ACTA ZOOLOGICA BULGARICA

Acta Zool. Bulg., 73 (3), September 2021: 393-400

Aquatic Ecology
Research Article

Published online 30 August 2021
http://www.acta-zoologica-bulgarica.eu/2021/002467

Morphometry and Diet Analysis of the Kashmir Catfish *Glyptothorax kashmirensis* Hora, 1923 (Siluriformes: Sisoridae) from the Jhelum, Poonch and Kurram Rivers, Pakistan

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Abstract: Kashmir catfish *Glyptothorax kashmirensis* Hora, 1923 is a critically endangered species endemic to Pakistan and the Indian administered Kashmir, with limited information available on its basic biology. The present study explored the morphology and feeding habits of the Kashmir catfish from the Jhelum, Poonch and Kurram Rivers of Pakistan. To study feeding habits (n=19), the stomach fullness index, gastro-somatic index (GaSI) and relative gut length (RGL) were calculated. Gut contents were analysed to assess the percentage of occurrence of prey items. The maximum body length was 160 mm, while the minimum length was 82 mm (av. 122.84 mm). The most distinctive character was the thoracic adhesive apparatus, with its prominent streaks and oval central pit. The calculated GaSI of the studied specimens were 8.48±0.68, while RGL was 0.054±0.003. Members of the family Hydropsychidae were the most preferred prey (46%), followed by pupae and unidentified prey items (17.2%), Baetidae (8.6%), Chironomidae (8.3%), Psychodidae (6.4%), Elmidae (4.7%), Stenopsychidae (3.6%), Perlidae (2.5%), Tipulidae (1.9%) and Pleidae (1.1%). The present study provides information about the basic biology of the Kashmir catfish and provides a platform for future studies and conservation.

Key words: Kashmir catfish, *Glyptothorax kashmirensis,* Critically Endangered, endemic and morphology, diet, gastro somatic index, Hydropsychidae

Introduction

The genus *Glyptothorax* Blyth, 1860 is among the most species-rich and widely distributed genera of the family Sisoridae (Kumar 2016). Several species of this genus, i. e. the cave catfish (*G. cavia*), the Kashmir catfish (*G. kashmirensis*), the Naziri catfish (*G. naziri*), the Punjab catfish (*G. punjabensis*), the copper catfish (*G. telchitta*), the river catfish (*G. pectinopterus*) and the Kordestan catfish (*G. platypogonoides*) have been reported from Pakistan (Javed et al. 2013). The Kashmir catfish is a benthopelagic freshwater fish (Singh et al. 2015). It has been reported for the first time from the Jhelum valley (Hora 1923a) and, later, from the Jhelum and Poonch Rivers (Rafique 2013). The Kashmir catfish

