An abnormal specimen of threatened Ryukyu-ayu (*Plecoglossus altivelis ryukyuensis*) with an additional pelvic fin

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Abstract. The present study describes a fish individual of *Plecoglossus altivelis ryukyuensis* with three pelvic fins collected from a hatchery-reared population in Amami-oshima Island, Japan. The additional pelvic fin had the same number of rays as the neighboring pelvic fins (eight fin rays), and its girdle was fused with that of the neighbor. The comparison of morphometric measurements and meristic counts of the abnormal specimen with those of the normal specimens revealed that this abnormality was not a syndrome when all other characteristics were in the expected ranges. The condition factor of abnormal fish was close to the ideal value, suggesting that this abnormality did not severely affect the activities of the fish under rearing conditions. Well-managed rearing conditions might contribute significantly to the survival and growth of abnormal fish. The cause of this abnormality is unclear, but the limited diversity in the genome of the subspecies is assumed to be a probable factor.
Key Words: abnormality, accessory fin, ayu, deformity, genetic stress.

Introduction. Morphological abnormalities have been well documented in fish, occurring in both wild and rearing conditions (Komada 1980), affecting various characteristics such as body shape, jaws, operculum, scales, spinal column, or fins (Divanach et al 1996). Generally, abnormalities have been found at a higher incidence in hatchery-reared populations than in wild populations, partly because a large percentage of such abnormal fish do not survive in the wild environment as in rearing conditions (Houde 1971; Komada 1980; Verhaegen et al 2007).

Several types of abnormalities in ayu (*Plecoglossus altivelis*), an amphidromous fish that is widely cultured in Japan, have been documented (e.g., bend of jaws, loss of operculum, loss of dorsal fin, loss of pelvic fin, spinal curvature, shortened spin, dumpy, and hump-backed (Komada 1974, 1980). However, no publications on Ryukyu-ayu (*Plecoglossus altivelis ryukyuensis*), an insular and endangered subspecies of ayu endemic to the Ryukyu Archipelago of Japan, have reported abnormalities in this subspecies (even abnormalities are frequently observed in culture tanks; first author's personal observation). From the collection of hatchery-reared Ryukyu-ayu specimens produced by Amami City on Amami-Oshima Island, a specimen possessing three pelvic fins was detected that was brought to the notice of the authors.

To the authors' knowledge, only a few cases of additional fins among fishes have been reported. These include the additional dorsal fin in *Labeo calbasu* (Sathyanesan 1962), *Salvelinus malma* (Blackett & Armstrong 1965), *Salmo trutta* (Raadik 1993); additional caudal fin lobe in *Esox lucius* (Quigley 2015); additional pectoral fin and additional pectoral fin filaments in the genus *Setipinna* (Gangan et al 2018); and an additional fin at an uncommon position in *Esox lucius* (Lawler 1964, 1966). However, no studies have recorded the presence of additional pelvic/ventral fins in fish. This article is the first record of additional pelvic fins in fish, especially for this species.

Material and Method. One fish specimen of Ryukyu-ayu with three pelvic fins (abnormal specimen, henceforth) was obtained from the collection of fish specimens of hatchery-reared Ryukyu-ayu that were artificially bred and reared at the Ayu Observation Center, Amami City, Amami-oshima Island, Japan (a small hatchery and nursery facility with less than ten thousand fish cultured each year for conservation purposes). Nineteen normal specimens from the same collection were examined for comparison. The specimens were collected after natural death in the culture tank in December 2019, fixed in 10% formalin solution for two days, and preserved in 70% ethanol.

The morphometric dimensions were measured to the nearest 0.01 mm using a digital caliper following the method by Nishida (1985), and body weight was measured using an analytical balance to the nearest 0.01 g. The sex of the specimens was determined based on the shape of the anal fin (Iguchi et al 1991). Meristic characteristics were determined using a dissecting microscope (Nikon C-LEDs; Nikon Corporation, Tokyo, Japan). The relative body condition factor (K) was calculated according to Le Cren (1951). Specimens were stained and cleared using Alcian blue, alizarin red, and trypsin (Potthoff 1984) to visualize insight structures such as the vertebral column, fin girdles, and pterygiophores. Since the specimens were not completely cleared for unknown reasons, they were dissected to observe the osteological structures. Morphological abnormalities were photographed using a digital camera.

To examine whether this type of abnormality was observed in this species, the authors verbally surveyed ayu aquaculturists in Japan on whether they had encountered a fish with an additional fin during their career.

Results and Discussion. The abnormal specimen was a male with a standard length of 150.45 mm and body weight of 45.08 g (Table 1).

