

FIRST RECORD OF THE CHINESE FANRAY, *PLATYRHINA SINENSIS* (ELASMOBRANCHII: MYLIOBATIFORMES: PLATYRHINIDAE), IN THE SEAWATERS OF ZHUJIAJIAN, ZHOUSHAN, CHINA

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Abstract. There are several studies of the ichthyofauna of the East China Sea, however, the knowledge of fishes from this region still remains incomplete. This report confirms the occurrence of Chinese fanray, *Platyrrhina sinensis* (Bloch et Schneider, 1801), in the seawaters of Zhoushan by analysing nine specimens of the Chinese fanray obtained in Zhujiajian from May 2017 to December 2017. The morphometric measurements and meristic counts were taken. The principal characters that are conclusive for the species were consistent with *P. sinensis* and were sufficient for separation of the examined specimens from other *Platyrrhina* species. In addition, the fragment of 12S rRNA was also sequenced for the purpose of classification. The mean genetic distance within *P. sinensis* group was 0.71%, group mean distance between *P. sinensis* and other *Platyrrhina* species ranged from 6.01% to 7.71%. Species were also separated from each other at the genetic level. Given the Chinese fanray has not been reported to exist in this region, our findings represent the first record from Zhoushan and extend the distribution range of this species into the north of the East China Sea. The reason behind the observed northward migration of some *P. sinensis* individuals from their southern habitat might be global warming. Collection of many additional specimens is needed to better define the geographic limits of the Chinese fanray.

Keywords: *Platyrrhina sinensis*, seawaters of Zhoushan, first record, 12S rRNA, global warming, zoogeography, fish taxonomy

INTRODUCTION

The marine fish fauna of the East China Sea is characterized by considerable species richness and diversity (Zhao et al. 2012, 2016), and have a significant place in Chinese ichthyology due to the presence of numerous endemic species with restricted distribution ranges. The fish fauna checklist of Zhu and Meng (2001) on the East China Sea includes 647 species, but now the marine fish species of the area lists contains as many as 732 species (Zhao et al. 2016). The increased number of species is a consequence of several factors, such as the introduction of exotic species, range extension of the

species, and identification of new species (Zhao et al. 2012, 2016).

It is difficult, however, to make new findings for rare and economically insignificant species such as rays, which have always been overlooked. The Chinese fanray, *Platyrrhina sinensis* (Bloch et Schneider, 1801), is a benthic ray (Bloch and Schneider 1801) occurring from the south of China to Vietnam (Zhang et al. 1955, Chu 1960, Chen et al. 1997, Chu and Wu 1984, Iwatsuki et al. 2011). The knowledge of the distribution of Chinese fanray is still insufficient to define their potential distributional range. In particular, data on their distribution in the East China

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Sea are still lacking, even though this sea is closest to the south of China. Apart from the Taiwan Strait and the South China Sea, specimens of “*P. sinensis*” has been recorded in other areas including the East China Sea, however, these records in China were the misidentification of *Platyrrhina tangi* Iwatsuki, Zhang et Nakaya, 2011 as *P. sinensis* (see Tang 1933, Compagno et al. 1999, 2005, Nakabo 2002, Iwatsuki et al. 2011, Zhao et al. 2012, 2016).

MATERIAL AND METHODS

From May 2017 to December 2017, nine samples of *Platyrrhina sinensis* were collected in the seawaters of Zhujiajian (122°25.109'E, 29°52.551'N), Zhoushan. Therefore, this paper is the first reliable report from this area and extends the distribution range of the Chinese fanray into the north of the East China Sea.

In order to better preserve the original morphology of the presently reported *P. sinensis* samples, we kept specimens in a freezer prior to examination. The body colour and pigmentation of specimens were pictured, together with all counts and measurements handled. All collected specimens were identified based on morphological characteristics used by Tang (1933), Last et al. (2006), Iwatsuki et al. (2011), and White and Last (2016). The majority of the measurements followed the existing convention for rays and sharks and were taken directly (point to point) unless otherwise stated. Detailed guidance of counts and measurements was listed as follows: total length (TL); snout length (defined as direct length from the snout tip to the firm nasal capsule adjacent the orbit–forward of eye socket); spiracle length (greatest length of the main cavity); pre-oral length (direct length from the snout to the posterior edge of upper jaw at its symphysis); mouth width (taken across the exposed width); pelvic fin insertion to dorsal fin origin–horizontal distance from the pelvic insertion to the origin of the first dorsal fin.

