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Review

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Molecular mechanisms of adaptation and evolution of odor perception in fishes with special reference to the evolutionary dynamics of chemosensory receptor gene families

Yasuyuki Hashiguchi and Mutsumi Nishida

Abstract Recent research on the molecular diversity and evolutionary dynamics of fish odorant/pheromone receptor gene families are reviewed in terms of achievements and future perspectives. Because olfaction plays a crucial role in various biological activities of fishes, such as foraging, migration and mating, an understanding of the evolutionary dynamics of their odorant/pheromone receptors may provide important insights into the mechanisms of fish adaptive evolution. Genes encoding the main odorant receptors (ORs) and type 2 vomeronasal receptors (V2Rs), two of the three main classes of odorant receptors in teleost fishes, have markedly increased copy numbers, being used for detecting a diverse array of odor chemicals. In addition, many homologs of trace amine-associated receptor (TAAR) genes, a class of chemosensory receptors recently identified in mice, have also been found in fish genomes, being considered to function as chemosensory receptors. Repertoires of ORs, V2Rs and TAARs are highly divergent among species, suggesting species-specific adaptation to different odor environments. Recent electrophysiological studies have suggested functional differences between ORs and V2Rs in fishes. In particular, it is likely that fish ORs perceive amino acids, bile acids, sex steroids and prostaglandins, whereas fish V2Rs detect mainly amino acids and peptides. Based on these findings, future directions are proposed for further exploration of the adaptive evolution of fish olfaction.

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Full Papers

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Comparative morphology related to reproductive strategy in two chum salmon (*Oncorhynchus keta*) populations in the Yurappu River

Naotaka Imai, Hideaki Kudo, and Masahide Kaeriyama

Abstract Differences in age composition, body size and morphology in adult chum salmon (*Oncorhynchus keta*) collected from mid October 2005 to early January 2006 in lower- and upper-reaches in the Yurappu River were investigated. Although no difference in age composition was evident between upper- and lower-reach populations, latter was characterized by significantly larger male body size than the former, there being no such difference in female body size. Stepwise discriminant function and allometric analyses revealed that the lower-reach males had greater body depth and smaller head proportions, including snout and upper-jaw lengths, than their upper-reach counterparts, lower-reach females having a larger caudal peduncle and smaller head proportions than those in the upper-reach. These morphological differences between populations may reflect plasticity and a trade-off between escapement risk and breeding competition in the reproductive strategy of chum salmon.

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The status of four native and three alien bitterling species (Acheilognathinae) in Lake Kasumigaura, Japan

Takahiro Morosawa and Masahiro Fujioka

Abstract Information about the habitats and status of bitterling species (Acheilognathinae) is limited even though 9 out of 14 native species in Japan are listed

in the Red Data Book. To collect such information, we captured fish in Mondori traps and measured environmental features such as current velocity and electric conductivity at 126 points in 13 sites along the shore of Lake Kasumigaura. An alien species, *Rhodeus ocellatus ocellatus*, was most abundant, followed by two native species, *Acheilognathus tabira* subsp. 1 and *A. melanogaster*. Of the two other native species that have been recorded in the lake, *Tanakia lanceolata* was captured in only 4 sites, and *A. typus* was not captured at all. On the other hand, *A. macropterus*, which was introduced in about 2000 at an estuary along the southern shore, was captured in 7 widely scattered sites in the lake. Generalized linear mixed models revealed that the occurrence of two native species, *A. tabira* and *A. melanogaster*, was negatively related to electric conductivity and/or concrete ditches, while the occurrence of *R. o. ocellatus*, the most successful alien bitterling species, was positively related to electric conductivity and concrete ditches, suggesting that *R. o. ocellatus* may have a higher tolerance than native bitterlings to habitat degradations such as eutrophication and concrete ditches.

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Discovery of the Japanese rosy bitterling, *Rhodeus ocellatus kurumeus*, in Nara Prefecture

Takuya Miyake, Kouichi Kawamura, Kazumi Hosoya, Toshio Okazaki, and Tadao Kitagawa.

