

Comparison of vomerine tooth rows in juvenile and adult *Hynobius guabangshanensis* (Urodela: Hynobiidae)

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Abstract

In this note, the vomerine tooth rows of juveniles and adults *H. guabangshanensis* are described and compared. The vomerine tooth rows are long and posteriorly directed, and arranged in a V-shape in adults while short and slightly arched, parallel to the premaxilla and maxilla in juveniles. The vomerine tooth rows of juveniles are similar to the aquatic salamanders, which feed by suction in water, and the vomerine tooth rows function in hindering escape of prey. The vomerine tooth rows of adults are resemble with terrestrial salamanders, which use inertial feeding or protruding tongue to capture prey, and the vomerine tooth rows function in holding and delivering the prey. Hence, the differences of the vomerine tooth rows reflect the differences of life habit, feeding modes and function among different development period.

Key words

Hynobiid salamander, ontogeny, morphology, evolution.

Introduction

Like other lissamphibians, most salamanders exhibit two different life histories, and undergo a metamorphosis transformation in body form and substantial internal remodeling (ROSE, 2003). Hence, the ontogenesis can classify into three periods: larval, metamorphic and post-metamorphic periods (including juvenile, subadult and adult stages). The vomerine tooth is the tooth bears in the vomer. The spatial position, shape and length of vomerine tooth rows in adult salamanders are varied among different salamanders, so it was regarded as an important diagnosis (e.g., LIU, 1950; WAKE, 1966; ZHAO *et al.*, 1988; FEI *et al.*, 2006; BUCKLEY *et al.*, 2010). Same as

other cranial skeleton, vomerine tooth rows undergoes a pronounced change in spatial position, shape and length during metamorphosis. However, the transition during postmetamorphic period has received a little attention in hynobiid salamanders (e.g., ROSE, 2003; LEBEDKINA, 2004; JÖMANN *et al.*, 2005).

Hynobius guabangshanensis, a poorly known hynobiid salamander, was described by SHEN *et al.* (2004) using specimens from Guabangshan Nature Reserve, Qiyang County, Hunan Province. It is one of the endemic salamanders of China, only distributed in the type locality. Because the information on its extent of occurrence, sta-

tus and ecological requirements is lacking, it was assessed as data deficient species (DD) in IUCN (the International Union for Conservation of Nature) by XIE & JIANG (2006). Recently, some researches on *H. guabangshanensis* have been reported in Chinese. MI *et al.* (2007) reported the early embryonic development that the early embryonic development can divide into 21 periods, and it takes 1134 h for the fertilized eggs to hatch. GUO *et al.* (2008) reported the breeding ecology that the adults lived on land during non-breeding season and in water during breeding season; the male had the behaviors of display and sperm competition. XIONG *et al.* (2008) reported the karyotype that the diploid number is 56 (with four groups), with nine large pairs, four biarmed medium pairs, five biarmed microchromosome pairs, and ten uniarmed microchromosome pairs. XIONG *et al.* (2010) reported the morphological features of the hyobranchial apparatus in adult and subadult (juvenile in this note) and pointed that the differences of morphological characters may be caused by the function of hyobranchial apparatus in adult and subadult. GUO *et al.* (2010) reported the morphological structure of the spermatozoa of *H. guabangshanensis* that the spermatozoa share some characteristic morphological features and synapomorphies with that of Hynobiidae, but the sperm differ significantly from other species of Hynobiidae. REN *et al.* (2010) reported the length of CO I is 1551bp and D-loop is 803 bp.

Fortunately, we have had the opportunity to gain some adults and juveniles specimens of *H. guabangshanensis*, and have the chance to throw light on the changes of vomerine tooth rows during postmetamorphic period. So, in this note, the vomerine tooth rows of two developmental stages juvenile and adult *H. guabangshanensis* are described and compared.

