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A New Species of Lentic Breeding Korean Salamander of the Genus *Hynobius* (Amphibia, Urodela)

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ABSTRACT—A new species of salamander, *Hynobius yangi*, is described from Kori (=Hyoam-ri), Busan-shi, southeastern Korea. It is a lentic breeder belonging to the *H. nebulosus* species group, and has long been confused with another Korean species *H. leechii*. The new species, however, is genetically substantially differentiated from Korean and Japanese relatives. *Hynobius yangi* is morphologically very similar to *H. leechii*, but could be differentiated from it by the tail shape, degree of limb separation, shape of vomerine teeth series, shape of egg sac, and dorsal coloration.

Key words: cryptic species, *Hynobius yangi*, *Hynobius leechii*, *Hynobius quelpaertensis*, speciation

INTRODUCTION

In the recent decade, presence of several unique populations among a Korean salamander *Hynobius leechii* Boulenger, 1887 has been repeatedly reported in South Korea (Lee and Jung, 1993; Cha and Lee, 1995; Yang *et al.*, 1997, 2000; Lee *et al.*, 1998) and distinct specific status of two forms from the nominal species has been suggested (Yang *et al.*, 1997). One of which from Jeju (=Cheju) Island and peripheral islands has already an available name, *H. quelpaertensis* (Mori, 1928), but another from Kori (=Hyoam-ri and Gilcheon-ri), Busan-shi, has never been formally evaluated taxonomically.

Our morphological survey including the population from Kori and many populations of south Korean *H. leechii* and *H. quelpaertensis*, as well as two Japanese relatives, *H. nebulosus* from Nagasaki and *H. tsuensis* from Tsushima, proved the former assumption that the salamander from Kori could be constantly distinguished from other species by the combination of several morphological characteristics. In this short article, we describe the salamander from Kori, South Korea as a new species on the basis of the results of morphological analysis and hitherto accumulated information.

SYSTEMATICS

Hynobius yangi sp. nov.

[Korean name : Kori-dorongnyong]

[Japanese name : Kori-sansyouo]

Fig. 1, Fig. 2

Hynobius leechii (part): Dunn, 1923. p. 456.

Hynobius leechii (part): Sato, 1943. p. 33.

Hynobius leechii (part): Cha and Lee, 1995. p. 89.

Hynobius leechii Form-C: Yang *et al.*, 1997. p. 253, Fig. 3C.

Hynobius leechii (part): Lee *et al.*, 1998. p. 155.

Hynobius sp.: Yang *et al.*, 2000, p. 44.

Holotype: KUHE (Graduate School of Human and Environmental Studies, Kyoto University) 29842, an adult male from Hyoam-ri, Jangan-eup, Gijang-gun, Busan-shi (129°17'E, 35°19'N, alt. 45m), collected by Jong-Bum Kim on 26 February 2002.

Paratypes: KUHE 29843-29849 and 29851-29865, 17 males and 3 females, with the same data as the holotype.

Diagnosis: A member of the lentic breeder of the *Hynobius nebulosus* group of Sato (1943): living on hilly regions and breeding in still waters in ditches; most similar to some populations of *H. leechii* in appearance, but is distinguished from it by longer and higher tail relative to snout-vent length (SVL), greater separation of adpressed limbs, slightly deeper vomerine teeth series (Fig. 3A, B), egg sac of coil shape (Fig. 4), light brownish dorsum, and distinct genetic differences. It has smaller SVL, relatively longer head, higher tail, and shorter trunk, and more costal grooves than