Abstract The Japanese rosy bitterling (*Rhodeus ocellatus kurumeus*) is a cyprinid subspecies endemic to Japan. It is now on the verge of extinction due to hybridization with its subspecies *R. o. ocellatus*, introduced from mainland China. In this study, we examined 13 populations of *R. ocellatus* from Nara Prefecture, Japan, using morphological analysis and molecular analyses of mtDNA and microsatellites. One of the populations in Nara Park (Stn. 9) closely matched the morphological characteristics of *R. o. kurumeus*, while other populations more closely resembled *R. o. ocellatus*. The

former population possessed mitotypes of *R. o. kurumeus* only, while the latter possessed those of both subspecies or those of *R. o. ocellatus* only. Notably, mitotypes of *R. o. kurumeus* were widely observed in the Nara Basin. In addition to the presence of mitotypes reported in an Osaka population of *R. o. kurumeus*, the low genetic diversity of the population at Stn. 9 was comparable to that of the Osaka *R. o. kurumeus* population. In a principal component analysis of microsatellites data, three clusters were recognized; 1. *R. o. kurumeus* (including the population at Stn. 9), 2. *R. o. ocellatus* and 3. their probable hybrids. These results suggest that *R. o. kurumeus* was once distributed widely in the middle reaches of the Yamato River, but the remaining populations have hybridized with *R. o. ocellatus*, except for the population at Stn. 9. Low genetic diversity in the Nara Park population is considered a result of a population bottleneck in a small isolated habitat.

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Distribution of indigenous and non-indigenous mtDNA haplotypes of *Biwia zezera* (Cyprinidae) in northern Kyushu, Japan

Marina Horikawa, Jun Nakajima, and Takahiko Mukai

Abstract The Japanese cyprinid fish *Biwia zezera*, naturally distributed on the Nobi Plain and in the Lake Biwa–Yodo River system, is also distributed in northern Kyushu, although it has been unclear whether or not that population was indigenous. Sixty-five specimens of *B. zezera* was sampled from four localities in northern Kyushu (Ima River, Naka River, Chikugo River and Kase River) and the mitochondrial cytochrome *b* gene sequenced for a comparison with sequences from Nobi Plain and Lake Biwa populations. The phylogenetic and population genetic analyses of mtDNA sequences suggested that the northern Kyushu populations of *B. zezera* were naturally distributed in the area, although three of the 11 mtDNA haplotypes in the Kyushu populations may have been artificially introduced from Lake Biwa.

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Seasonal migration of Hotoke-dojo loach *Lefua echigonia* in a spring-derived water stream of Nishikinugawa District, Tochigi Prefecture, Japan

Takumi Moriyama, Masakazu Mizutani, and Akira Goto

Abstract The Yagawa stream, a primarily canal-fed agricultural waterway in the west Kinugawa district, Tochigi Prefecture, Japan, and an upstream, spring-derived waterway were surveyed from October 2002 to May 2005, using trapping and marking and recapture methods, to establish the migration characteristics of Hotoke-dojo loach between the upper and lower portion of the waterway system. The ecology of the loach was also studied, considering the relationships between migration and seasonal changes in environmental factors and maturation of individuals. Although the Yagawa stream flowed throughout the year, the spring and associated waterway dried up for about two months and one month, respectively, every winter. Trapping resulted in the capture of many individuals having each spring (from late March to early June), the number having increased progressively over autumn and winter (October to February). Upstream migration in the Yagawa stream to the spring-fed waterway began immediately after water started to flow in the latter in spring, mature individuals comprising 64.7% of the migrating individuals in 2004 and 70.7% in 2005. The numbers of migrating loach in autumn and winter was positively correlated with differences in water temperature between the Yagawa stream and the spring-fed waterway. Marking and recapture confirmed that individuals migrated from the latter downstream to the Yagawa stream during this period. The results indicated that Hotoke-dojo living in the survey area spawned in the spring-fed waterway, a temporary aquatic area, and overwintered in the constantly flowing Yagawa stream.

