

# Food and feeding habits of the seahorses *Hippocampus spinosissimus* and *Hippocampus trimaculatus* (Malaysia)

M.Y. YIP<sup>1,2</sup>, A.C.O. LIM<sup>1,2,3</sup>, V.C. CHONG<sup>1,3</sup>, J.M. LAWSON<sup>4</sup> AND S.J. FOSTER<sup>4</sup>

<sup>1</sup>Faculty of Science, Institute of Biological Science, University of Malaya, Kuala Lumpur 50603, Malaysia, <sup>2</sup>Save Our Seahorses Malaysia, No. 2, Jalan 6/24, Seksyen 6, 46000 Petaling Jaya, Selangor Darul Ehsan, Malaysia, <sup>3</sup>Institute of Ocean & Earth Sciences, C308, Institute of Postgraduate Studies Building, University of Malaya, 50603 Kuala Lumpur, Malaysia, <sup>4</sup>Project Seahorse, Fisheries Centre, The University of British Columbia, 2204 Main Mall, Vancouver, BC V6T 1Z4, Canada

*Two seahorse species, Hippocampus spinosissimus and Hippocampus trimaculatus, sampled in east and west coastal waters of Peninsular Malaysia, fed mostly on crustacean prey; small caridean shrimps and amphipods as adults (both species), and copepods and larval meroplankton as juveniles (for H. trimaculatus only). The similar short relative gut length (~0.4) of both species is consistent with a carnivorous diet. Both species are considered specialists in prey selection, focusing on slow-moving epibenthic, hyperbenthic and canopy-dwelling crustaceans that dwell on the mud-sand seabed, or are associated with seagrass or mangrove areas. In this light, seahorses with their juveniles in shallow waters are vulnerable to coastal reclamation and development.*

**Keywords:** Syngnathidae, food habits, stomach content, crustacean prey, preponderance index, PCA, diet overlap, ontogenetic shift, relative gut length

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## INTRODUCTION

Seahorses are globally traded in large volumes in the aquarium and marine curio trade, and in traditional Chinese medicine (Lourie *et al.*, 2004). Demersal trawl fishing has also greatly affected their habitats (Baum *et al.*, 2003). As a result, wild seahorse populations appear to be declining (Perry *et al.*, 2010), prompting concern and their listing in the International Union for Conservation of Nature (IUCN) Red List of Threatened Species. They are now described as endangered or vulnerable (Baillie *et al.*, 2004).

Malaysia is among the tropical and subtropical regions where seahorse diversity and abundance is high (Lourie *et al.*, 2004), with at least 12 species of seahorses being found in the region along with 50 species of other related members in the family Syngnathidae (Lim *et al.*, 2011). Unfortunately, there are very few studies on Malaysian seahorses, particularly on their ecology. These include the first documented report on seahorse diversity and its distribution in Malaysia (Choo & Liew, 2003, 2004), later updated by Lim *et al.* (2011) and Lawson *et al.* (2014).

No studies have been conducted to determine the feeding habits of any species of seahorse or their relatives in Malaysian waters, but such studies have been conducted in other regions. However, studies conducted elsewhere have indicated that the type of food consumed by seahorses depends

on the species and habitat. In the Aegean Sea, *Hippocampus guttulatus* Cuvier, 1829 and *Hippocampus hippocampus* Linnaeus, 1758 were reported to commonly feed on decapod larvae, mysids, amphipods and other unidentified prey (Gurkan *et al.*, 2011). A study in north-eastern Brazil revealed that *Hippocampus reidi* Ginsburg, 1933 and *Hippocampus subelongatus* Castelnau, 1873 consumed cyclopoid copepods, amphipods and caridean shrimps (Castro *et al.*, 2008). Another study in New Zealand reported the dietary items of *Hippocampus abdominalis* Lesson, 1847 as being largely composed of crustaceans, especially amphipods, caridean shrimps and peracarids (Woods, 2002). The size of the seahorse's snout apparently determines the diet of the seahorse; for instance, the small snout of the lined seahorse, *Hippocampus erectus* Perry, 1810, is adapted to take small or slender-bodied amphipods living in seagrass and seaweed beds, such as *Ampithoe longimana* S.I. Smith, 1873, *Gammarus mucronatus* (Say, 1818) and *Caprella penantis* Leach, 1814 (Teixeira & Musick, 2001). Diets shift significantly as seahorses move from juvenile to adult life stages. *Hippocampus hippocampus*, *H. guttulatus*, *Hippocampus mohndikei* Bleaker, 1854a and *H. reidi* prefer smaller planktonic animals as juveniles, while adults consume larger pelagic prey (Kanou & Kohno, 2001; Castro *et al.*, 2008; Gurkan *et al.*, 2011).