Materials and Methods

Six adults (SCUM051201XJL–SCUM051206XJL, three male and three female) specimens of *H. guabangshanensis* were collected from the type locality, Guabangshan Nature Reserve (26.37.755°N, 111.58.106°E, altitude 720 m), Qiyang County, Hunan Province, China from 25 to 26 November (breeding season), 2005. Six juvenile's specimens (SCUM051221XJL, SCUM051222XJL, HNUST00239–HNUST00242, which distinguished according to the characteristics of JÖMANN *et al.* (2005) and LEBEDKINA (2004) that the external gills of specimens have resorbed, sex was not identifiable, only genital ridges were seen when dissected) were from two egg sacs collected in the field, and larvae hatched and reared under laboratory condition until juvenile.

All specimens (adults and juveniles) were fixed in 10% formalin, and the total length (TOL, from tip of snout to tip of tail) was obtained using digital calipers

to the nearest 0.1 mm (the total length of adults ranged from 105.49 to 153.31 mm, and juveniles from 51.36 to 78.55 mm). Then specimens (all adults and two juveniles, and other four juveniles only examined the shape of vomerine tooth rows under dissecting microscope) were skinned, eviscerated (juveniles not), cleared and double-stained using a bone-cartilage staining procedure (HANKEN & WASSERSUG, 1981). The prepared skulls were examined with a LEICA MZ6.0 binocular dissecting microscope. The figures were draw by the method of LIU & XIONG (2013), which summarized as follows: achieve the picture of specimens through camera; open the picture in ViewGIS software and form the layer; add a line layer, and related to the picture layer; depict according to the outline of each skull elements; modify the drawing with the button of line separate, node connect, line break point modify; invisible of picture layer after modified; output the picture and save. Vouchers specimens are deposited in the Museum of Sichuan University (SCUM) and Museum of Henan University of Science and Technology (HNUST). For the elements of the skull, we followed the terminology of JÖMANN *et al.* (2005).

Results

In adults (Fig. 1A), the vomerine tooth bears in the middle-to-posterior portion of vomer on its medial edge. The vomerine tooth rows are long and posteriorly directed, bent in a V-shape, but left and right vomerine tooth rows are not connected medially. Tooth rows form inner and outer series, inner series beginning at end of vomer and outer series ending at the posterior internal nares, the inner series obviously longer than the outer series. The anterior portion of the tooth row does not exceed the anterior edge of the choana. In juveniles (Fig. 1B), the vomerine tooth locates on posterior portion of the vomer, and the vomerine tooth rows are not connected at the midline. The tooth row exceeds the anterior edge of the choana. The vomerine tooth rows are short and slightly arched, parallel to the premaxilla and maxilla.

Variability of vomerine tooth rows occurs in both adults and juveniles. In adults, left and right vomerine tooth rows are not connected medially in most specimens, but they are connected medially in one specimens (SCUM051204XJL, Fig. 2A), and are overlapped in another specimens (SCUM051206XJL, Fig. 2B). In juveniles, the vomerine tooth rows are short and slightly arched, parallel to the premaxilla in most specimens (SCUM051221XJL, SCUM051222XJL, HNUST00239, HNUST00242), but obviously arched and allowing for distinction of an inner and outer series in other two specimens, the inner series nearly equal to the outer series (HNUST00240, HNUST00241, Fig. 3B, C). The variation is independent from the total length in juveniles,