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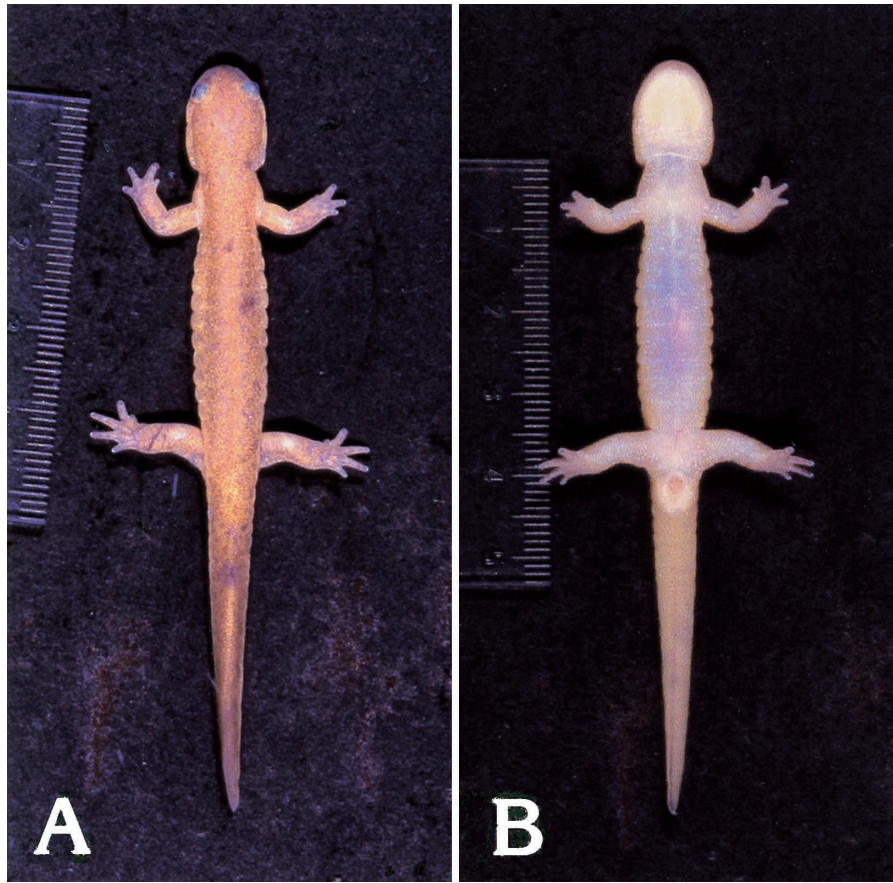


Fig. 1. Dorsal (A) and ventral (B) views of holotype of *Hynobius yangi* sp. nov. (KUHE 29842). Scale=1 mm.



Fig. 2. Living holotype of *Hynobius yangi* sp. nov. (KUHE 29842), male, 49.1 mm SVL.

H. quelpaertensis from Jeju. From *H. quelpaertensis* from peripheral islands, *H. yangi* can be differentiated by relatively longer and higher tail and wider head, and by having more costal grooves and narrower separation of limbs. It differs from *H. tsuensis* by smaller SVL, higher tail, much shallower vomerine teeth series (Fig. 3A, D), and usually unspotted dorsum. From *H. nebulosus*, *H. yangi* can be differentiated by smaller SVL, longer tail and wider head relative to SVL, less costal grooves, smaller separation of limbs, much shallower vomerine teeth series, and lack of yellowish light stripes on top and base of the tail.

Description and variation: The following description is based on the maximum number of 18 adult males and 3 adult females. Morphometric data are summarized in Table 1 together with those of the allied species, *H. leechii*, *H. quelpaertensis*, *H. tsuensis*, and *H. nebulosus*. Head longer than broad. Males with relatively larger head in length (HL) than in females, when each dimension is converted to percentage ratio to SVL (Mann-Whitney *U* test, $U_{18,3}=3$, $p<0.05$; Table 1). Females with relatively longer trunk than males ($U_{18,3}=3$, $p<0.05$). Number of costal grooves including axillary groove 12 to 14, the modal number being 14 in both sexes (Table 2). Limbs short, and when adpressed, they mainly do not overlap. Separation is greater in females (median=2.5 folds) than in males (median=0 fold; $U_{18,3}=2$,

