ResearchGate

See discussions, stats, and author profiles for this publication at: https://www.researchgate.net/publication/225432193

Stable isotope analysis reveals ontogenetic migration and the importance of a large mangrove estuary as a feeding ground for juvenile John's snapper Lutjanus johnii

Article *in* Fisheries Science · September 2011

DOI: 10.1007/s12562-011-0396-x

CITATIONS
31

READS
328

8 authors, including:

Katsuhisa Tanaka
National Research Institute of Far Seas Fisheries
32 PUBLICATIONS **638** CITATIONS
SEE PROFILE

Ving Ching Chong
University of Malaya
181 PUBLICATIONS **4,396** CITATIONS
SEE PROFILE

Satoshi Watanabe
National Research Institute of Fisheries Science Japan Fisheries Research and Ed…
37 PUBLICATIONS **564** CITATIONS
SEE PROFILE

Alias Man
Department of Fisheries Malaysia
26 PUBLICATIONS **436** CITATIONS
SEE PROFILE

All content following this page was uploaded by Katsuhisa Tanaka on 11 May 2016.

The user has requested enhancement of the downloaded file.

Fish Sci (2011) 77:809–816
DOI 10.1007/s12562-011-0396-x

ORIGINAL ARTICLE

Biology

Stable isotope analysis reveals ontogenetic migration and the importance of a large mangrove estuary as a feeding ground for juvenile John's snapper *Lutjanus johnii*

Katsuhisa Tanaka · Yukio Hanamura · Ving Ching Chong · Satoshi Watanabe · Alias Man · Faizul Mohd Kassim · Masashi Kodama · Tadafumi Ichikawa

Received: 8 March 2011 / Accepted: 25 July 2011 / Published online: 13 August 2011
© The Japanese Society of Fisheries Science 2011

Abstract Stable carbon (δ^{13}C) and nitrogen (δ^{15}N) isotope ratios were measured to investigate the migration of John's snapper *Lutjanus johnii* and its dependence on the food resources provided within the large Matang Mangrove Forest Reserve (40,151 ha), Malaysia. John's snapper, and its main prey food such as penaeids, *Acetes* shrimps and mysids, showed generally depleted δ^{13}C values in the inner mangrove area but more enriched values in the river mouth and coastal area. Some juveniles migrated into the inner mangrove areas, although they were also distributed near the river mouth areas. Isotopic signatures of snapper fish and prey reveal the ontogenetic migration of the youngest juvenile fish (<5 cm in total length) from the coastal area into the mangrove area, shifting their dependence from the coastal food web to the inner mangrove food web with their growth. The study shows the importance of the complex interconnected mangrove waterways and associated prey animals present in the large mangrove system to juvenile John's snapper.

Keywords John's snapper · *Lutjanus johnii* · Mangrove estuary · Ontogenetic migration · Prey animals · Stable isotope

K. Tanaka (✉) · S. Watanabe
Japan International Research Center for Agricultural Sciences, Owashi, Tsukuba 305-8686, Japan
e-mail: katuhi@affrc.go.jp

Y. Hanamura · M. Kodama · T. Ichikawa
National Research Institute of Fisheries Science, Fisheries Research Agency, Yokohama, Kanagawa 236-8648, Japan

V. C. Chong
Institute of Biological Sciences, Faculty of Science, University of Malaya, 50603 Kuala Lumpur, Malaysia

A. Man
Fisheries Research Institute, Kampung Acheh, 32000 Sitiawan, Perak, Malaysia

F. Mohd Kassim
Japan International Research Center for Agricultural Sciences (Penang Office), Batu Maung, 11960 Penang, Malaysia

Introduction

Mangroves contribute substantially to coastal fisheries in terms of providing trophic and refuge support as well as larval retention [1]. Typically, the adults of many euryhaline, tropical fish species spawn in the offshore area, producing eggs which develop into planktonic larvae that move or are carried by currents into the inshore and estuarine waters, including mangroves [2–4]. The young fish spend a period of time in these waters, where they grow before returning back to the offshore areas or adjacent reefs. Recently, evidence has been mounting of inter-habitat connectivity between mangroves and reefs, based on studies comparing fish from these habitats in terms of length frequencies [5–7], densities [8], stable isotopes [9], otolith structures and microchemistries [10, 11], and implanted tags [12]. However, the mangrove areas surveyed in almost all these studies are shoreline fringe type or small-scale mangroves that are normally less than 1 km in width; there are few studies focusing on the ontogenetic migration of fish into large mangrove estuaries.

Stable isotope ratios can provide useful information on the origins and migration paths of organisms if the isotopic compositions of their prey differ following the movement of the organisms to a new habitat [13]. The isotope ratios

serve as natural tags for distinguishing recent immigrants from those that have partially or fully equilibrated with the isotopic composition of their prey consumed in the new habitat [14]. After migration, the stable isotope ratios of the animal's tissues gradually converge toward the value of its consumed food or the organic sources at the new habitat. This implies that comparisons of stable isotope ratios can be used to distinguish between recent immigrants and individuals that have inhabited the sampling site for a relatively long period of time [15, 16].

