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Carbon and nitrogen sources for juvenile blue crabs *Callinectes sapidus* in coastal wetlands

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ABSTRACT: We used a combination of field and laboratory techniques to examine the relative importance of food webs based on marsh detritus, benthic algae, or phytoplankton in supporting growth of the blue crab *Callinectes sapidus*. We conducted a laboratory experiment to compare the growth of newly metamorphosed juveniles fed natural diets from potential settlement habitats such as marshes. The experimental diets consisted of zooplankton, *Uca pugnax* and *Littoraria irrorata* tissue, a mixture of plant detritus and associated meiofauna and detritus only. Crabs fed the zooplankton diet showed the fastest growth and reached a mean dry weight of 32.4 mg, from an initial dry weight of 0.8 mg, during a 3 wk period. Based on the isotopic composition, juvenile crabs obtain carbon and nitrogen from various food sources. For example, crabs fed zooplankton obtained their nutrition from phytoplankton-derived organic matter, consistent with zooplankton feeding on phytoplankton. The mean $\delta^{13}C$ values for juveniles fed detritus and detritus-plus-meiofauna were considerably lighter ($\delta^{13}C = -19‰$), than that of their respective diets ($\delta^{13}C = -16‰$), suggesting that crabs were selectively ingesting prey items that obtain their nutrition from an isotopically lighter carbon source like phytoplankton. Conversely, crabs fed *U. pugnax* or *L. irrorata* had isotopic ratios ($\delta^{13}C = -16$ to $-14‰$) consistent with these species feeding on isotopically heavier marsh grass carbon. Isotopic ratios of crabs collected in the field appeared to corroborate the experiment and suggest that either *Spartina alterniflora* detritus or benthic algae-based food webs supported juvenile crab growth in marsh environments, whereas phytoplankton-based food webs dominate habitats more closely associated with the main estuary.

KEY WORDS: *Callinectes sapidus* · Juveniles · Growth · Stable isotopes · Carbon and nitrogen · Salt marsh · Food web

INTRODUCTION

Salt marshes and other coastal wetlands are critical nursery areas for many species of fish and invertebrates. The importance of these nurseries has been attributed to at least 2 factors: refuge from predators and high food abundance (Orth et al. 1984). The refuge value of marshes has been well documented and is related to the structured nature of the habitat (e.g. Heck & Thoman 1984, Hines et al. 1987, Orth & van Montfrans 1987). However, the role of autochthonous marsh production in providing high food abundance is still unclear. While early studies (Odum & de la Cruz 1967) had suggested that vascular marsh plants support the nutrition of fish and invertebrates via detritus-based food webs, more recent investigations have come to varying conclusions about the respective importance of vascular plant detritus and algal carbon (e.g. Peterson & Howarth 1987, Stoner & Zimmerman 1988, Sullivan & Montcreiff 1990). At present, there is no consensus in the literature concerning the relative value of different sources of primary production that might ultimately support the growth and development of species that use the marsh during their early life-history stages.

Of particular interest are predatory species that may provide top-down effects on community structure in estuaries and that may support large commercial fisheries with obvious societal importance. For example, the blue crab *Callinectes sapidus* is a major predator along the Atlantic coast from New Jersey (USA) to

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Argentina (Virnsten 1977, 1979, Holland et al. 1980, Williams 1984) and supports a large fishery through much of its range. The species exploits both estuarine and neritic ecosystems during its complex life cycle. Adult blue crabs are found primarily in estuaries, but the larval forms undergo development in the adjacent coastal ocean with subsequent transport of the postlarvae (megalopa stage) back to the estuary, wherein they settle and undergo metamorphosis in an appropriate juvenile habitat (Dittel & Epifanio 1982, Epifanio 1995, Jones & Epifanio 1995). While it is well known that seagrass beds and salt marshes are among the most important of these nursery areas, the sources of food for postlarvae and juveniles in these habitats are poorly understood.

Numerous laboratory studies have addressed the growth and development of crustacean larvae and postlarvae. Most of these have depended on live prey items such as newly hatched brine shrimp larvae (*Artemia* spp.) or marine rotifers (e.g. *Brachionis plicatilus*) (Mootz & Epifanio 1974, Sulkin 1975, Epifanio et al. 1991). These foods are rich in energy and support development of larval crabs, shrimp, and lobsters; however, they are surrogates for natural prey organisms and may not provide maximum growth (e.g. Anger & Darwis 1981, Epifanio et al. 1991, Harvey & Epifanio 1997). Other studies have utilized analysis of stomach contents to assess the dietary preferences and have concluded that postlarvae and juveniles consume a variety of items, including small invertebrates and plant detritus (Chong & Sasekumar 1981, Laughlin 1982, Wassenberg & Hill 1987, Robertson 1988, Stoner & Zimmerman 1988). However, this type of investigation does not address the ultimate source of primary production that supports the growth and development of these early life-history stages.