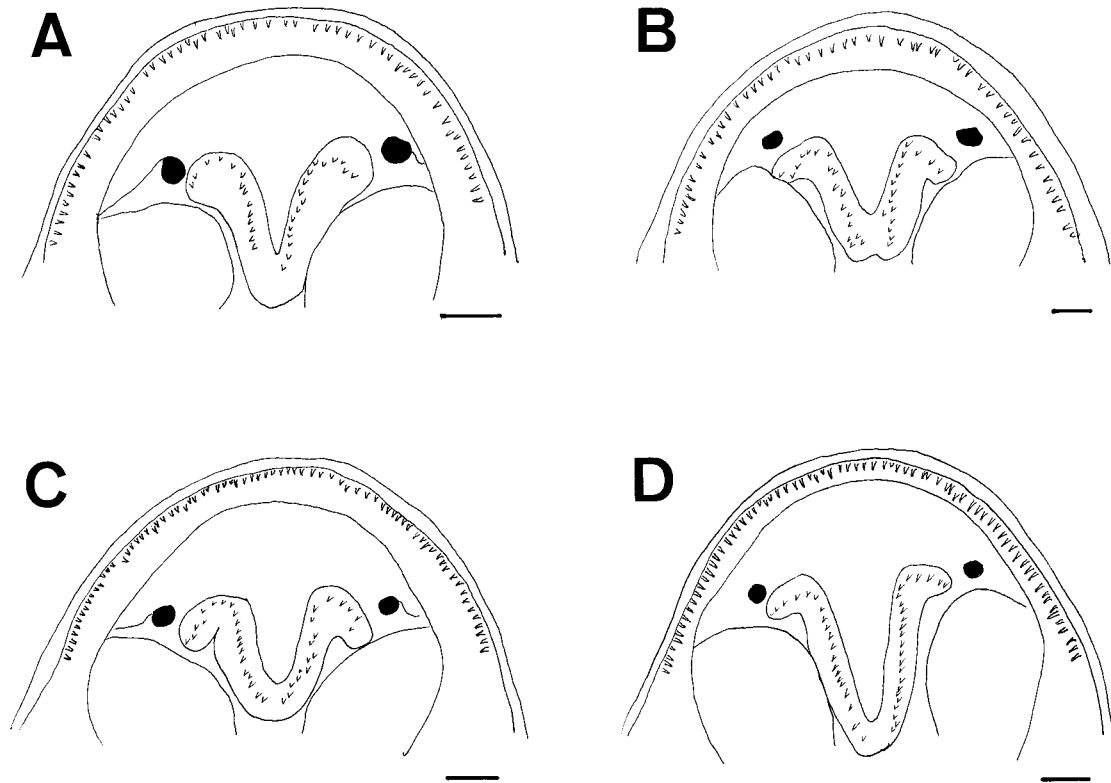


Fig. 3. Open mouths of *Hynobius* species showing variations in shape of vomerine teeth series. A: *H. yangi* sp. nov.; B: *H. leechii*; C: *H. quelpaertensis* from Jeju Isl.; D: *H. tsuensis*. Scales=1mm.