Table 1

Comparison of morphometric dimensions and body condition factor between the abnormal three-pelvic fin specimen and normal specimens of Ryukyu-ayu; body condition factor (K) was calculated using the relative condition factor (Le Cren 1951) (NA indicates that data is not available)

Measurements	*Abnormal specimen*	*Normal specimens (n = 19)*	
		Range	*Mean±SD*
Standard length (SL)	150.45	108.83-183.55	150.16±18.18
Body weight	45.08	17.54-80.19	48.00±15.35
Body condition factor (K)	0.96	0.82-1.26	1.01±0.09
Ratio between morphometric dimensions and standard length (%)			
Head length/SL	19.72	15.96-26.61	20.88±2.47
Snout length/SL	6.25	5.67-8.17	6.55±0.70
Postorbital distance/SL	9.23	6.58-13.30	10.02±1.53
Eye diameter/SL	4.23	3.60-5.66	4.72±0.51
Upper jaw length/SL	11.00	7.31-11.42	9.36±0.95
Lower jaw length/SL	8.99	7.26-10.86	8.99±0.89
Prepectoral distance (right)/SL	20.17	15.19-25.66	20.21±2.72
Prepectoral distance (left)/SL	20.22	15.89-26.50	20.68±2.83
Prepelvic distance (right)/SL	48.64	36.82-58.09	48.49±5.14
Prepelvic distance (left)/SL	49.24	36.73-59.53	48.48±5.31
Prepelvic distance (additional fin)/SL	43.53	NA	NA
Predorsal distance/SL	55.84	17.21-57.14	44.82±11.02
Preanal distance/SL	68.90	53.76-84.57	70.24±7.27
Depressed dorsal fin length/SL	25.72	13.89-32.57	23.28±5.51
Dorsal fin base length/SL	16.45	7.64-18.05	13.99±3.12
Anal fin length/SL	7.21	7.53-12.02	9.49±1.23
Anal fin base length/SL	22.40	11.13-25.16	18.76±3.73
Right pectoral fin length/SL	14.50	10.94-20.17	15.71±3.04
Left pectoral fin length/SL	15.15	11.02-20.16	15.93±2.98
Right pelvic fin length/SL	15.79	9.19-18.76	14.66±3.16
Left pelvic fin length/SL	14.44	9.13-19.70	14.98±3.54
Additional pelvic fin length/SL	10.95	NA	NA

The additional fin was attached anteriorly to the original pelvic fin on the left side (dorsal view) (Figure 1). Except for the additional pelvic fin, other morphometric dimensions and meristic counts of the abnormal specimens were in the range of those of normal specimens (Tables 1 and 2). Observation of the osteological structure did not reveal any other abnormalities in the specimen.

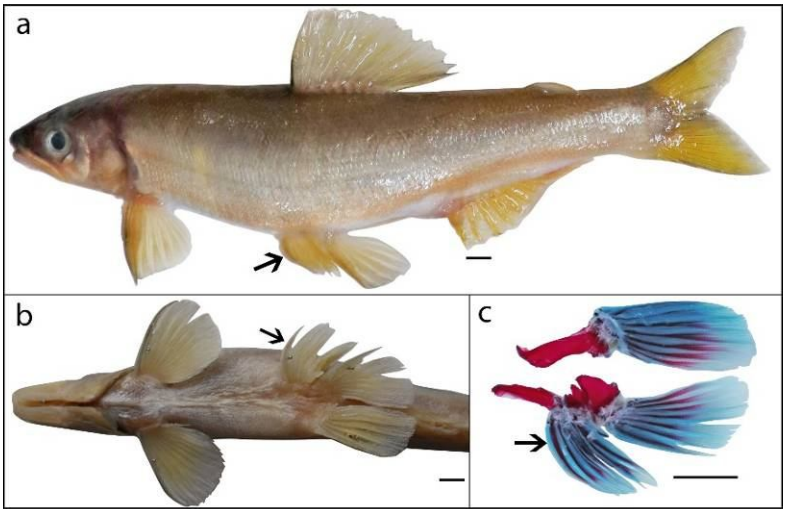

Figure 1. Abnormal specimen of Ryukyu-ayu with an additional pelvic fin; (a) lateral view, (b) ventral view, (c) skeletal structure of pelvic fins; arrows indicate the position of the additional pelvic fin, scale bars are 1 cm.

Table 2

Comparison of meristic counts between the abnormal three-pelvic-fin specimen and normal specimens of Ryukyu-ayu (NA indicates that data is not available)

Meristic characteristics	*Abnormal specimen*	*Normal specimens (n = 19)*		
		Range	*Mean±SD*	*Mode*
Dorsal fin rays	12	10-12	11.17±0.46	11
Dorsal fin pterygiophores	12	11-12*	11.60±0.55	11
Anal fin rays	17	16–19	17.70±0.7	18
Anal fin pterygiophores	16	16*	16.00±0.00	16
Pectoral fin rays (right)	13	12-14	12.77±0.50	13
Pectoral fin rays (left)	13	12-14	12.93±0.45	13
Pelvic fin rays (right)	8	7-8	7.56±0.50	8
Pelvic fin rays (left)	8	7-8	7.76±0.43	8
Pelvic fin rays (additional fin, right side)	8	NA	NA	NA
Gill raker (right)	38	33-39	36.03±1.43	37
Gill raker (left)	35	33-38	36.00±1.36	37
Comb-like teeth on lower jaw (right)	13	11-13	11.57±0.57	12
Comb-like teeth on lower jaw (left)	13	10-14	11.57±0.73	13
Comb-like teeth on upper jaw (right)	13	11-13	11.26±0.58	11
Comb-like teeth on upper jaw (left)	13	10-12	11.20±0.71	11
Scale above the lateral line	14	11-15	13.23±1.25	14
Scale below the lateral line	11	8-13	10,67±1.30	10
Scale on longitudinal row	118	105-130	116.03±5.33	115
Pored scale on the lateral line	68	64-67	65.67±0.96	65

*n = 5.