In contrast, stable isotope ratios (e.g. $\delta^{13}C$ and $\delta^{15}N$) offer a powerful method of examining the ultimate sources of primary production supplying food energy for secondary and tertiary consumers. This approach has been used to examine the carbon and nitrogen sources for estuarine fauna in habitats adjacent to coastal wetlands such as salt marshes (for a review see Fry & Sherr 1984). However, studies have come to varying conclusions about the relative importance of vascular plant production in supporting higher trophic levels (Peterson & Howarth 1987). For example, results of some studies along the Gulf and southeast Atlantic coasts of the USA have indicated that detrital food chains are of primary importance to juvenile crustaceans (Fry & Parker 1979, Haines & Montague 1979), while other studies have pointed to the preeminence of algae-based sources (Hackney & Haines 1980, Hughes & Sherr 1983, Sullivan & Montcreiff 1990). Furthermore, the relative importance of detrital food chains depends on location within the marsh. For example, Peterson et al. (1985) reported that ribbed mussels in the interior portions of the marsh derived 80% of their diet from detrital sources, whereas in the main marsh channels 70% of the diet consisted of plankton.

In the present study we examined the hypothesis that blue crabs switch from a phytoplankton-based food chain to a detritus-based food chain after settling in the estuary and that growth through the juvenile stages is supported by this detritus-based food chain. To test our hypothesis, we conducted a laboratory experiment to compare the growth of newly metamorphosed juveniles fed natural diets from potential settlement habitats such as marshes. The isotopic composition (carbon and nitrogen) of the organisms and their respective diets was used to determine the carbon and nitrogen sources for blue crab growth.

MATERIALS AND METHODS

We conducted a 3 wk experiment to measure growth and isotopic composition of juvenile blue crabs fed either a control diet consisting of newly hatched brine shrimp *Artemia* sp. or 1 of several experimental diets based on prey organisms from the natural environment of the crabs. Juveniles used in the experiments were obtained by collecting blue crab postlarvae from open water adjacent to the marsh and holding them in the laboratory without feeding for 2 to 4 d until they molted to the first juvenile stage. Newly metamorphosed juveniles were haphazardly assigned to one of the control or experimental groups and immediately subjected to experimental conditions.

Newly hatched brine shrimp *Artemia* sp. were used as the control diet because these are the standard food organism used in the laboratory culture of decapod crustaceans (e.g. Welch & Epifanio 1995, Dittel et al. 1997). Experimental diets were chosen based on their accessibility to juvenile crabs that use local marshes as nursery areas and based on our assumptions concerning the different trophic origins of the diets. Diets 1 to 3 (below) were expected to be part of a detrital food chain originating in vascular marsh plants, while Diets 4 and 5 were anticipated to be part of food chains respectively based on benthic algae or phytoplankton.

Components of the experimental diets were collected from the Great Marsh, located near the mouth of Delaware Bay along the Atlantic coast of the USA (Fig. 1). The Great Marsh is part of a larger system of salt marshes that extends more or less continuously along the 150 km western shoreline of the bay. The dominant emergent vegetation at our study site is marsh cord grass *Spartina alterniflora*. The experimental diets are described below.

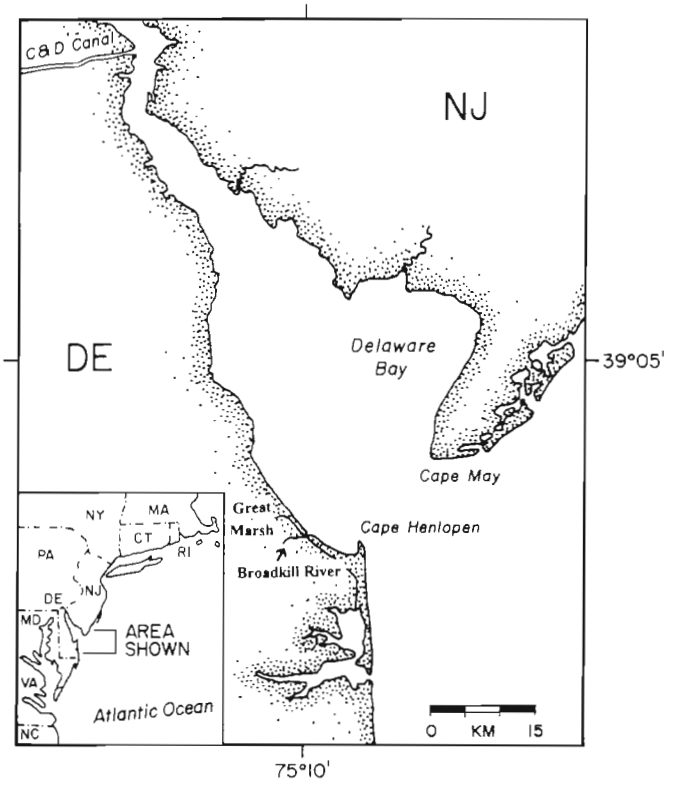

Fig. 1. Location of the study area in the Delaware Bay

(1) Detritus. Sediment samples from the marsh were elutriated in order to separate organic detritus from associated meiofauna and inorganic marsh sediments. This was done by suspending the sample in filtered seawater and decanting the suspended materials onto a 253 µm sieve. This retained the organic detritus, but allowed meiofauna and inorganic sediment to pass through. The organic detritus was resuspended in filtered seawater, and the resultant mixture was termed a *feeding suspension*. Microscopic examination was performed daily to assure that the diet was devoid of meiofauna.

