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First report of the Taiwan sardinella *Sardinella hualiensis* (Clupeiformes: Clupeidae) in the Philippines

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A range expansion of the Taiwan sardinella *Sardinella hualiensis* to the Philippines is reported. The data suggested that the southern translocation of *S. hualiensis* occurred across oceanographic features that are typically barriers to small, pelagic fish dispersal. With this addition, the Philippines is among the most diverse countries for *Sardinella* biodiversity in the world, second only to India.

Key words: Cagayan; Luzon Strait; new report; sardines.

Sardinella (Clupeiformes: Clupeidae) is a genus of small, coastal pelagic, planktivorous fishes that form large schools over the continental shelf shallower than 200 m. The 21 species of *Sardinella* are found in tropical, subtropical and temperate waters to 40° N and S; however, the highest diversity of *Sardinella* is in the tropical Indo-Pacific region (Whitehead, 1985).

Sardinella is socioeconomically and commercially valuable, particularly in developing countries where they serve as a cheap source of animal protein for millions of people (FAO, 2011).

Whereas many species of *Sardinella* have broad geographical known ranges, the range of Taiwan sardinella *Sardinella hualiensis* (Chu & Tsai 1958) was previously restricted to Taiwan and adjacent mainland China south to Hong Kong (Whitehead, 1985; Froese & Pauly, 2010) (Fig. 1). Overlapping distributions and morphological similarities between *Sardinella* species make them difficult to differentiate; however, several unique characters distinguish *S. hualiensis* from other *Sardinella* species that occur over the same geographical area [white sardinella *Sardinella albella* (Valenciennes 1847), fringescale sardinella *Sardinella fimbriata* (Valenciennes 1847), goldstripe sardinella *Sardinella gibbosa* (Bleeker 1849), Bali sardinella *Sardinella*

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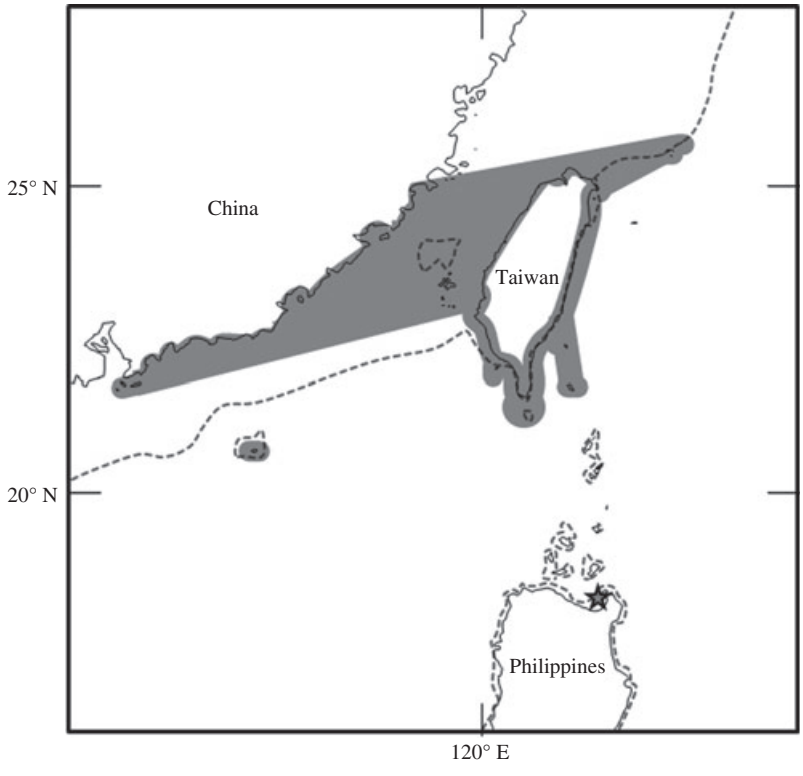

FIG. 1. Geographical distribution of *Sardinella hualiensis* in the north-west Pacific Ocean (■ area) based on Froese & Pauly (2010), location of current record (☆) and 200 m water depth demarcation (----) based on images from NAMRIA – Philippines (1988), Centurioni *et al*. (2004) and Google Earth (2011).

lemuru (Bleeker 1853), Richardson's sardinella *Sardinella richardsoni* (Wongratana 1983), freshwater sardinella *Sardinella tawilis* (Herre 1927) and Japanese sardinella *Sardinella zunasi* (Bleeker 1854)]. *Sardinella hualiensis* and *S. tawilis* have overlapping or continuous vertical striae on the scales and black tips on the caudal fin (Table I), features other Philippine and Taiwan-occurring *Sardinella* species lack. On the other hand, *S. tawilis* is endemic to the freshwater habitat of Taal Lake and is geographically isolated from the marine *S. hualiensis* (Herre, 1927; Chu & Tsai, 1958). In addition to geographical separation, Chu & Tsai (1958) state that the body depth of *S. hualiensis* is overlapping but generally less than *S. tawilis*, and that the origin of the *S. hualiensis*'s ventral fin is below the anterior portion of the dorsal fin, whereas *S. tawilis* has a more posterior origin of the ventral fin.

