Development of the Scutes in Eightspine Stickleback, *Pungitius sinensis kaibarae* (Gasterosteidae, Pisces) from Korea

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The developmental process of scutes in the eightspine stickleback, *Pungitius sinensis kaibarae* (Tanaka), was investigated. The scutes appeared first on the caudal peduncle at 13mm and then on the thoracoabdominal part at 16mm. The scutes continued to develop and then the trachurus type of scute arrangement was completed at about 20mm. Finally the number of scutes was stabilized as 32 – 33 in the fish over 22mm. The developmental state of scutes was different according to part of body but most scutes except those of anteriormost part entered into late stage at about 25mm. The scutes of anteriormost part remained as scute of middle stage even though the fish became adults. All scutes were formed around neuromasts of lateral line. Few anterior plates(up to five plates) had additional neuromasts above the plates. But scale material was not accumulated around the additional neuromasts. Relationship between Korean and Japanese eightspine stickleback and the taxonomic meaning of the additional neuromasts were discussed.

Introduction

Sticklebacks have only a row of scutes on the body side. Pattern of arrangement and morphology of the scutes varied and these characters are used as important key in their classification. The developmental process of scutes of sticklebacks from Japan was studied by Igarashi (1962, 1963, 1964, 1965, 1968, 1969a, b). Of those reports, however, full descriptions were not presented on the eightspine stickleback, *Pungitius sinensis kaibarae*(Tanaka); particulary on the first appearance of scutes in juveniles. More studies on Japanese eightspine stick-

lebacks were not possible because the species was already extinguished in Japan(Miyadi et al., 1976). Recently the inhabitation of *P. sinensis kaibarae* in southern Korean Peninsula was revealed and some studies on their morphology, distribution, classification and ecology were carried out(Jeon, 1982, 1987; Chae and Yang, 1988, 1989, 1990, 1993; Kim et al., 1989; Yang and Min, 1990). But the developmental process of scutes on the Korean eightspine sticklebacks was not studied. Tanaka et al.(1982) reported some differences in morphology between Korean and Japanese eightspine sticklebacks. Then additional investigations on them are nece-

ssary. In present study, the developmental process of scutes in Korean eightspine stickleback from the Chayang Stream was investigated.

Materials and Methods

All specimens of eightspine stickleback, *Pungitius sinensis kaibarae*, were collected at the Chayang Stream(Yanghang – ri, Imgo – myon, Yongchon – gun, Kyongsangbuk – do province) in April and May, 1985. Fish were fixed in 10% formalin immediately. Two hundred and sixty individuals ranged 5.0 – 26.8mm(TL 6.2 – 31.1 mm) were used. They were divided into standard body length groups by one mm interval and were

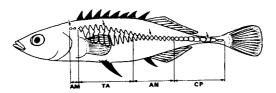


Fig. 1. Four different parts of the row of scute in eightspine stickleback, Pungitius sinensis kaibarae. Abbreviations are as follows: am, anteriormost part; an, anal part; cp, caudal peduncle part; ta, thoracoabdominal part. Arrows are indicate the scutes that monitored their developing process.

treated seperately. They were cleared and stained with Alizarin – red S(Park and Kim, 1984) and preserved in glycerin. Right side of the cleared specimens was skinned and then mounted on a slide glass with glycerin. Observations and drawings of scutes were made with a stereoscopic microscope(Nikon, SMZ – 10, with a camera lucida). To compare the developmental state of scutes among parts of the body, the row of scute divided into four parts(Fig. 1). The developmental states were described as early, middle and late stages after Igarashi(1952). In the text, all of the body length are expressed as standard body length except that noted as TL which represents total body length.