(2) Detritus-plus-meiofauna. Samples were collected daily from the intertidal zone of a creek within the marsh. Meiofauna was obtained by elutriation of sediments with a 53 µm sieve, rather than the 253 µm sieve mentioned above. This smaller sieve retained both meiofauna and detritus, while again allowing inorganic marsh sediment to pass through. The actual feeding suspension consisted of a mixture of plant detritus, associated microbes, and a meiofaunal assemblage. Meiofauna consisted mainly of nematodes, harpacticoid copepods, and ostracods.

(3) Fiddler crabs Uca pugnax. This species is indigenous to our study site and appears to consume detritus. Male crabs were collected daily from the marsh. Fresh tissue was removed from the major claw and cut into small pieces before use in the experiment.

(4) Marsh periwinkle snails Littoraria irrorata. Marsh periwinkle snails are grazers that feed on epiphytic algae and fungi or on benthic algae growing on the marsh surface. Snails were collected twice a week from the marsh. Fresh tissue from the foot was cut into small pieces and stored in a refrigerator before use in the experiment.

(5) Zooplankton. Zooplankton was collected daily from open water adjacent to the marsh using a 253 µm mesh plankton net. Samples were passed through a 500 µm sieve to remove large organisms and returned to the laboratory where they were left undisturbed for 15 to 20 min to allow the detritus and inorganic sediment to settle to the bottom of the container. Living zooplankton was then siphoned into a beaker, and the density (ind. ml^{-1}) was determined with a dissecting microscope. The zooplankton assemblage used in the experiments was always dominated by the copepod *Acartia tonsa*, but also included other copepod species as well as larval forms of local mollusks and crustaceans.

Growth experiments. Experiments were conducted in replicated (n = 10) 200 ml glass bowls under controlled conditions of salinity, temperature, and light (25°C; 30‰; 14 h light/10 h dark). At the beginning of the experiment, 20 megalopae and 10 newly metamorphosed juveniles were frozen for determination of initial size and isotopic composition. Each experimental bowl contained 1 newly metamorphosed juvenile crab. Each day juvenile crabs were transferred to clean filtered seawater and fed one of the various diets. Daily rations of each diet were sufficient to allow the crabs to feed ad libitum, and excess food always remained in the bowls at the end of each 24 h period. Thus, any differences in growth were due to quality of diet, rather than quantity of ration. The daily ration for those diets consisting of discrete prey organisms (i.e. brine shrimp, meiofauna, zooplankton) was 800 prey items $crab^{-1}$ d^{-1} and the feeding suspension in the detritus treatment was added in daily units of 10 ml. The daily ration in the fiddler crab and snail diets was presented as 1 small piece of fresh tissue (~0.25 cm^2). At the end of the experiment, all surviving crabs were frozen for later determination of dry weight and isotopic composition. Treatment effects on final dry weights were compared by Model I ANOVA followed by Tukey's multiple comparison test ($\alpha = 0.05$). Dry weights and carapace widths of the crabs were transformed to log ($x + 1$) to normalize distribution and de-correlate the mean and variance.

We also determined mean growth rates of crabs for each of the dietary treatments. Growth in carapace

width (G_C) was assumed to be linear (Epifanio et al. 1998) and was calculated as:

$$G_C = (C_2 - C_1)/T$$

where C_1 is the mean length (carapace width, including lateral spines) in mm of crabs at the initiation of the experiment, C_2 is mean length at the end of the experiment, and T is the duration of the experiment in days (21 d). Growth in dry weight (G_W) was assumed to be exponential (Welch & Epifanio 1995, Epifanio et al. 1998) and was determined by:

$$G_W = (\ln W_2 - \ln W_1)/T$$

where W_1 is the mean dry weight (mg) of the crabs at the initiation of the experiment, W_2 is the mean dry weight at the end of the experiment, and T is the duration of the experiment in days.

Field collections. In addition to conducting an analysis on material from the growth experiment, we also determined isotopic ratios of a number of samples from different habitats utilized by postlarval and juvenile blue crabs (see Table 2). Samples included primary producers, juvenile crabs, and potential prey items. Particulate organic matter (POM) was collected by filtering 1 l of seawater through a pre-combusted 0.7 µm glass fiber filter. Phytoplankton was collected by filtering seawater through a 3 µm glass fiber filter (Fantle et al. 1999). Benthic algae were collected at low tide by scraping the marsh surface carefully to exclude as much non-algal material as possible. The different habitats are shown in Fig. 1 and included: (1) the Great Marsh (described above); (2) the Broadkill River, a secondary estuary that drains the marsh; (3) Roosevelt Inlet, the connection between the marsh system and the main Delaware estuary; and (4) Henlopen Flats, a sand flat located approximately 7 km seaward of the marsh.