In addition to the morphological study, a DNA barcoding approach was also employed to support the taxon of *P. sinensis* at the genetic level. For genetic study, a piece of muscle tissue was obtained to carry out DNA extraction. The classical phenol–chloroform method was used for DNA extraction. PCR was subsequently conducted. The sequences of MiFish-E-F and MiFish-E-R primers used for 12S rRNA amplification were 5'-GTTGGTAAATCTCGTGCCAGC-3' and 5'-CATAGTGGGGTATCTAATCCTAGTTTG-3' (Miya et al. 2015), respectively. PCR was carried out in a 25 μ L reaction mix containing DNA template (1 μ L, 50 ng \cdot μ L⁻¹), forward primer (1 μ L, 10 μ M \cdot L⁻¹), reverse primer (1 μ L, 10 μ M \cdot L⁻¹), dNTPs (2 μ L, 2.5 mM \cdot L⁻¹ each), EasyTaq DNA Polymerase (0.15 μ L, 5 U \cdot μ L⁻¹), and 10 \times PCR buffer (2.5 μ L, 25 μ M \cdot L⁻¹). A Biometra thermal cycler (Gottingen, Germany) with the following given procedure: an initial denaturation (95°C, 5 min), 35 cycles consisting of denaturation (94°C, 15 s), annealing (54°C, 15 s) and extension (72°C, 15 s), and a final extension (72°C, 3 min), was employed for PCR amplification. PCR products were preserved at

4°C. After agarose gel electrophoresis, the PCR products were sequenced by Shanghai Majorbio Bio-Pharm Technology Co., Ltd. All original sequences were then revised by DNASTAR software (DNASTAR, Madison, WI, USA). One 12S rRNA sequence of *Platyrrhina hyugaensis* Iwatsuki, Miyamoto et Nakaya, 2011 (LC277746) downloaded from NCBI was included in phylogenetic tree study. *Anoxypristis cuspidata* (Latham, 1794) (KP233202) was chosen as the out-group to root the tree. MEGA 5.0 (Tamura et al. 2011) was used to construct the neighbor-joining (NJ) tree under the Kimura 2-parameter (K2P) model.

RESULTS

The general morphological features of *Platyrrhina sinensis* are shown in Fig. 1 and Table 1. The most significant character distinguishing this species from others was there were two rows of hooked thorns on mid-dorsum of tail (vs. only one row of hooked thorns on mid-dorsum of tail in *P. tangi* and *P. hyugaensis*) (Iwatsuki et al. 2011). Thorns on orbital, nape and scapular regions were not encircled by light yellow or white pigment, which was also different in *P. tangi*. The detailed counts and measurements of *P. sinensis* were listed in Table 1, which verified the identification of this species in the north of East China Sea.

The neighbor-joining phylogenetic tree is shown in Fig. 2. Specimens of *P. sinensis* in the presently reported study clustered in a group, and those of *P. tangi* and *P. hyugaensis* clustered in another two groups, respectively. Groups of all species including *A. cuspidata* were well defined based on the distance of the 12S rRNA sequence. The genetic distance of the 12S rRNA sequence within *P. sinensis* group was only 0.71%, and the mean distance between *P. sinensis* and other two *Platyrrhina* species (*P. tangi* and *P. hyugaensis*) was up to 6.01% and 7.71%, respectively. The interspecific distance was about 8.5–10.6 times larger than intraspecific distance. Together, both the morphological and genetic analysis strongly supported the validity of *P. sinensis* as a new record in Zhoushan.

