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The distribution and habitat preferences of the zebrafish in Bangladesh

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The present study presents the results of a survey of a wide range of water bodies in Bangladesh to identify and describe *Danio rerio* habitat preferences. Field-based experiments were conducted to determine the vertical distribution of *D. rerio* in the water column, together with five other fish species commonly found in association with *D. rerio*. *Danio rerio* is a floodplain rather than riverine species, being most abundant in shallow lakes, ponds and ditches, typically in open locations with relatively clear water and abundant vegetation at the margins. It is commonly found in water bodies with a connection to rice cultivation and is more common in the north of Bangladesh than the south. *Danio rerio* occupies the whole of the water column and is observed as frequently in open water as amongst aquatic vegetation.

Key words: behaviour; Cyprinidae; *Danio rerio*; ecology; model species.

INTRODUCTION

The zebrafish, *Danio rerio* (Hamilton) is a small (*c*. 30 mm standard length, L_S) cyprinid fish, first described in the Ganges delta. It is a popular aquarium species and is a model organism of enormous significance in developmental genetics (Kimmel, 1989; Granato & Nüsslein-Volhard, 1996; Grunwald & Eisen, 2002). Over the next decade, *D. rerio* is expected to play a fundamental role in biomedical research. The zebrafish genome project, based at the Sanger Institute in Cambridge, began in 2001 and will shortly be completed (www.sanger.ac.uk). Over 300 laboratories worldwide now routinely use *D. rerio* in fundamental and applied research, in studies ranging from understanding organ function to the action of disease, and the importance and the probable impact of this animal on biological research cannot be overstated (Gerlai *et al.*,

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2000; Darland & Dowling, 2001; Nüsslein-Volhard & Dahm, 2002; Tropepe & Sive, 2003; Wright *et al.*, 2003).

Remarkably, little is known about the natural biology or behaviour of *D. rerio*, and few studies have been conducted on natural populations. The purpose of this study was to identify and describe the habitat preferences of *D. rerio,* assess its relative abundance within local fish assemblages, and observe its interactions with other fish species as a precursor to more detailed research into its natural ecology and behaviour.

The natural range of *D. rerio* is centred around the Ganges and Brahmaputra River basins in north-eastern India, Bangladesh, Nepal and northern Myanmar (Laale, 1977; Barman, 1991), though it has also been reported from southern India (collection of the Swedish Natural History Museum), and Jammu in Kashmir (Dutta, 1993). Although wild *D. rerio* have been collected previously for taxonomic (Pritchard, 2001; http://artedi.nrm.se/nrmfish/) and population genetic analysis (Gratton *et al.*, 2004; Wright, 2004), no systematic attempt has been made to describe its natural ecology, other than brief descriptions in fish identification guides. *Danio rerio* is typically described as inhabiting slow-moving or standing water bodies, particularly rice fields (Sterba, 1962; Talwar & Jhingran, 1991; Jayaram, 1999), although it is also reported as inhabiting rivers and hill streams (Daniels, 2002). The present study concentrated on its habitat preferences in the Ganges and Brahmaputra drainages in Bangladesh.

Bangladesh has a monsoon climate with wide seasonal variation in the extent of freshwater habitats. Even in the dry season there is a wide range of aquatic environments. The main river systems, the Ganges and Brahmaputra, rise in the Himalayas, and run through low-lying areas known as 'hoars', which flood extensively during the monsoon months from June to September. Rivers in this region continually change their course over the floodplain, leaving networks of oxbow lakes and blind channels or 'beels', which may have seasonal connections to the main river. In addition, the country has extensive areas of man-made lakes, ponds and irrigation channels, constructed for fish and rice cultivation. The present study involved sampling across the full range of these habitats. In addition, an experiment was conducted to observe its vertical distribution in the water column, under semi-natural conditions, together with five other fish species with which it was commonly found.

MATERIALS AND METHODS

GENERAL DISTRIBUTION

Sample sites

A survey was conducted during the dry season, in January 2005. This period was chosen to facilitate access to a wide range of sites and identify discrete populations. Surveys were conducted in two areas, Khulna District in the south of Bangladesh in the River Ganges drainage (GPS reading 22° N, 90° E), and Mymensingh District, to the north, in the Brahmaputra drainage (GPS reading 24° N, 90° E). A total of 23 sites were sampled in January 2005. In addition, a further three sites were sampled in Mymensingh District in August 2005 during the monsoon season. Sites sampled in August were temporary habitats, connected to inundated areas and representing additional habitat types unique to the monsoon season. A description of all study sites is summarized in Table I.