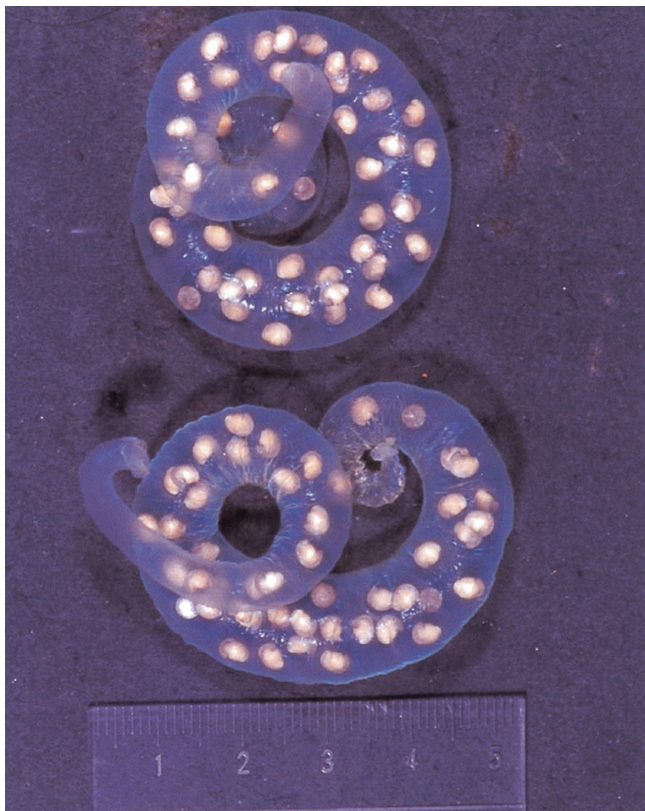


Fig. 4. A pair of egg sacs of *Hynobius yangi* sp. nov.

$p < 0.01$, Table 3). Statistically significant sexual dimorphism in tail shape: males with relatively longer ($U_{18,3}=0$, $p < 0.01$) and higher ($U_{18,3}=0$, $p < 0.01$) tail than in females. Fifth toe usually present but rudimentary in some specimens. Vomerine teeth in two small, obliquely arched series, nearly touching at midline, and forming shallow "V" shape. Combined series distinctly wider than long (VTW/VTL 1.24–1.89 in males and 1.30–1.52 in females). Shape of vomerine teeth series are not sexually different as shown by comparison of the ratio of VTW to VTL ($U_{16,3}=9$, $p > 0.05$, Table 4).

Color in life: Dorsum olive without dark speckles (Fig. 2), but some specimens have dark brownish dorsum dotted very finely with yellow speckles. These speckles tend to fuse on sides of the tail. Underside of body lighter than dorsum.

Color in preservative: Dorsum changes into gray or dark gray color in alcohol.

Measurements and counts of the holotype: An adult male with the following measurements (mm; in preservative): Head width (HW) 10.9, snout to gular fold (HL) 11.8, head depth at posterior angle of jaw 4.2, eyelid length 3.0, anterior rim of orbit to snout 4.2, horizontal orbit diameter 2.1, interorbital distance 3.3, snout to insertion of forelimb 17.4, distance separating internal nares 3.1, distance separating external nares 3.1, snout to anterior angle of vent (SVL) 49.1, axilla to groin 23.4, anterior angle of vent to tip of tail (tail length, TAL) 38.8, tail width at base 5.7, tail height at base 5.6, tail height at middle 5.9, maximum tail height

Table 1. Comparisons of SVL measurements (means±SD, followed by ranges in parenthesis, in mm) and percentage ratios of each character dimension to SVL (medians, followed by ranges in parenthesis) in *Hynobius yangi* sp. nov., *H. leechii*, *H. quelpaertensis*, *H. tsuensis*, and *H. nebulosus*.

