

Jens-Peter Herrmann I would like to thank for helping me with experimental setups, designs and the experiments themselves. For many apparently unsolvably questions he developed perfectly operating setups.

Thank you Sven Stäcker and Jochen Lütke for helping me with the experiments, feeding the animals and helping me out on several week ends. Further thanks to all the IHF members and especially the weekend cheklisters.

Prof. Fritz Buchholz, Dr. Reinhard Saborowski and the AWI Helgoland members I would like to thank for support during the performance of the growth experiments and three nice, productive and warm summer month on the island.

Moreover I would like to thank Adreas Dänhardt for taking me along on several interesting, helpful, workloaded and blithe surveys and for collecting the shrimps from the power station.

Carmen Czserwinski, Inga Röwer, Anneke Denda, Julia Herbolzheimer, Antje Krohn, Claudia and all the HiWis employed in the project, thank your for measuring and preparing shrimp samples.

Chris Rückert, Matthias Bernreuther, Alexander Kempf and Robert Perger I would like to thank for a very very good time, scatterbraining coffee and lunch brakes, Ehrensenf sessions, long discussions, support and fun.

I would also like to honor all shrimps that lost their lives in the name of science hopefully they swim in endless prey fields or will be reborn one day as scientists.

I am further in true gratitude to my parents who enabled and always supported me in all kinds of ways during my study and who never questioned, but always encouraged my decision becoming a scientist.

Finally I am deeply grateful to my wonderful girlfriend Annika Schrader her love, patience and ability to always find the right words to motivate me and cheer me up. She always remembered me about the life beyond science and without her I would have gone lost in my office.