In Southeast Asia, John's snapper *Lutjanus johnii* is an important commercial fish species, both in capture fisheries and in aquaculture. The average annual catch of this species in Malaysia was 4,056 t from 1997 to 2007, while aquaculture production is 1,762 t annually. It has been reported that young John's snapper continuously recruit into the estuaries of the Matang Mangrove Forest Reserve (MMFR), staying there for approximately a year, before they start to migrate back to the open sea [17]. The diet of John's snapper in the estuaries mainly consists of hyperbenthic crustaceans (penaeid prawns, *Acetes* shrimps and mysids) [17, 18]. Juvenile prawns in the estuaries of the MMFR showed a marked shift in their tissue stable isotope ratios with distance from the river mouth; those in mangrove creeks and waterways have generally depleted $\delta^{13}C$ values, while those located offshore show more enriched values, thus producing distinctive isotopic signatures between these habitats [19]. Such shifts in the $\delta^{13}C$ values of macroinvertebrates from the mangroves to the adjacent sea were also observed in Gazi Bay, Kenya [20]. These observations of isotopic changes in prey organisms as a function of location indicate that stable isotope ratios could potentially be used as a tracer of the ontogenetic migration of John's snapper from the coastal area into the mangrove estuary, and to elucidate its dependence on the food sources provided within the MMFR.

In this study, stable carbon and nitrogen isotope ratios of juvenile John's snapper and its potential prey animals collected from the estuaries of MMFR were measured to investigate (1) the ontogenetic migration of the juveniles into the mangrove estuaries, and (2) the dependence of the fish on the food sources provided in the mangrove nursery area.

Materials and methods

Study area

The Matang Mangrove Forest Reserve (MMFR), situated on the northwestern coast of Peninsular Malaysia, is reputed to be the world's best managed mangrove forest. The reserve is the largest single tract of mangrove forest in Peninsular Malaysia (40,151 ha), measuring 52 km between the extreme ends and 13 km wide in the middle, and has been managed as a sustainable production forest since 1905 [21]. The average annual rainfall at Kuala Sepetang (Fig. 1) is 2,109 mm. Rainfall occurs throughout the year, but there are two peaks of heavy rainfall coinciding with the onset of the southwest and northeast monsoons in April and November, respectively [22]. MMFR is a riverine forest type mangrove that is inundated during most spring high tides. In contrast, water is mainly confined to the channels during neap tides. Tides in Kuala Sepetang are typically semi-diurnal, with mean high water springs of 2.65 m [23].

Sampling locations in the northern part of the MMFR are shown in Fig. 1. MMFR is deltaic in origin, being drained by a complex system of interconnected waterways comprising large permanent channels and their smaller branches and creeks that dry out during low tide. The whole system is strongly tidal and estuarine, although the large channels are topographically termed rivers.

Offshore collections were made at the station OFF approximately 10 km from the shore. Stations R1 and R2 were situated near the coastal mudflat at 4 and 2 km offshore from the mouth of the Sangga Besar River, respectively. Stations R3 (river mouth) to R5 (9 km upstream) were located along the Sangga Besar River, which is the widest river system in the estuary. Station S3 was located 13 km upstream from the river mouth (S1) of the Selinsing River, which joins the Sangga Besar River at the fishing village of Kuala Sepetang. Stations C1–C3 were located in a creek area. The two river systems were interconnected by a creek which has outlets at station S2 and between stations R3 and R4.

The depth of the river basin of the Sangga Besar River ranged from 4 to 6 m between stations R3 and R5, while the coastal and mudflat areas (OFF, R1, R2) were shallower (1–3 m). Stations along the Selinsing River (S1–S3) had water depths of 4–5 m, while the creek stations (C1–C3) had depths of 2–3 m. The mean water temperature in the study area was 29.6°C (range 26.6–31.1°C). In the Sangga Besar River, the water during spring tides was vertically well mixed, while the water was partially stratified during neap tides [24]. In the Selinsing River and creek water, the salinity range was relatively small compared to the the Sangga Besar River, with a maximum in the bottom layer of the mouth of the Selinsing River (S1) [24]. The mean salinity and standard deviation in the bottom water of the Sangga Besar River (R1–R5) and the Selinsing River and creek stations (S1, S2, C1–C3) ranged from 26.0 ± 2.9 (R1) to 19.3 ± 6.0 (R5) and 22.5 ± 2.1 (S1) to 20.2 ± 2.8 (C3), respectively [24].