DISCUSSION

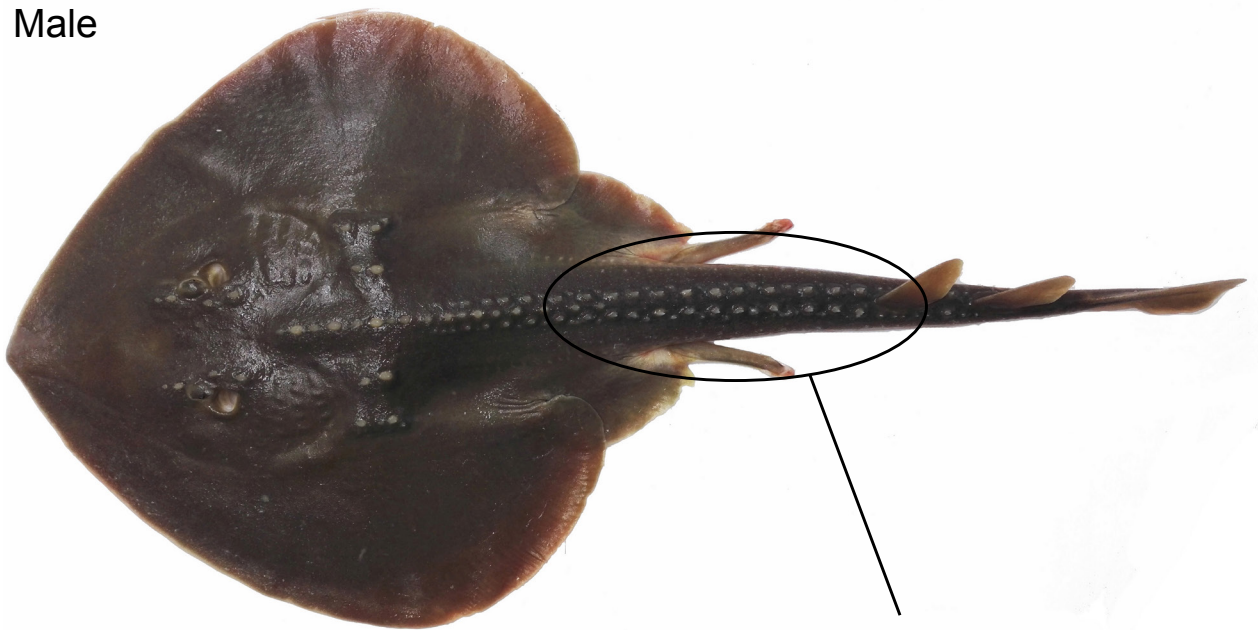
Although *Platyrrhina sinensis* has little economic value, it shows significance from the aspect of ichthyofaunal conservation and overall fish diversity. The globe is heating up now with warmer lands and oceans than when record keeping began in 1880, and temperatures are still ticking upward (Brander et al. 2003). In response to climate change, long-term trends and variations in abundances and distribution of many marine fish have been observed over the past few years, such as cod, flatfish, salmonids, and so on (Drinkwater 2005, Farrell 2009, Hermant et al. 2010). The previous report showed that *P. sinensis* mainly existed in the Taiwan Strait and the South China Sea (Iwatsuki et al. 2011) and no *P. sinensis* has ever been collected from other sea areas in China (Iwatsuki et al. 2011). In this study, nine *P. sinensis* individuals were collected from

Zhoushan. This paper extends the distribution range of this species into the north of East China Sea and represents the northernmost record of *P. sinensis* in China. Our findings indicate that global warming might prompt some *P. sinensis* individuals to migrate northward from its southern habitat like other fishes mentioned above. Attention should be paid to the new region fish species including *P. sinensis* expanded to because their appearance might indicate climatic change, which may make a big difference for local fishery production and fish diversity.

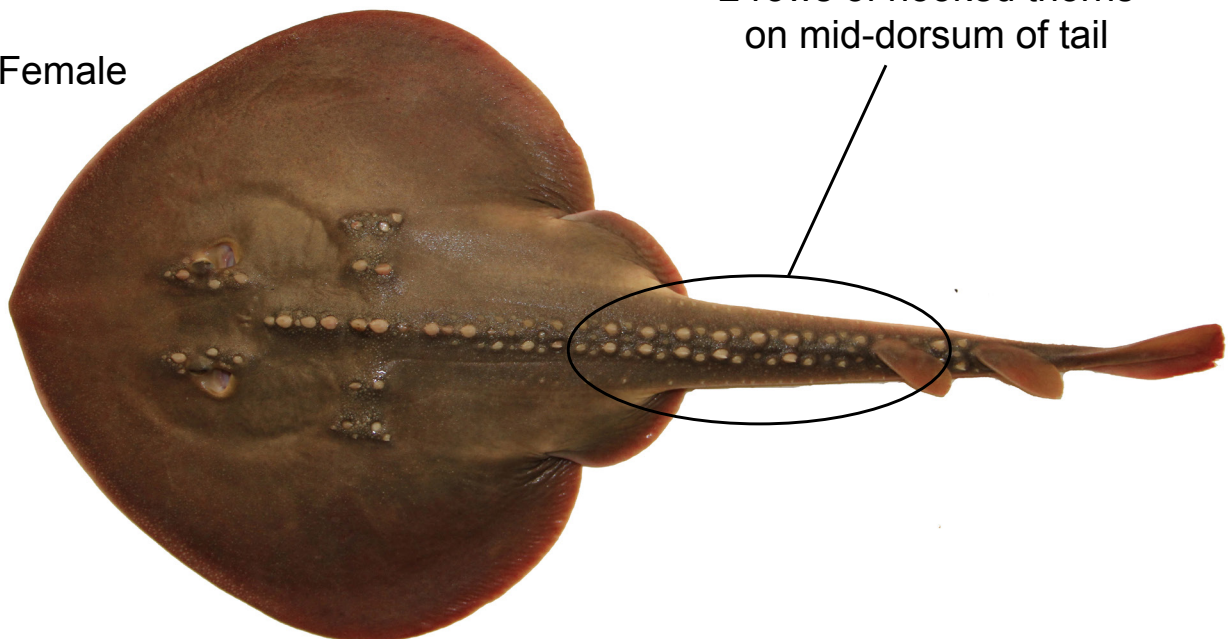
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Male