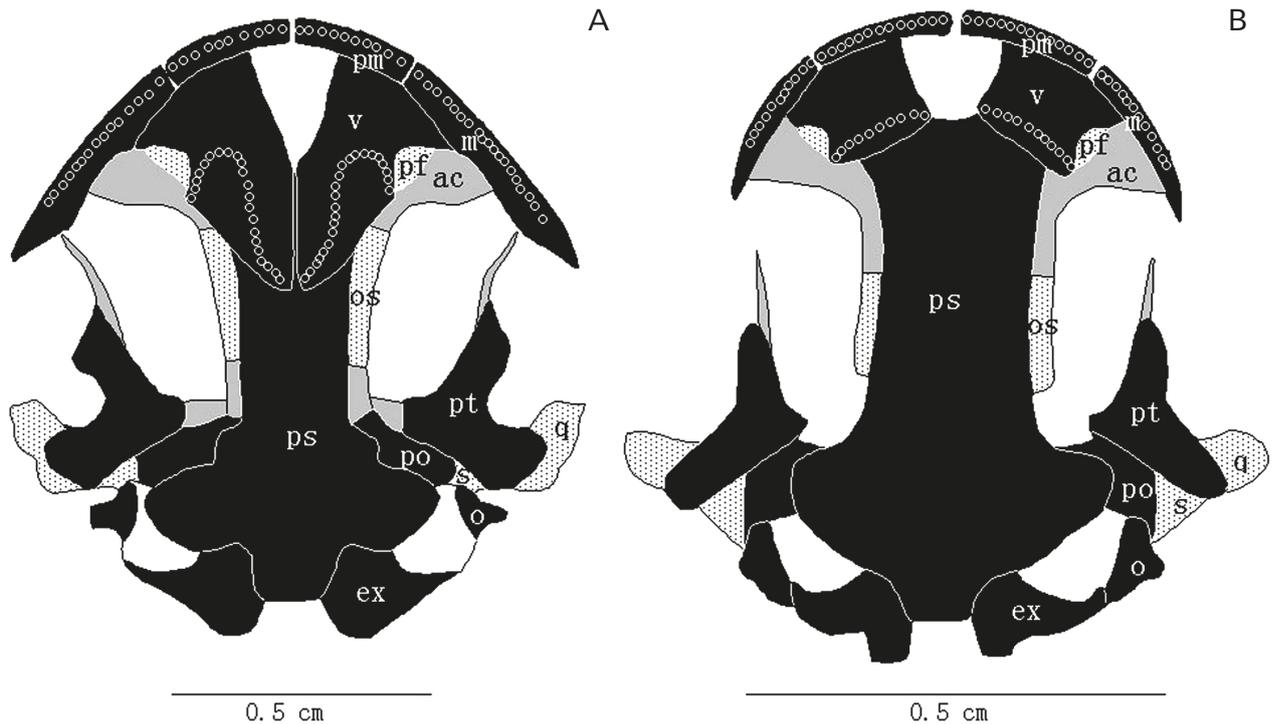


Fig. 1. The skull of *Hynobius guabangshanensis* in ventral view. **A:** The skull of adult (SCUM051202XJL; **B:** The skull of juvenile (SCUM051221XJL). The bones in white or white with black dots (in deeper zones of the skull), cartilage in gray, teeth in open circles, and gaps or holes in white. antorbital cartilage (ac), exoccipital (ex), maxillary (m), nasal (n), operculum (o), orbitosphenoid (os), prefrontal (pf), premaxilla (pm), prootic (po), parasphenoid (ps), pterygoid (pt), quadrate (q), squamosal (s), and vomer (v).

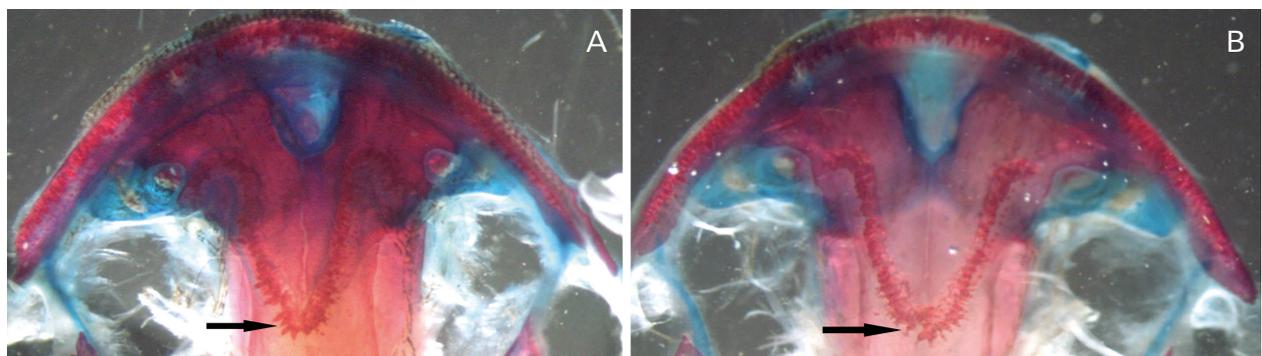


Fig. 2. The variation of the vomerine tooth rows in adults *Hynobius guabangshanensis*. **A:** The vomerine tooth rows of SCUM051204XJL; Arrows show the vomerine tooth rows are connected medially. **B:** The vomerine tooth rows of SCUM051206XJL. Arrows show the vomerine tooth rows are overlapped.

because the variation present in median size individuals, and most individuals (including minimal and maximal) have the typical vomerine tooth rows.