Two species of seahorses, *Hippocampus spinosissimus* Weber, 1913 and *Hippocampus trimaculatus* Leach, 1814, are commonly found in Malaysian waters (Choo & Liew, 2003; Lawson *et al.*, 2014). They are commonly traded for traditional medicine and are vulnerable to trawl fishing and habitat destruction (Choo & Liew, 2005; Perry *et al.*, 2010). These seahorses are known to occur in variable bottom

Corresponding author:  
V.C. Chong  
Email: chong@um.edu.my

habitats from shallow (5 m) to deep waters of up to 100 m (Lourie *et al.*, 2004). Knowledge of feeding habits for these two species can help to identify the important prey and niches of seahorses. Such knowledge can also contribute to understanding the patchy distribution of seahorse populations (Foster & Vincent, 2004), as well as to improve seahorse breeding according to ontogenetic development.

In this study, the hedgehog seahorse *H. spinosissimus* and the three-spot seahorse *H. trimaculatus* were examined for their diet composition which was analysed for differences due to species, ontogenetic development (juvenile, small and large adult) and location (east and west coast of Peninsular Malaysia). We hypothesize that these factors influence diet variability in the seahorses.

## MATERIALS AND METHODS

### Seahorse collection sites

Seahorse samples were largely obtained from trawl bycatch from the east (Endau) and west (Langkawi, Teluk Bahang, Hutan Melintang, Kesang Laut, Pontian) coasts of Peninsular Malaysia (Figure 1). Bathymetric charts show that trawl vessels, which operate at least five nautical miles offshore by law, mostly fish in depths of 20–50 m on the west coast and 20–40 m on the east coast. Samples collected off Kesang Laut were seahorses collected from shallow waters

(5–10 m) by artisanal fishermen using drift nets and cast nets. Bottom substrates from the northern half of the west coast, from Langkawi to Hutan Melintang, consist of muddy substrates (largely  $<62.5 \mu\text{m}$  grain size) (Mohammad Shaari *et al.*, 1974). To the south of it, the sediment consist of sandy bottoms ( $>62.5 \mu\text{m}$  grain size) from Pangkor but progressively shifting to muddy or clayey ( $<4 \mu\text{m}$  grain size) substrates towards Pontian (Mohammad Shaari *et al.*, 1976). In contrast, the bottom substrates in the east coast consist of predominantly sand to the north of Endau, but the substrates progress to muddy sand to the south of it (Pathansali *et al.*, 1974).

### Sampling of specimens

Seahorse samples from the east coast were collected in March and April 2010. On the west coast, specimens at Hutan Melintang and Pontian landing sites were collected in July and August 2013, at Teluk Bahang and Kesang Laut in June 2013, and at Langkawi in January 2013. Collected specimens were immediately fixed in 10% formaldehyde.

### Seahorse measurements and gut content analysis

In the laboratory, all seahorses were sexed before morphometric measurements were made according to Lourie *et al.* (2004).



Fig. 1. Known fish landing sites in Peninsular Malaysia where seahorses were collected for present study. (1) Langkawi; (2) Penang; (3) Hutan Melintang; (4) Kesang laut; (5) Pontian; (6) Endau.

A total of 34 individuals of *Hippocampus spinosissimus* and 49 individuals of *Hippocampus trimaculatus* from the east coast were sampled for gut content analysis. Their heights ranged from 114.0–191.0 and 121.0–186.0 mm, respectively. On the west coast, a total of eight individuals of *H. spinosissimus* and 27 individuals of *H. trimaculatus* were sampled for gut content analysis with heights ranging from 63.0–145.0 and 121.0–186.0 mm, respectively.