TABLE I. List of sampling sites showing location, basic site description and approximate size of the area in which sampling was conducted

Site	Location	Description	Approximate area (m)
	Khulna District		
1	Khulna University Campus	Ditch surrounding a series of ponds	3 × 90
2	Bagerhat	Large artificial lake	600 × 600
3	Khulna University Campus	Seasonally isolated arm of a canal	10 × 500
4	Khulna University Campus	Small pond treated with lime and rotenone	8 × 10
5	Khulna University Campus	Semi-natural pond	10 × 15
6	Khulna University Campus	Natural ponds	4 × 8
7	Golamari	Village pond near Golamari Bridge	25 × 50
8	Bhatiaghata River	A tributary of the Ganges	150 (width)
9	Golamari	Isolated channel of the Golamari River	200 × 1500
10	Khulna University Campus	Large artificial pond	15 × 30
11	Golamari	Isolated channel adjacent to site 9	100 (width)
12	Golamari	Established irrigation channel	10 (width)
13	Golamari	Isolated channel	100 (width)
	Mymensingh District		
14	Sutiakhali	Pond used for pearl mussel culture	17 × 20
15	Sutiakhali	Shallow cultivated pond connected to paddy field	30 × 50
16	Sutiakhali	Isolated pond	10 × 12
17	Sutiakhali	Isolated pond	10 × 10
18	Sutiakhali	Isolated pond	10 × 12
19	Brahmaputra River	An old arm of the Brahmaputra River	150 (width)
20	Dhuno River, Balikhala	A large river in a low-lying 'hoar' region	200 (width)
21	Haibatnagar	Old fish farm	30 × 30
22	Bangladesh Agricultural University field station	Semi-natural pond	150 × 150
23	Bangladesh Agricultural University field station	Ditch connecting to paddy fields	10 (width)
	Further sites sampled in August		
24	Bangladesh Agricultural University field station	Small semi-natural pond	8 × 15
25	Bangladesh Agricultural University field station	Channel adjacent to campus, feeding into field station	8 (width)
26	Brahmaputra River	A creek flowing out from the main river channel into extensive flooded area	15 (width)

Sampling procedure

Site position was recorded using a GPS handset. Sampling was conducted using a fine mesh seine measuring 5·0 m × 1·3 m, with mesh-size varying from 2 to 7 mm. Between three and five hauls over a distance of 5 m were taken at each site and all fishes captured were retained live. A representative sub-sample of the fishes caught at each site was retained and fixed in 4% formalin. Specimens of any newly encountered species were also preserved in 4% formalin. All species collected were identified (Sterba, 1962; Talwar & Jhingran, 1991; Roberts, 1994, 1998; Jayaram, 1999; Daniels, 2002) and specimens in sub-samples were counted and their L_S measured to the nearest 1 mm.

At each site the following environmental variables were recorded: geographic location, connection with other water bodies, dimensions, mean depth, Secchi depth, temperature, pH, ammonia or ammonium, salinity and substratum type. Specimens of aquatic vegetation were collected, pressed and later identified using the botanical collection in the herbarium of the University of Leicester.

VERTICAL DISTRIBUTION

An experiment was conducted at the Fisheries Field Laboratory of the Faculty of Fisheries at the Bangladesh Agricultural University, Mymensingh, in August 2005. Experiments were conducted during the monsoon season, which corresponds with the spawning season of *D. rerio* (Daniels, 2002). Experiments were performed in glass-sided outdoor aquaria (246 × 30 × 84 cm) filled with pond water to a depth of 60 cm. Environmental conditions (light, temperature, rainfall and water quality) were identical to those in the surrounding ponds from which experimental fish were collected (sample sites 22, 23, 24 and 25). Water temperature ranged from 29 to 33° C. During mid-afternoon, rice-straw screens (108 × 208 cm) were placed on top of the aquaria to provide shade.

Four species that were shown from spatial distribution surveys to commonly occur with *D. rerio* were used in the experiment: *Aplocheilus panchax* (Hamilton), *Esomus danricus* (Hamilton), *Parambassis lala* (Hamilton) and *Colisa lalia* (Hamilton). In addition, the study used *Oreochromis niloticus* L., a widely introduced species in Bangladesh. All fishes were collected from among the same group of ponds and were randomly assigned to test aquaria, with no specific sex ratio being used. Seven replicates were conducted over 2 days. Each aquarium was stocked with 10 *D. rerio*, five *A. panchax*, six *E. danricus*, five *P. lala*, three *C. lalia* and five *O. niloticus*, the numbers of fishes being chosen to match the mean relative abundance of each species from previous surveys. The mean ± S.D. L_S (mm) of fish used in the experiment were as follows: *D. rerio,* 24·4 ± 3·6; *A. panchax,* 29·4 ± 5·4; *C. lalia,* 34·1 ± 4·3; *P. lala,* 21·3 ± 2·6; *E. danricus,* 34·5 ± 5·1; *O. niloticus,* 28·4 ± 6·1.