Discussion

The vomerine tooth undergo considerable transformation during ontogeny in salamanders (e.g., VASSILIEVA & SMIRNOV, 2001; ROSE, 2003; LEBEDKINA, 2004; JÖMANN

et al., 2005; GREVEN & CLEMEN, 2009; VASSILIENVA & SERBINOVA, 2013), which including the tooth number and arrangement, the number of tooth rows and tooth morphology. The development and evolution of salamander dentition is mediated by thyroid hormones (ROSE, 1995, 2003; SMIRNOV & VASSILIEVA, 2003; GREVEN & CLEMEN, 2009), and SMIRNOV & VASSILIEVA (2003) point out that the TH-dependence is reduced to zero in postmetamorphosis. In this note, the vomerine tooth rows shows an obviously change from juveniles to adults *H. guabangshanensis*, especially in shape. This case is also found in other salamanders. Such as in the juvenile *R. sibiricus*, the vomerine tooth rows arranged in a straight line shape, and formed a short slight arch allowed the differentiation

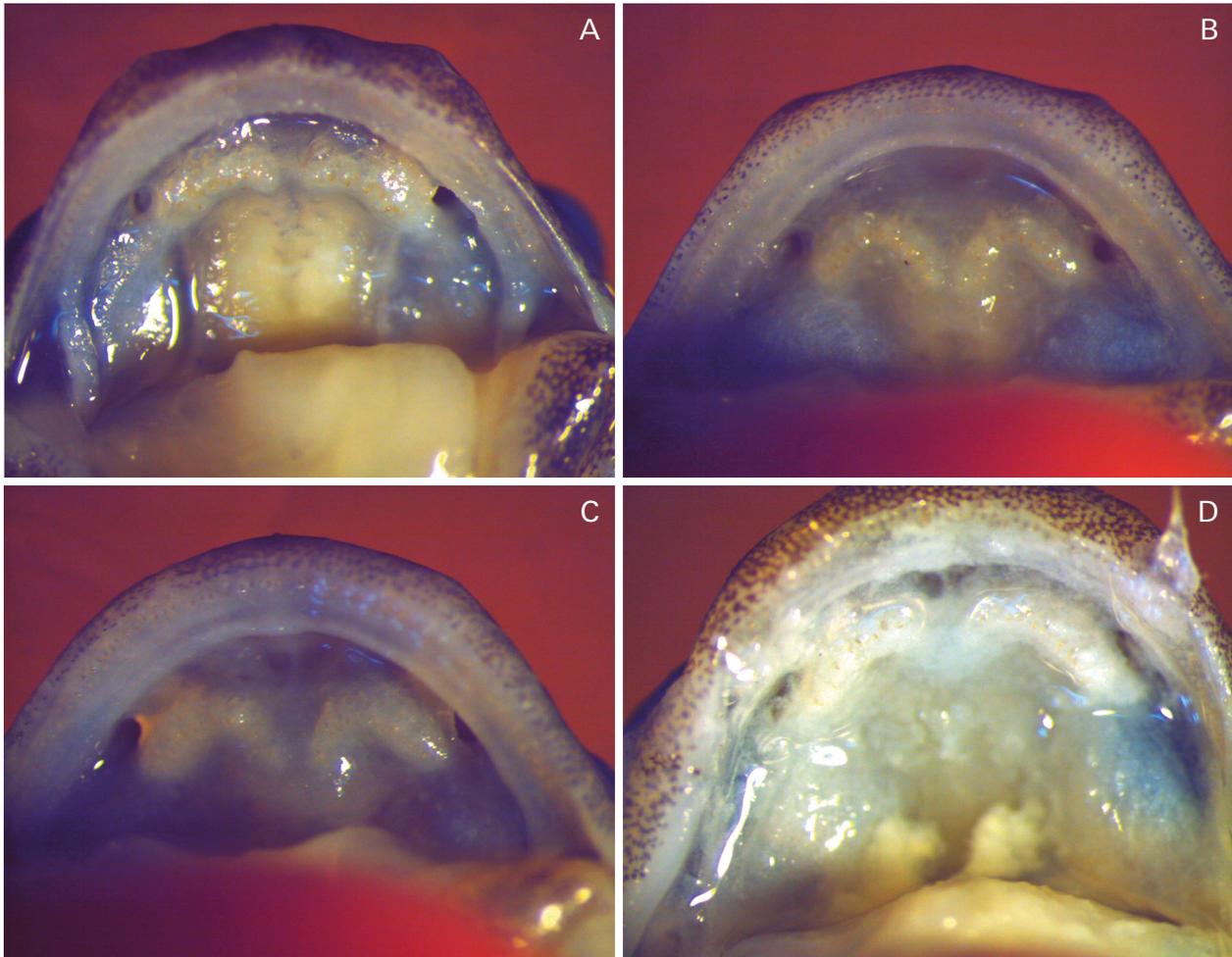


Fig. 3. The variation of the vomerine tooth rows in juveniles *Hynobius guabangshanensis*. **A:** The vomerine tooth rows of HNUST00239; **B:** The vomerine tooth rows of HNUST00240; **C:** The vomerine tooth rows of HNUST00241; **D:** The vomerine tooth rows of HNUST00242. The vomerine tooth rows of A and D same to SCUM051221XJL and SCUM051222XJL; and those of B and C are obviously arched allowing for distinction of an inner and outer series.

of an inner series and an outer series of teeth in subadults and adults (LEBEDKINA, 2004; JÖMANN *et al.*, 2005). These showed that the development of the vomerine tooth may be last until subadults or adults.

ZHAO & HU (1988) separated hynobiids into two natural groups, the *Ranodon* and *Hynobius* groups based on the features of dentition (the shape of the vomer, the spatial relationship between the vomerine tooth row and the choana, the shape of the vomerine tooth row), length of larval state, numbers of eggs laid, and fontanelle. The vomerine tooth rows of *Hynobius* group (including the species of genus of *Hynobius* and *Salamandrella*) are long, posteriorly directed and obviously arched allowing for distinction of an inner and outer series (inner series is longer than outer); those of *Ranodon* group (species of other genera) are short, transversely oriented and slightly arched or obviously arched allowing for distinction of an inner and outer series (inner and outer are almost equal or outer series is longer than inner). The vomerine tooth row of juveniles of *H. guabangshanensis* is resembled to those of *Ranodon* group, and that of adults is similar to *Hynobius* group. In the view of hae-

ckel's law (biogenetic law), the vomerine tooth row of *Ranodon* group is considered as an ancestral state, and that of *Hynobius* group is a derived state. This is consistent with the result of ZHAO & HU (1998). Though the division of two groups did not supported by the ancestral state reconstruction analysis because this division requires those characters in *Salamandrella* and some species of *Hynobius* evolved independently, the ancestral state of the vomerine tooth row in *Ranodon* group is verified as an ancestral state (ZHANG *et al.*, 2006).