A ventral incision along the keel line of the seahorse's abdomen and a horizontal incision from the anal fin towards the lateral trunk ridge were made to expose the digestive system. The start of the oesophagus and end of the rectum were cut and the entire digestive system was removed from the abdominal cavity. The total length of the gut was then measured (mm) from anterior to posterior. The 'stomach' was identified from a slight constriction of the foregut (at about 1/3 distance from oesophageal opening to the anus) indicative of the pyloric sphincter that separates it from the midgut or intestine. A small cut was made at the constriction, and the upper portion from it was slit open so as to extract the stomach contents for further analysis.

The entire stomach contents were gently washed out into a glass cavity block using a water jet from a glass pipette. Next, the stomach contents were pipetted out onto a gridded ( $10 \times 10$  one mm<sup>2</sup> grid) Sedgewick rafter cell for viewing and enumeration under a stereo light microscope. Prey items were identified to the lowest possible taxonomic level and quantified. Those prey items that were partially digested and impossible to identify were not included. Quantification of each food item was done using two methods following Hyslop (1980), namely, frequency of occurrence and percentage volume. Percentage frequency of occurrence (%FO) of each food item was computed based on the proportion of examined stomachs that contained the particular food item. Percentage volumetric composition (%VO) of each food item was estimated using the eye estimation method (Chong, 1977) by examining the number of grids occupied by each food item under the microscope.

## Data analysis

The height of the fish was used to separate the sampled seahorses into three developmental stages, juvenile, small adult and large adult. Although juveniles can be distinguished from adults based on the absence of a brood pouch in the male (Perante *et al.*, 1998; Wilson & Vincent, 1998), male *H. trimaculatus* was reported to reach sexual maturity size at 120 mm despite having developed a brood pouch earlier (80–90 mm) (Cai *et al.*, 1984). Lourie *et al.* (2004) also reported an average size of 120 mm for this species. Lawson *et al.* (2014) however reported 90.5 and 99.6 mm as the height at physical maturity ( $H_{t_m}$ ) and 121.8 and 123.2 mm as height at reproductive activity ( $H_{t_r}$ ) for *H. trimaculatus* and *H. spinosissimus*, respectively. Hence, for the purpose of the present study, 120 mm was considered the definite size of sexual maturity for the adult of both species. Small and large adults were arbitrarily grouped by first ordering their heights and dividing the number of individuals equally between small and large adults. This arbitrary division gave the following sizes for *H. spinosissimus*: small adults 120–156 mm, large adults 162–191 mm. For *H. trimaculatus*, small adults measured 121–158 mm, large adults 159–183 mm. Juvenile *H. trimaculatus* ranged from

63–110 mm height. No juvenile *H. spinosissimus* was sampled.

The relative gut length (RGL) was calculated by dividing the total gut length by the height (GL/H) of each fish. The preponderance index (PI) of the seahorse diet (Natarajan & Jhingran, 1961) was calculated as follows:  $PI = [(\%VO_i \times \%FO_i) / \sum (\%VO_i \times \%FO_i)] \times 100$ , where  $i$  indicates the  $i$ th food item. Schoener's index of diet overlap, given as  $C_{AB} = 1.0 - 0.5 (\sum |I_{A,i} - I_{B,i}|)$ , was calculated between species by developmental stage, where  $I$  is the preponderance index estimated from %VO and %FO of prey  $i$  in the diets of species A and B (Schoener, 1970).

Principal Component Analysis (PCA) of the preponderance index by species and developmental stage (juvenile, small and large adults) and location (west and east) was carried using CANOCO ver. 4.5 (Microcomputer Power, Ithaca, NY, USA). In CANOCO, Aitchison's (1990) log-ratio analysis of compositional data (PI) was selected by centring log-transformed data by samples as well as by species (Braak & Smilauer, 2002). A  $t$ -test using Statistica 10.0 software (StatSoft Inc., Tulsa, OK, USA) (Statsoft, 2001) was conducted on prey items to test for significant difference between species.