Two bundles of aquatic plants were placed in one half of each aquarium and four plastic boxes (150 × 100 × 40 mm), filled with gravel, were placed on the bottom, *c.* 50 cm apart. Wild and domesticated *D. rerio* readily use boxes of gravel for oviposition; the eggs fall between the gravel, which prevents egg cannibalism. Three vertical sections (top, middle and bottom) were marked on each aquarium with a horizontal line at 20 and 40 cm. Fishes were fed three times each day with zooplankton collected from adjacent ponds, together with commercial dry food.

Behavioural observations of fishes in each aquarium were conducted twice; at dawn (*c.* 0530 hours) when *D. rerio* spawn, and after spawning between 1100 and 1200 hours. During each observation, the position in the aquarium of every individual of each species was recorded five times. There were six possible positions: top, vegetated; top, open; middle, vegetated; middle, open; bottom, vegetated; bottom, open. The total duration of each observation period was 15 min, the position of every individual of each species being recorded every 3 min. In addition, any aggressive interactions among species and feeding in spawning boxes were recorded. After the second observation period, the spawning boxes were checked for the presence of *D. rerio* eggs. At the end of the experiment, all fishes were removed, their L_S measured to the nearest mm, and returned to the ponds from which they had been collected.

DATA ANALYSES

All data were tested for normality using a Kolmogorov–Smirnov test and for equality of variance using a Bartlett's test. Where data did not respond to standard transformations, they were ranked. A principal component analysis (PCA) was used to identify relationships among the following variables at each sample site: locality; flow; depth; Secchi depth; substratum; presence of vegetation; salinity; pH; ammonia or ammonium; presence of predators; connection with rice cultivation. The correlation between each of the first two principal components and *D. rerio* $\log_{10}$ abundance was then examined.

Paired *t*-tests were used to compare the distribution of each species between the two observation periods in the vertical distribution experiment. A log-linear contingency table (Everitt, 1977) was used to test for the effects of height in the water column and vegetation cover on the distribution of each species in aquaria. In all but one species, there was no significant difference in distribution between the two time periods, so a two-way contingency table was used. In the case of the remaining species, a three-way contingency table was used, taking account of time period in addition to height and vegetation cover. The analysis of each species separately made the untested assumption that their distribution was independent of the rest of the assemblage. The aim, however, was to test whether apparent patterns of distribution of each species were statistically significant and no inferences are made about the basis for the observed distributions.

Hurlbert's measure of niche breadth was also calculated for each species in the vertical distribution experiment. This is a measure of the uniformity of distribution of individuals among resource states or habitats and is given by the equation: $B' = \{\Sigma[(pj^2)(aj)^{-1}]\}^{-1}$ where p = proportion of individuals, j = resource states and a = available resources. Thus, the smaller the number, the narrower the niche breadth and the more specialized the species.

RESULTS

GENERAL DISTRIBUTION

Danio rerio was found in nine out of the 26 sites sampled, two in Khulna District and seven in Mymensingh District. The environmental variables recorded for each sampling site, together with $\log_{10}$ abundance of *D. rerio,* are shown in Table II. *Danio rerio* was more abundant in the north (Mymensingh) and was not found in any of the rivers sampled. This result was reflected in the PCA. The first two principal components accounted for 32 and 18% of the variance, respectively (Fig. 1). The first component (PC1, loadings in parentheses) related to depth (−0·47), flow (−0·46), substratum (−0·46), transparency (−0·42) and the presence of predators (−0·39). The second component (PC2) related to a connection to water bodies associated with rice cultivation (−0·58), the presence of vegetation (−0·48) and locality (−0·34). There was no correlation between PC1 and the occurrence of *D. rerio* across all sampling sites (Pearson correlation, $r_p = 0{\cdot}29$, $n = 26$, $P > 0{\cdot}05$). In the nine sites where *D. rerio* was found, however, PC1 significantly correlated with *D. rerio* $\log_{10}$ abundance (Pearson correlation, $r_p = 0{\cdot}85$, $n = 9$, $P < 0{\cdot}01$). PC2 was negatively correlated with the $\log_{10}$ abundance of *D. rerio* across all sampling sites (Pearson correlation, $r_p = 10{\cdot}46$, $n = 26$, $P < 0{\cdot}05$).