Results

The developmental order of scutes in eightspine stickleback was presented in Fig. 2. The scutes appeared first on the caudal peduncle and then forward gradually. There were 4 scutes at about 13mm and 7-8 scutes at 14mm. Thoracoabdominal scutes appeared at about 16mm. In this time there were no scutes on the anal part. The scutes continued to develop and then the

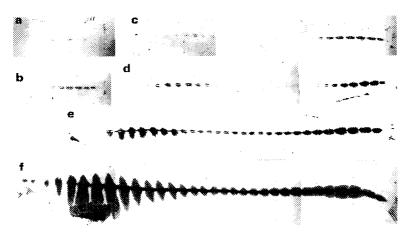


Fig. 2. The developmental order of scutes in eightspine stickleback, *Pungitius sinensis kaibarae*. Figure a is that of 13mm individual, b is 14mm, c is 17mm, d is 18mm, e is 21mm and f is 25mm. Figures a and b are presented only caudal peduncle part because there are no scutes forward from there.

trachurus type of scute arrangement was completed at about 18 – 20mm. The developmental state of scutes at about 25mm showed close resemblance to that of adults.

Scutes were formed around the neuromasts of lateral line. There was a set of three neuromasts under each plate. The first few anterior plates(up to fifth plate) had an additional set of neuromast above the plates. Each additional set consisted of



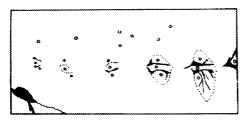


Fig. 3. Anteriormost part of an individual of eightspine stickleback, Pungitius sinensis kaibarae, with body length 22mm showing a scute row and additional set of neuromast. Upper: photograph, lower: schematic drawing.

one to three neuromasts. Scutes were not developed around this additional neuromasts even in adult specimens(Fig. 3).

Some individuals at about 13.8mm(about TL 16mm) had 4 to 5 scutes on the caudal peduncle part. In this time, only main ridges were developed and scale material was not accumulated. It was observed in one individual of 13.0mm (TL 14.8mm) that a rudiment of scute appeared around the neuromasts of lateral line on the caudal peducle. The rudiment scute was crescent shaped and was not stained with alizarin - red. At about 16mm, scale material was accumulated around the main ridge and the scutes were broadened(early stage). Main ridge began to branch in a few individuals of 17mm and most individuals of 18mm had lateral ridge on their scutes(middle stage). Becoming over 20mm, the two main ridges developed inward and connected each other and the mucuous tube was formed completely. Lateral ridges branched out and thickened simultaneously(late stage) (Fig. 4 and Table 1-D). All individuals over 24mm had scutes of late stage.

Thoracoabdominal scutes appeared first at about 16mm(Fig. 5). Accumulation of the scale

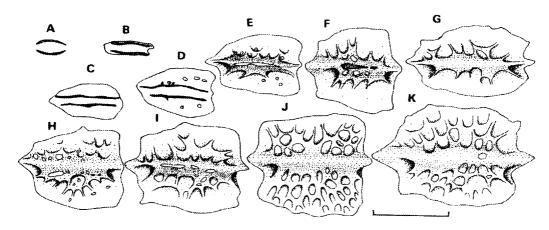


Fig. 4. The developmental process of scutes on the caudal peduncle part of eightspine stickleback, *Pungitius sinensis kaibarae*. Standard body length of individuals with each scute is as follows. a: 15mm, b: 16mm, c: 17mm, d: 18mm, e: 19mm, f: 21mm, g: 22mm, h: 23mm, i: 24mm, j: 25mm and, k: 26mm. Scale bar indicates 0.5mm.

Table 1. Relation between body length and the developmental state of the scute in Yongchon population of eightspine stickleback, *Pungitius sinensis kaibarae*. The data represent the percentage of the occurance of scute with specific developmental stages in each group of fish having indicated body length. Stage E, M and L indicate early, middle and late stage, respectively. Numbers in parentheses are number of individuals used in observation.