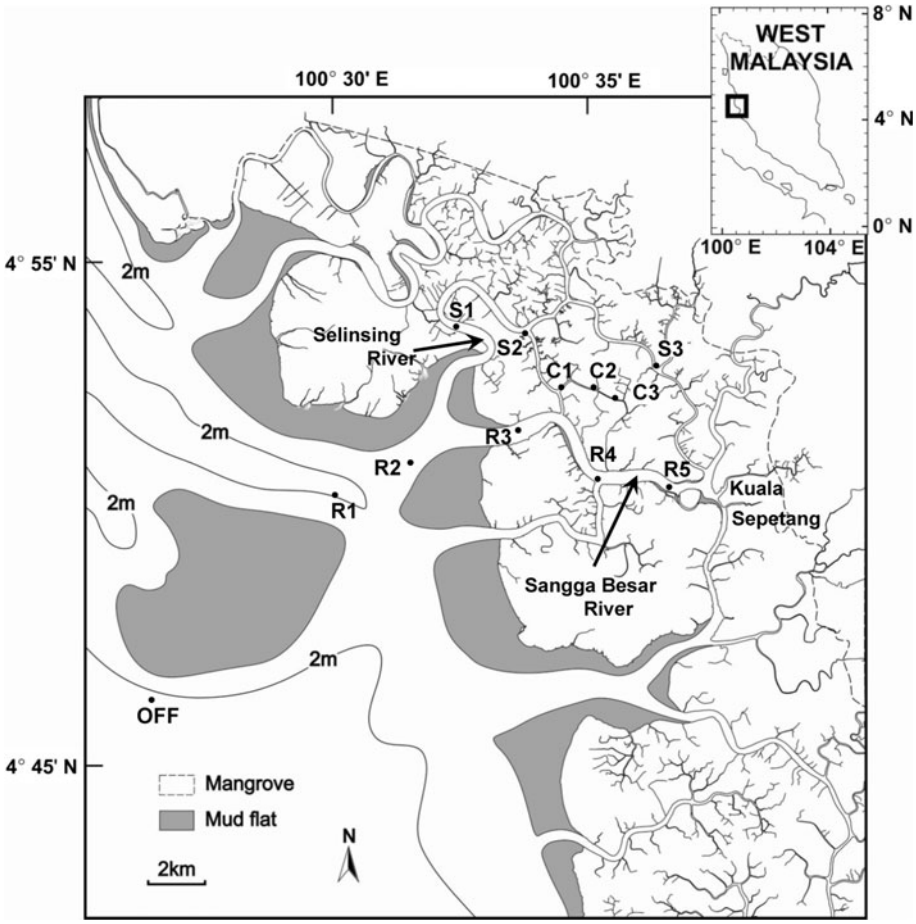

Fig. 1 Map of the sampling stations (*filled circles*) in the estuaries of the Matang Mangrove Forest Reserve, on the west coast of Peninsular Malaysia

Sample collections

John's snapper sampling was carried out mainly at four stations (S1, S2, R3 and R4) monthly from July to November 2007. Additional fish sampling was also conducted in November 2008 and February 2009 at stations S2–S3 and C1, respectively. The fish were caught by a 2 m wide otter trawl net (R3, R4, S1–S3) or set traps (C1). Mysids and *Acetes* shrimps were sampled in November 2007 and from November to December 2008 using a small hand dip net (30 × 30 cm mouth area; 0.77 mm mesh openings) during the morning at low tide, when the shrimps were observed to form large aggregations at the mangrove margin. In addition, collections were made using a sledge net (60 × 40 cm mouth width, 0.77 mm mesh openings) in the coastal area (OFF, R1, R2). Juvenile penaeid prawns were obtained using an otter trawl net on Oct. 24, 2007 in the lower Sangga Besar River (R3, R4) and the Selinsing River (S1, S2).

At each sampling time, salinity and temperature were measured with a multiple sensor (Alec Electronics Co., ASTD687). All of the John's snapper and hyperbenthic crustaceans collected were kept on ice in the field and frozen in the laboratory until examination. In the laboratory, the total lengths (TL) and wet weights (WW) of the fish were measured. TL was used as the standard length measurement for mysids, while carapace length (CL) was used for *Acetes* shrimps and prawns. However, at the sampling in November 2007, TL was measured for *Acetes* samples, and this was converted to CL using the relationship between TL and CL.

Hyperbenthic crustaceans

From previous studies in the estuaries of MMFR, it is known that the main prey items of John's snapper are *Acetes* shrimps (89% occurrence rate in examined fish stomachs), followed by penaeid prawns (33%) and mysids (10%), while the volumetric composition of *Acetes* shrimps constituted 37–43% of total stomach contents [17, 18]. These three groups of crustacean prey also comprised more than 95% of food items in the stomach based on numerical abundance [17].

In MMFR, penaeids constituted 48.5%, followed by *Acetes* (33.6%) and mysids (12.4%), of the total biomass of

hyperbenthic crustaceans [25]. The dominant mysids included *Mesopodopsis tenuipes*, *Acanthomysis* spp., and *Rhopalophthalmus* sp., while the three species of *Acetes* found were *Acetes sibogae*, *A. japonicus*, and *A. indicus* [25]. Among the three mysid species, acanthomysid species (*Acanthomysis thailandica* + *Notacanthomysis hodgarti*: collectively called *Acanthomysis* spp. in this paper) occurred mainly in the coastal and mudflat areas (OFF, R1–R3), while *Mesopodopsis tenuipes* and *Rhopalophthalmus* sp. were distributed in the upper Sangga Besar River (R5) and in the inner creek stations (S2, C2, C3). *Acetes* were also frequently found in the studied area, where *Acetes japonicus*, *A. indicus,* and *A. sibogae* were the major components. *A. japonicus* and *A. indicus* were abundant in the river mouth, although they invaded slightly more inner estuarine areas, while *A. sibogae* tended to increase in the middle to upper reaches of the MMFR [25]. *Penaeus merguiensis* (=*Fenneropenaeus merguiensis*) is known to be closely associated with the mangrove habitat [19], and was the most abundant prawn species in the Selinsing River, while another species, *Metapenaeus brevicornis*, was equally dominant in the Sangga Besar River [26].