A total of 48 fish species from 16 families were captured across the 26 sites (Table III). Cyprinids were the dominant group in the assemblage, with *D. rerio* among the most abundant species. *Parambassis lala, Chanda nama* Hamilton, *E. danricus*, *A. panchax, Glossogobius giuris* (Hamilton), *Oryzias carnaticus*

TABLE II. Environmental variables for each sampling site: Khulna District: 1, ditch; 2, large artificial lake; 3, seasonally isolated arm of a canal; 4, pond treated with lime and rotenone; 5, semi-natural pond; 6, natural ponds; 7, village pond; 8, Bhatiaghata River; 9, isolated river channel; 10, artificial pond; 11, isolated river channel; 12, irrigation channel; 13, isolated river channel. Mymensingh District: 14, cultivated pond; 15, cultivated pond; 16, isolated pond; 17, isolated pond; 18, isolated pond; 19, Brahmaputra River; 20, Dhuno River; 21, old fish farm; 22, semi-natural pond; 23, ditch; 24, small semi-natural pond; 25, channel; 26, creek

Site	*Danio rerio* $\log_{10}$ abundance	Locality	Flow	Depth (cm)	Secchi depth (cm)	Substratum	Vegetation	Temperature (° C)	pH	Ammonia or ammonium (mg l^{-1})	Salinity	Connection with paddy	Predators
1	1	Khulna	Still	80	51	Mud		20	8	0·01	0		Yes
2	0	Khulna	Still	100	92	Mud	Yes	22	8	0·01	0		Yes
3	0	Khulna	Still	80	16	Mud		18·5	8	0·1	0·4		
4	0	Khulna	Still	73	37	Mud		20	7·4	0·1	0·2		Yes
5	0	Khulna	Still	50	50	Mud	Yes	17·5	7·6	0·1	0·2		
6	0	Khulna	Still	35	35	Mud	Yes	20	8	0·1	0	Yes	Yes
7	0	Khulna	Still	85	37	Mud		18	7·4	0·1	0·8		
8	0	Khulna	Running	125	75	Mud		21·5	8	0·1	0·6		Yes
9	1	Khulna	Still	50	19	Mud	Yes	20	8	0·1	0		
10	0	Khulna	Still	71	64	Mud	Yes	20·5	8	0·1	0		
11	0	Khulna	Still	73	52	Mud	Yes	20·3	8	1	0·4		
12	0	Khulna	Still	50	44	Mud		19	8	0·1	0·4		
13	0	Khulna	Still	76	23	Mud		22	8	0·1	0·2		
14	0	Mymensingh	Still	30	16	Mud	Yes	20	7·6	0·01	0·6		
15	3	Mymensingh	Still	15	15	Mud	Yes	20·5	8	0·01	0·6	Yes	
16	2	Mymensingh	Still	40	15	Mud		19·5	8	0·01	0·6		
17	2	Mymensingh	Still	103	30	Mud		16·5	8	0·01	0·6		
18	0	Mymensingh	Still	25	32	Mud		19	8	0·01	0·6		
19	0	Mymensingh	Running	200	197	Sandy Mud	Yes	21	8	0·01	0·4		Yes
20	0	Mymensingh	Running	130	46	Sandy Mud	Yes	21	7·6	0·01	0·4		Yes
21	0	Mymensingh	Still	80	12	Mud		22	8	0·01	0·4		
22	1	Mymensingh	Still	96	31	Mud	Yes	21	8	0·01	0·4		
23	2	Mymensingh	Still	50	50	Mud	Yes	23	8	0·01	0·4	Yes	
24	2	Mymensingh	Still	65	15	Mud	Yes	33	8	0·01	0		
25	2	Mymensingh	Still	75	15	Mud	Yes	33	8	0·01	0	Yes	
26	0	Mymensingh	Running	120	15	Mud	Yes	30	8	0·01	0	Yes	Yes