Together with tongue, the vomerine tooth rows functions in feeding, but the tongue is the mainly role, and the vomerine tooth row play an auxiliary role (REGAL, 1966). It is confirmed that the feeding mode are different among salamanders because of the diversity of life habit. Terrestrial salamanders use inertial feeding or protruding tongue to capture prey, and aquatic salamander feed by suction (LARSEN *et al.*, 1996; DEBAN & WAKE, 2000; WELLS, 2007). Then, the functions of the vomerine tooth rows are different among different feeding mode. The vomerine teeth in aquatic salamanders may hinder escape of prey when water is released from the mouth; whereas

the vomerine teeth rows in the terrestrial salamanders can hold and deliver the prey (REGAL, 1966; DEBAN & WAKE, 2000). Hynobiid salamanders can divide into two groups, aquatic (e.g., the species of genus *Liua*, *Batrachuperus*, *Pachyhynobius*, *Paradactylodons*) and terrestrial (the species of genus *Hynobius* and *Salamandrella*) groups according to their life habit (FEI *et al.*, 2006). The vomerine tooth rows of aquatic group are short, transversely oriented and slightly arched, and those of terrestrial group are long, posteriorly directed and obvious arched to distinguish into two inner and outer series (ZHAO & HU, 1998; FEI *et al.*, 2006; ZHANG *et al.*, 2006). The vomerine tooth rows of juveniles are similar to the aquatic salamanders, this reflects that the juveniles feed by suction in water, and the vomerine tooth rows function in hindering escape of prey. The vomerine tooth rows of adults are resemble with terrestrial salamanders, which use inertial feeding or protruding tongue to capture prey, and the vomerine tooth rows function in holding and delivering the prey. Hence, the difference of vomerine tooth rows in juvenile and adult of *H. guabangshanensis* reflect the differences of life habit, feeding modes and function.

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References

- BUCKLEY, D., WAKE, M.H. & WAKE, D.B. (2010): Comparative skull osteology of *Karsenia koreana* (Amphibia, Caudata, Plethodontidae). – *Journal of Morphology*, **271**: 533–558.
- DEBAN, S.M. & WAKE, D.B. (2000): Aquatic feeding in salamanders. In: SCHWENK, K. (eds.), *Feeding: Form, Function, and Evolution in Tetrapod Vertebrates*, Academic Press, San Diego, CA, pp. 65–94.
- FEI, L., HU, S.Q., YE, C.Y. & HUANG, Y.Z. (2006): *Fauna Sinica Amphibia*, Vol. 1. Beijing: Science Press (In Chinese).
- GREVEN, H. & CLEMEN, G. (2009): Early tooth transformation in the paedomorphic Hellbender *Cryptobranchus alleganiensis* (Daudin, 1803) (Amphibia: Urodela). – *Vertebrate Zoology*, **59**(1): 71–79.
- GUO, K.J., MI, X.Q. & DENG, X.J. (2010): Morphological structure of the spermatozoa in *Hynobius guabangshanensis* (Urodela: Hynobiidae). – *Life Science Research*, **14**(1): 38–43 (In Chinese).
- GUO, K.J., MI, X.Q. & DENG, X.J. (2008): Breeding ecology of *Hynobius guabangshanensis*. – *Chinese Journal of Ecology*, **27**: 77–82 (In Chinese).