Part of body	Stage	Body length (mm)														
		<13 (54)	13 (9)	14 (19)	15 (19)	16 (14)	17 (25)	18 (20)	19 (22)	20 (18)	21 (23)	22 (8)	23 (11)	24 (10)	25 (6)	26 (2)
M												9.1	10.0	16.7	50.0	
L																
B. Thoracoabdominal	E					100.0	100.0	95.0	40.0	16.7	4.3					
	M							5.0	60.0	83.3	91.4	87.5	54.5	40.0		
	L										4.3	12.5	45.5	60.0	100.0	100.0
C. Anal	E							100.0	100.0	100.0	78.3	50.0				
	M										21.7	50.0	100.0	70.0	16.7	
	L													30.0	83.3	100.0
D. Caudal peduncle	Е		100.0	100.0	100.0	100.0	96.0	60.0	18.2							
	M						4.0	40.0	81.8	100.0	65.2	12.5	9.1			
	L										34.8	87.5	90.9	100.0	100.0	100.0

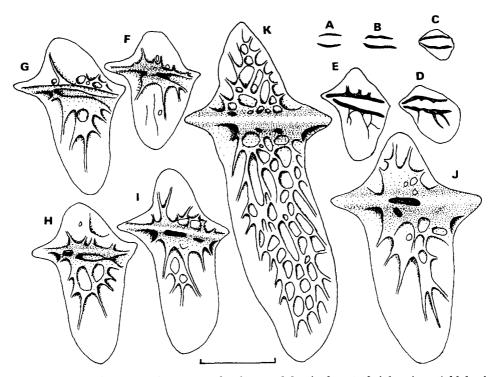


Fig. 5. The developmental process of scutes on the thoracoabdominal part of eightspine stickleback, *Pungitius sinensis kairarae*. Standard body length of individuals with each scute is as follows. a: 17mm, b:18mm, c:19mm, d and e:20 mm, f:21mm, g:22mm, h:23mm, i:24mm, j:25mm and k:28mm. Scale bar indicates 0.5mm.

material was started at about 18mm in many specimens. Main ridges began to branch in a few individuals of 18mm. Most individuals of 20mm had scutes of middle stage. The scutes of late stage began to appear in some individuals of 21mm. All individuals over 25mm had scutes of this stage only(Table 1 ~ B).

Some individuals of about 18mm had crescent – shaped structures around the neuromast of lateral line on the anteriormost part. They were not stained with alizarine – red until the fish reach 21mm. Almost all individuals had scutes of early stage until 22 – 23mm. Scutes of middle stage started to appear in a few individuals of about 23mm. But even over 26mm most individuals had scutes of early and middle stage simultaneously. Although the fish grew into adults, those scutes were very small and rarely attained late stage(Fig. 6 and Table 1 – A).

Scutes on the anal part started to appear at about 18mm and remained as early stage untill 20mm. After 21mm, they developed lateral ridges and became middle stage scutes. Then at about 25mm mucuous tube was formed completely, most scutes entered late stage(Fig. 7 and Table 1-C). It seemed that 14th to 16th

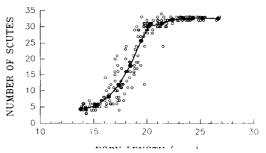


Fig. 8. A relationship between standard body length and number of scutes of eightspine stickleback, *Pungitius sinensis kaibarae*.

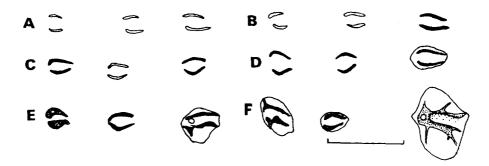


Fig. 6. The developmental process of scutes on the anteriormost part of eightspine stickleback, *Pungitius sinensis kaibarae*. Standard body length of individuals with each scute is as follows. a: 18mm, b: 20mm, c: 21mm, d: 23mm, e: 24mm, and f: 26mm. Scale bar indicates 0.5mm.