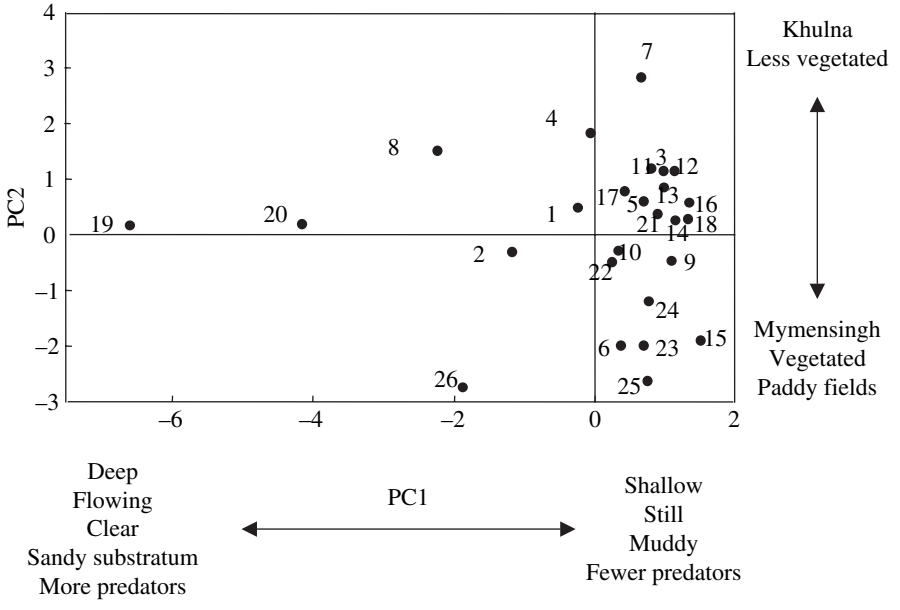

FIG. 1. Scores for each sampling site (see Table II) on the two principal components.

(Jerdon) together with *Puntius* spp. and *Colisa* spp., while less abundant, were more widely distributed than *D. rerio*. Some sites had been used for aquaculture and contained commercial species, such as *Aristichthys nobilis* (Richardson), *Catla catla* (Hamilton), *Cirrhinus cirrhosus* (Bloch), *Hypothalmichthys molitrix* (Valenciennes), *Labeo rohita* (Hamilton) and *O. niloticus*. The main predatory taxa captured were *Channa* spp. and *Xenentodon cancila* (Hamilton).

VERTICAL DISTRIBUTION

The only species to show a significant difference in its distribution between dawn and midday observation periods was *A. panchax* (paired *t*-test, square-root transformed data: d.f. = 12, $P < 0{\cdot}001$). In both time periods, there was a significant effect of height in the water column on fish distribution but not of vegetation cover (χ^2, d.f. = 7, $P < 0{\cdot}01$; Fig. 2).

There was no significant difference in the distribution of the other five species between the two observation periods ($P > 0{\cdot}05$). There were no significant effects of either height in the water column or vegetation cover on the distribution of *D. rerio* (χ^2, d.f. = 2, $P > 0{\cdot}05$). Similarly, *E. danricus* showed an even distribution across all sections of the aquaria (χ^2, d.f. = 2, $P > 0{\cdot}05$). For *C. lalia* there was a significant effect of vegetation, but not of height in the water column and there was no interaction (χ^2, d.f. = 2, $P < 0{\cdot}001$). In contrast, for *P. lala* there was a significant effect of height in the water column (χ^2, d.f. = 2, $P < 0{\cdot}01$) but not of vegetation and there was no interaction. For *O. niloticus* there was a significant effect of height in the water column (χ^2, d.f. = 2, $P < 0{\cdot}05$) but not of vegetation cover and there was no interaction.

TABLE III. Fish species recorded in each sampling site (see Table II)

	Khulna District													Mymensingh District											
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26
Ambassidae																										
Chanda nama		x	x			x	x	x	x	x		x	x									x			x	x
Parambassis lala	x		x		x	x		x	x	x		x	x			x	x				x	x	x		x	x
Anabantidae																										
Anabas testudineus	x											x														
Badis badis	x	x	x			x							x									x	x			x
Channa punctatus	x	x		x								x														x
Channa orientalis																			x							x
Colisa chun										x															x	
Colisa lalia	x	x	x		x	x			x	x	x	x	x		x	x	x					x	x		x	
Colisa fasciatus	x																									
Anguillidae																										
Anguilla bengalensis	x																									
Aplocheilidae																										
Aplocheilus panchax	x	x	x		x	x			x	x	x	x	x	x	x		x					x	x	x	x	x
Oryzias carnaticus	x		x	x	x	x	x	x	x	x	x	x	x						x		x					x
Bagridae																										
Mystus vittatus																			x							x
Balitoridae																										
Acanthocobitis botia																			x							x
Nemacheilus sp.																										x
Belonidae																										
Xenedonton cancila						x		x	x										x	x						