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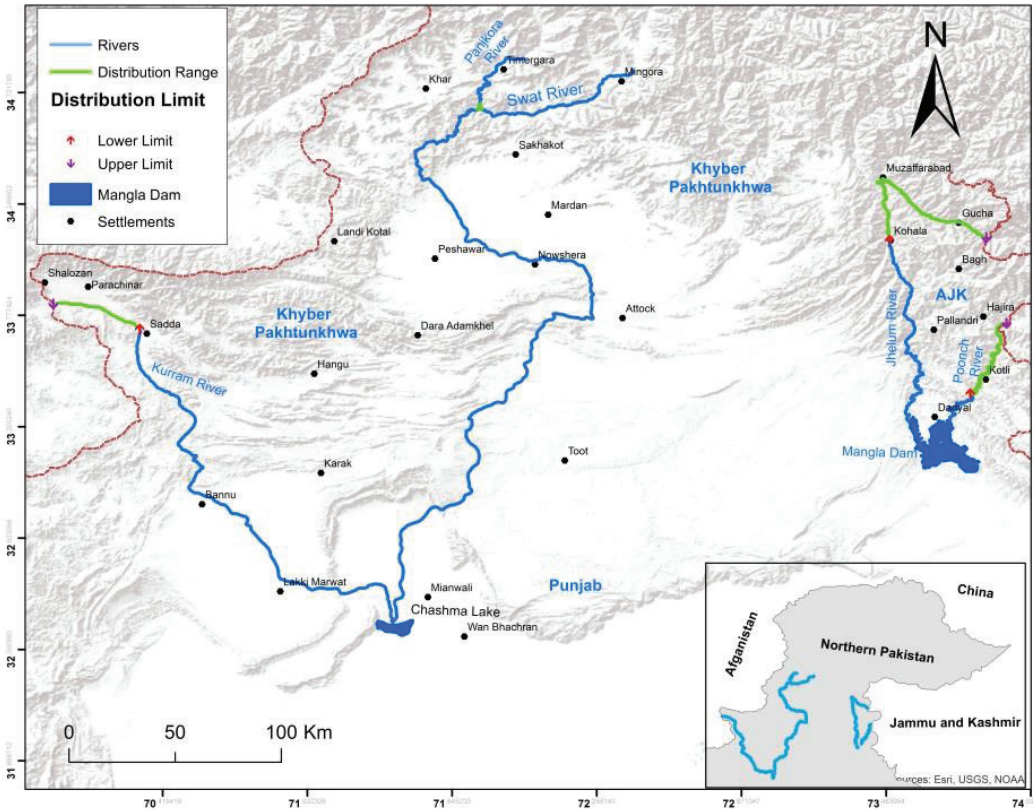

Fig. 1. Study area map showing the upper and lower limits of the distribution of the Kashmir catfish.

has been declared as critically endangered by IUCN (2010). An annual decline of c. 5 % is predicted due to habitat destruction as a result of dam construction in its distribution range (Ng 2010).

The success of good scientific planning and management of fish populations largely depends on the knowledge of their biological aspects, among which food and feeding habits contribute a valuable portion (Sarkar & Deepak 2009). Unfortunately, most of the species are still in need of proper in-depth studies, including taxonomic evaluation (Dahanukar et al. 2011, Jayaram 2010), which is the primary step towards the development of conservation strategies. In addition to body shape and size, morphological studies and meristic counts give useful information about the taxonomic status of a species (Ihssen et al. 1981).

The feeding habit of fish is the search for food and ingesting it, while food habits and diet are the organisms that are habitually eaten (Gerking 1994). Food is one of the key factors that promote growth. Any change in the food items may affect the well-being of the fish. Therefore, it is necessary to understand the relationship between the fish and their prey organisms for predicting not only the feeding habits of fish populations but also to assist in exploring their distribution, which is of utmost importance for the successful management of fisheries (Rao & Durga 2002).

Throughout the distribution range, various studies have been conducted on morphology and feeding habits of different species of the genus *Glyptothorax* (see Javed et al. 2013, Dahanukur et al. 2011). Only a few studies about the morphology of this threatened species are available throughout its entire distribution range (Hora 1923, Talwar & Jhingran 1991, Rashida et al. 1996). The feeding ecology of the Kashmir catfish has been neglected, although it has been reported as an omnivore fish (Singh et al. 2015); detailed information on its food composition is lacking. Such crucial and basic information on the Kashmir catfish is necessary for the identification of its prey.

The aim of the present study is to get information on the morphometric characteristics, to conduct an analysis on the diet of the Kashmir catfish in the Jhelum, Poonch and Kurram Rivers and to provide baseline guidelines for conservation strategies.

Materials and Methods

Study area

The present study was conducted in the Jhelum and the Poonch Rivers of Azad Jammu and Kashmir Province (AJK) and the Kurram River of the Khy-

ber Pakhtunkhwa Province of Pakistan**.** The Jhelum River is a large eastern tributary of the Indus River; the distribution range of the Kashmir catfish in this river is from Chakothi to Kohala (Fig. 1). The ichthyofauna of the Jhelum River is diverse, with 51 reported species (Mirza et al. 2011). The distribution range of the Kashmir catfish in the Poonch River is from Jawra to Gulpur. The Poonch River joins the River Jhelum just above the Mangla Reservoir. Totally, 37 fish species have been recorded from the Poonch River (Rafique 2011). The catchment of the Kurram River is in the western part of the Khyber Pakhtunkhwa, with distribution of the Kashmir catfish between Sadda and Shaloazan. Seventeen fish species have been reported from the Kurram River (Shahjehan & Khan 1997).