Isotope analysis. Samples were dried at 50°C, in an oven that was flushed with N_2 gas, before being ground to a fine powder and sealed in a glass desiccator with concentrated HCl vapors to remove carbonates. Juvenile crabs were subjected to an additional step of acidification in 0.1 N HCl in order to remove calcium carbonate present in the chitinous exoskeleton (Fantle et al. 1999). Samples were analyzed isotopically by a modified Dumas combustion that converts organic carbon and organic nitrogen to CO_2 and N_2 gas for mass spectral analysis (Macko 1981). The samples were placed in quartz tubes with Cu and CuO and sealed off under vacuum (Macko et al. 1982). The quartz tubes were heated to 900°C for 1 h and cooled to room temperature at a rate of 60°C h^{-1}. The slow cooling cycle ensured that any oxides of nitrogen were decomposed to N_2. CO_2 gas was separated from N_2 gas by cryogenic distillation and analyzed by mass spectrometry. A Nier-Johnson type double-focusing mass spectrometer was used to run the nitrogen samples whereas the isotopic composition of carbon dioxide was measured on a Finnigan MAT-252 mass spectrometer. Stable isotope ratios are presented in standard notation:

$$\delta Z = (R_{sample}/R_{standard} - 1) \times 1000‰$$

where Z denotes the heavier isotope (either ^{13}C or ^{15}N), and R represents the ratio (either ^{13}C:^{12}C or ^{15}N:^{14}N). The standard for nitrogen was atmospheric N_2 and the standard for carbon was PeeDee Belemnite. For $\delta^{15}N$ and $\delta^{13}C$ the precision was ±0.2 and ±0.02‰, respectively. Carbon and nitrogen concentrations were measured on a Carlo Erba EA-1108 Elemental Analyzer.

RESULTS

We conducted a laboratory experiment in which newly molted *Callinectes sapidus* juveniles were fed natural diets in order to examine the relative importance of plant detritus and other prey items in supporting growth. We measured the isotopic composition of wild-caught megalopae and newly molted juveniles at the initiation and at the end of the experiment in order to determine the carbon and nitrogen source for blue crab growth.

There was a significant effect of diet on the final size of juvenile blue crabs in terms of both carapace width (ANOVA; $F_{5,46} = 48.9$; $p < 0.01$) and dry weight (ANOVA; $F_{5,46} = 59.9$; $p < 0.01$). At the initiation of the experiment, juveniles had a mean carapace width of 2.6 mm and a mean dry weight of 0.8 mg. Crabs fed the zooplankton diet showed the fastest growth and reached a mean carapace width of 10.5 mm and a mean dry weight of 32.4 mg over a period of 21 d (Fig. 2). This high rate of growth suggests that water-column zooplankton are a potential dietary component for newly settled juvenile crabs. Crabs fed the control brine-shrimp diet showed intermediate growth and attained a dry weight of approximately 21 mg by the end of the experiment. Juveniles fed the *Uca pugnax* or *Littoraria irrorata* diets grew more slowly, and both groups exhibited dry weights of approximately 10 mg at termination of the investigation. Crabs fed the detritus diet or the detritus-plus-meiofauna diet grew very slowly and reached only 2 to 4 mg in mean dry weight over the same time period. These dietary effects were reflected in the calculated growth rates of the crabs, which ranged from 0.07 to 0.37 mm d^{-1} for carapace width and from 0.03 to 0.17 d^{-1} for dry weight, and in the mean number of molts, which ranged from 1.3 to 4.0 over the experimental period (Table 1). In spite of

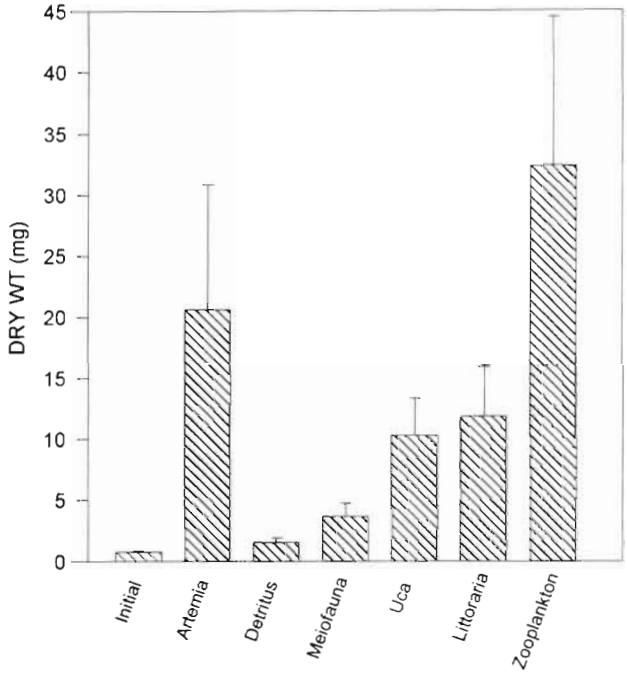

Fig. 2. Mean dry weights of *Callinectes sapidus* juveniles fed various diets after 21 d. Initial = dry weight of juvenile crabs at $T = 0$; *Artemia* spp. nauplii = control diet

the strong effects of diet on all parameters of growth, there was no apparent effect on survivorship, which was uniformly high in all groups.