TABLE III. Continued

	Khulna District													Mymensingh District											
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26
Cichlidae																										
Oreochromis niloticus				x											x									x		
Clupeidae																										
Corica soborna									x																	
Cobitidae																										
Botia rostrata																			x							x
Lepidocephalichthys guntea																										x
Neoeucirrhichthys maydelli																			x							
Cyprinidae																										
Amblypharyngodon mola	x		x											x		x										x
Aristichthys nobilis	x																				x					
Catla catla				x																						
Cirrhinus cirrhosus	x																									
Danio rerio	x								x						x	x	x					x	x	x	x	
Devario devario																										x
Esomus danricus	x		x	x	x	x	x		x	x	x	x	x	x	x	x	x				x	x		x	x	x
Hypophthalmichthys molitrix	x																									
Labeo calbasu																										x
Labeo rohita				x															x							x
Puntius chola	x											x			x											x
Puntius conchonius											x	x	x													x
Puntius gelius	x	x	x																		x					x
Puntius phutunio	x		x	x	x	x			x	x	x										x					x
Puntius sophore			x																							x

TABLE III. Continued

	Khulna District													Mymensingh District											
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26
Puntius terio	x	x							x		x															x
Unidentified *Puntius* spp.						x																				x
Salmostoma phulo														x			x			x						
Eleotridae																										
Eleotris sp.										x																
Gobiidae																										
Brachygobius nunus									x		x	x	x						x							
Glossogobius giuris	x	x	x	x	x		x			x	x	x	x	x			x		x		x					x
Gobiopterus chuno									x			x	x													
Mastacembalidae									x																	
Macrognathus sp.																										x
Macrognathus aculeatus																										x
Macrognathus pancalus																			x							x
Mastacembelus armatus																										x
Nandidae																										
Nandus nandus																										x
Siluridae																										
Wallago attu																										x

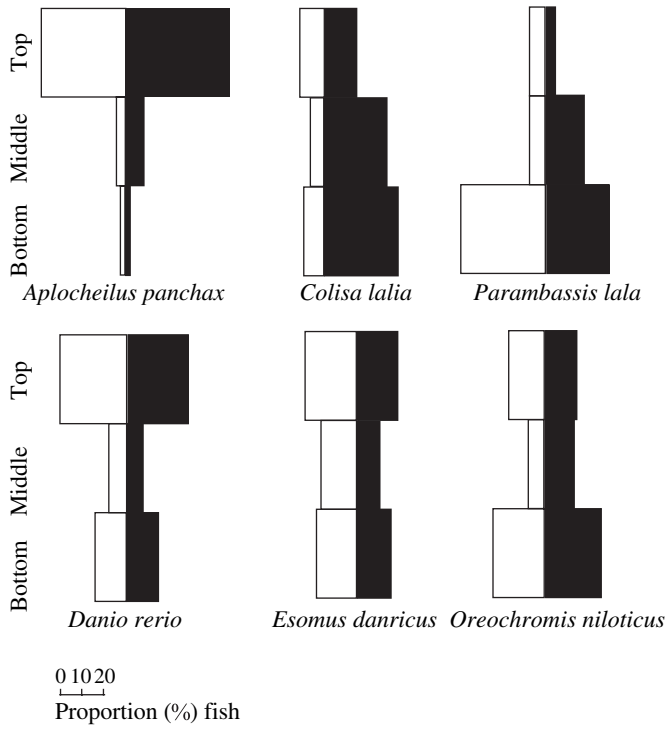

FIG. 2. Mean proportion (%) of each species in vegetated (■) and unvegetated (□) sides of experimental aquaria, at different heights in the water column, averaged over seven replicates and two 15 min observation periods.

The Hurlbert's B' scores for each species, together with estimates of their 95% CL, are shown in Table IV. Of the six species, *A. panchax* was the most specialized in its habitat use and *E. danricus* the most generalist.