Sampling and preservation of fish

Totally, 19 specimens were captured through cast netting for morphometric and diet analysis in repeated surveys in March – October 2017. Sampling was not carried out in winter (November to February) because of a previous study (Nagina et al. 2020) demonstrating that the species remains inactive during winter. Specimens were collected at different times from Chakothi to Kohala (Jhelum River), Jawra to Gulpur (Poonch River) and Sadda to Shalozan (Kurram River). Samples were preserved in labelled jars containing 70% ethanol or 10% formalin following Ng (2005) and Jayaram (2010) and deposited at the Taxonomic Laboratory, Pakistan Museum of Natural History (PMNH), Islamabad.

Morphometric measurements and meristic counts

Identification of the Kashmir catfish was done based on the shape of the adhesive apparatus, barbels position and length (Talwar & Jhingran 1991). All body measurements were made by using a measuring scale (mm) and a dial vernier calliper with an accuracy of 0.05 mm (Ng 2005, Jayaram 2010). Features like fin rays and sucker streaks were observed using a magnifying glass (50 mm Dia) and a light microscope (LEICA DFC 295 1.6X-4X). Morphometric measurements were taken, including total length, standard length, head length and head depth, maximum body depth, cleft of mouth, eye diameter, snout length, pre-dorsal length, post dorsal length, dorsal fin length, the height of adipose dorsal, pre pectoral length, pelvic length and anal fin length. Meristic counts were made by observing the streaks on the adhesive apparatus, barbels length and the number of fin rays. Fin ray counts were presented in the form of a fin formula.

Study of feeding habits

All specimens were weighed with an electronic balance (Amini et al. 2014). Each specimen was dissected viscerally from mouth to anus to expose the internal organs. For the study of gut contents, the gut was removed intact from the oesophagus to the anus and its length (GL) and weight (GW) were measured. For examination of the stomach condition, the degree of stomach fullness was categorised into full, half-filled and empty while the degree of digestion was categorised into digested, semi-digested and undigested. The gut was placed in a Petri dish containing distilled water and excised to take out the stomach contents with twisters. The stomach content was preserved in 70% ethanol. The stomach content was examined visually and microscopically to identify food items. Large food items were identified visually, while microscopic examination (6X to 400X) was done for the smaller food items to identify them to the lowest possible taxon level (Defaye 1988). After identification, food items were counted to estimate their numerical contribution to the diet of the fish. The frequency of occurrence index was determined using the total number of individuals of each prey item in each stomach and the calculation of their mean for all stomachs (Oso et al. 2006).

Statistical analysis

Minitab (17.3.1) was used for the analysis of data on different morphometric characters. Data on morphometric and meristic characters are presented as the mean ± standard error of the mean (SEM). All the characters calculated were taken as a percentage of standard length. For the study of feeding habits, collected samples were presented as percentage, mean and standard deviation (Gwana et al. 2014). The total number of prey items of each food category was recorded for all stomachs, expressed as a percentage and the mean the number of individuals per stomach in each food category was calculated.

Feeding intensity was calculated using the Fullness Index (FI) of the stomach as follows:

FI = [(Number of stomachs with the same degree of fullness) / (Total number of stomachs examined)] X 100

The variations in feeding intensity were expressed by the Gastro-Somatic Index (GaSI) using the equation (Amini et al. 2014):

GaSI = [(Weight of the gut) / (Weight of fish)] X 100

Relative Gut Length (RGL) was calculated using the formula:

RGL = (Gut length) / (Total length)

Table 1. Metrical characters of the Kashmir catfish (n = 19) from Jhelum, Poonch and Kurram River.