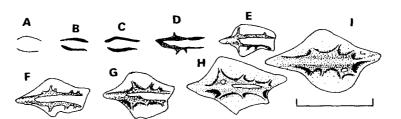


Fig. 7. The developmental process of scutes on the anal part of eightspine stickleback, *Pungitius sinensis kaibarae*. Standard body length of individuals with each scute is as follows. a: 18mm, b: 19mm, c: 20mm, d: 21mm, e: 22mm, f: 23mm, g: 24mm, h: 25mm, and i: 26mm. Scale bar indicates 0.5mm.

scutes from posterior end appeared last of all scutes.

Fig. 8 represents a relationship between body length and total number of scutes. The pattern of increase in the number of scute was sigmoid. The scutes appeared first at 13mm. The number of scutes hardly increased until 16mm but rapidly increased since then. At about 20mm, the number of scutes exceeded 30 near the number of scutes in adults. After that the rate of increment decreased and finally the numbers were stabilized as 32 – 33 in the fish over 22mm.

Discussion

Adults of eightspine stickleback had 30 to 34 scutes(mean number 33). Their scutes were very well developed as a trachurus type and there were no individuals lacking scutes even in part (Chae and Yang, 1988, 1989).

Scutes appeared in this species first at caudal peduncle and then at thoracoabdominal part. Since then scutes are formed back and forth from these two parts of fish body and are connected at anal parts. This fact shows that the origin of scute formation is two parts and agrees with the observation in Japanese eightspine stickleback(Igarashi, 1969a). Threespine stickleback, Gasterosteus aculeatus aculeatus make scutes in different order with eightspine stickleback but they also have two origins of scute formation(Igarashi, 1964). It seems common to gasterosteid fishes that there are two centers for scute formation. But Igarashi(1962) reported that ninespine stickleback, P. sinensis sinensis, had only one center for scute formation at caudal peduncle and then the scutes were formed forward from there. It is considered, therefore, that a comparison on the development of scute in ninespine stickleback from Korea and Japan is necessary.

Igarashi(1969a) pointed out that the first appearance of scutes in Japanese eightspine stickle-back would be observed in the fishes less than TL 18mm. In present investigation, it was observed that the scutes appeared first in some specimens with 13mm(TL 16mm) and specimens with 15 – 16mm(TL 17 – 18mm) had 4 to 10 scutes. This result accords well with prediction of Igarashi(1969a). So it seems that time of first appearance of scutes is almost same between Korean and Japanese eightspine stickle-back

It was known that in Japanese eightspine stickleback the numbers of scutes reached in the adult's mean at TL 28mm and most scutes became late stage at TL 31mm(Igarashi, 1969a). The same states were reached at 20 - 22mm(TL 23 - 25mm) and 25mm(TL 28 - 29mm), respectively, in Korean eightspine stickleback. Also we pointed out previously that the size of scutes on thoracoabdominal part of Korean fish was larger than that of Japanese fish(Chae and Yang, 1989). It seems, therefore, that the rate of scute development is more rapid and the size of scutes is larger in Korean eightspine sticklebacks than Japanese one. It is notable that the scutes of most individuals become late stage at 25mm as well as the least sized individual with mature eggs is 25mm(Chae and Yang, 1993).

It was known that there were two forms of tenspine stickleback, *P. pungitius*, from Japan: common(semiarmatus) and specialized(trachurus) forms. They were originated from the difference of time of neotenization(Igarashi, 1969b). Applying this hypothesis to eightspine stickleback, it may be thought that Korean fish was neotenized later than Japanese fish, but the situation is contrary to that. That is, Korean eightspine stickleback become adults at 30 to 35mm (Chae and Yang, 1993) but Japanese one matures sexually at about 45mm(Kobayashi, 1933).

Therefore, it seems that there is no relation between the time of sexual maturation and the developmental state of scutes in eightspine stickleback at least. It is not known whether this difference between Korean and Japanese eightspine stickleback represents the geographic variation among populations, the difference of time of land-locking(Mizuno, 1962), or that they are distinct taxa.