Specimens examined here were captured in a drift gillnet by a local fisher in the Babuyan Channel off of Santa Ana (18° 30′ N; 122° 8′ E), Cagayan Province, Philippines (Fig. 1), on 18 December 2010. Ten specimens were transported to the National Fisheries Research and Development Institute for identification and measurements (Fig. 2). Diagnostic features included body depth, number of scutes, number of lower gill rakers, scale features and colouration and are included in Table II. Measurement and meristic data placed the specimens within the previously described limits for

TABLE I. Morphology and meristics of *Sardinella* species of Taiwan and the Philippines based on Herre (1927), Whitehead (1985) and Froese & Pauly (2010). Shading indicates measurements and features shared or with overlapping frequency between *Sardinella hualiensis* and other *Sardinella* species

Biometrics/meristics/colouration	*S. hualinesis*	*S. richardsoni*	*S. zuanis*	*S. albella*	*S. gibbosa*	*S. lemuru*	*S. fimbriata*	*S. tawilis*
L_s (mm)	100–125	100–120	100–130	100–140	120–170	150–230	110–130	100
Body depth:L_s (%)	29–34	26–32	24–33	25–40	24–30	<30	25–34	29–31
Scutes	30–32	30–32	29–32	29–33	32–34	NR	29–33	28–30
Lower gill rakers	51–66	63–74	42–58	41–68	45–59	77–188	54–82	61–74
Pelvic fin rays (branched and unbranched)	8	8	8	8	8	9	8	8
Enlarged last two anal fin rays	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Scales with overlapping/continuous striae	Yes	Yes	Yes	No	No	No	No	Yes
Scales with no, few or many small perforations	Many	Many	Few	Few	Many	No	Many	Many
Scale protrudes posteriorly	Somewhat	Yes	Somewhat	Somewhat	Yes	Somewhat	Yes	Somewhat
Scales fimbriated	Yes	No	No	Yes	Yes	Yes	Yes	Yes
Black spot at dorsal fin origin	Yes	No	No	Yes	Yes	No	Yes	Yes
Tips of caudal fin black	Yes	No	No	No	Yes	No	Yes	Yes
Dorsal fin blackish	Yes	No	No	No	Yes	Yes	Yes	Yes
Black spot at posterior margin of operculum	Yes	Yes	Yes	Yes	Yes	Yes	No	Yes
Golden midline and spot behind gill cover	No	No	No	No	Yes	Yes	No	No
		Sardinella spp. of Taiwan			*Sardinella* spp. of the Philippines			

L_s, standard length.

FIG. 2. Photograph of specimen of *Sardinella hualiensis* (standard length 100 mm) captured in the Babuyan Channel, Cagayan Province, Philippines.

S. hualiensis. A single specimen exhibited 29 scutes (outside the range of 30–32). *Sardinella hualiensis* is described in Whitehead (1985) as having 51–66 lower gill rakers. The 68–94 lower gill rakers, however, were counted on the lower half of the first gill arch as described by Whitehead (1985) on these specimens (Table II), a range that no described *Sardinella* species possesses. Although co-occurring *S. lemuru* has the closest comparable lower gill raker count (77–188), the species share few other features (Table I) and misidentification was unlikely. Instead, it is inferred that variability in gill raker counts in *Sardinella* may be influenced by the fish's natal environment. In *Sardinella aurita*, sampled along the eastern seaboard of the U.S.A. gill raker counts in young fish were correlated with regions; however, this was lost as the fish grew in size (Kinsey *et al*., 1994). Likewise, Kinsey *et al*. (1994) also found the number of gill rakers of *S. aurita* from some locations fell below the diagnostic range in Whitehead (1985). This plasticity in the gill raker number has been described as more common in planktivores than with other feeding strategies because of the strong dependency on this organ for feeding (Hessen *et al*., 1988).

Furthermore, 10 *S. hualiensis* were sampled from Yilan County, along the eastern coast of Taiwan on 21 April 2011. Fish were measured for diagnostic features (Table II) and vouchers were preserved at the Academia Sinica, Taipei, Taiwan. These Taiwanese specimens also showed a slightly higher number of gill rakers than described by Chu & Tsai (1958), and, considering the aforementioned plasticity in gill raker counts, this could be because these fish were captured from a different location than the type location (Hualien County, Taiwan).