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**Annual changes in gonadal histology and serum profiles of sex steroids in KAJIKA,
Cottus sp. SE (small egg type), under rearing conditions**

Kentaro Fukui, Ryoji Fujii, Daisuke Tahara, Youichi Hayakawa, and Yasunori Koya

Abstract Gonadal development, kidney weight and serum profiles of sex steroids in reproductive KAJIKA, *Cottus* sp. SE (small egg type), were monitored over a 12-month period. The female reproductive cycle was divided into the following 5 periods based on oocyte development: recovery period (May to August), cortical alveoli formation period (September), vitellogenic period (October to December), reproductive period (January and February) and spent period (March and April). Based on ovarian histological observations, females may spawn 2 or 3 times in one reproductive period. Serum levels of estradiol-17 β increased during the vitellogenic period, indicating regulation of the vitellogenic progress. Serum 17,20 β -dihydroxy-4-pregnen-3-one in females exhibited high levels only during the reproductive period, suggesting a role in inducing final oocyte maturation. The male reproductive cycle was divided into the following 5 periods based on testicular histological observations: resting period (June to August), spermatogonial proliferation period (September), spermatogenic period (October and November), reproductive period (December to February) and spent period (March to May). Parasperm formation, which is known in some cottidae species, was noted, occurring in the sperm ducts during the spermatogenic period, but only in the main testicular lobes during the reproductive period. Serum 11-ketotestosterone levels in males increase of continually during the spermatogenic period, suggesting a role regulation of spermatogenesis in this species.

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A method for identifying individual Japanese charr, *Salvelinus leucomaenoides*, using parr marks

Masayuki Yagyu, Hiroshi Nakamura, and Toshitaka Miyazaki

Abstract To develop a method for harmlessly identifying individual Japanese charr, *Salvelinus leucomaenoides*, the distribution and shape of parr marks were examined in fluvial specimens. Fish captured in six mark-and-recapture samples from a tributary of the Tenryu River (35°41'N, 138°07'E, El.1,045 m) between June 2005 and May 2006 were photographed, and the linear distance and width of each parr mark on the lateral line measured from digital images. Similarities between and within individuals during growth were analyzed using square Euclidean distances. A photo-matching test was also conducted.

The number of parr marks and the parr mark ratio increased with body length (BL) up to 50 mm BL. The square Euclidean distance was not correlated with the number of days until recapture, although there was a negative correlation with initial BL (distribution: $r=-0.25$, $P=0.029$, width: $r=-0.24$, $P=0.038$). The similarity analysis discriminated 74% and 95% of parr mark distribution and width, respectively, in the same individuals. In the photo-matching test, 8 of 10 testers identified 20 individuals from 40 photographs (average accuracy 97%), the exercise requiring about 2 h per tester.

This method, utilizing parr marks, enables the identification of individuals with visible parr marks on the base of tail ($BL > 32$ mm) without harming the fish. This should be useful for identifying fish with parr marks, especially in endangered charr populations.

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Short Reports

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Record of a platytroctid fish, *Maulisia argipalla* (Argentiniformes: Platytroctidae), from off northeastern Japan

Makoto Okamoto, Hiroya Sugisaki, Kazuhisa Uchikawa, Masatoshi Moku, and Yuji Okazaki.

Abstract Two specimens of a platyptroctid fish, *Maulisia argipalla* Matsui and Rosenblatt, 1979 (96–188 mm in standard length), were collected by midwater trawl (300–750 m) from east of Aomori and Miyagi Prefectures, in the transition waters between the Kuroshio and Oyashio fronts off northeastern Japan. *Maulisia argipalla* is distinguished from other congeners by having the following combination of characters: a shoulder pit behind supracleithrum, photophores present ventrally on body, supraorbital extending laterally beyond the widest part of frontal, thoracic organ (THO) small and round. This is the first record of the species from Japan, and it is given a new Japanese name, Kuzuboshi-hanameiwashi.