Character	Range (mm)	Min (mm)	Max (mm)	Standard error of mean (SEM)		Standard deviation	Percentage of standard length
				Mean	Standard error		
Total length	78	82	160	122.84	4.43	19.308	-
Standard length	67	64	131	101.47	4.014	17.497	-
Pre-pectoral length	14	16	30	23.84	0.821	3.579	23.49
Pectoral length	15	18	33	26.47	0.87	3.791	26.10
Pre-pelvic length	34	35	69	51.42	2.069	9.02	50.67
Pelvic length	16	10	26	18.58	0.886	3.863	18.31
Pre-anal length	50	42	92	68.42	2.854	12.442	67.42
Anal fin length	10	17	27	22.00	0.592	2.582	21.68
Pre-dorsal length	23	26	49	37.79	1.548	6.746	37.24
Dorsal fin length	13	17	30	22.53	0.792	3.454	22.2
Post-dorsal length	13	16	29	22.32	0.937	4.083	21.99
Adipose fin length	15	8	23	12.84	0.852	3.716	12.65
Max. body depth	25	11	36	21.84	1.697	7.396	21.52
Caudal peduncle length	13	15	28	20.89	0.837	3.65	20.58
Snout length	9	9	18	13.68	0.472	2.056	13.48
Head length	24	8	32	19.89	1.873	8.164	19.6
Eye diameter	2	2	4	3.16	0.175	0.765	3.11
Head width	18	12	30	20.11	0.982	4.28	19.18

Results

Morphometric characteristics

The data on morphometric characteristics of the Kashmir catfish and the percentage of the standard length of all measured characters are presented in Table 1.

In all the specimens studied, the dorsal body profile was rising from the snout tip to the end of the dorsal fin and then becoming straight at the end of the caudal peduncle, while having a straight ventral side. Dorsal and lateral sides dark brown, abdominal region at the base of fins light yellowish. Skin with black dots with even granulation on dorsal side due to the presence of tubercles, ventral profile smooth.

Mouth inferior and wide, with a semicircular shape; lips papillated with cleft of mouth (CM) 4.1% of standard length (SL). Eyes very small, rounded and subcutaneous, dorsally in the posterior region of head. Nostrils close to each other and nearer to the snout. Occipital process reaching basal bone of the dorsal fin. Snout rounded, depressed and close to the tip of the mouth. Adhesive apparatus longer than broad, with 30-36 prominent streaks. Adhesive apparatus with distinctive creamy oval central pit**.**

Total body length 82-160 mm (mean 122.84 mm) and standard length 64-131 mm (mean 101.47 mm). Maximum body depth (MBD) at the base of the origin of the dorsal fin behind the operculum. Head depressed, longer than broad. Dorsal fin (DF) present near the tip of the snout. Adipose dorsal fin (AdDF) originating opposite to the origin of anal fin, deeper than long. Pectoral fin originating from the end of the head at anterior side; pelvic fin originating opposite to the origin of DF on ventral side. Anal fin originating before the origin of AdDF on ventral side of body. Anal fin length nearly one-third of pre-anal region, 42-92 mm (mean 67 mm). Caudal fin forked, with lower lobe slightly longer than upper lobe and strong and muscular caudal peduncle.

Table 2. Meristic characters of the Kashmir catfish (n= 19) from Jhelum, Poonch and Kurram Rivers.

Character	Number
Pectoral fin rays	P.I, 7-8
Pelvic fin rays	V.I, 5
Anal fin rays	A.III, 6
No. of barbels	4 pairs
Dorsal fin rays	D.1, 5
Caudal fin rays	C. 17-18
Adhesive organ streaks	13

The data on meristic characters of the Kashmir catfish are shown in Table 2.

Table 3. General body and gut measurements for feeding habits of the Kashmir catfish collected from the Jhelum and Poonch Rivers, Azad Jammu and Kashmir.

Parameter	Min	Max	Mean	Standard error	Standard deviation
Fish body length (mm)	112	162	134.13	3.745	14.505
Gut length (mm)	5.10	9.2	7.04	0.4144	1.6048
Relative gut length	0.04	0.08	0.054	0.00271	.01049
Fish body weight (mg)	12.90	51.60	24.999	3.67863	14.24729
Gut weight (mg)	0.70	6.29	2.364	0.48382	1.87382
Gastro-somatic index	4.86	14.98	8.485	0.68631	2.658075

Table 4. Feeding frequency of different prey items (average number of individuals per stomach) per specimen of the Kashmir catfish collected from the Jhelum and Poonch Rivers, Azad Jammu and Kashmir.