DISCUSSION

Danio rerio was more abundant in samples from the northern than from southern areas. Khulna probably represents the southern limit of its range in

TABLE IV. Hurlbert's B' measures of niche breadth for six species, based on the proportion of each species in vegetated and unvegetated sides of experimental aquaria, at different heights in the water column

Species	Score	95% CL
Aplocheilus panchax	0·46	0·28
Colisa lalia	0·79	0·70
Parambassis lala	0·61	0·38
Danio rerio	0·79	0·36
Esomus danricus	0·96	0·26
Oreochromis niloticus	0·86	0·45

Bangladesh; further south is the northern fringe of the Sundarban mangrove forest, and here the water becomes increasingly saline and *D. rerio* is probably absent (C. Smith, unpubl. data). *Danio rerio* appears to be a floodplain rather than a true riverine species. It was not found in rivers or temporary creeks that opened during the monsoon season. Genetic analysis, however, has shown that there is considerable mixing of populations (Gratton *et al.*, 2004) and some of the sites where *D. rerio* was found may connect with the main river channel during the monsoon. Sample site descriptions by Fang (http://artedi.nrm.se/nrmfish/) largely correspond with the present findings, although *D. rerio* was also found in streams with moderate and, in one case, fast flow.

All the sites where *D. rerio* was captured were shallow, relatively clear and frequently with aquatic vegetation. Predatory species were found in only one site with *D. rerio* (site 1), which also had the lowest abundance of *D. rerio*. Two predatory fish taxa were captured, *Channa* spp. and *X. cancila,* which may feed on *D. rerio.* The sampling protocol, however, may have failed to capture other potential fish predators such as nocturnal catfishes. Avian predators such as the Indian pond heron *Ardeola grayii* and the common kingfisher *Alcedo atthis* were also ubiquitous in the area and may feed on *D. rerio*.

All the sample sites were subject to a degree of human influence. Some of the cultivated ponds had been treated with lime and rotenone prior to the introduction of commercial species. Introduced species such as *O. niloticus* were common in these sites. Many ponds contained commercial and introduced species as well as small indigenous species, although none of the sites sampled were the subject of active fish cultivation at the time of sampling. Every site sampled was actively exploited for fishery purposes; small indigenous species (fishes with a maximum size <25 cm L_S) form a substantial constituent of the protein intake of poor households in Bangladesh (Wahab, 2003). *Danio rerio* is among the smallest of these small species and is not, therefore, considered an important food resource even by subsistence fishermen.

Although *D. rerio* was relatively widespread and abundant among sampling sites, in two sites (15 and 23) it was the most abundant species. It was not as widespread as some of the other species in the assemblage, particularly *E. danricus*, another cyprinid and the sister group to *Danio* sp. (Fang, 2003). One notable feature of the results was that *D. rerio* tended to be abundant in habitats connected to rice cultivation at the time of sampling, though not in rice fields themselves. There is no clear explanation for this association with rice cultivation. It could be related to the use of fertilizers, which may promote the growth of zooplankton on which *D. rerio* chiefly feeds (Dutta, 1993; R. Spence, unpubl. data).

Females collected in January from one site (site 23) were found, on dissection, to contain mature ova. This finding contradicts anecdotal evidence that *D. rerio* has a discrete breeding season only during the monsoon (Daniels, 2002). It may be that breeding is dependent on food availability rather than season *per se*. *Danio rerio* specimens ranged in size from 15 to 34 mm L_S. It is probable that the few large specimens caught in January represent a different year class. Thus, *D. rerio* is probably predominantly an annual species, breeding primarily during the monsoon, but spawning continuously after achieving maturity (R. Spence, unpubl. data).

The results of the experimental study of vertical distribution show that *D. rerio, E. danricus* and *O. niloticus* were generalist in their habitat use, while *A. panchax, P. lala* and *C. lalia* were more specialized (Fig. 2). *Aplocheilus panchax* tended to spend a greater proportion of their time near the water surface, while *P. lala* and *O. niloticus* spent more time towards the bottom. This result is reflected in the scores for Hurlbert's B', *Danio rerio*'s score being in the middle of the range for the six species. *Danio rerio* did not spawn during the experiment although it did spawn in the same experimental aquaria in a separate experiment (R. Spence & C. Smith, unpubl. data). It swims close to the bottom during spawning (Spence & Smith, 2005), although courtship involves males actively pursuing females, which utilize the entire water column. No aggressive interactions with other species were observed during the experiment.

In conclusion, *D. rerio* was found to be relatively abundant in Bangladesh. It was most commonly found in shallow ponds and standing water bodies in open locations with aquatic vegetation in the margins. It was also frequently encountered in water bodies with a connection to rice cultivation and was more common in the samples from the north of Bangladesh than the south. *Danio rerio* occupied the whole of the water column and was observed as frequently in open water as amongst aquatic vegetation.