We also measured organic carbon and nitrogen of the diets in order to determine whether differences had an effect on growth. There was a significant difference in the C:N ratio among the experimental diets (ANOVA; $F_{5,18} = 128.0$; $p < 0.01$). The C:N ratio was highest in the 2 detritus-based diets, reflecting their high carbohydrate contents (Table 1). The C:N ratio of the animal-based diets was 3- to 4-fold lower, which undoubtedly indicates a higher proportion of protein.

Table 1. Growth rates and average number of molts of *Callinectes sapidus* juveniles fed various diets during a 3 wk period. n: number of crabs that survived in each treatment

Diet	C:N ratio	Dry weight growth (d^{-1})	Carapace growth (mm d^{-1})	Average no. of molts	n
Artemia spp.	6.2	0.15 ± 0.02	0.32 ± 0.07	4.0 ± 0.0	9
Zooplankton	5.6	0.17 ± 0.02	0.37 ± 0.07	4.0 ± 0.0	10
Littoraria irrorata	4.6	0.13 ± 0.02	0.26 ± 0.06	3.6 ± 0.5	8
Uca pugnax	5.9	0.12 ± 0.01	0.24 ± 0.04	3.0 ± 0.8	7
Meiofauna	16.6	0.07 ± 0.02	0.15 ± 0.04	2.2 ± 0.6	10
Detritus	23.2	0.03 ± 0.01	0.07 ± 0.02	1.3 ± 0.5	9

The daily rations of the control, zooplankton, and detritus-plus-meiofauna diets all contained equal numbers of prey organisms. Expressed mathematically:

$$N_c = N_z = N_m$$

where N_c is the number of brine shrimp in the daily control ration, N_z is the number of zooplankters in the zooplankton ration, and N_m is the number of meiofauna organisms in the detritus-plus-meiofauna ration. Because the individual organisms that comprised each of the diets were of similar size, the approximate quantity of protein in the rations of the respective diets was probably similar, and the detritus-plus-meiofauna differed only in its added component of high-carbon detritus. Expressed mathematically:

$$P_c \approx P_z \approx P_m$$

where P is the amount of protein in the daily rations of the respective diets. Nevertheless, the growth supported by the detritus-plus-meiofauna diet was much less than that supported by either brine shrimp or wild zooplankton. Moreover, the crabs fed the *Uca pugnax* and *Littoraria irrorata* diets also grew more slowly than the control and zooplankton groups, even though the C:N ratios of the 4 diets were very similar.

Over the 3 wk course of the experiment, the isotopic signature of the juvenile crabs changed as a function of diet. Crabs fed *Uca pugnax* and *Littoraria irrorata* had the highest $\delta^{13}C$ values (−16 to −14‰), which is consistent with the composition of these prey species (Table 2, Fig. 3A). On the other hand, the mean $\delta^{13}C$ values for juveniles in the detritus and detritus-plus-meiofauna groups were lower (= lighter) than those of their respective diets (Fig. 3A). For example, $\delta^{13}C$ of juveniles fed detritus-plus-meiofauna was approximately −19‰, whereas the average $\delta^{13}C$ of the diet was about −16‰. This large difference (fractionation) between the isotopic composition of the crabs and the detritus diet suggests that crabs were selectively ingesting prey items and were not using the detritus directly. Furthermore, the ingested prey appear to have obtained their nutrition from an isotopically lighter carbon source such as phytoplankton (Fig. 3A). Similarly, crabs fed detritus showed a large negative fractionation (3‰) in $\delta^{13}C$ relative to the detritus diet, suggesting that these crabs did not utilize the detritus directly.

There was also variation in fractionation of $\delta^{15}N$ by crabs grown on the different food items. Crabs fed *Uca pugnax*, *Littoraria irrorata* and zoo-

Table 2. Isotopic composition (‰). (A) Experimental diets, (B) juvenile crabs after 21 d, and (C) key factors from the study sites. n: number of samples analyzed; values are mean ± 1 SD. POM: particulate organic matter