Prey items	Mean No.	Standard deviation	Standard error of mean
Hydropsychidae	11.00	7.874	2.033
Baetidae	2.07	1.486	0.384
Elmidae	1.13	1.302	0.336
Perlidae	0.60	0.632	0.163
Chironomidae	2.00	3.836	0.990
Tipulidae	0.47	0.915	0.236
Stenopsychidae	0.87	0.834	0.215
Pleidae	0.27	0.458	0.118
Psychodidae	1.53	1.807	0.467
Pupa and unidentified	4.13	3.226	0.833
Total	2.41	4.298	0.351

Four pairs of barbels were observed: nasal and maxillary on the posterior side and outer and inner mandibular on the anterior tip of the mouth. Maxillary barbels with maximum length extending beyond base of pectoral fin; nasal barbells smallest of all. Inner mandibular barbels originating from base of lower jaw, longer than nasal barbels. Outer mandibular barbels slightly longer than inner mandibular barbels, reaching thoracic adhesive apparatus. Dorsal fin sharp and with pointed serrations. Pectoral spine longer and stronger than dorsal spine, with 10-11 serrations on right and left sides, respectively. Anal fin with sharp serrations on upper side and with fin rays. Caudal fin deeply forked, upper lobe slightly longer than lower.

Fin formula: D. I, 5; P. I, 7-8; V. I, 5; A.III, 6; C. 16-17

Feeding characteristics

The Kashmir catfish has an inferior and transverse mouth with villiform teeth on the roof of the mouth. Stomach is distinct and sac-shaped. Intestine is thin and short. For gut measurements, see Table 3.

Gut analysis

The Fullness Index values was 60% for filled and 40% for partially filled stomachs. The gut length was 5.22% of total body length, while gut weight was 9.44% of total body weight. Relative Gut Length (RGL) was 0.054±0.003 on average and the Gastro-Somatic Index was 8.48±0.68 (Table 3). Of all the studied specimens, the stomach was mostly found as half full or filled and swollen. There were no significant variations in the diet of individuals in the smallest (112 mm), intermediate (134 mm) and largest (162 mm) specimens.

Diet composition

Identification of the prey species was made to the family level or the ordinal level; the average number of prey species per stomach for each food category was calculated (Table 4).

The primary food items observed were aquatic insects: Trichoptera [Hydropsychidae (11 specimens per fish on average), Stenopsychidae (1), Plecoptera [Perlidae (1)], Ephemeroptera [Baetidae (2)], Coleoptera [Elmidae (1)] and Diptera [Chironomidae (2), Tipulidae (1) and Blephariceridae (0.5)]. The most preferred prey was of the family Hydropsichidae (46%), followed by Baetidae (8.6%), Chironomidae (8.3%), Psychodidae (6.4%), Elmidae (4.7%), Stenopsychidae (3.6%), Perlidae (2.5%), Tipulidae (1.9%) and Pleidae (1.1%). A great percentage (17.2%) of the prey items were recorded as unidentified species due to partially digested unidentifiable body parts or being at larval stages.

Discussion

Despite recent studies, documenting the diversity within the genus, the taxonomy of *Glyptothorax* is still poorly resolved (Ng 2005). There are limited data on the critically endangered Kashmir catfish, from only a few studies describing its morphol-

ogy based of a limited number of specimens. The original description was based on two specimens only (Hora 1923). No previous comprehensive evidence on its feeding habits has been published. The present study is the first to provide detailed information on the morphometrics of the Kashmir catfish from the Jhelum, Poonch and Kurram Rivers of Pakistan.

