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Identification and growth rates comparison of divergent migratory contingents of Japanese eel (*Anguilla japonica*)

Wann-Nian Tzeng[a,*], Yoshiyuki Iizuka[b], Jen-Chieh Shiao[a],
Yoshiaki Yamada[c], Hideo P. Oka[c]

[a]*Department of Zoology, College of Science, National Taiwan University, Taipei 106, Taiwan, ROC*
[b]*Institute of Earth Sciences, Academia Sinica, Nankang, Taipei 106, Taiwan, ROC*
[c]*IRAGO Institute, 377 Ehima-Shinden, Atsumi-Cho, Atsumi-gun, Aichi-Ken 441-3605, Japan*

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Abstract

The strontium (Sr) and calcium (Ca) concentrations in the otoliths of the Japanese eels *Anguilla japonica* collected from China, Japan and Taiwan were measured by electron probe micro-analyzer. The Sr/Ca ratios indicated that the eels beyond elver stage can be classified into three types of migratory contingents. Type 1 (seawater), the Sr/Ca ratios from approximately 150 μm from primordium to edge of the otolith maintained at the level of approximately 4–10 ‰, indicating that the eel after elver stage stayed in sea water until the silver eel stage. Type 2 (freshwater), the ratios were lower than 4 ‰, indicating that the eel stayed in freshwater from elver stage to the silver eel stags. Type 3 (estuarine), the ratios fluctuated between those of Types 1 and 2, indicating that eel migrated between freshwater and sea water before the silver stage. The estuarine contingents constituted the majority of the eel population and grew faster than the freshwater contingents.

Keywords: Japanese eel; Otolith; Sr/Ca ratio; Environmental history; Growth

1. Introduction

Japanese eel *Anguilla japonica* is a catadromous fish. It is widely cultivated in the inland freshwater of Taiwan, China and Japan. Due to rapid development of the eel aquaculture,

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[*] Corresponding author. Fax: +886-2-23636837.
E-mail address: wnt@ccms.ntu.edu.tw (W.-N. Tzeng).

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the underground water reserves in Taiwan were overdrawn, which has led to land subsidence and seawater ingress in coastal areas. If the eel can be cultivated in seawater, the use of freshwater can be reduced and the land subsidence can be prevented.

The eel's life cycle has five principal phases, i.e. leptocephalus, glass eel, elver, yellow eel and silver eel (Bertin, 1956). Japanese eel was presumed to be a panmictic population (Sang et al., 1994), spawning in the waters west of the Mariana Islands, 15°N 140°E (Tsukamoto, 1992). Their larvae, leptocephali, drifted with the North Equatorial Current to the west and turned to the north into the Kuroshio current in the eastern coast of the Philippines. Upon arriving in the continental shelf of north Asian countries, they metamorphose to glass eel and become pigmented elvers in the estuary. They spend approximately 5–6 months migrating from spawning ground to the estuary (Tzeng, 1990; Tzeng and Tsai, 1992; Cheng and Tzeng, 1996). Elvers have been overfishing for aquaculture in Asian countries and thus the eel population is obviously decreased (Tzeng, 1983, 1984a,b, 1985, 1986, 1997). The yellow eels live in the rivers approximately 5–8 years (Tzeng et al., 2000a), then metamorphose to silver eel, migrate downstream to spawn and die (Tesch, 1977).

In the recent years, the trace element strontium (Sr) in the otoliths was used to discriminate the divergent salinity history of the European eels (Tzeng et al., 1997, 1999). Tsukamoto et al. (1998) and Tzeng et al. (2000b) further indicated that some of the European and Japanese eels may skip the freshwater phase and can grow up in the brackish water and sea water until silver eel stage. These lead us to speculate that the eel may have divergent migratory contingents with different life-cycle pathway particularly in the yellow-phase growth stage. The migratory contingents are the intrapopulation migratory groups (Clark, 1968), which were recently intensively studied in the anadromous striped bass *Morone saxatilis* (Secor, 1999, 2001). But the mechanism of the formation of migratory contingents is still not clear.