Stable isotope analysis

Frozen specimens were thawed and rinsed with distilled water before their tissues were filleted and dried in an oven at 60°C for 24–48 h. The tissues were then ground to a fine powder with a mortar and pestle. Fish white muscle tissue was used for isotope analysis because of its slow turnover rate, which should reflect the isotopic composition of food assimilated over periods of several weeks to months [14]. Abdominal muscle tissues of prawns were used for analysis. Small crustaceans (*Acetes* shrimps and mysids) were analyzed whole. To eliminate the effect of lipids on δ^{13}C measurements, powdered samples were defatted by adding 2:1 chloroform–methanol solution (v/v) and centrifuged. The defatted samples were oven-dried, and an aliquot of each sample (ca. 0.8 mg) was put in a tin container for the analyses. Because of their small sizes, 30–100 individuals of mysids and 2–5 individuals of *Acetes* shrimps were pooled to make a single sample, rinsed with distilled water, and treated with 1 M hydrochloric acid to remove inorganic carbonate.

The carbon and nitrogen isotope ratios (δ^{13}C and δ^{15}N) were analyzed using an EA-1108 elemental analyzer (Carlo Erba) coupled with an isotope ratio mass spectrometer (Finnigan Mat ConFlo II, Mat 252). The isotope ratios were expressed as the per mil (‰) deviation from international standards (i.e., fossil calcium carbonate for C and air for N) as follows: δ^{13}C and $\delta^{15}\mathrm{N} = [(R_{\mathrm{sample}}/R_{\mathrm{standard}}) - 1] \times 10^3$. Instrumental precision was 0.2‰ [27].

Results

Mysids and *Acetes* shrimps

Isotopic compositions of mysids and *Acetes* shrimp samples obtained from the Matang Mangrove estuaries are listed in Table 1. There were significant differences between the δ^{13}C values of *Acanthomysis* spp. residing in the coastal mudflat and the other two mysid species (*Mesopodopsis tenuipes* and *Rhopalophthalmus* sp.) residing inside the mangrove (*t* test, $P < 0.001$); the latter showed depleted (lower) δ^{13}C values in the upper rivers and creek stations (−28.0 to −24.5‰), while the former gave more enriched (higher) values at the mouth of the Sangga Besar River and coastal stations (−21.9 to −17.5‰). Similarly, *Acetes japonicus* and *A. indicus* distributed mainly in the coastal (R1–R2) and lower Sangga Besar River (R3–R4) stations showed enriched δ^{13}C values (−21.8 to −17.8‰), while those in the Selinsing River (S1, S2) had slightly lower values (−23.2 to −22.1‰). On the other hand, *Acetes sibogae* found mainly in the upper Sangga Besar River (R5), upper Selinsing River (S2, S3) and creek stations (C1−C3) had significantly depleted δ^{13}C values (−27.7 to −23.9‰) compared with other *Acetes* species (*t* test, $P < 0.001$).

The average δ^{15}N values for *Acetes* shrimps and mysids were 10.8 ± 0.5 and 10.3 ± 0.7‰, respectively. Unlike δ^{13}C, the δ^{15}N variations in *Acetes* shrimps and mysids were small, with no significant differences being detected among the coastal area, river and creek stations. However, *Acetes indicus* had slightly higher δ^{15}N values than *A. japonicus* (*t* test, $P < 0.05$), which may reflect the larger body size of *A. indicus* and possibly its higher trophic level.

Penaeids

More enriched δ^{13}C values were observed in *Penaeus merguiensis* and *Metapenaeus brevicornis* occurring in the Sangga Besar River (R3, R4) compared to those in the Selinsing River (S1, S2), while *M. brevicornis* showed 1–3‰ lower δ^{13}C values than *P. merguiensis* at all stations (Table 2). In the Selinsing River (S1, S2), the average δ^{13}C values ranged from −23.7 (*P. merguiensis*) to −26.7‰ (*M. brevicornis*), whereas the values were more enriched in the Sangga Besar River (R3, R4), ranging from −20.2‰ (*P. merguiensis*) to −24.3‰ (*M. brevicornis*). These data for both species exhibited a gradual seaward δ^{13}C enrichment from the channel of the Selinsing River (S2, S1) through the lower main river of the Sangga Besar (R4) to the river mouth of the Sangga Besar River (R3).