The phylogenetic relationship between *S. hualiensis* specimens from Taiwan and the northern Philippines was examined by amplifying a *c*. 543 bp fragment of the mitochondrial DNA (mtDNA) 16S region *via* polymerase chain reaction (PCR) using primers 16Sar and 16Sbr (Palumbi, 1996). Reactions consisted of 14 μl of 10× PCR buffer, 2·0 μl of 25 mM $MgCl_2$, 2·5 μl of each 10 mM dNTP, 1·25 μl of each primer, 0·2 units of Taq DNA polymerase and 1 μl template DNA in a final volume of 25 μl. The PCR parameters were an initial denaturation at 94° C for 10 min, 38 cycles of 94° C for 30 s, 45° C for 30 s, 72° C for 45 s and a final extension of 72° C for 10 min. The PCR product was purified for sequencing using ExoSap-It (USB Corp.; www.affymetrix.com) at 37° C for 30 min and 80° C for 20 min then sent to MacroGen, Inc. (www.macrogen.com) for sequencing. Sequences were cleaned and

TABLE II. Morphology and meristics of the 10 specimens of *Sardinella hualiensis* from Cagayan Province, Philippines. Means of diagnostic features of Taiwanese specimens collected from Yilan County, Taiwan, in April 2011 are also shown

Morphology/meristics/colouration	Specimen number										Mean	Mean of 10 Taiwan specimens
	1	2	3	4	5	6	7	8	9	10		
L_S(mm)	109	118	104	110	109	109	108	108	112	110	109·7	170·2
Body depth:L_S (%)	29	30	32	30	30	32	32	33	32	31	31·0	34·7
Head length (mm)	27	30	27	29	27	27	30	29	28	29	28·3	37·8
Snout (mm)	7	9	8	8	8	8	8	9	7	9	8·1	10·5
Eye diameter (mm)	8	8	8	8	7	8	8	8	8	8	7·9	10
Post orbital length (mm)	12	13	11	13	12	13	14	12	13	12	12·5	17·3
Pectoral fin length (mm)	21	23	20	21	20	22	21	20	21	21	21·0	32·0
Weight (g)	20·5	27·9	18·5	23·3	21	21·9	21·9	22·1	24·1	20·3	22·2	119·0
Scutes	31	30	30	31	31	30	29	31	30	31	30·4	31·2
Lower gill rakers	75	84	68	70	88	94	76	85	94	78	81·2	70
Dorsal fin rays	17	18	18	18	18	18	18	17	18	18	17·8	17·3
Pelvic fin rays	8	8	8	8	8	8	8	8	8	8	8·0	7·9
Pectoral fin rays	14	15	14	14	14	14	14	14	14	15	14·2	14·5
Anal fin rays	18	18	18	19	18	18	19	18	18	18	18·2	18·6
Enlarged last two anal fin rays	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes		
Scales with overlapping/continuous striae	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes		
Scales with numerous perforations	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes		
Black spot at dorsal fin origin	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes		
Tips of caudal fin black	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes		
Dorsal fin blackish	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes		
Black spot at margin of operculum	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes		

aligned in Sequencher v4.8 (GeneCode; www.genecodes.com) and ClustalX (Larkin *et al*., 2007), and sequence divergence and neighbour-joining phylogenetic tree were inferred in MEGA v5 (Tamura *et al*., 2011) using the Kimura two-parameter model (Kimura, 1980). Intraspecific genetic distance for *S. hualiensis* was 0·1%, whereas interspecific distance between *S. hualiensis* and the outgroups of *S. gibbosa* and the spotted sardine *Amblygaster sirm* (Walbaum 1792) were 9·2 and 17·8%, respectively. Neighbour-joining analysis revealed a single, strongly supported clade containing all *S. hualiensis* sequences from Taiwan and the northern Philippines that is distinct from the out-group congener *S. gibbosa* and *A. sirm* (Fig. 3). Sequences were submitted to GenBank and assigned the accession numbers JN580476–JN580491.