This study is to identify the different migratory contingents of the eel by their otolith Sr/Ca ratios and to compare their growth rates among different contingents.

2. Material and method

A total of 212 Japanese eels were collected from China, Japan and Taiwan, respectively (Table 1). The eels were collected for artificial maturation and thus all of them were silver eels except those collected from Taiwan. The Pearl River is the third largest river in China, and has a large freshwater volume for eels. The eels from China were collected at Fan-Yu in the lower Pearl River (113°30′ E and 23°N), ca. 200–250 km from South China Sea, in the fall of 1996 during their downstream spawning migration (Tzeng et al., 2000a). The eels from Japan were collected from the brackish Mikawa Bay (137°E and 34°45′ N) on the east coast and from freshwater Shinjiko lake and brackish Naka-umi inland sea (132°45′ –133°45′ E and 35° 30′ –35°35′ N) on the north coast of western Japan, in June 1998 to June 1999. Mikawa means three rivers in Japanese, because there are three small rivers flowing into the bay. The eels from Taiwan included yellow and silver eels. They were collected from Kao-Ping River estuary on the southwest coast of Taiwan (120°50′ E and 22°40′ N), in November 1998 to November 1999.

Table 1
Environmental history of the eels collected from China, Japan and Taiwan. (Type 1, seawater; Type 2, freshwater; Type 3, estuarine)

Country	Area	Salinity (0/00)	Maturation stage	Sample size			Total length (cm)		Age (year)		Composition of life history types (%)		
				Male	Female	Total	Male	Female	Male	Female	Type 1	Type 2	Type 3
Japan (east)	Mikawa Bay	Brackish (24.6–32.7)	Silver	–	42	42	–	73.6±7.67	–	6.05±2.14	22.8	18.2	59.0
Japan (west)	Shinjiko and Naka-umi	Freshwater and Brackish (0–32)	Silver	45	–	45	52.88±7.29	–	5.02±1.64	–	20.0	42.2	37.8
China	Pearl River	Freshwater (0)	Silver	38	36	74	48.32±4.48	61.44±4.05	6.38±1.62	8.30±1.62	–	100.0	–
Taiwan	Kao-Ping River	Brackish (0–32)	Silver	3	15	18	45.1±7.79	60.32±9.10	5.33±1.53	6.43±1.79	13.3	6.7	80.0
			Yellow	–	33	33	–	46.57±6.52	–	4.76±1.37	9.1	27.3	63.6
			Silver + yellow	3	48	51	45.1±7.79	50.56±9.49	5.33±1.53	5.25±1.65	9.8	23.5	66.7
Total				86	126	212							

Eels were collected using both set net and eel tubes. For each eel collected, total length was measured to the nearest 0.5 cm. Then, the eels were dissected and the sexes were determined from the histology of the gonad. The largest pair of otoliths (sagitta) of the eels were extracted for age determination and Sr/Ca ratio measurement.

Electron probe micro-analyzer (EPMA, JEOL, and JXA-800 M) was used to analyze the weight ratio (wt.%) of Sr to Ca in eel otoliths. The procedure of the preparation of otolith and the beam condition for the analysis of Sr and Ca concentration are similar to the previous study (Tzeng et al., 1997). According to the level of Sr/Ca ratios in the otolith, the eels were classified into sea water, freshwater and estuarine contingents.

After electron microprobe analysis, the otoliths were repolished and etched with 5% ethylene diamine tetra-acetate (EDTA, PH adjusted with NaOH to 7.4) for 3 min to enhance the annuli (Tzeng et al., 1994). The annuli were discriminated using a transmitted light microscope. Age was determined by counting the annuli in the otoliths.