## RESULTS

### Fish height and relative gut length (GL/H ratio)

*Hippocampus spinosissimus* and *Hippocampus trimaculatus* for the east and west coast had significantly different mean heights ( $t(82) = 13.32$ ,  $P < 0.05$ ) tested according to developmental stage (Table 1). The mean height of adult *H. spinosissimus* (138 mm) was shorter than that of *H. trimaculatus* (147 mm). East coast specimens were generally larger than west coast specimens for both species. No juvenile of either species was sampled from the east coast, while 11 juvenile *H. trimaculatus* were sampled from the west coast. Out of these, six juveniles were from Kesang Laut in shallow waters, and all seahorses caught here were juveniles. No significant size difference existed between females and males for adult *H. spinosissimus* ( $t(30) = 0.92$ ,  $P > 0.05$ ), while *H. trimaculatus* males were larger than females ( $t(34) = -2.75$ ,  $P < 0.05$ ).

The mean RGLs of *H. spinosissimus* from the east coast and west coast were not significantly different (0.38), as were the RGLs of *H. trimaculatus* from the west coast (0.40) and east coast (0.36) ( $t(22) = 1.42$ ,  $P > 0.05$ ). When species were compared, the RGLs of *H. spinosissimus* and *H. trimaculatus* were not significantly different ( $t(73) = 0.42$ ,  $P > 0.05$ ).

### Prey's frequency of occurrence (%FO)

Empty gut content limited sample sizes for diet analysis to 29 individuals of *H. spinosissimus* and 36 individuals of *H. trimaculatus* from the east coast. Similarly, four individuals of *H. spinosissimus* and 16 individuals of *H. trimaculatus* from the west coast with filled stomachs were analysed.

A total of 14 and 16 prey taxa were recorded from the stomachs of sampled *H. spinosissimus* and *H. trimaculatus*, respectively. Stomachs of both *H. spinosissimus* and *H. trimaculatus* from the east coast contained the highest frequency of small caridean shrimps as prey food at 16%FO and 29%FO, respectively. Other prey items were present at fairly low frequencies (Table 2). About 12–18% of the stomachs examined

**Table 1.** Descriptive statistics of the height (mm) and gut length of *Hippocampus spinosissimus* and *Hippocampus trimaculatus* by location, developmental stage and sex (Peninsular Malaysia). GL/H = ratio of gut length to height.

Location, species developmental stage, sex	Number	Height		Length	Gut	GL/H
		Range	Mean $\pm$ SE (mm)			
East coast						
<i>Hippocampus spinosissimus</i>	24	120–191	156.50 $\pm$ 19.93	37–84	59.92 $\pm$ 11.75	0.38 $\pm$ 0.06
Small adult	11	120–152	138.27 $\pm$ 11.33	37–67	54.45 $\pm$ 9.51	0.39 $\pm$ 0.06
Large adult	13	159–191	171.95 $\pm$ 9.61	48–84	64.54 $\pm$ 11.77	0.38 $\pm$ 0.07
Sex						
Male	11	120–176	154.55 $\pm$ 19.80	40–69	59.18 $\pm$ 12.16	0.38 $\pm$ 0.05
Female	13	120–191	161.33 $\pm$ 20.07	37–84	60.54 $\pm$ 11.85	0.39 $\pm$ 0.08
<i>Hippocampus trimaculatus</i>	35	121–183	157.26 $\pm$ 14.55	15–89	56.94 $\pm$ 19.31	0.36 $\pm$ 0.12
Small adult	18	120–158	146.94 $\pm$ 12.17	15–77	56.50 $\pm$ 17.12	0.38 $\pm$ 0.11
Large adult	17	159–183	168.18 $\pm$ 6.79	35–89	57.41 $\pm$ 21.94	0.34 $\pm$ 0.13
Sex						
Male	19	148–183	163.21 $\pm$ 9.43	36–89	51.35 $\pm$ 18.53	0.32 $\pm$ 0.11
Female	16	121–176	150.19 $\pm$ 16.58	15–83	63.31 $\pm$ 18.81	0.42 $\pm$ 0.11
West coast						
<i>Hippocampus spinosissimus</i>	3	130–175	155.67 $\pm$ 23.16	53–72	59.67 $\pm$ 23.16	0.38 $\pm$ 0.07
Small adult	1	130	130.00	54.00	54.00	0.42
Large adult	2	162–175	168.50 $\pm$ 9.19	53–72	62.50 $\pm$ 13.44	0.37 $\pm$ 0.10
Sex						
Male	2	130–175	152.50 $\pm$ 31.82	53–54	53.50 $\pm$ 0.707	0.36 $\pm$ 0.08
Female	1	162	162.00	72.00	72.00	0.44
<i>Hippocampus trimaculatus</i>						
Juvenile	11	63–110	84.64 $\pm$ 14.95	11–46	34.09 $\pm$ 10.55	0.41 $\pm$ 0.14
Sex						
Male	3	89–99	93.0 $\pm$ 5.29	28–39	34.33 $\pm$ 5.69	0.37 $\pm$ 0.05
Female	8	63–110	81.5 $\pm$ 12.24	11–46	34.0 $\pm$ 12.24	0.43 $\pm$ 0.16