Female



2 rows of hooked thorns
on mid-dorsum of tail

Fig. 1. Dorsal (upper) views of *Platyrhina sinensis* from Zhoushan, China

Table 1

Comparative counts and measurements of *Platyrrhina sinensis* and other *Platyrrhina* species from Zhoushan, China

Character	Species			
	<i>P. sinensis</i> (n = 9)		<i>P. tangi</i> (n = 2)	
	Range	Mean ± SD	Range	Mean ± SD
Total length [mm]	210–438		316–423	
Number of hooked thorns rows on mid-dorsum of tail	2		1	
Disc length	45.24–52.17	49.23 ± 1.75	43.59–51.45	47.52 ± 2.38
Disc width	45.69–55.48	51.85 ± 2.31	44.98–51.49	48.34 ± 3.21
Body depth	7.08–8.24	7.31 ± 0.84	5.26–7.01	6.14 ± 0.52
Snout to maximum disc width	39.36–42.19	42.12 ± 1.24	36.58–41.29	38.94 ± 2.96
Head length	28.31–29.96	29.16 ± 0.56	21.68–25.86	23.77 ± 1.08
Snout to cloaca	43.58–45.02	44.29 ± 0.21	39.66–44.81	42.24 ± 1.09
Pre-oral length of snout	15.02–18.43	17.01 ± 0.42	11.52–12.67	12.10 ± 0.22
Pre-orbital length of snout	15.23–18.66	16.45 ± 0.33	11.89–13.21	12.55 ± 0.11
Prenarial length of snout	11.09–12.57	12.25 ± 0.14	9.65–12.04	10.85 ± 0.48
Orbit diameter	3.84–4.97	4.42 ± 0.26	3.70–4.56	4.13 ± 0.32
Eye diameter	2.10–3.02	2.45 ± 0.12	2.52–3.01	2.77 ± 0.54
Spiracle length	2.11–2.98	2.56 ± 0.21	2.11–3.58	2.85 ± 0.87
Interorbital width	5.89–7.02	6.45 ± 0.43	4.99–6.02	5.51 ± 0.45
Inter-spiracular width	6.97–7.76	7.59 ± 0.31	6.34–7.28	6.81 ± 0.58
Internarial width	3.04–3.92	3.52 ± 0.19	2.41–3.83()	3.12 ± 0.49
Mouth width	9.06–11.31	10.19 ± 0.75	8.54–9.37	8.96 ± 0.87
Nostril length	1.88–2.23	2.06 ± 0.09	1.02–1.96()	0.99 ± 0.21
Nasal flap length	1.71–2.97	2.19 ± 0.23	0.98–2.06	1.52 ± 0.17
Distance across anterior nasal apertures	7.95–8.87	8.47 ± 0.11	6.39–7.95()	7.17 ± 0.44
Snout to first gill opening	22.09–22.95	22.52 ± 0.09	19.74–20.82	20.29 ± 0.36
First gill opening width	1.09–1.23	1.17 ± 0.05	1.17–1.58()	1.38 ± 0.29
Fifth gill opening width	0.98–1.17	1.08 ± 0.08	1.01–2.01	1.51 ± 0.52
Distance between first gill openings	16.48–17.67	17.24 ± 0.12	14.46–15.66	15.06 ± 0.39
Distance between fifth gill openings	10.58–13.11	11.79 ± 0.56	12.86–13.98	13.4 ± 0.48
Pelvic fin length	14.67–18.84	16.84 ± 0.87	16.33–18.02	17.18 ± 0.62
Posterior base of pelvic fin to first dorsal fin origin	15.72–24.35	20.46 ± 2.54	17.84–19.20	18.52 ± 0.71
First dorsal fin length	5.89–6.98	6.45 ± 0.42	6.46–8.79	7.62 ± 0.62
First dorsal fin height	4.11–5.26	4.69 ± 0.63	3.28–5.96	4.62 ± 0.25
First dorsal fin base length	3.92–5.41	4.67 ± 0.29	3.69–4.56	4.13 ± 0.62
Inter-dorsal distance	4.11–5.72	5.06 ± 0.40	4.73–6.17	5.45 ± 0.24
Second dorsal fin length	7.03–9.18	8.11 ± 0.83	8.16–9.10	8.63 ± 0.60
Second dorsal fin height	4.21–4.78	4.61 ± 0.34	4.39–5.46	4.93 ± 0.94
Second dorsal fin base length	3.92–6.85	4.96 ± 0.48	4.06–4.77	4.42 ± 0.48
Snout to first dorsal fin origin	62.29–69.85	66.15 ± 2.17	55.49–67.71	61.60 ± 4.69
Snout to second dorsal fin origin	69.96–77.47	73.98 ± 1.47	72.09–77.93	75.01 ± 2.88
Posterior base of second dorsal fin to tip of caudal fin	16.71–18.91	17.86 ± 0.29	19.18–23.04	21.11 ± 1.74
Caudal fin length (dorsal margin)	15.51–17.16	16.26 ± 0.61	16.53–19.21	17.87 ± 1.45
Caudal fin length (abdominal margin)	12.14–15.41	13.57 ± 0.48	16.99–19.62	18.31 ± 1.31
Tail length	53.45–65.75	58.23 ± 2.19	55.62–59.03	57.33 ± 2.93
Tail width at pelvic axil	7.75–8.89	8.38 ± 0.42	6.95–7.87	7.41 ± 0.66
Ventral fold width at first dorsal fin origin	0.45–1.24	0.83 ± 0.19	0.12–0.97	0.55 ± 0.10
Clasper length	13.65–14.42	13.94 ± 0.11	12.44–18.09	15.27 ± 1.32