In the developmental process of scutes, it was observed that additional neuromasts were present above the first few anterior plates but they have no scale material around them. The number of neuromast in additional set is fewer than that of the main row. These results can be interpreted as that the additional set of neuromasts had been degenerated from the state having both scutes and three neuromasts. Eightspine stickleback, therefore, might be derived from an ancestor that have an additional scute row. In brook stickleback, Culaea inconstans, the first few anterior plates are paired with up to five additional plates which are out of line with the main row(Nelson, 1969). It is considered that this fact supports our interpretation. It was known that of the gasterosteid fishes 3 genera Culaea, Gasterosteus and Pungitius had the most close relationship(Bertin, 1925; Leiner, 1934; Wootton, 1976). Whether the genus Gasterosteus has the additional neuromast sets is until not known. It is considered, however, as an important evidence which reveals kinship of these genera that eightspine stickleback and brook stickleback have additional neuromasts and plates respectively and also they have same number of additional ones.

References

Bertin, L. 1925. Recherches bionomiques, biométriques et systématiques sur les Epinoches(Gast-

- érostéides). Ann. Inst. Oceanogr. Monaco 2: 1 204.(not cited directly)
- Chae, B. S. and H. J. Yang. 1988. Comparison of the morphology between two populations of eight-spine stickleback, *Pungitius sinensis kaibarae* (Tanaka). Kor. J. Lim. 21(2): 79-91.
- Chae, B. S. and H. J. Yang. 1989. Geographic variation of scutes in eightspine stickleback *Pungitius sinensis*(Gasterosteidae) from Korea. Korean J. Ichthyol. 1(1/2): 42 53.
- Chae, B. S. and H. J. Yang. 1990. Sexual dimorphism in eightspine stickleback, *Pungitius sinensis*: Gasterosteidae. Korean J. Zool. 33: 260 265.
- Chae, B. S. and H. J. Yang. 1993. Spawning and growth of eightspine stickleback, *Pungitius sinensis kaibarae*(Tanaka) (Gasterosteidae, Pisces), in the Chayang stream, a tributary of Kumho River, Korea. Korean J. Zool. 36: 181-192.
- Igarashi. K. 1962. Observation on the development of the scutes in ten-spined stickleback, *Pungitius* sinensis(Guichenot). Bull. Japan. Soc. Sci. Fish. 28(4): 393-398.
- Igarashi, K. 1963. Observation on the development of the scutes in ten-spined stickleback, *Pungitius* pungitius(Linnaeus). Bull. Japan. Soc. Sci. Fish. 29(4): 342 - 348.
- Igarashi, K. 1964. Observation on the development of the scutes in landlocked form of three spined stickleback, *Gasterosteus aculeatus aculeatus* Linnaeus. Bull. Japan. Soc. Sci. Fish. 30(2): 95 - 103.
- Igarashi, K. 1965. Observation on the development of the scutes in landlocked form of three - spined stickleback, *Gasterosteus aculeatus microcephalus* Linnaeus. Bull. Japan. Soc. Sci. Fish. 31(1): 33 -40.
- Igarashi, K. 1968. Observation on the development of the scutes in a ten – spined stickleback, Musashi Tomiyo, *Pungitius* sp. Bull. Japan. Soc. Sci. Fish. 34(12): 1083 – 1087.
- Igarashi, K. 1969a. Studies on the lateral scutes of stickleback(Gasterosteidae) in Japan \(\mathbb{N} \). An observation on the development of the scutes in ten spined stickleback, Pungitius kaibarae