Species	Sex	N	SVL	HL/SVL	TRL/SVL	TAL/SVL	HW/SVL	MTAH/SVL
<i>H. yangi</i>	M	18	52.6±4.4 (42.4–62.6)	24.1 (22.0–26.5)	75.9 (73.5–78.0)	82.2 (77.8–90.8)	20.0 (17.8–22.2)	11.6 (9.2–15.2)
	F	3	57.8±2.4 (56.8–61.4)	22.1* (21.8–22.3)	77.9* (77.7–78.2)	73.4* (68.3–75.9)	18.4 (18.3–19.4)	10.2* (9.2–10.3)
<i>H. leechii</i>	M	99	56.2±7.4 (43.1–75.4)	23.9 (20.4–27.1)	76.1 (72.9–79.6)	79.7* (61.1–93.5)	19.2 (16.3–23.5)	9.5* (7.4–12.6)
	F	4	59.8±4.6 (55.8–65.2)	23.9 (22.7–25.4)	76.1 (74.6–77.3)	73.1 (68.4–86.4)	16.5 (15.9–18.3)	7.4 (6.8–9.0)
<i>H. quelpaertensis</i>								
(Jeju)	M	34	60.4±3.7* (51.2–66.6)	23.0* (21.5–26.1)	77.0* (73.9–78.5)	87.1 (72.9–94.8)	20.1 (16.6–21.7)	10.4* (7.65–13.3)
	F	2	62.3±4.5 (59.1–65.5)	23.3 (22.8–23.9)	76.7 (76.1–77.3)	53.4 (50.8–56.0)	15.9 (15.1–16.8)	7.6 (6.6–8.6)
(Islands)	M	41	52.5±3.8 (43.6–64.4)	24.7 (20.2–27.1)	75.3 (72.9–79.8)	80.8* (65.7–91.1)	18.1* (16.0–22.1)	10.1* (7.05–14.0)
	F	4	61.3±4.1 (56.2–67.6)	23.9 (21.9–24.4)	76.1 (75.6–78.1)	73.0 (72.0–81.0)	16.7 (14.8–17.5)	7.8 (7.6–8.4)
<i>H. tsuensis</i>	M	9	61.9±6.2* (52.6–73.8)	23.1 (22.5–24.7)	76.9 (75.3–77.5)	82.3 (71.3–85.9)	19.5 (17.3–20.7)	10.7* (9.6–11.6)
	F	4	62.1±3.3 (56.1–63.4)	23.0 (22.2–23.7)	77.0 (76.3–77.8)	70.0 (66.1–73.9)	18.7 (18.6–19.9)	8.9
<i>H. nebulosus</i>	M	31	59.3±4.5* (50.9–66.9)	23.6* (21.0–25.7)	76.4 (74.3–79.0)	70.5* (62.2–80.4)	18.9 (17.2–20.4)	12.5 (9.6–14.5)
	F	1	66.9	22.0	78.0	63.1	17.2	11.1

SVL=snout-vent length; HL=Head length; TRL=trunk length; TAL=tail length; HW=head width; MTAH=maximum tail height.

* significantly different from male *H. yangi* at $p<0.05$.**Table 2.** Variation in the number of costal grooves, including the axillary groove, in *Hynobius yangi* sp. nov., *H. leechii*, *H. quelpaertensis*, *H. tsuensis* and *H. nebulosus*.

Species	Sex	N	Number of costal grooves		
			12	13	14
<i>H. yangi</i>	M	18	3	4	11
	F	3			3
<i>H. leechii</i>	M	98	10	49	39
	F	3	1	2	
<i>H. quelpaertensis</i>					
(Jeju)	M*	20	1	18	1
	F	3	3		
(Islands)	M*	40	13	18	9
	F	4	1	2	1
<i>H. tsuensis</i>	M	8		3	5
	F	4			4
<i>H. nebulosus</i>	M*	31	3	26	2
	F	1		1	

* significantly different from male *H. yangi* at $p<0.05$.

(MTAH) 5.8, axilla to tip of out-stretched forelimb 13.9, groin to tip of out-stretched hindlimb 15.3, width of vomerine teeth series (VTW) 2.79, length of vomerine teeth series (VTL) 1.77. The holotype has 54 upper jaw teeth, 31 vomerine teeth, 13 costal grooves between axilla and groin, 1.5 costal folds between adpressed limbs, and 5 digits on both hindlimbs.

Etymology: The specific name “yangi” is a patronymic noun in the genitive singular, honoring Dr. Suh-Yung Yang. He contributed much to the study of South Korean animals.

Range: Known so far from the type locality Hyoam-ri and nearby Gilcheon-ri, both in Jangan-eup (=Changan-eub), Gijang-gun (=former Yangsan-gun), Busan-shi (formerly included in Kyungsangnam-do), southeastern South Korea (Fig. 5).

Morphological comparisons: Limited number of available female specimens prohibited detailed interspecific comparisons and only male specimens were compared. In SVL, *H. yangi* was significantly smaller than *H. quelpaertensis* from Jeju, *H. tsuensis*, and *H. nebulosus* (Tukey test, $p<0.05$; Table 1).

By comparing percentage ratios of each character dimension to SVL, the following significant differences (*U*-

Table 3. Variation in the number of costal folds between addressed limbs in *Hynobius yangi* sp. nov., *H. leechii*, *H. quelpaertensis*, *H. tsuensis*, and *H. nebulosus*.