Sample type	$\delta^{13}C$	n	$\delta^{15}N$	n	Site
(A) Experimental diets					
Artemia spp. control	−22.3 ± 0.2	3	6.9 ± 0.1	2	
Littoraria irrorata	−14.6 ± 0.7	4	10.3 ± 0.2	2	
Uca pugnax	−15.7 ± 0.6	10	7.4 ± 0.4	4	
Detritus-plus-meiofauna	−16.1 ± 0.3	5	5.5 ± 1.1	4	
Detritus	−15.4 ± 0.3	5	6.7 ± 0.4	4	
Zooplankton	−16.6 ± 0.7	5	10.8 ± 0.5	5	
(B) Juvenile crabs					
$T = 0$ juveniles	−17.1 ± 0.4	3	10.0 ± 0.1	2	
Artemia spp. control	−21.3 ± 0.4	3	8.4 ± 0.3	5	
Littoraria irrorata	−14.4 ± 0.4	4	11.1 ± 0.3	5	
Uca pugnax	−15.8 ± 0.1	2	8.3 ± 0.2	6	
Detritus-plus-meiofauna	−19.5 ± 0.4	2	8.6 ± 0.9	5	
Detritus	−18.5 ± 0.5	2	9.0 ± 0.1	4	
Zooplankton	−16.7 ± 0.4	2	10.9 ± 0.3	5	
(C) Key factors					
Phytoplankton	−22.0 ± 0.3	5	7.4 ± 0.7	4	Roosevelt Inlet
Benthic algae	−17.6 ± 0.3	5	5.8 ± 1.2	2	Great Marsh
Spartina alterniflora detritus	−15.7 ± 1.3	8	5.8 ± 1.2	6	Great Marsh
POM	−22.1 ± 0.6	3	5.1 ± 1.7	3	Great Marsh
Meiofauna-ostracods	−19.3	1	6.8	1	Great Marsh
Mytilus edulis	−20.2 ± 0.3	5	9.1 ± 1.1	3	Broadkill River
Geukensia demissa	−18.5 ± 0.7	5	8.0 ± 0.5	3	Great Marsh
Ilyanassa obsoleta	−13.0 ± 0.2	3	9.7	1	Great Marsh
Zooplankton (>500 µm)	−16.8 ± 0.3	2	10.1 ± 0.0	2	Roosevelt Inlet
Copepods (253–500 µm)	−18.2 ± 0.01	3	11.2	1	Roosevelt Inlet
Crab larvae (>500 µm)	−17.1 ± 0.6	3	10.0	1	Roosevelt Inlet
Callinectes sapidus	−16.6 ± 0.8	11	11.2 ± 0.6	10	Broadkill River
C. sapidus	−16.0 ± 0.8	2	11.4 ± 1.0	3	Henlopen Flats
C. sapidus	−15.1 ± 0.7	9	10.7 ± 1.2	6	Great Marsh
C. sapidus megalopae	−19.3 ± 0.1	2	8.6 ± 0.2	2	Roosevelt Inlet

plankton showed small fractionations of the diets (0.1 to 0.9‰), whereas crabs fed detritus and meiofauna fractionated their diets to a greater extent (2 to 3‰ respectively) (Fig. 3B). Although $\delta^{15}N$ values of crabs fed *L. irrorata* (11.1‰) reflected those of the diet (10.3‰), the ^{15}N isotopic composition of the *L. irrorata* diet was higher than has been reported in previous investigations (Peterson & Howarth 1987, Currin et al. 1995).

In order to place the results of our experiment into a natural context, we measured the isotopic composition of a number of key species from the marsh and from adjacent habitats (Table 2). Wild-caught blue crab megalopae had $\delta^{13}C$ values (−19‰) consistent with a phytoplankton-based food web. The $\delta^{13}C$ isotopic composition of the zooplankton (−17 to −18‰) indicates that phytoplankton (−22 ± 0.3‰) were the food source for the zooplankton. In turn, isotopic ratios of megalopae resembled those of the zooplankton. Also, juvenile crabs sampled in open-water environments and in the marsh reflected various contributions of phytoplankton and *Spartina alterniflora* detritus with open-water crabs showing isotopic signatures that were substantially lighter than marsh crabs.

The nitrogen isotope composition of primary producers in the bay was significantly different (*t*-test; $t_6 = 2.4$; $p < 0.01$) than that of primary producers from the Great Marsh (Table 2). In the marsh, POM from the water column had the lowest $\delta^{15}N$ values (5.1‰), while benthic algae and marsh detritus were only slightly enriched ($\delta^{15}N = 5.8$‰), suggesting that primary producers in the marsh derive their N from related sources. Phytoplankton collected in Roosevelt Inlet were considerably enriched in $\delta^{15}N$ (7.4‰) relative to primary producers in the marsh. These differences in the N isotope composition of primary producers in the inlet and the marsh were reflected in the isotopic composition of crabs fed the various diets. In general, crabs fed the marsh-based diets (8 to 9‰) were enriched by 2 to 3‰ in ^{15}N relative to the detritus and benthic algae. In contrast, crabs fed zooplankton were enriched in ^{15}N (10.9‰) relative to the phytoplankton ($\delta^{15}N = 7.4$‰), which is consistent with the hypothesis that these crabs were subsisting on a phytoplankton-based food web. Primary consumers (e.g. *Uca pugnax*, *Geukensia demissa*) collected in the marsh also reflected the $\delta^{15}N$ values of the marsh primary producers.