- (Tanaka). Japan. J. Ichthyol. 16(1): 10 16.
- Igarashi, K. 1969b. On the distribution and variation of *Pungitius pungitius* in the Mogami basin with particular regard to its scutes. Zool. Mag. 78: 340-350.
- Jeon, S. R. 1982. On the new distribution site of the eightspine stickleback, *Pungitius sinensis kaibarae*, from Korea. Freshwater Fishes(Tansuigyo) 8: 69.
- Jeon, S. R. 1987. Studies on the key and distributions of the cottid and gasterosteid peripheral fresh water fishes from Korea. Coll. Thes. Sang Myung Women's Univ. 19: 549 576.
- Kim, I. S., C. H. Yoon and J. H. Lee. 1989. Morphological variation and karyo-type of the Korean species of the sticklebacks(Pisces, Gasterosteidae) in Korea. Korean J. Syst. Zool. 5: 107-120.
- Kobayashi, J. 1933. Ecology of a stickleback, Pungitius sinensis var. kaibarae(Tanaka). J. Sci. Hiroshima Univ. Ser. B, Div. 2, 2:71-89.
- Leiner, M. 1934. Die drei europäischen Stichlinge (Gasterosteus aculeatus L., Gasterosteus pungitius L. und Gasterosteus spinachia L.) und ihre Kreuzungsprodukte. Z. Morph. Ökol. Tiere 28: 107 154.(not cited directly)
- Miyadi, D., H. Kawanabe and N. Mizuno. 1976. Colored Illustrations of the Freshwater Fishes of Japan. Hoikusha Publ. Co. Ltd., Osaka: 289 -

291.

- Mizuno, N. 1962. Distributions of Cottus japonicus Okada(Cottidae) and Tukugobius flumineus Mizuno(Gobiidae), with special references to their peculiarities in both the land locking and the speciation from amphidromous ancestors.

 Mem. Osaka Univ. Liberal Arts and Educ. B 11: 129 161.
- Nelson, J. S. 1969. Geographic variation in the brook stickleback, *Culaea inconstans*, and notes on nomenclature and distribution. J. Fish. Res. Bd. Canada 26: 2431-2447.
- Park, E. H. and D. S. Kim. 1984. A procedure for staining cartilage and bone of whole vertebrate larvae while rendering all tissues transparent. Stain Technology 59: 269-272.
- Tanaka, S., K. Hirai and S. R. Jeon. 1982. Comparison of the morphology between eightspine sticklebacks, *Pungitius sinensis* f. *kaibarae*(Tanaka), from Korea and Japan. Freshwater Fishes(Tansuigyo) 8:70-72.
- Wootton, R. J. 1976. The Biology of the Sticklebacks. Academic Press, London. 387 pp.
- Yang, S. Y. and M. S. Min. 1990. Genetic variation and systematics of the sticklebacks(Pisces, Gasterosteidae) in Korea. Korean J. Zool. 33: 499 – 508.

한국산 잔가시고기 Pungitius sinensis kaibarae (Gasterosteidae, Pisces)의 인판의 발달

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금호강의 한 지류인 자양천에 서식하고 있는 잔가시고기 Pungitius sinensis kaibarae(Tanaka)의 인판 발생과정을 조사하였다. 인판의 발생 순서는 체장 13mm경에 미병부에서 최초로 나타났으며 체장 16mm 경에는 배지느러미가시의 상부에 흉부인판이 나타났다. 그후 이 두 점을 기점으로 하여 앞뒤로 인판이 차례차례 발생하여 체장 20mm경에 서로 연결되어 trachurus형의 배열이 완성되었다. 체장 22mm경에는 인판의 수가 32 - 33개로서 성체와 거의 같아졌다. 인판의 발달 상태는 어체의 부위에 따라 다르지만 체장 25mm경에는 최전방부를 제외한 대부분의 인판이 후기의 상태로 되었다. 최전방의 인판은 성체가 되어도 대부분이 중기 상태에 머물러 있었다. 모든 인판은 측선의 neuromast 주위에서 발생하였다. 그러나 최전 방의 5개까지의 인판 위에는 인판이 발생하지 않는 추가적인 neuromast가 있음이 발견되었다. 일본산 잔가시고기와의 관계 및 추가적인 neuromast 발견의 계통분류학적 중요성에 대하여 논의하였다.