in both species contained unidentified food items which were either masticated or semi-digested. Also about 30% of stomachs examined invariably contained inorganic sediment in small amounts.

On the west coast, *H. spinosissimus* and *H. trimaculatus* consumed six and 13 prey taxa respectively, varying less than their east coast counterparts. *Hippocampus trimaculatus* from the west coast recorded the highest frequency of

**Table 2.** Stomach content of *Hippocampus spinosissimus* and *Hippocampus trimaculatus* from the East and West Coast, Peninsular Malaysia.

Prey taxa	East coast				West coast			
	<i>H. spinosissimus</i>		<i>H. trimaculatus</i>		<i>H. spinosissimus</i>		<i>H. trimaculatus</i>	
	%VO	%FO	%VO	%FO	%VO	%FO	%VO	%FO
Crustacea								
Caridea	28.89	16.47	36.02	29.41	–	–	4.06	8.24
Mysidae	3.60	2.35	5.46	5.88	32.60	2.35	–	–
Brachyura	3.89	4.71	5.47	5.88	–	–	12.90	3.53
Anomura	4.87	2.35	1.51	3.53	–	–	1.07	1.18
Amphipoda	3.65	4.71	11.01	10.59	34.66	3.53	4.43	5.88
Copepoda	–	–	–	–	2.65	1.18	68.55	12.94
Harpacticoida	1.45	2.35	–	–	–	–	0.07	1.18
Stomatopoda	1.36	1.18	1.75	2.35	–	–	–	–
Ostracoda	–	–	0.43	1.18	–	–	2.87	1.18
Gastropoda	0.39	1.18	1.58	5.88	–	–	0.14	1.18
Bivalvia	1.18	2.35	1.72	7.06	–	–	–	–
Polychaeta	0.08	1.18	0.05	1.18	–	–	–	–
Fish Larvae	–	–	0.42	1.18	–	–	–	–
Cephalopoda	0.31	1.18	–	–	–	–	–	–
Foraminifera	2.12	5.88	10.41	20.00	–	–	–	–
Unidentified egg	–	–	1.11	3.53	0.22	1.18	0.01	1.18
Unidentified food	19.81	17.65	6.95	12.94	25.00	1.18	5.35	4.71
Detritus	–	–	0.44	1.18	–	–	–	–
Sediment	28.39	28.24	15.67	30.59	4.87	1.18	0.55	2.35

%VO, Percentage Volume; %FO, Frequency of Occurrence.

copepods (13%FO) followed by amphipods (6%FO) and unidentified food (5%FO), while *H. spinosissimus* showed the highest frequency of amphipods (4%FO), mysids (2%FO), copepod (1%FO), unidentified eggs (1%FO) and unidentified food (1%FO).

**Percentage volume of food items (VO%)**

Caridean shrimps dominated the food composition of both *H. spinosissimus* and *H. trimaculatus* from the east coast comprising 29 and 36% of %VO respectively. *Hippocampus trimaculatus* also consumed amphipods (11%VO) and foraminiferans (10%VO). The west coast's *H. spinosissimus* showed higher ingestion of amphipods (35%VO) and mysids (33%VO) although with a large portion of unidentified food (25%VO). In contrast, the west coast's *H. trimaculatus* mostly consumed copepods (69%VO) and brachyuran larvae (13%VO).

**Principal component analysis of ingested stomach items**

The first PCA axis (horizontal) explains 55.3% of the total variability (eigenvalue = 0.553) while the second PCA axis (vertical) explains 27.3% of the total variability (eigenvalue = 0.272); thus, the first two PCA axes accounted for 82.6% of the total variability, providing a good representation of the data structure for seahorse diets.