The present study provides a morphometric description of the Kashmir catfish in addition to the parameters used in the earlier studies (Hora 1923, Jayram 1999, Talwar & Jhingram 1991, Rashida et al. 1996). We have recorded higher maximum body length (160 mm) than the preciously reported lengths of 117 mm (Hora 1923) and 145 mm (Javed et al. 2013). In addition, we provide morphometric measurements of the cleft of the mouth, pre- and post-dorsal length, pre-pectoral, pre-pelvic and pre-anal lengths, the height of the adipose fin and the origin of the adipose dorsal and the anal fins.

In addition to the original description (D I-5, V I-5, P I-8-9, A. II-6) of the Kashmir catfish (Hora 1923), the present study has shown variations of meristic counts such as dorsal (D I-5), pectoral (P I-7-8), pelvic (V I-5), anal (A. II-6) and caudal-fin rays (C 17-18). The number of caudal-fin rays and point of origin of adipose dorsal and anal fins observed in the present study have not been reported in the previous study (Hora 1923). Fin rays count reported for the Kashmir catfish in other studies is D. I, 6; P.I, 8-9; V.I, 5; A.III, 7-8; C.17 (Javed et al. 2013) and D I-5, P I-8-9, V I-5, A. II-6, C 17-18 (Singh et al. 2015). Differences in meristic characters in different populations of the same species due to genetic or environmental factors have already been described (Watanable 1998). Variations in meristic counts with changes in body length and weight have been reported for angelfish (Bibi & Sekarian 2008) and Gangetic latia *Crossocheilus latius* (see Brraich & Saima 2015). In contrast to the earlier report (Javed et al. 2013), the nasal barbels are the smallest of all.

We provide information on the dietary habits of the Kashmir catfish. Owing to the lack of data about the approximate number of this species in the wild, its global status (Critically Endangered) and the restricted distribution range, we examined a limited number of specimens for the diet analysis. Furthermore, this species is most active in turbid water and is difficult to be captured in clear water. By this reason the diet analysis of the Kashmir catfish has been based on only 19 specimens. It has been reported that RGL value is generally low in carnivorous fish, higher in omnivorous fish and highest in herbivorous fish (Das & Moitra 1963). The results of the present study indicate that the Kashmir catfish has a lower relative gut length (RGL 0.054±003), which is indicative of its carnivorous nature. The relative gut length observed in the present study was lower than that recorded for *Glyptothorax silviae* (0.66), another congeneric species (Amini et al. 2014).

The prey species recorded in the stomach of the Kashmir catfish are mostly benthic macroinvertebrates, indicating that the Kashmir catfish is a benthic feeder. Benthic macroinvertebrates have previously been reported as the most common prey species for the genus *Glyptothorax* (see Dahanukar et al. 2011). The gut content analysis shows that the Kashmir catfish is a strict carnivore species.

The prey items observed in the gut of the Kashmir catfish were mostly benthic macroinvertebrates. The higher percentage of the Hydropsychidae indicates the preference of the Kashmir catfish for this group or the maximum availability of this species as a prey item. Similar feeding behavior has been reported for *G. silviae* in the Marun River, whose diet included Plecoptera, Ephemeroptera, Diptera, Hemiptera and Trichoptera (Amini et al. 2014). In contrast to the present study, the Kashmir catfish was reported earlier as an omnivore (Singh et al. 2015); however, that study lacks a detailed description of the prey items in the gut.

Conclusions

The present study describes the morphometric features of the Kashmir catfish for the first time and adds useful knowledge to the morphometric base identification of this species in Pakistan. Besides, this study provides data on the diet preference of the Kashmir catfish, which could be useful in developing a future conservation plan. The study also confirmed that the Kashmir catfish is a strict carnivore species. In addition to its description, more studies are recommended to examine different life aspects such as habitat, population status and breeding ecology.

Acknowledgments: We thank the Pakistan Museum of Natural History for providing equipment and chemicals for this study. We are especially grateful to Hagler Bailly Pakistan for arranging the field trips for sample collection at the study sites. All guidelines for the care and use of animals were followed.

Conflict of interest: Authors declare no conflict of interest in this article.

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Received: 11.06.2020
Accepted: 06.11.2020