- HANKEN, J. & WASSERSUG, R. (1981): The visible skeleton. A new double-stain technique reveals the native of the “hard” tissues. – *Functional Photography*, Hempstead, **16**: 22–26.
- JÖMANN, N., CLEMEN, G. & GREVEN, H. (2005): Notes on cranial ontogeny and delayed metamorphosis in the hynobiid salamander *Ranodon sibiricus* Kessler, 1866 (Urodela). – *Annals of Anatomy*, **187**: 305–521.
- LARSEN, J.H., BENESKI, J.T. & MILLER, B.T. (1996): Structure and function of the hyolingual system in *Hynobius* and its bearing on the evolution of prey capture in terrestrial salamanders. – *Journal of Morphology*, **227**: 235–248.
- LEBEDKINA, N.S. (2004): *Evolution of amphibian skull* (Translated and edited by SMIRNOV, S.V.). Pensoft Publishers, Sofia–Moscow.
- LIU, C.C. (1950): Amphibians of western China. *Fieldiana: Zoology Memoirs*, **2**: 1–400.
- LIU, X.Y. & XIONG, J.L. (2013): A method of drawing biological black line chart using the ViewGIS software. – *Chinese Journal of Zoology*, **48**: 200–205 (In Chinese).
- MI, X.Q., DENG, X.J., GUO, K.J., NIU Y.D. & ZHOU Y. (2007): Early embryonic development of *Hynobius guabangshanensis*. – *Sichuan Journal of Zoology*, **26**: 377–378 (In Chinese).
- REGAL, P.J. (1966): Feeding specializations and the classification of terrestrial salamanders. – *Evolution*, **20**: 392–407.
- REN, W., NIU, Y.D., ZHOU, Y., WANG, X. & DENG, X.J. (2010): Discussion on the Taxonomic status of *Hynobius guabangshanensis* based on mitochondrial DNA *CO I* gene and D-loop region. – *Life Science Research*, **14**(4): 327–330 (In Chinese).
- ROSE, C.S. (1995): Skeletal morphogenesis in the urodela skull: II. Effect of developmental stage in thyroid hormone-induced remodelling. – *Journal of Morphology*, **223**: 149–166.
- ROSE, C.S. (2003): The developmental morphology of salamander skulls. In: HEATWOLE, H. & DAVIES, M. (eds.): *Amphibian Biology*, Vol. 5. Osteology. – Surrey Beatty & Sons, Chipping Norton, pp. 1684–1781.
- SHEN, Y.H., DENG, X.J. & WANG B. (2004): A new hynobiid species *Hynobius guabangshanensis* from Hunan Province, China (Amphibia: Hynobiidae). – *Acta Zoologica Sinica*, **50**: 209–215 (In Chinese).
- SMIRNOV, S.V. & VASSILIEVA, A.B. (2003): Skeletal and dental ontogeny in the smooth newt, *Triturus vulgaris* (Urodela: Salamandridae): Role of thyroid hormone in its regulation. – *Russian Journal of Herpetology*, **10**(2): 93–100.
- VASSILIEVA, A.B. & SERBINOVA, I.A. (2013): Bony skeleton in the Caucasian salamander, *Mertensiella caucasica* (Urodela: Salamandridae): Ontogeny and embryonization effect. – *Russian Journal of Herpetology*, **20**(2): 85–96.
- VASSILIEVA, A.B. & SMIRNOV, S.V. (2001): Development and morphology of the dentition in the asian salamander, *Ranodon sibiricus* (Urodela: Hynobiidae). – *Russian Journal of Herpetology*, **8**(2): 105–116.
- WAKE, D.B. (1966): Comparative osteology and evolution of the lungless salamanders, family Plethodontidae. – *Memoir of Southern California Academy of Sciences*, **4**: 1–111.
- WELLS, K.D. (2007): *The ecology and behavior of amphibians*. The University of Chicago Press, Chicago and London.