Both species from the east and west coast displayed different food preferences for the type and amount of prey items consumed (Figure 2). Generally, the small and large adults of both species of seahorses from the east coast had similar diet composition. Adult *H. spinosissimus* and *H. trimaculatus* from the east coast consumed seven taxa of prey animals, namely Foraminifera, Ostracoda, Stomatopoda, Caridea, Bivalvia, Gastropoda and Brachyura. The prey animals were either small organisms, or were the larvae of large forms. Small or larval stages of caridean shrimps were the most abundantly consumed by the adult seahorses. On the west coast, adult *H. spinosissimus* showed preference for mysid shrimps and amphipods. Juvenile *H. trimaculatus* tended to feed more on planktonic prey such as copepods and ostracodes.

**Diet overlap**

The dietary preferences for both seahorse species obtained from the east coast overlapped with one another, for both small and large adults (Table 3). The measured dietary overlap ( $C_{AB}$ ) ranged from 57–94%. Diets were most similar for small and large adults of *H. trimaculatus* and *H. spinosissimus*, as indicated by a large diet overlap. In contrast, diet differences were most distinct between east and west coast seahorses ( $C_{AB} = 3–12\%$ ), even for the same species; and between juvenile and adult *T. trimaculatus* on the east coast ( $C_{AB} = 8–9\%$ ).

**DISCUSSION**

The study revealed that both species of seahorse from the east and west coast of the Malaysian peninsula consumed a wide range of prey organisms, with the widest variety of prey

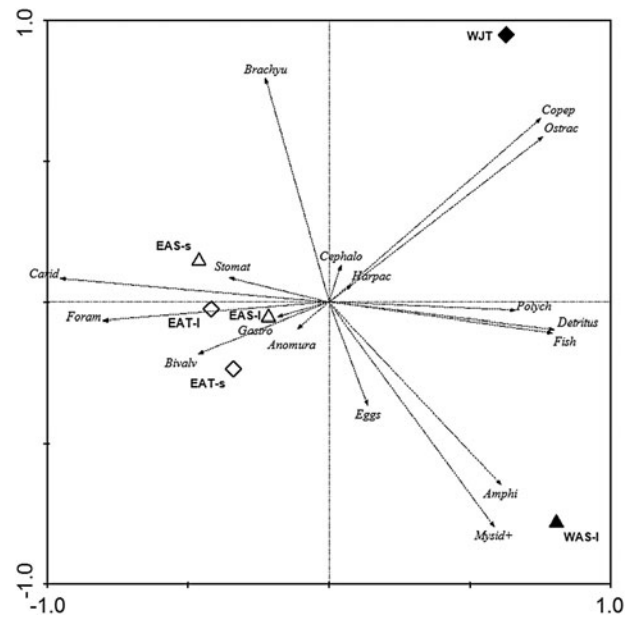


Fig. 2. Principal component analysis (PCA) of prey's preponderance index of *Hippocampus spinosissimus* (triangles) and *Hippocampus trimaculatus* (diamonds) from the east (unfilled symbols) and west coast (filled symbols) of Peninsular Malaysia. Arrows point to the gradient of importance (higher preponderance index) of food items: (Foram) Foraminifera; (Polych) Polychaeta; (Amphi) Amphipoda; (Harpac) Harpacticoida; (Copep) Copepoda; (Anomura) Anomuran larvae; (Stomat) Stomatopod larvae; (Carid) Caridea; (Mysid+) Mysidae & Acetes; (Brachyu) Brachyuran larvae; (Ostrac) Ostracoda; (Gastro) Gastropod larvae; (Cephalo) Cephalopod larvae; (Bivalv) Bivalve larvae; (Fish) Fish larvae; (Detritus) Detrital fragments; (Eggs) eggs of unidentified taxa. Symbol identifier: (E) =east coast, (W) =west coast, (A) =large adult (l) or small adult (s), (J) =juvenile, (T) =*Hippocampus trimaculatus*, (S) =*Hippocampus spinosissimus*.

items being found for east coast seahorse species. Crustaceans constituted the bulk of the prey food in both species (>60%VO). Adults of *Hippocampus spinosissimus* and *Hippocampus trimaculatus* favoured epibenthic, small caridean shrimps and amphipods, while juvenile *H. trimaculatus* preferred copepods and other planktonic forms such as meroplanktonic larvae of crustaceans.