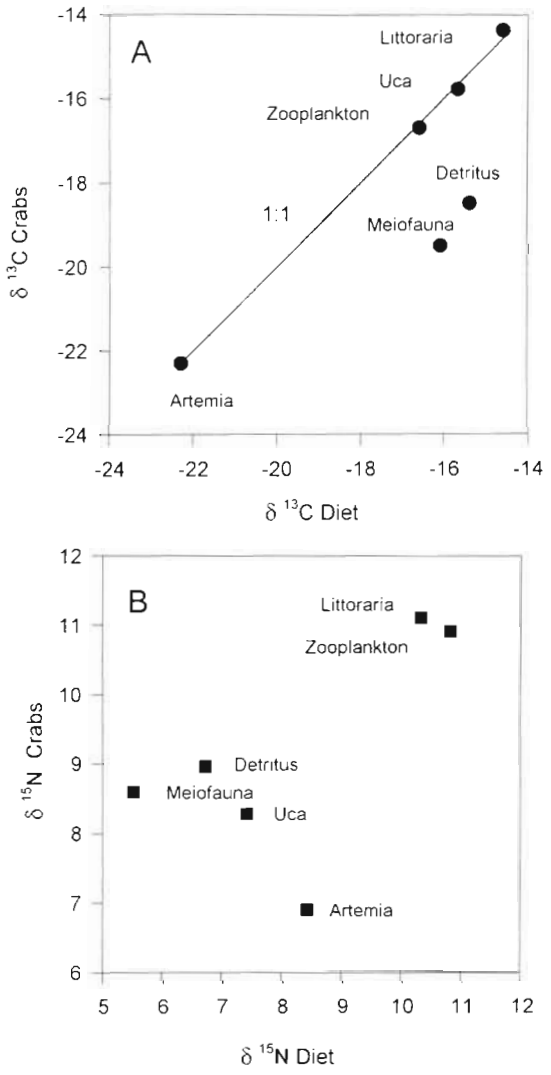

Fig. 3. (A) Stable carbon ($\delta^{13}C$) isotope ratios of juvenile blue crabs versus their respective diets. Line indicates 1:1 fractionation (i.e. crab $\delta^{13}C$ = diet $\delta^{13}C$). (B) Nitrogen isotopic composition of juvenile crabs versus their respective diets

DISCUSSION

Results of our investigation have shown that blue crab juveniles grew rapidly on diets consisting of zooplankton or brine shrimp and at moderate rates on diets consisting of *Uca pugnax* or *Littoraria irrorata*. Crabs fed zooplankton had a 40-fold increase in dry weight over the 3 wk duration of the experiment, whereas juveniles fed *U. pugnax* or *L. irrorata* grew at least 10-fold over the same period. However, growth rates of crabs fed meiofauna-plus-detritus were significantly less than those fed zooplankton. This was probably due to the fact that the crabs were ingesting both meiofauna and detritus, thus reducing the overall nutritional value of the ration. The exceptionally high growth rates observed in our zooplankton treatment indicate that juveniles are able to capture and ingest water-column zooplankton and that these organisms constitute a potential dietary component for newly settled juvenile crabs. Juveniles in the control and zooplankton treatments increased in dry weight at an exponential rate of between 0.15 and 0.17 d^{-1}, which is comparable to the rapid growth observed in larval crabs under optimum conditions in the laboratory (Welch & Epifanio 1995). This potential for very rapid growth in the earliest juvenile stages may be an adaptation to the high levels of predation experienced at this point in their life history (Seed & Brown 1978, Baily & Houde 1989, Meekan & Fortier 1996).

Blue crab juveniles fed detritus grew significantly less than those fed other diets. Nonetheless, the high survival (90%) and positive growth rates of juveniles fed this diet indicate that detritus may provide an ancillary source of nutrition for crabs when other food sources are scarce. While our isotope data showed that crabs did not assimilate carbon directly from the detritus, it is likely that these juveniles derive nutrition from microbial assemblages associated with the detritus and egest the refractory detrital carbon as feces.

Results of our experiment also provided explicit information concerning isotopic fractionation of carbon and nitrogen derived from known food sources. Generally our results support the idea that the $\delta^{13}C$ values of consumers are similar to the isotopic composition of their diet (Haines & Montague 1979) or are higher by 1 or 2‰ (DeNiro & Epstein 1978, Fry & Sherr 1984), while the $\delta^{15}N$ values of consumers tend to be higher than the food by as much as 2 to 4‰ per trophic level (Minagawa & Wada 1984).

The isotopic composition of the zooplankton used in our experiment was consistent with zooplankton feeding on phytoplankton. In turn, crabs were isotopically similar to the zooplankton diet. Conversely, crabs fed *Uca pugnax* or *Littoraria irrorata* had isotopic ratios consistent with these species feeding on isotopically heavier marsh carbon (e.g. benthic algae or *Spartina alterniflora* detritus). The $\delta^{13}C$ values determined for *U. pugnax* and *L. irrorata* in our investigation are similar to those reported in previous studies that have demonstrated the importance of *S. alterniflora* and benthic algae as food sources for fiddler crabs and marsh periwinkles (e.g. Haines & Montague 1979, Montague 1980, Currin et al. 1995).

In contrast to other diets, crabs fed detritus and detritus-plus-meiofauna were depleted in $\delta^{13}C$ by 2.3 and 3.1‰ relative to the detritus and meiofauna diets, respectively. This discrepancy in $\delta^{13}C$ between the crabs and the diets indicates that the $\delta^{13}C$ values of the bulk material did not reflect the material actually con-

sumed by the crabs. For example, juvenile crabs fed the detritus-plus-meiofauna diet probably picked the meiofauna from the detritus. Nematodes, ostracods and harpacticoid copepods, which characterized the meiofauna diet, are known to be among the preferred prey items of small juvenile blue crabs (Laughlin 1982, Stoner & Buchanan 1990). Furthermore, the $\delta^{13}C$ isotope ratios of the crabs fed detritus-plus-meiofauna (–19.5‰) suggest that these crabs were ingesting prey items that obtained their nutrition from a food web based on an isotopically lighter carbon source such as benthic algae rather than marsh detritus.