The radii from primordium to elver mark (r_o), annuli (r_n) and the edge of otolith (R) were measured. The total lengths at elver stage (l_o) and at annulus formation period (l_n) were estimated, respectively, by Dahl–Lea formula (Francis, 1990):

$$l_n = r_n L R^{-1} \tag{1}$$

where L is total length of the eel at capture. The mean l_o and l_n were compared among contingents by sexes because the growth rates of the eel were significantly different between males and females (Tzeng et al., 2000b). To avoid the effect of geographic interaction, only the 87 eels collected from Japan were selected for the comparison of growth rate. The females were collected from Mikawa Bay, and the males from Shinjiko and Naka-umi. The significance of difference in mean values l_o and l_n of the eel among contingents was tested by Kruskal–Wallis one way analysis of variance on ranks (Zar, 1984).

3. Result

3.1. Otolith Sr/Ca ratio patterns

The ratios in the otoliths of the eels before elver stage approximately 150 μm from primordium were similar among individuals, indicating that the migratory environmental history was similar among individuals during the marine leptocephalus stage. However, the Sr/Ca ratios in the otolith beyond elver stage were obviously classified into three types of migratory contingents (Fig. 1). The Sr/Ca ratios reached a peak (>16‰) at metamorphosis from leptocephalus to glass eel, decreased to approximately 4‰ at the time when glass eel became elver and migrated from seawater to freshwater water. The temporal change of Sr/Ca ratios in the otoliths of the eels before elver stage was similar to our previous study (Tzeng and Tsai, 1994). The three different types of Sr/Ca ratios in the otoliths of the eels beyond the elver stage were briefly described as follows.

Type 1 (seawater), the ratios in the otolith in the layer approximately 150 μm from primordium to the edge of the otolith averaged approximately 6‰, indicating the elvers