Species	Sex	N	Overlap of adressed limbs shown by number of costal folds													
			−3.5	−3	−2.5	−2	−1.5	−1	−0.5	0	0.5	1	1.5	2	2.5	3.0
<i>H. yangi</i>	M	18				2		5	2	7	1	1				
	F*	3		1	1		1									
<i>H. leechii</i>	M*	99		1		7	3	14	2	22	3	32	7	6	1	1
	F	4	1	1	1	1										
<i>H. quelpaertensis</i>																
(Jeju)	M	33				2	3	13	2	12		1				
	F	2		1		1										
(Islands)	M*	41		1	1	7	5	16	1	5		4		1		
	F	4	1	2		1										
<i>H. tsuensis</i>	M	8				2	1	3		2						
	F	4		1	1	1		1								
<i>H. nebulosus</i>	M*	31		4	6	8	5	4	3	1						
	F	1		1												

* significantly different from male *H. yangi* at $p < 0.05$.**Table 4.** Comparisons of width-length ratio of vomerine teeth series (median) in *Hynobius yangi* sp. nov., *H. leechii*, *H. quelpaertensis*, *H. tsuensis*, and *H. nebulosus*. Figures in parenthesis indicate variation range.

Species	Sex	N	VTW/VTL
<i>H. yangi</i>	M	16	1.62 (1.24–1.89)
	F	3	1.37 (1.30–1.52)
<i>H. leechii</i>	M*	55	1.78 (1.41–2.17)
	F	3	1.64 (1.26–1.91)
<i>H. quelpaertensis</i>			
(Jeju)	M	18	1.73 (1.28–2.13)
	F	2	1.73 (1.67–1.78)
(Islands)	M	24	1.50 (1.00–1.82)
<i>H. tsuensis</i>	M*	6	1.13 (0.98–1.27)
	F	3	1.09 (0.94–1.18)
<i>H. nebulosus</i>	M*	31	1.04 (0.85–1.32)
	F	1	1.00

* significantly different from male *H. yangi* at $p < 0.05$.

test, $p < 0.05$) in body shape were detected. *Hynobius yangi* was greater in relative TAL ($U_{18,82}=430$, $p < 0.01$) and relative MTAH ($U_{18,8}=204.5$, $p < 0.01$) than *H. leechii*. It had greater values in relative HL ($U_{18,34}=202$, $p < 0.05$) and relative MTAH ($U_{18,34}=121$, $p < 0.01$), but had smaller value in relative TRL ($U_{18,34}=202$, $p < 0.05$) than *H. quelpaertensis* from Jeju. Compared with *H. quelpaertensis* from peripheral islands, *H. yangi* had greater values in relative TAL ($U_{18,36}=212.5$, $p < 0.05$), relative MTAH ($U_{18,34}=105$, $p < 0.01$), and relative HW ($U_{18,41}=108.5$, $p < 0.01$). *Hynobius yangi* was greater in relative MTAH than *H. tsuensis* ($U_{18,6}=15$,

$p < 0.05$), and greater in relative TAL ($U_{18,19}=5$, $p < 0.05$) and relative HL ($U_{18,31}=109$, $p < 0.01$) than *H. nebulosus* (Table 1).

In males, *H. yangi* had limbs more separated than in *H. leechii* ($U_{18,99}=538$, $p < 0.01$), but less so than in *H. quelpaertensis* from peripheral islands ($U_{18,41}=247.5$, $p < 0.05$) and *H. nebulosus* ($U_{18,31}=72.5$, $p < 0.01$) (Table 3).

Hynobius yangi had vomerine teeth series (as expressed by the ratio of width and length) slightly deeper than *H. leechii* ($U_{15,57}=209$, $p < 0.01$), but much shallower than *H. tsuensis* ($U_{15,5}=1$, $p < 0.05$) and *H. nebulosus* ($U_{15,31}=2$, $p < 0.01$) (Table 4).

Hynobius yangi had costal grooves more than *H. quelpaertensis* from Jeju ($U_{18,34}=134$, $p < 0.01$) and peripheral islands ($U_{18,39}=199.5$, $p < 0.01$) and *H. nebulosus* ($U_{18,31}=159.5$, $p < 0.01$) (Table 2, Fig. 3A, D).