Table 1 Isotopic compositions of mysids and *Acetes* shrimps

Sample	Sampling date	Sampling gear	Size (mm)	Sampling site	δ^{13}C (‰)	δ^{15}N (‰)
Mysids						
Mesopodopsis tenuipes	Nov. 26, 2008	Sledge net	3.5–8.0 TL	R5	−24.5	11.7
	Dec. 2, 2008	Hand net	5.5–8.0 TL	C2	−27.9	10.1
	Dec. 2, 2008	Hand net	4.0–8.0 TL	C3	−27.9	9.9
	Nov. 26, 2008	Hand net	5.0–9.0 TL	R5	−27.3	9.9
	Nov. 28, 2007	Hand net	5.0–8.0 TL	R5	−25.8	9.7
Acanthomysis spp.[a]	Nov. 25, 2008	Sledge net	4.0–7.0 TL	OFF	−17.5	9.0
	Nov. 25, 2008	Sledge net	3.0–7.0 TL	R1	−19.1	10.1
	Nov. 25, 2008	Sledge net	4.0–7.0 TL	R3	−21.7	10.9
	Nov. 25, 2008	Sledge net	3.0–7.0 TL	R2	−21.0	10.4
	Nov. 28, 2007	Sledge net	5.0–8.0 TL	R1	−21.9	10.8
Rhopalophthalmus sp.	Nov. 26, 2008	Sledge net	4.5–10.0 TL	S2	−28.0	10.4
Acetes						
Acetes sibogae	Nov. 26, 2008	Hand net	4.0–5.0 CL	S2	−26.4	10.4
	Nov. 26, 2008	Sledge net	3.5–4.5 CL	S2	−27.7	10.1
	Dec. 2, 2008	Hand net	3.5–4.5 CL	C1	−25.8	10.6
	Dec. 2, 2008	Hand net	2.5–3.0 CL	C1	−23.9	10.9
	Dec. 2, 2008	Hand net	1.5–2.5 CL	C2	−24.4	10.7
	Dec. 2, 2008	Hand net	2.5–3.0 CL	C3	−25.0	10.7
	Nov. 26, 2008	Sledge net	2.0–5.5 CL	R5	−25.8	11.1
	Nov. 26, 2008	Hand net	1.5–3.5 CL	R5	−26.5	10.0
	Dec. 2, 2008	Hand net	3.0–3.5 CL	R5	−26.7	11.0
	Nov. 28, 2007	Hand net	3.5–4.7 CL	R5	−25.2	11.1
	Nov. 26, 2008	Hand net	3.0–4.5 CL	S3	−27.4	10.0
Acetes japonicus	Nov. 25, 2008	Sledge net	3.5–4.5 CL	R1	−20.4	10.8
	Nov. 25, 2008	Sledge net	2.0–5.5 CL	OFF	−17.8	10.4
	Nov. 26, 2008	Hand net	2.8–4.0 CL	S2	−22.1	10.7
Acetes indicus	Nov. 25, 2008	Sledge net	5.0–7.5 CL	R1	−20.2	11.3
	Nov. 25, 2008	Sledge net	4.0–6.5 CL	R2	−21.4	11.0
	Nov. 25, 2008	Sledge net	5.0–7.0 CL	R3	−20.0	11.6
	Nov. 25, 2008	Sledge net	3.5–6.0 CL	R4	−21.8	11.0
	Nov. 26, 2008	Sledge net	3.5–6.5 CL	S1	−23.2	11.4

For sampling sites, see Fig. 1

CL carapace length, *TL* total length

[a] *Acanthomysis thailandica* + *Notacanthomysis hodgarti*

Table 2 Isotopic compositions of juvenile penaeids obtained by a trawl on Oct. 24, 2007

Sample	Carapace length (mm)	Sampling site	*n*	δ^{13}C, ‰ (mean ± SD)	δ^{15}N, ‰ (mean ± SD)
Penaeus merguiensis	13.1–15.7	R3	5	−20.2 ± 2.5	10.5 ± 0.6
	21.4–27.5	R4	5	−22.3 ± 0.7	11.2 ± 0.6
	13.3–20.4	S1	5	−23.7 ± 0.8	10.2 ± 0.8
	14.4–20.6	S2	5	−24.3 ± 0.7	10.6 ± 0.9
Metapenaeus brevicornis	13.1–15.7	R3	5	−21.7 ± 2.5	10.4 ± 0.4
	10.5–14.8	R4	5	−24.3 ± 0.8	10.3 ± 0.5
	8.0–15.3	S1	4	−24.9 ± 0.5	10.1 ± 0.7
	9.8–13.0	S2	4	−26.4 ± 1.0	9.5 ± 0.8

For sampling sites, see Fig. 1

n sample size, *SD* standard deviation

Table 3 Isotopic compositions of *Lutjanus johnii*

Sampling site	*n*	δ^{13}C, ‰ (mean ± SD)	δ^{15}N, ‰ (mean ± SD)	Total length (mm) range (average)	Wet weight (g) range (average)
R3	3	−19.8 ± 0.5	13.1 ± 0.2	126–191 (144)	35.4–125.6 (58.1)
R4	37	−20.8 ± 0.6	13.1 ± 0.5	55–144 (98)	2.5–50.5 (19.2)
C1	8	−21.5 ± 0.5	12.9 ± 0.2	77–123 (99)	6.9–28.8 (16.7)
S1	51	−22.5 ± 0.8	12.6 ± 0.4	37–161 (76)	0.7–68.6 (10.6)
S2	35	−24.0 ± 0.5	12.3 ± 0.4	53–185 (104)	2.3–123.0 (23.6)
S3	5	−25.9 ± 0.3	11.9 ± 0.4	89–127 (111)	11.4–57.8 (26.1)

For sampling sites, see Fig. 1

n sample size, *SD* standard deviation

John's snapper

A total of 139 young and juvenile John's snapper were collected by the otter trawl net (R3, R4, S1–S3) and set traps (C1) (Table 3). The range of bottom salinity where *L. johnii* were caught was 15.0–27.5, which was within the reported bottom salinity range (12.4–28.3) of *L. johnii* caught in the Sangga Besar River estuary and in the coastal areas by Kiso and Mahyam [17]. The ranges of TL and WW in *L. johnii* were 37–191 mm and 0.7–125.6 g, respectively. The largest number, 51, was obtained at the Selinsing River mouth (S1), followed by the upper river mouth stations of the Selinsing River (S2) and the Sangga Besar River (R4). Individuals of <50 mm TL were collected only at station S1.