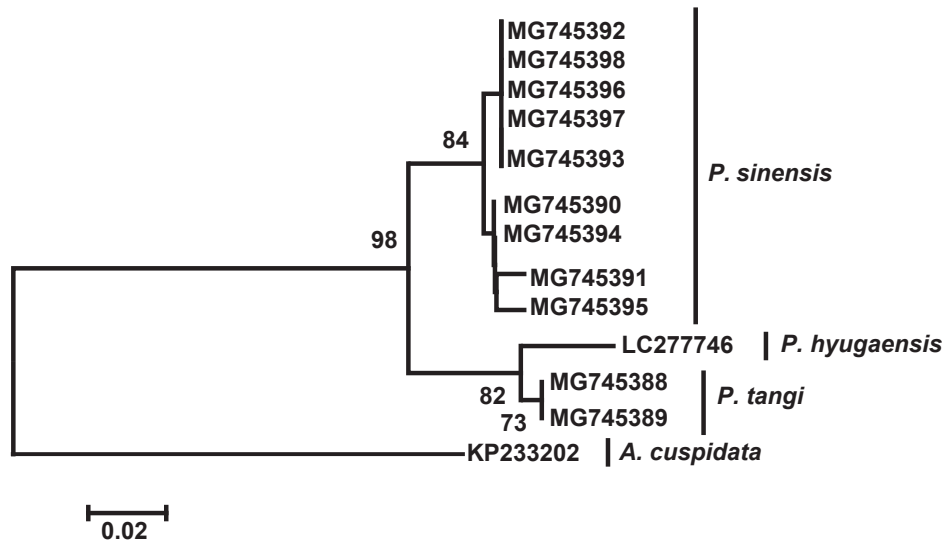


Fig. 2. Phylogenetic tree of three *Platyrrhina* species based on neighbor-joining analysis of 12S rRNA sequence; numbers above branches indicate neighbor-joining bootstrap percentages; only bootstrap values of > 70% are shown in the above NJ tree

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