Biochemical characteristics: *Hynobius yangi* differs from *H. leechii* completely at Ldh-1 allele and nearly completely at Ldh-2, Gp-2, and Xdh-2 alleles. From *H. quelpaertensis*, it differs completely at Ldh-1 and Ldh-2 alleles and nearly completely at Aat-2 and Xdh-2 alleles (Yang *et al.*, 1997). In partial sequence of cytochrome b gene (246 bp), *H. yangi* (as *H. leechii* from Yangsan) is reported to have mean Kimura's two-parameter distance (Kimura, 1980) of 7.8% from *H. leechii* and 10.9% from *H. quelpaertensis* (Lee *et al.*, 1998).

Karyotype: $2N=56$ with Ag-NORs constantly at 23rd chromosome, in contrast to great intrapopulation variations in *H. leechii* (Cha and Lee, 1995).

Fecundity and natural history: The clutch size of one female was 86. The mean diameter of ten ova just oviposited by one female ranged from 2.70–2.84 (mean \pm SD = 2.78 ± 0.05) mm. The animal pole is dark brown and vegetal

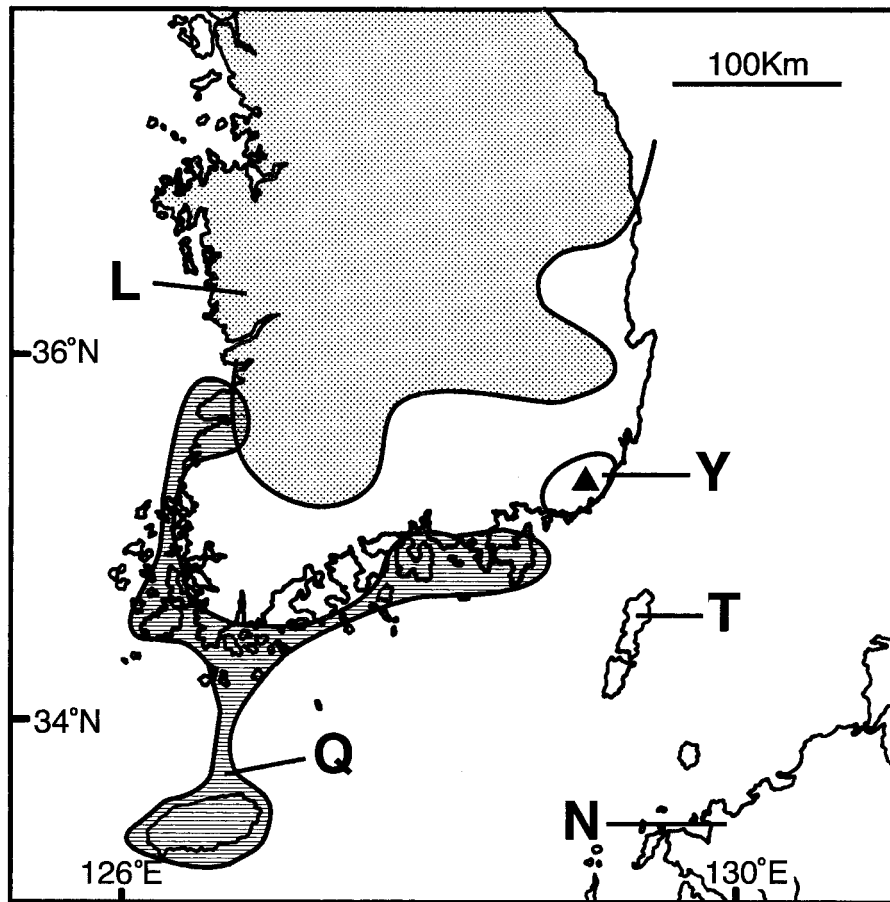


Fig. 5. Map of South Korea and north western Kyushu showing distribution of the *Hynobius leechii* complex. Y: *H. yangi* sp. nov.; L: *H. leechii*; Q: *H. quelpaertensis*; T: *H. tsuensis*; N: *H. nebulosus*. A triangle indicates the type locality of *H. yangi*.

pole is grayish brown in color. Breeding occurs from late February to late March. Egg sacs of coil shape (Fig. 4) lack striations on the surface and are attached to water plants or fallen branches in the still-water.