The δ^{13}C and δ^{15}N values for John's snapper ranged from −26.4 to −19.4‰ and from 11.3 to 14.2‰, respectively. Although we could not examine the variability over an annual cycle because of the limited samples, there was no significant difference in δ^{13}C and δ^{15}N at station S1 between the dry season (July–September: $n = 18$) and wet season (October–November: $n = 33$) (t test, $P > 0.05$). The differences in the δ^{13}C isotope ratios between the sampling sites of John's snapper were remarkable, indicating that the δ^{13}C isotope ratios of John's snapper varied depending on the location of the sampling site (Table 3). There were significant differences in δ^{13}C among stations (ANOVA, $P < 0.001$; Tukey, $P < 0.005$), except between stations R3 and R4 and between R4 and C1 (Tukey, $P > 0.05$), showing a decreasing tendency from the lower estuarine stations of the Sangga Besar River (R3, R4) to the inner stations of the creek connecting the Sangga Besar River and the Selinsing River (C1) and to the upper Selinsing River (S1–S3).

Figure 2 shows the relationship between δ^{13}C values and TL for John's snapper. Individuals caught in the large Sangga Besar River (R3, R4) generally had δ^{13}C values of over −22.1‰ which was about the highest δ^{13}C value of their crustacean prey in the smaller Selinsing River and creek stations (Tables 1, 2). On the other hand, in the smaller Selinsing River, the fish had depleted δ^{13}C values

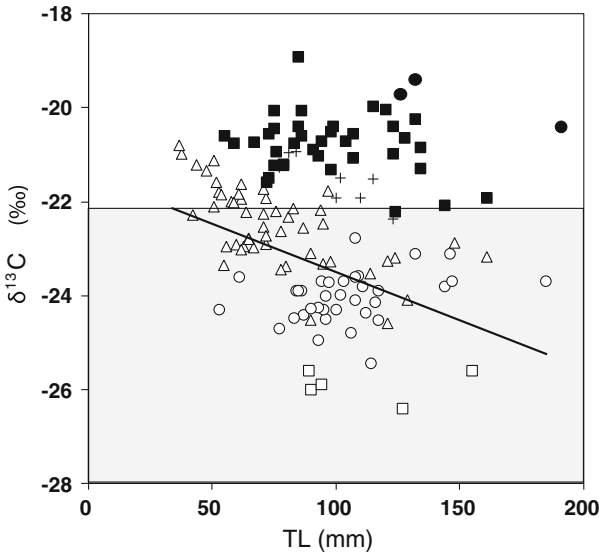

Fig. 2 Relationship between δ^{13}C values and total length (TL) of *Lutjanus johnii* collected from the Sangga Besar River (R3, R4), Selinsing River (S1–S3), and creek (C1) stations. Symbols indicating the stations: *filled circle* R3, *filled square* R4, *plus sign* C1, *open triangle* S1, *open circle* S2, *open square* S3. The *shaded area* shows the range of δ^{13}C values ($-28.0 < \delta^{13}\text{C} < -22.1$‰) for mangrove-associated prey animals from the upper Sangga Besar River (R5), the Selinsing River (S1–S3), and creek stations (C1–C3). TL and δ^{13}C for the samples collected from the Selinsing River (S1–S3) have a significant correlation: $\delta^{13}\text{C} = -0.0218\text{TL} - 21.4$ ($r^2 = 0.297$, $n = 91$, $P < 0.001$)

that were generally below −22.1‰, except for small individuals caught at the most downstream station S1. At station S1, many of the smaller individuals of TL <50 mm showed more enriched δ^{13}C values of greater than −22.1‰. Individuals collected from the Selinsing River (S1–S3) showed a tendency for δ^{13}C to decrease with increasing TL ($r^2 = 0.297$, $n = 91$, $P < 0.001$).

Discussion

Kiso and Mahyam [17] carried out sampling in the Sangga Besar River estuary and obtained the largest number of

juvenile John's snapper in the river mouth area, while we also obtained John's snapper from the smaller Selinsing River (S1–S3) and the creek station (C1) (Table 3). These results show that the juveniles migrate throughout large areas of the MMFR, not only in the main Sangga Besar River estuary but also into many small channels and creeks, and some juveniles migrate to the upstream area as far as 13 km from the river mouth (S3), although they are also distributed in areas near the river mouth.

Mysids and *Acetes* shrimps in the MMFR showed distinctive δ^{13}C signatures between habitats (Table 1) and could be divided into two groups: one group with generally enriched δ^{13}C values (*Acanthomysis* spp., *Acetes japonicus* and *A. indicus*) were closely associated with coastal and lower estuarine areas, while the other group (*Acetes sibogae*, *Mesopodopsis tenuipes* and *Rhopalophthalmus* sp.) with more depleted δ^{13}C values were captured mainly in the mangrove areas of the upper Sangga Besar River, Selinsing River, and the creek (mangrove-associated prey animals). Similarly, the δ^{13}C values of prawn species in the estuaries of MMFR are site dependent, as previously indicated by Chong et al. [19], showing a gradual seaward δ^{13}C enrichment in which the lower δ^{13}C values in *M. brevicornis* as compared to *P. merguiensis* were also consistent with the data of Chong et al. [19] (Table 2).

At the lower Sangga Besar River (R3, R4), the average δ^{13}C and δ^{15}N values of John's snapper were −20.8 to −19.8 and 13.1‰, respectively (Table 3), while the average δ^{13}C and δ^{15}N values of prey animals (*Acetes indicus*, *Penaeus merguiensis* and *Metapenaeus brevicornis*) at stations R3 and R4 ranged from −21.7 to −20.2‰ and 10.5 to 11.6‰, respectively (Tables 1, 2). These values showed approximately 1‰ enrichment in δ^{13}C and 2–3‰ enrichment in δ^{15}N as compared to the δ^{13}C and δ^{15}N ranges of its crustacean prey at the same locality. These results indicate that John's snapper fed mainly on these prey animals caught in the same locality, adopting the generally accepted trophic enrichment values (1.0‰ for δ^{13}C and 2–4‰ for δ^{15}N).