- XIE, F. & JIANG J.P. (2006): *Hynobius guabangshanensis*. In: IUCN 2013. IUCN Red List of Threatened Species. Version 2013.2. <www.iucnredlist.org>. Downloaded on 19 January 2014.
- XIONG, J.L., QING, L.Y., ZENG, X.M. & ZHAO, E.M. (2008): Karyotype of *Hynobius guabangshanensis* (Urodela: Hynobiidae). – *Sichuan Journal of Zoology*, **27**: 236–238.
- XIONG, J.L., SUN, P., ZHU, W.W. & LIU X.Y. (2010): The morphological features of hyobranchial apparatus in adult and subadult *Hynobius guabangshanensis*. – *Chinese Journal of Zoology*, **45**: 138–142 (In Chinese).
- ZHANG, P., CHEN, Y.Q., ZHOU H., LIU, Y.F., WANG X.L., PAPENFUSS T.J., WAKE, D.B. & QU, L.H. (2006): Phylogeny, evolution, and biogeography of Asiatic Salamanders (Hynobiidae). – *Proceeding of the National Academy of Sciences*, **103**: 7360–7365.
- ZHAO, E.M. & HU, Q.X. (1988): Studies on Chinese tailed amphibians. In: ZHAO, E.M., JIANG, Y.M., HU, Q.X. & YANG, Y.H. (eds.), *Studies on Chinese Salamanders*. Society for the Study of Amphibians and Reptiles, Ohio, Oxford.