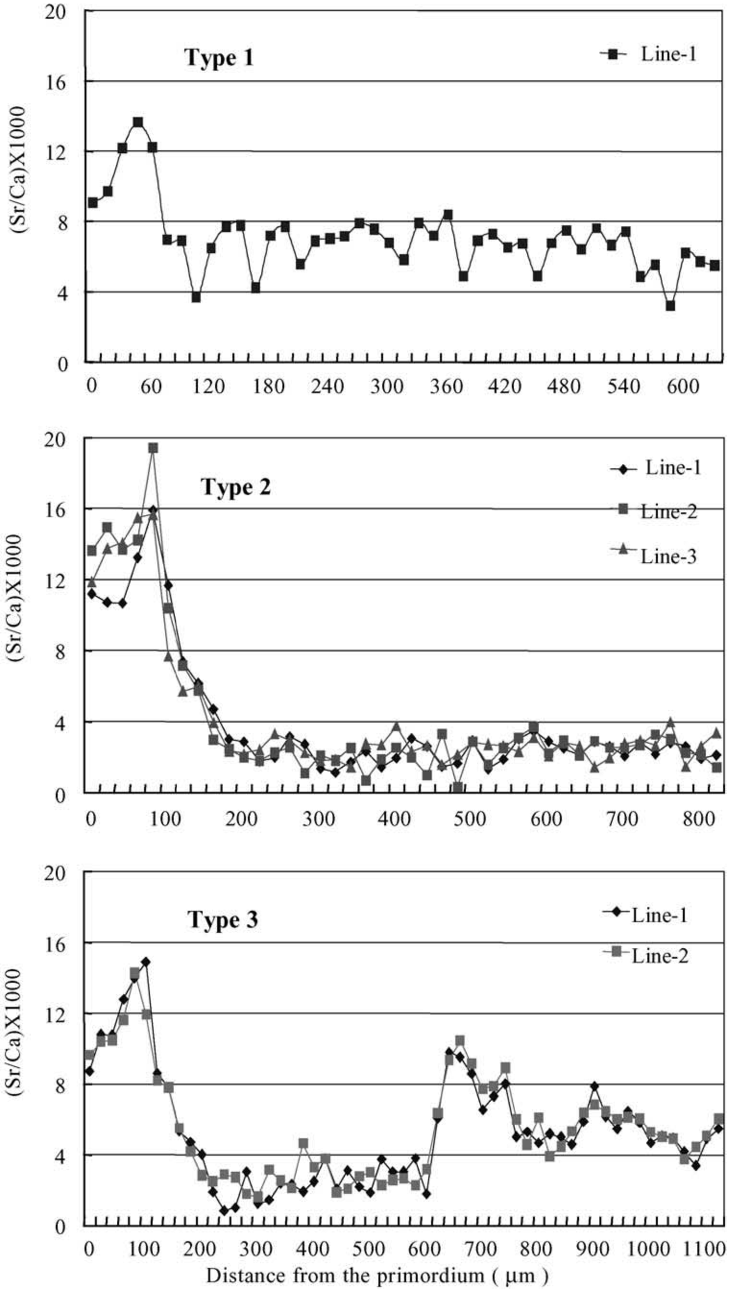

Fig. 1. Environmental history of the eel as indicated by the temporal pattern of the Sr/Ca ratios in their otoliths. Type 1: seawater; Type 2: freshwater; Type 3: intermediate between freshwater and seawater. Lines 1–3 are replicate measurements.

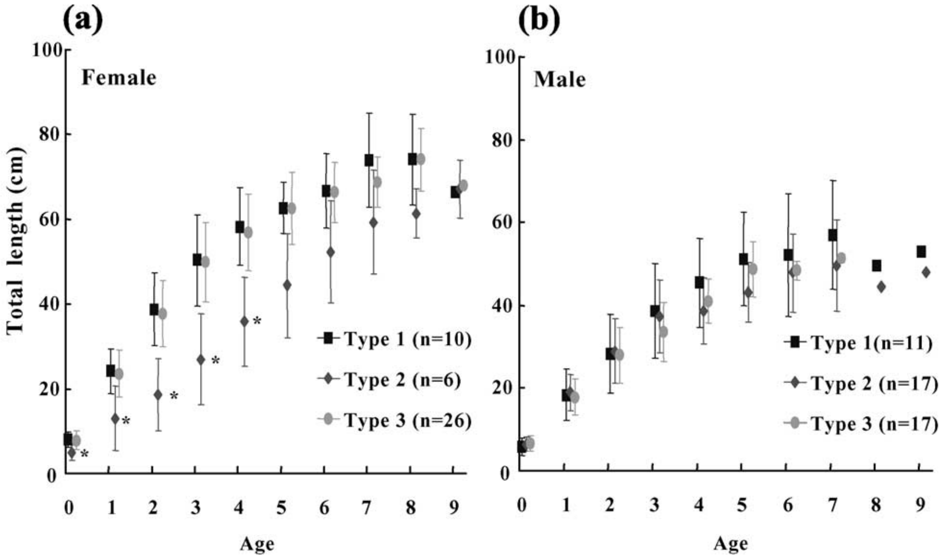

Fig. 2. Comparison of mean (±S.D.) growth rates among life history types. (a) Females collected from Mikawa Bay, (b) males collected from Shinjiko and Naka-umi. Type 1 = sea water, Type 2 = freshwater, Type 3 = estuarine; *n* = sample size; *, the mean total length was significantly smaller at the significant level $P < 0.05$. The data of females at age 9 and males beyond age 8 were excluded in the comparison because of small sample size.

had not been migrating to freshwater but stayed in the brackish water to grow until the stage at metamorphosis to silver eel (Fig. 1a).

Type 2 (freshwater), the ratios in the otolith in the layer approximate 150 μm from primordium to the edge of the otolith fluctuated between 0 ‰ and 4 ‰, indicating that the eels after elver stage had been migrating to freshwater to grow until the stage at metamorphosis to silver eel (Fig. 1b).