There were significant differences in the δ^{13}C values of the John's snapper found in the Sangga Besar River (R3, R4) and the Selinsing River (S1–S3). In the Selinsing River (S1–S3), the average values of John's snapper were −22.5 to −25.9‰, while the δ^{13}C values of prey animals were −22.1 to −28.0‰. These remarkable differences observed in the δ^{13}C values between sampling sites reflect the difference in food sources between the two habitat groups, and imply that the juveniles in the lower Sangga Besar River (R3, R4) do not migrate frequently to forage in the Selinsing River estuary. At station C1, which was located in the creek that interconnects the two river channels, John's snapper had intermediate δ^{13}C values (−22.4 to −20.9‰), which may indicate that the nearby Sangga Besar River estuary is included in their foraging area.

Because it could take several weeks to months for juveniles to acquire the signature of the food from a new habitat [13, 14, 16], juvenile John's snapper are likely to remain resident in the Selinsing River stations (S1–S3) until they grow larger and move offshore. However, small individuals of John's snapper (<5 cm TL) caught from the lower Selinsing River station (S1) had higher δ^{13}C values, which reflect the relatively high δ^{13}C values of the prey animals collected in the coastal and lower river stations. This suggests that the small fish had recently migrated into the lower estuary from the coastal area (Fig. 2). Studies have reported that small John's snapper of TL <5 cm were caught only in the coastal and river mouth areas [17]. Linear regression analysis based on TL–δ^{13}C data indicates that larger John's snapper collected from the Selinsing River (S1–S3) showed more depleted values with increasing TL (Fig. 2). These results suggest that the juveniles migrate from the coastal area into the mangrove area, and that some juveniles migrate upstream as far as 13 km from the river mouth, shifting their dependence from the coastal food web to the inner mangrove food web with their growth. Thereafter, after a period of residence in these areas, they migrate out of the estuary upon reaching a size of 20–22 cm TL [17]. Such ontogenetic shifts in habitat usage have also been reported in several species of Lutjanidae that migrate between seagrass beds and fringing mangrove forests [28, 29].

The MMFR is a major nursery area for shrimps, and shrimp densities are highest inside mangrove waterways [1]. Moreover, the predominant mysid species (*Mesopodopsis orientalis*) in the upstream mangrove formed aggregations close to the littoral zone [25]. It is considered that the existence of such prey animals in the upstream mangrove areas is one of the major factors that prompt the upstream migration of juvenile John's snapper. Although further information is needed because we could not examine the variability over an annual cycle in the δ^{13}C and δ^{15}N of both fish and prey animals, the findings of the present study have important implications for the fishery management of John's snapper. This study shows the importance of conserving sufficiently large single areas of mangrove with their complex interconnected waterways (including estuary, river, rivulet and creek)—exemplified by the large MMFR—as habitat and feeding grounds for highly migratory fish species and their diverse and abundant prey species.

Acknowledgments The authors gratefully acknowledge permission by the Department of Fisheries, Government of Malaysia, to carry out the research in the mangrove areas. Appreciation is also expressed to Mr. Mohammed Noordin, director of the Fisheries Research Institute, for providing various facilities for this study. The present study was

funded by the Japan International Research Center for Agricultural Sciences (JIRCAS) and in part by Grants-in-Aid for Scientific Research from the Japan Society for the Promotion of Science (nos. 22405030 and 20570101).