Protection: Because the new species has been confused with wide-ranging *H. leechii*, no conservation measures have been taken. However, since the habitats of *H. yangi* are limited and being destroyed, its immediate protection is needed.

DISCUSSION

As a by-product of genetic divergence during speciation process, most animal species acquire morphological differences that are suitable for diagnostic purpose, but cryptic species fail to acquire such differences (Mayr and Ashlock, 1991). Korean salamanders of the *H. leechii* complex (= *H. leechii*, *H. quelpaertensis*, and *H. yangi*) seems to correspond to such a case.

From results of studies using different methods, some Korean authors have already reported that South Korean *H. leechii* actually includes three putative cryptic species that are morphologically very difficult to distinguish (Yang *et al.*, 1997, 2000; Lee *et al.*, 1998). Of these three species, *H.*

leechii was originally described from Wonsan (Gensan), north Korea (Boulenger, 1877) and actual taxonomic assignment of South Korean populations remains undone because of current political problems. The second species, now called *H. quelpaertensis*, was originally described from Jeju Island as a subspecies of *H. leechii* by Mori (1928), but has long been synonymized with the nominotypical subspecies (e.g., Sato, 1943). Close affinities of the populations from islands peripheral to the Korean Peninsula to *H. quelpaertensis* (see Yang *et al.*, 2000) has been clarified by biochemical studies (Lee and Jung, 1993; Yang *et al.*, 1997). The third putative species represented by the population from Kori (Yang *et al.*, 1997=Yangsan in Lee *et al.*, 1998) had no name until present study.

Among lines of evidence proposed, these species are especially clearly split by several diagnostic allelic differences, and the population from Kori (now *H. yangi*) is split from *H. leechii* and *H. quelpaertensis* with Nei (1972) D of 0.25 and 0.30, respectively (Yang *et al.*, 1997). The minimum genetic distance reported for seven closely related species of lentic-breeding *Hynobius* is 0.22 (Matsui, 1987), and compared with interspecific genetic differentiations in these Japanese members, genetic divergences among three forms of the Korean *H. leechii* complex seem to be

substantially large to regard each of them as heterospecific.

Also, an analysis of mitochondrial cytochrome b gene partial sequence divergence of the Korean *H. leechii* complex revealed that *H. yangi* was separated from *H. leechii* and *H. quelpaertensis* at the level ordinary found between different species (Kimura's two-parameter distance >7.8%; Lee *et al.*, 1998). Further, the three putative species differ significantly in some skeletal characters. The population from Kori has been reported to be different from *H. leechii* and *H. quelpaertensis* in the numbers of vomerine teeth and coccyx (Yang *et al.*, 1997). In the present comparisons of external morphology of salamanders from Korea and Japan proved that *H. yangi* could be constantly separated from the other species by the combination of several characters. Thus, distinct specific position of *H. yangi* is now undoubtful.

Hynobius yangi is isolated in the Milyang sub-basin of the Kyeongsang basin in southeastern Korea, and is split in the north from a population of *H. leechii* by the Taehwa river and in the south from an islet population of *H. quelpaertensis* by a small strait (Fig. 5). River basin is regarded as one of intra-continental barriers that might have played an important role in amphibian speciation processes (e.g., Arntzen and Garcia-Paris, 1995; Garcia-Paris *et al.*, 1998), and the Taehwa river in the Milyang sub-basin might have been an important barrier for genetic interchange between *H. yangi* and *H. leechii* that inhabit hilly or montane regions. Although distributional and divergence patterns similar to *H. yangi* and *H. leechii* have never been reported, future studies of various lower vertebrates in this area might contribute to the understanding of their speciation processes within South Korea.

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