Small seahorses with correspondingly small snout sizes may be restricted in their ability to capture larger and more mobile prey (Teixeira & Musick, 2001), explaining the differences we observed between juvenile and adult diets. A study done by Woods (2002) reported that smaller individuals of *Hippocampus abdominalis* (<13.75 mm) take in amphipods,

Table 3. Schoener's index of diet overlap for *Hippocampus spinosissimus* (HS) and *Hippocampus trimaculatus* (HT) and their developmental stage\*.

		East coast				West coast		
		HS		HT		HS	HT	
		s	l	s	l	l	j	
East	HS	s	-	57.12	79.80	79.98	6.95	12.00
		l	57.12	-	78.79	73.62	3.28	4.71
	HT	s	79.80	78.79	-	93.82	9.97	8.53
		l	79.98	73.62	93.82	-	8.22	8.86
West	HS	l	6.95	3.28	9.97	8.22	-	5.76
	HT	j	12.00	4.71	8.53	8.86	5.76	-

\*l, large adult; s, small adult; j, juvenile.



while adults (>13.8 mm) consumed mostly caridean shrimps. A similar dietary partitioning is found in *H. trimaculatus*. Since the consumed copepods and brachyuran zoal larvae are planktonic forms, we speculate that our collected juvenile *H. trimaculatus* came from shallow waters on the west coast; indeed, the shallow waters of Kesang Laut substantiate this. Rich diversity and abundance of copepods and other zooplankton have been recorded in shallow coastal waters (Chew & Chong, 2011), and around offshore islands (Chew *et al.*, 2008) in the Straits of Malacca. Since *H. trimaculatus* have been reported to inhabit shallow habitats (Lourie *et al.*, 2004) including around reefs (Masuda *et al.*, 1984) and muddy estuaries near mangroves (Kuitert & Tonozuka, 2001), presumably, the larger or adult seahorses would have moved on to offshore areas of greater depths. Hence, habitat shift in *H. trimaculatus* is accompanied by developmental changes and an ontogenetic shift in diet. Foster & Vincent (2004) reported that seahorses undergo ontogenetic shift in their diet as a result of morphological changes that affect the snout length and diameter. Species with longer snouts are more of a specialist feeder relying on particular prey, whereas species with a shorter snout tend to be generalist with a larger prey range (Kendrick & Hyndes, 2005; Van Wassenbergh *et al.*, 2011).

Consumed food items confirm the bottom feeding habit and behaviour of the seahorses. Interestingly, a small intact juvenile cephalopod was found in the stomach of an east coast *H. spinosissimus* specimen. Cephalopods like squids and cuttlefish lay egg capsules attached to bottom substrates including seaweeds, gorgonids, shells, rocks and sandstones, or even insert these capsules into the muddy substrate (Reid *et al.*, 2005; Chembian & Saleena, 2011). Their newly hatched paralarvae are planktonic but may remain closer to the sea bottom for some time (Nabhitabhata, 1996). Seahorses have been observed to forage by orally pumping forceful jets of water onto the sediment substratum thereby suspending their prey in the water column (Foster & Vincent, 2004). This is followed by oral suction of the

suspended prey along with the intake of water and suspended sediment. This feeding behaviour thus explains the ubiquitous presence of fine sediment in the seahorse stomachs, particularly the adults.

Various works have reported that relative gut length (RGL) is correlated with the feeding habits of fish, where carnivores are found to have a short RGL (1 or less) and herbivores or detritivores a longer RGL (>3) (Al-Hussaini, 1947; Horn, 1989; Kramer & Bryant, 1995). The similar RGLs (<0.4) of both species of seahorses in the present finding supports the assumption that these fish are carnivorous. The RGL also supports the need for constant prey consumption (Foster & Vincent, 2004), in contrast to studies that reported a digestion time of approximately 1.3–1.5 h for *H. trimaculatus* in captivity (Murugan *et al.*, 2009).