References

1. Chong VC (2007) Mangroves-fisheries linkages—the Malaysian perspective. Bull Mar Sci 80:755–772
2. Gillanders BM, Able KW, Brown JA, Eggleston DB, Sheridan PF (2003) Evidence of connectivity between juvenile and adult habitats for mobile marine fauna: an important component of nurseries. Mar Ecol Prog Ser 247:281–295
3. Blaber SJM (2007) Mangroves and fishes: issues of diversity, dependence, and dogma. Bull Mar Sci 80:457–472
4. Sarpedonti V, Chong VC (2008) Abundance and distribution of *Stolephorus baganensis* Hardenberg 1933 and *Thryssa kammalensis* (Bleeker 1849) larvae in relation to ontogeny and environmental factors in a Malaysian estuary. Trop Zool 21:195–208
5. Nagelkerken I, Dorenbosch M, Verberk WCEP, Cocheret de la Moriniere E, van der Velde G (2000) Importance of shallow-water biotopes of a Caribbean bay for juvenile coral reef fishes: patterns in biotope association, community structure and spatial distribution. Mar Ecol Prog Ser 202:175–192
6. Serafy JE, Faunce CH, Lorenz J (2003) Mangrove shoreline fishes of Biscayne Bay, Florida. Bull Mar Sci 72:161–180
7. Mumby PJ, Edwards AJ, Arias-Gonzalez JE, Lindeman KC, Blackwell PG, Gall A, Gorczynska MI, Harborne AR, Pescod CL, Renken H, Wabnitz CCC, Llewellyn G (2004) Mangroves enhance the biomass of coral reef fish communities in the Caribbean. Nature 427:533–536
8. Halpern BS (2004) Are mangroves a limiting resource for two coral reef fishes? Mar Ecol Prog Ser 272:93–98
9. Nakamura Y, Horinouchi M, Shibuno T, Tanaka Y, Miyajima T, Koike I, Kurokura H, Sano M (2008) Evidence of ontogenetic migration from mangroves to coral reefs by black-tail snapper *Lutjanus fulvus*: stable isotope approach. Mar Ecol Prog Ser 355:257–266
10. Aburto-Oropeza O, Dominguez-Guerrero I, Cota-Nieto J, Plomozo-Lugo T (2009) Recruitment and ontogenetic habitat shifts of the yellow snapper (*Lutjanus argentiventris*) in the Gulf of California. Mar Biol 156:2461–2472
11. Yamada H (2010) Age and growth during immature stages of the mangrove red snappers *Lutjanus argentimaculatus* in waters around Ishigaki Island, southern Japan. Fish Sci 76:445–450
12. Luo JG, Serafy JE, Sponaugle S, Teare PB, Kieckbusch D (2009) Movement of gray snapper *Lutjanus griseus* among subtropical seagrass, mangrove, and coral reef habitats. Mar Ecol Prog Ser 380:255–269
13. Hobson KA (1999) Tracing origins and migration of wildlife using stable isotopes: a review. Oecologia 120:314–326
14. Herzka SZ (2005) Assessing connectivity of estuarine fishes based on stable isotope ratio analysis. Estuar Coast Shelf Sci 64:58–69
15. Maruyama A, Yamada Y, Yuma M, Rusuwa B (2001) Stable nitrogen and carbon isotope ratios as migration tracers of landlocked goby, *Rhinogobius* sp. (the orange form), in the Lake Biwa water system. Ecol Res 16:697–703
16. Suzuki KW, Kasai A, Isoda T, Nakayama K, Tanaka M (2008) Distinctive stable isotope ratios in important zooplankton species in relation to estuarine salinity gradients: potential tracer of fish migration. Estuar Coast Shelf Sci 78:541–550
17. Kiso K, Mahyam MI (2003) Distribution and feeding habits of juvenile and young John's snapper *Lutjanus johnii* in the Matang mangrove estuary, west coast of Peninsular Malaysia. Fish Sci 69:563–568
18. Amy Then YH, Chong VC, Moh HH, Hanamura Y (2006) Size frequency abundance and feeding habits of young snappers (*Lutjanus* spp.) and groupers (*Epinephelus* spp.) in the Matang Mangrove Estuary, Malaysia. JIRCAS Working Rep 44:1–5
19. Chong VC, Low CB, Ichikawa T (2001) Contribution of mangrove detritus to juvenile prawn nutrition: a dual stable isotope study in a Malaysian mangrove forest. Mar Biol 138:77–86
20. Marguillier S, van der Velde G, Dehairs F, Hemminga MA, Rajagopal S (1997) Trophic relationships in an interlinked mangrove–seagrass ecosystem as traced by $\delta^{13}C$ and $\delta^{15}N$. Mar Ecol Prog Ser 151:115–121
21. Gan BK (1995) A working plan for the Matang Mangrove Forest Reserve, Perak, 4th Rev. State Forestry Department of Perak, Ipoh
22. Ahmad-Adnan N, Loneragan NR, Connolly RM (2002) Variability of, and the influence of environmental factors on, the recruitment of postlarval and juvenile *Penaeus merguiensis* in the Matang mangroves of Malaysia. Mar Biol 141:241–251
23. Sasekumar A, Chong VC, Singh H (1994) The physical and chemical characteristics of Matang mangrove waters. In: Proc 3rd ASEAN-Australia Symp Living Coastal Resources, Bangkok, Thailand, 16–20 May 1994, 2:446–453
24. Okamura K, Tanaka K, Siow R, Man A, Kodama M, Ichikawa T (2010) Spring tide hypoxia with relation to chemical properties of the sediments in the Matang Mangrove Estuary, Malaysia. JARQ 44:325–333
25. Hanamura Y, Siow R, Chee PE (2007) Abundance and spatio-temporal distribution of hyperbenthic crustaceans in the Merbok and Matang mangrove estuaries, Malaysia. JIRCAS Working Rep 56:35–41
26. Low CB, Chong VC, Lim LHS, Hayase S (1999) Prawn production of Matang and Dinding river mangroves: species distribution and seasonal recruitment. In: Kiso K, Choo PS (eds) Proceedings of Fourth Seminar on Productivity and Sustainable Utilization of Brackish Water Mangrove Ecosystems. Fisheries Research Institute, Penang, pp 89–101
27. Toyokawa M (2001) Investigation of the measurable range of sample amount and bias of the carbon and nitrogen stable isotope ratio analyzed by elemental analyzer–isotope ratio mass spectrometer system. Bull Nat Res Inst Fish Sci 16:27–37
28. Nagelkerken I, van der Velde G (2004) Relative importance of interlinked mangroves and seagrass beds as feeding habitats for juvenile reef fish on a Caribbean island. Mar Ecol Prog Ser 274:153–159
29. Faunce CH, Serafy JE (2007) Nearshore habitat use by gray snapper (*Lutjanus griseus*) and bluestriped grunt (*Haemulon sciurus*): environmental gradients and ontogenetic shifts. Bull Mar Sci 80:473–495

View publication stats