In addition to bulk isotopes, recent studies (Hayes et al. 1990, Fogel et al. 1997) have used compound-specific isotope analysis to investigate trophic dynamics in natural studies. In one such investigation, we have used the C isotope composition of individual amino acids to clarify trophic relationships of juvenile blue crabs and to define fractionation related to metabolic processes (Fantle et al. 1999). As with bulk isotopes, $\delta^{13}C$ values of various essential amino acids (isoleucine, valine, phenylalanine, leucine) revealed that when offered diets consisting of marsh detritus juvenile blue crabs assimilated isotopically lighter carbon that probably originated in microorganisms rather than the detritus itself (Fantle et al. 1999). These results are in agreement with bulk-isotope studies which have shown that planktonic and benthic algae are important contributors of carbon for the dominant meiofaunal taxa like nematodes, harpacticoid copepods and ostracods (e.g. Gleason & Zimmerman 1984, Montagna 1984, Riera et al. 1996). In contrast, Couch (1989) reported $\delta^{13}C$ values for the meiofauna (–13.9 to –15‰) intermediate between the benthic algae (–12 to –13.5‰) and *Spartina alterniflora* detritus (–15.9 to –16.7‰) and concluded that detrital *S. alterniflora* may be the main source of C for the meiofaunal populations in the North Inlet Estuary, South Carolina. However, the isotopic similarity of the meiofauna to both food sources does not rule out the assimilation of benthic algae.

The results of our experiment were used to interpret the isotopic composition of juvenile blue crabs collected in the field and ultimately to determine the relative contributions of marsh and algal carbon. For example, the $\delta^{13}C$ values of crabs found in the Great Marsh (–15‰) resembled those of crabs fed the *Uca pugnax* and *Littoraria irrorata* diets, indicating that food webs based on heavy marsh carbon are an important dietary component for crabs feeding in the marsh. On the other hand, crabs collected at open-water sites (–16 to –17‰) had $\delta^{13}C$ values similar to those fed the zooplankton diet (–17‰). These results seem to indicate that very small (2.2 to 3.2 cm) juvenile crabs collected in the bay consumed organisms that derived their nutrition from an isotopically lighter carbon source such as phytoplankton.

Isotope analysis also indicated that primary producers in open-water and marsh habitats derive their N from different sources. Marsh primary producers had ^{15}N values which ranged between 5.1 and 5.8‰, whereas those of phytoplankton were about 7.4‰. Previous studies have shown that a major source of N for *Spartina alterniflora* in marshes is ammonium generated by bacterial remineralization of marsh detritus (Haines 1977, Fogel et al. 1989). The higher ^{15}N values of the phytoplankton may be indicative of N derived from agricultural and municipal wastes that end up in the estuary from point sources or land run-off. These differences in the N isotopic composition of primary producers were reflected in the ^{15}N values of the experimental crabs and the organisms collected in the field. With the exception of crabs fed *Littoraria irrorata*, those fed marsh diets in the laboratory were enriched (2 to 3‰) in ^{15}N relative to the detritus and benthic algae. Crabs fed zooplankton had considerably higher ^{15}N values (10.9‰) and were enriched by 3.5‰ relative to the phytoplankton. The nitrogen composition of crabs fed periwinkles (10.7‰) reflected that of their diet, although we lack an explanation for the unexpectedly high ^{15}N values of the periwinkle (10.3‰) diet, itself. In the field, primary consumers collected in the marsh also reflected the $\delta^{15}N$ values of the marsh primary producers. The higher $\delta^{15}N$ signatures of blue crab juveniles in the marsh (10.7‰) and in open-water habitat (11.4‰) compared to the experimental crabs may be attributed to a greater diversity in the types of prey items consumed by field crabs. Also, crabs in the field most likely fed on organisms from more than 1 trophic level, which would result in the higher $\delta^{15}N$ values.

Overall, our data show a considerable fidelity between the isotopic composition of juvenile blue crabs and their apparent food sources. Crabs fed controlled diets under laboratory conditions had isotopic signatures consistent with expected trophic fractionation. Moreover, the isotopic composition of crabs collected from the natural environment varied as a function of crab habitat. Crabs collected from open water appear to utilize phytoplankton-based production, while those found in marsh habitat appear to exploit food chains originating in marsh detritus or benthic algae. This dependence on marsh production may be particularly important in otherwise unvegetated estuaries like Delaware Bay, where salt-marsh creeks constitute the dominant nursery area for species like blue crabs. Our results offer strong evidence that these marsh areas provide more than simple refuge from predation for these juvenile forms; they provide the ultimate source of primary production that supports early growth and development. However, once the crabs move to open-water habitat, they appear to exploit food chains that are not dependent on outwelled marsh production.

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