The additional fin of the abnormal specimen consisted of eight rays, equal to the normal pelvic fin of the same individual; the length of the fin was 15.93 mm, which was shorter than the two original fins (22.97 and 21.01 for the right and left fins, respectively). Osteological examination revealed that the additional fin was articulated and flexible

(Figure 1c). The girdle structure of the additional fin was fused with that of the nearby fin to form one unit (Figure 1c).

Seemingly, the additional pelvic fin in the present case did not severely affect the activity of this individual (in rearing conditions), as its relative condition factor was still close to the ideal condition (the value of 1) at 0.96. Fish eggs of this hatchery, where the abnormal individual was collected, were incubated in relatively stable environmental conditions (clean freshwater, stable water temperature, and dissolved oxygen), which might enable embryos with the potential to develop abnormalities, as in this case, to survive. The rearing tank, supplied with sufficient food, made it easier for abnormal fish to obtain food. In addition, hatchery-reared fish did not have to face predation risks or risks in the migration route like fish in the wild, allowing abnormal individuals to live and grow at a rate similar to that of normal fish.

The cause of this abnormality in the present case is unclear. Abnormalities in animals are caused by various factors, including environmental contaminants, temperature disturbance, nutrient insufficiency, parasites, predators, inbreeding depression, and genetic mutations (Walker & Taylor 1965; Hore & Ahmad 2010; Tave et al 2011; Noble et al 2012; Jose et al 2020). In particular, the phenomenon of having extra limbs has been well documented in anurans, and the inducing factor was determined to be a parasite infection that interrupts the proper limb formation process (Johnson et al 2002). Although most abnormalities are non-heritable, they are commonly considered related to genetic degradation (Thomspon & Adams 1936; Nelson 1971; Tave 1993).

For the population where this abnormal specimen was collected, genetic diversity examined by mitochondrial DNA was very low due to genetic drift through inbreeding; thus, individuals in this population might have suffered from inbreeding depression (Ha et al 2021). Due to limited genetic diversity, recessive alleles are easier to express; therefore, deleterious errors occur more easily and may cause genetically induced abnormalities. In addition, when genetic diversity is low, the capability of the genome to self-repair errors that occur during ontogeny may decrease. In fact, the deleterious effect of low genetic diversity on this population was expressed by a high incidence of fluctuating asymmetry (non-directional differences between the left and right sides of bilateral traits, an indicator of developmental instability) (Ha et al 2021). We can assume that the abnormality in the present report is a type of fluctuating asymmetry but at a higher hierarchy (the meristic count is the fin unit).

Among the nominotypical subspecies in mainland Japan, hatchery-reared populations that have persisted for multiple generations without introducing any exotic broodstock can easily lose their genetic diversity (Iguchi et al 1999). However, although approximately two million individuals are cultured at each farm every year, fish with additional fins have never been observed (Tsuboi J., personal communication). This suggests that additional fin abnormalities are unique to Ryukyu-ayu. The uniqueness of this subspecies should be considered from two points of view: the population contains deleterious mutations or the population has an extremely limited genetic basement. However, having an additional fin is assumed to be a severe abnormality for fish in the wild; if a deleterious allele causes it, the allele will have already been purged during the evolutionary process. Therefore, we incline to the hypothesis that the genetic basement (which is not reflected in the diversity of mitochondrial DNA) of this subspecies is limited compared to that of the nominotypical subspecies.

The normal pelvic fins of fish act as static trimming in locomotion activities and function in stabilizing, braking, and diverting; thus, an abnormality in the pelvic fins may negatively affect the survival and welfare of fish (Standen 2011). If this type of abnormality occurs in fish in the wild environment, fish with an additional fin might have many difficulties in locomotion and controlling their balance, especially for migratory fish. Thus, their early mortality can be higher than expected and negatively affect population dynamics.

Conclusions. This is the first time a fish with an additional pelvic fin was reported. The abnormal fish in the present study might have difficulties in locomotion but remained alive by the well-managed cultural conditions. The additional pelvic fin seems to occur only in the Ryukyu-ayu subspecies. The cause of this abnormality might relate to the limited genetic basement of the Ryukyu-ayu.

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Conflict of interest. The authors declare that there is no conflict of interest.

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