The present study identifies clear differences in seahorse diet depending on location. Therefore, observed differences in diet are likely due to differences in site-specific resources and availability. The substrate on the northern side of the west coast is rather homogeneous, consisting largely of mud although grain size becomes progressively coarser (sandy) towards Pangkor Island (Mohammad Shaari *et al.*, 1974). Elsewhere on the west coast, the bottom substrate is similarly muddy (Mohammad Shaari *et al.*, 1976). On the other hand, the east coast area is characterized by a seabed overlain with mud, sandy mud, muddy sand to sandy substrates, strewn with patches of clay-mud, octocorals and giant cup sponges (Pathansali *et al.*, 1974; Higashikawa *et al.*, 1986). Thus, the higher number of prey taxa that were consumed by east coast seahorses may be a reflection of their more heterogeneous habitat. The substrate type also explains the higher composition of sediment and associated foraminiferan fauna found in the stomachs of both *H. spinosissimus* and *H. trimaculatus* from the east coast. Diet variability in these seahorses may be the result of location and ontogenetic development. However, there appears not to be a distinct difference between the diet of the adults (>70% diet overlap).

**Table 4.** Comparison of the dietary habits of worldwide seahorse species.

Species	$H_{\max}$ (cm)*	Authority	Place	Main dietary taxa	Authority
<i>Hippocampus breviceps</i>	10.0	Foster & Vincent (2004)	Port Fremantle, Australia	Amphipoda; Copepoda; Mysidaceae	Kendrick & Hyndes (2005)
<i>Hippocampus whitei</i>	13.0	Foster & Vincent (2004)	Port Hacking, New South Wales, Australia	Amphipoda	Burchmore <i>et al.</i> (1984)
<i>Hippocampus hippocampus</i>	15.0	Foster & Vincent (2004)	Aegean Sea, Turkey	Amphipoda; Mysidaceae	Gurkan <i>et al.</i> (2011)
<i>Hippocampus patagonicus</i>	17.0	Storero & González (2008)	Santiago Bay, Patagonia, Argentina	Amphipoda; Brachyura larvae	Storero & González (2008)
<i>Hippocampus reidi</i>	17.5	Foster & Vincent (2004)	Mamanguape, Paraiba, Brazil	Nematoda; Copepoda	Castro <i>et al.</i> (2008)
<i>Hippocampus guttulatus</i>	18.0	Foster & Vincent (2004)	Aegean Sea, Turkey	Mysidaceae; Decapoda larvae	Gurkan <i>et al.</i> (2011)
<i>Hippocampus trimaculatus</i>	18.3	This study	Coastal waters, Peninsular Malaysia	Caridean shrimps; Amphipoda; Copepoda; Brachyura larvae	This study
<i>Hippocampus erectus</i>	19.0	Foster & Vincent (2004)	Chesapeake Bay, USA	Amphipoda	Teixeira & Musick (2001)
<i>Hippocampus spinosissimus</i>	19.1	This study	Coastal waters, Peninsular Malaysia	Caridean shrimps; Amphipoda; Mysidaceae	This study
<i>Hippocampus abdominalis</i>	35.0	Foster & Vincent (2004)	Wellington Harbour, New Zealand	Amphipoda; Caridean shrimps; Mysidaceae	Woods (2002)

\*Maximum body height.

A comparison of the diet of *H. spinosissimus* and *H. trimaculatus* with other species of seahorses worldwide indicates a remarkably consistent diet of largely amphipods, mysids and caridean shrimps, with crustaceans dominating prey items (Table 4). Smaller seahorses (<15 cm) all consume amphipods, while the larger seahorses (>18 cm) were found to also consume caridean shrimps. Seahorses therefore appear to be specialists in their prey selection, focusing on slow-moving epibenthic, hyperbenthic or canopy-dwelling crustaceans. These crustaceans inhabit mud-sand bottoms and habitats in, or associated with mudflat, seagrass or mangroves areas (Zimmerman *et al.*, 1979; Gore *et al.*, 1981; Matheson *et al.*, 1999; Hanamura *et al.*, 2008; Ramarn *et al.*, 2014). This suggests that juveniles of deeper water adults may be vulnerable to impacts from development, meaning that deep-water refugia may not be enough to protect these seahorses from the impacts of sea-filling (land reclamation) and development, which increasingly threaten these habitats in most tropical regions.

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**Correspondence should be addressed to:**

V.C. Chong  
 Faculty of Science, Institute of Biological Science, University of  
 Malaya, Kuala Lumpur 50603, Malaysia  
 email: [chong@um.edu.my](mailto:chong@um.edu.my)