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References

Barman, R. P. (1991). A taxonomic revision of the Indo-Burmese species of *Danio rerio*. *Record of the Zoological Survey of India Occasional Papers* **137,** 1–91.

Daniels, R. J. R. (2002). *Freshwater Fishes of Peninsula India*. Hyderabad: Universities Press.

Darland, T. & Dowling, J. E. (2001). Behavioural screening for cocaine sensitivity in mutagenized zebrafish. *Proceedings of the National Academy of Sciences of the USA* **98,** 11691–11696.

Dutta, S. P. S. (1993). Food and feeding habits of *Danio rerio* (Ham. Buch) inhabiting Gadigarh stream, Jammu. *Journal of Freshwater Biology* **5,** 165–168.

Everitt, B. S. (1977). *The Analysis of Contingency Tables*. London: Chapman & Hall.

Fang, F. (2003). Phylogenetic analysis of the Asian cyprinid genus *Danio* (Teleostei, Cyprinidae). *Copeia* **2003,** 714–728.

Gerlai, R., Lahav, M., Guo, S. & Rosenthal, A. (2000). Drinks like a fish: zebrafish (*Danio rerio*) as a behaviour genetic model to study alcohol effects. *Pharmacology, Biochemistry and Behaviour* **67,** 773–782.

Granato, M. & Nüsslein-Volhard, C. (1996). Fishing for genes controlling development. *Current Opinion in Genetics and Development* **6,** 461–468.

Gratton, P., Allegrucci, G., Gallozzi, M., Fortunato, C., Ferreri, F. & Sbordoni, V. (2004). Allozyme and microsatellite genetic variation in natural samples of *D. rerio*. *Journal of Zoological Systematics & Evolutionary Research* **42,** 54–62.

Grunwald, D. J. & Eisen, J. S. (2002). Headwaters of the zebrafish – emergence of a new model vertebrate. *Nature Reviews Genetics* **3,** 717–724.
Jayaram, K. C. (1999). *The Freshwater Fishes of the Indian Region*. Delhi: Narendra Publishing House.
Kimmel, C. B. (1989). Genetics and early development of zebrafish. *Trends in Genetics* **5,** 283–288.
Laale, H. W. (1977). The biology and use of zebrafish, *Brachydanio rerio* in fisheries research. A literature review. *Journal of Fish Biology* **10,** 121–173.
Nüsslein-Volhard, C. & Dahm, R. (2002). *Zebrafish*. Oxford: Oxford University Press.
Pritchard, V. L. (2001). Behaviour and morphology of the zebrafish, *Danio rerio*. PhD Thesis, University of Leeds.
Roberts, T. R. (1994). Systematic revision of tropical Asian freshwater glassperches (Ambassidae), with descriptions of three new species. *Natural History Bulletin of the Siam Society* **42,** 263–290.
Roberts, T. R. (1998). Systematic observations on tropical Asian medakas or ricefishes of the genus *Oryzias*, with descriptions of four new species. *Ichthyological Research* **45,** 213–224.
Spence, R. & Smith, C. (2005). Male territoriality mediates density and sex ratio effects on oviposition in the zebrafish (*Danio rerio*). *Animal Behaviour* **69,** 1317–1323.
Sterba, G. (1962). *Freshwater Fishes of the World*. London: Vista Books, Longacre Press.
Talwar, P. K. & Jhingran, A. G. (1991). *Inland Fishes of India and Adjacent Countries*. Calcutta: Oxford and IBH Publishing.
Tropepe, V. & Sive, H. L. (2003). Can zebrafish be used as a model to study the neurodevelopmental causes of autism? *Genes, Brain and Behaviour* **2,** 268–281.
Wahab, M. A. (2003). Small indigenous fish species of Bangladesh: potentials for culture and conservation. In *Small Indigenous Species of Fish in Bangladesh, Technical Proceedings of BAU-ENRECA/DANIDA Workshop on Potentials of Small Indigenous Species of Fish (SIS) in Aquaculture & Rice-field Stocking for Improved Food & Nutrition Security in Bangladesh* (Wahab, M. A., Thilsted, Sl. H. & Hoq, M. E., eds), pp. 1–12. Mymensingh: Bangladesh Agricultural University.
Wright, D., Rimmer, L. B., Pritchard, V. L., Krause, J. & Butlin, R. K. (2003). Inter and intra-population variation in shoaling and boldness in the zebrafish (*Danio rerio*). *Naturwissenschaften* **90,** 374–377.
Wright, D. (2004). Genetic variation in zebrafish behaviour. PhD Thesis, University of Leeds.