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New records of a stichaeid fish, *Alectrias cirratus*, from Japan

Osamu Tsuruoka, Takuzo Abe, Nagaaki Sato, and Mamoru Yabe

Abstract Seven specimens of the stichaeid fish, *Alectrias cirratus* (Lindberg, 1938), were collected from the shallow waters off Shiretoko (eastern Hokkaido), Usujiri (southern Hokkaido), and Shizugawa (Miyagi Pref.), Japan. This species was previously known only from Primorskii, Russia, in the Sea of Japan. The present specimens represent the first records from Japan, the Sea of Okhotsk, and the Pacific Ocean for this species. Two specimens from Shizugawa are the southernmost record for the species. In addition, the coloration of fresh specimens and the reproductive ecology of the species are briefly commented upon.

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Records of two eelpouts, *Lycodes uschakovi* and *Lycodes concolor* (Perciformes:

Zoarcidae), from Japan

Satoshi Ikeda, Hisashi Imamura, and Kazuhiro Nakaya

Abstract Twenty specimens of an eelpout, *Lycodes uschakovi*, were collected from off Monbetsu, north-eastern Hokkaido on the coast of the Okhotsk Sea, and four specimens of *Lycodes concolor* from around Kitami-Yamato Bank in the Okhotsk Sea. *Lycodes uschakovi* is characterized by a scaleless belly, submental crests ill-developed and united anteriorly, lateral line mid-lateral in position, and body grey or greyish pink with 8–11 distinct or indistinct transverse white bars extending onto dorsal fin. *Lycodes concolor* is characterized by 22 pectoral fin rays, scaleless predorsal area, submental crests moderately developed and not united anteriorly, lateral line ventral, distantly spaced neuromasts on the mid-lateral part of the body, body and head uniformly blackish brown, and nostril tube brown. These two species are described in detail and reported as the first records from Japan.

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First record of a cardinalfish *Neamia articycla* (Perciformes: Apogonidae) from Amami-oshima Island, Ryukyu Islands, Japan

Koichi Shibukawa, Yohko Takata, and Gento Shinohara

Abstract Seven specimens of a cardinalfish *Neamia articycla*, collected from dead-coral rubble bottoms of shallow to relatively deep coral reefs (at 4–60 m depths) at the Ryukyu Islands, Japan, represent first record from Japanese waters and northernmost record for the species. A brief account of the morphology of the Japanese specimens is given, in addition to a photograph showing the coloration of a freshly-collected specimen. The species is also newly recorded from Lombok Island, Indonesia and Mabul Island (northeast of Borneo), Malaysia. The specimens are compared with the original description, and some discrepancies in scale condition and coloration noted. The new Japanese name “Kakushi-yatsutoge-tenjikudai” is proposed for the species.

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Distribution of mtDNA haplotypes of smallmouth bass (*Micropterus dolomieu*) in Japan

Chika Sato, Takahiko Mukai, Taiga Yodo, Toru Sakuma, and Katsuki Nakai

Abstract The geographical distribution of mtDNA haplotypes of non-indigenous smallmouth bass (*Micropterus dolomieu*) populations in Japan were examined utilizing nucleotide sequences of the mtDNA control region from 208 individuals collected from 20 localities. A total of three haplotypes (n, o and p), distinguished by a nucleotide substitution or an insertion/deletion of 294bp, were found in Japanese freshwater systems. In eastern Japan, most *M. dolomieu* populations had two haplotypes (n and p), the frequencies being similar among localities. On the other hand, haplotype n or p was fixed (or nearly fixed) in lakes and ponds in western Japan. These results suggested that the non-indigenous populations of smallmouth bass in Japan were initially established by an introduction into eastern Japan, the western Japanese populations being subsequently founded by relatively small numbers of individuals.

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