The Philippine specimens presented here share 14 of the 15 diagnostic traits with Chu & Tsai's (1958) original description of *S. hualiensis* (Table I) and with Taiwan *S. hualiensis* specimens in April 2011 from near the type location. The single exception is the gill raker count, and thus it is concluded that these are population-level differences and that these fish are *S. hualiensis*, particularly, as all other morphological characters clearly distinguish these specimens from all other *Sardinella* species in the Philippines. This is further confirmed by genetics showing a single clade of all

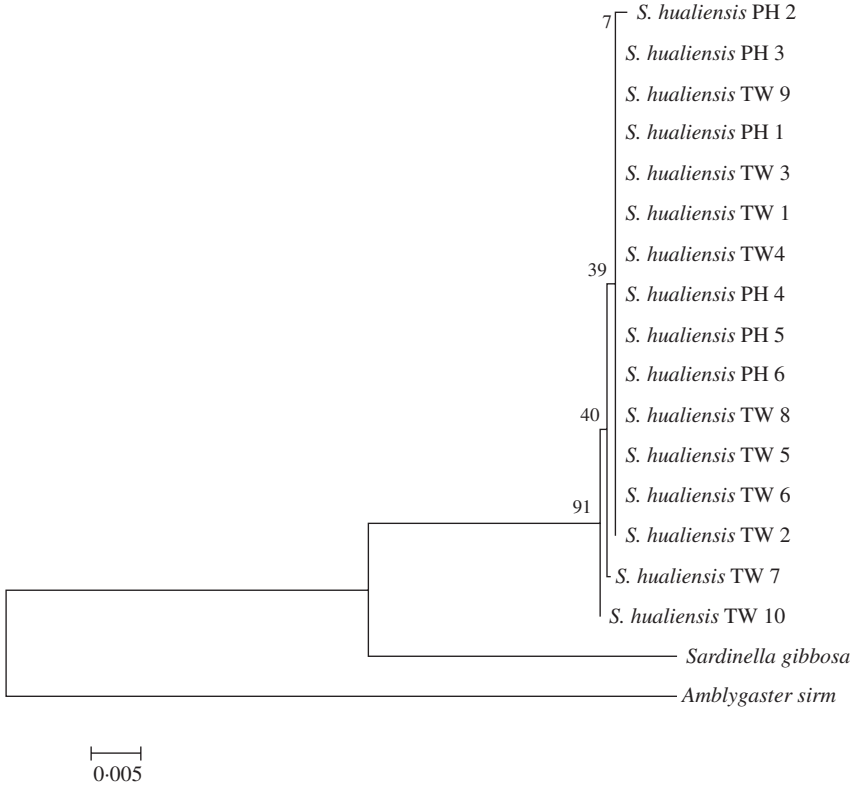

FIG. 3. Neighbour-joining tree of six *Sardinella hualiensis* from Cagayan, Philippines (PH), and 10 from Yilan County, Taiwan, (TW) for mtDNA 16S region showing a single clade. Node support based on 500 bootstrap replicates. The 16S sequences of *Sardinella gibbosa* and *Amblygaster sirm* from the Philippines (unpubl. data) were used as outgroups.

specimens supported by a high bootstrap value. Voucher specimens of the Philippines fish were fixed in formalin and are stored in 95% ethanol at the National Fisheries Research and Development Institute in Quezon City, Philippines.

These data raise interesting questions about the dispersal of small, pelagic planktivorous fishes. Cagayan Province, Philippines and Taiwan are separated by *c*. 360 km across the Luzon Strait. Lanyu and Hsiao Lanyu Island of Taiwan, and the southern tip of Taiwan previously represented the southernmost range limit of *S. hualiensis* (Froese & Pauly, 2010). Like other *Sardinella* species, *S. hualiensis* inhabits coastal waters with a depth $<$200 m, thus most of the Luzon Strait is an unsuitable habitat for *S. hualiensis* survival (Fig. 1). Furthermore, the prevailing north to north-west flow of the Kuroshio Current and less intense north-west Luzon Coastal Current create a south-to-north water regime that may limit the dispersal of *S. hualiensis* larvae south (Hu *et al*., 2000; Centurioni *et al*., 2004). An exception is during the summer monsoon when the seasonal Loop Current and north-west Luzon Cyclonic Eddy transport surface water from north to south towards the Philippines (Hu *et al.,* 2000). Although *Sardinella* embryonic development is short, eggs and newly hatched larvae typically float passively on the water surface for multiple days (Nair, 1959) and the late-larval stage of some clupeoids have been reported to have an exceptionally high survival rate in offshore eddy environments (Logerwell & Smith, 2001). Oceanographic data and life-history experiments support the working hypothesis that *S. hualiensis* was able to cross the Luzon Strait perhaps using the Taiwanese, Batanes and Babuyan Islands as stepping stones; however, it is unclear if the species originated in the Philippines and spread to Taiwan, or originated in Taiwan and moved to the Philippines. The present report confirms the presence of a sixth species of *Sardinella* in the Philippines and ranks the Philippines, alongside Taiwan and Indonesia, as second only to India for highest diversity of *Sardinella* species in the world.

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