Type 3 (estuarine), an intermediate type between types 1 and 2. The Sr/Ca ratios in the otolith beyond elver stage were lower than 4 ‰ in the early yellow eel stage, but thereafter increased to higher than 4 ‰ until the edge of otolith, indicating that the elvers had been migrating to freshwater, but returned to sea water at yellow eel stage and stayed there until silver eel stage (Fig. 1c).

The ratios of the three environmental histories of the eels varied with areas (Table 1). The eels from Kao-Ping River estuary were predominantly estuarine eels (66.7%), as were the eels from Mikawa Bay (59%). The freshwater eel from these two areas constituted only of 23.5% and 18.2%, respectively. The ratios of estuarine eels were slightly decreased (37.8%) in the Shinjiko and Naka-umi areas because the Shinjiko is a freshwater lake. The eels from the middle reach of the Pearl River were completely freshwater eels (100%). The estuarine rather than the freshwater contingents constituted the majority of the eel population in both Japan and Taiwan, i.e. a high percentage of eels did not migrate to freshwater during their yellow-phase stage.

3.2. Comparison of growth rates of different migratory contingents

Mean total length of the eels at elver mark (l_o) and annulus formation (l_n) was compared among three life-history types by sexes (Fig. 2). The mean total length of freshwater females (Type 2) at the younger ages (less than 5 years old) was significantly smaller than those of seawater and estuarine ones (Types 1 and 3) ($P < 0.05$), but it was not significantly different among the three types beyond 5 years old and between Types 1 and 3 through all age groups ($P > 0.05$). On the other hand, the mean total length of males was not significantly different among the three types ($P > 0.05$). These indicated that the difference in growth rates among types was sex-dependent.

4. Discussion and conclusion

The life-history scan of Sr/Ca ratios in the otoliths validated that the Japanese eels diverged into sea water, freshwater, and estuarine contingents during the yellow-phase stage (Fig. 1). The eel was a diadromous species, spawning in seawater and growing in freshwater (McDowll, 1988). However, we found that majority of the eel population inhabited in the estuarine brackish waters during growth phase (Table 1). This indicated that the habitat selection of the eel during growth phase may be opportunistic, not obligatory. Selection of the growth habitat may depend on the food availability and carrying capacity of the habitat. The rivers in both Japan and Taiwan were small, with limited freshwater, and thus most eels may be compelled to grow in the estuary. Although we did not examine the eels in the upper stream of the rivers in both Japan and Taiwan, the eels in the upper stream will be caught in the estuary during their downstream spawning migration. Accordingly, we believed that the ratios of life-history types of the silver eels collected in the estuary can represent those of the whole river system. The estuarine contingent contributed to approximately 60% of the eel population in both Japan and Taiwan. However, we do not know if the estuarine contingent is predominant in the eel population in the Pearl River System, because the fish caught in the estuary of the river were not examined.

Like other teleost fishes, the osmolality of the eel's body fluid is maintained at approximately 300–400 mosM. The osmolality of the sea water is 1000 mosM at the salinity of 35‰. In other words, the salinity of the eel's body fluid is approximately 10.5–14‰, which was close to that of brackish water. This implies that the energetic cost of the osmoregulation of the eel would be lower in brackish water than in an extreme environment such as freshwater or high saline sea water. Much energy is required to regulate the osmotic pressure of their body fluid to homeostasis (Hirai et al., 1999). The osmoregulation consumes energy requiring eels to consume more food to prevent a decrease in growth. The estuary is one of the most productive areas on earth comparable to that of coastal upwelling area (Haedrich and Hall, 1976). From the view point of osmoregulation and food availability, living in the estuary during growth phase is advantageous, and it may be the reason why there are a high percentage of eels staying in the estuary during growth phase (Table 1) and why the growth of the eel was faster in brackish water than in freshwater (Fig. 2). In aquaculture terms, it is suggested to culture eels with brackish water rather than with the traditional freshwater system. This can prevent the overuse of freshwater and slow down the land subsidence. The salinity necessary for optimal growth of the eel during culture needs to be evaluated.

The evolution of the migratory contingents of the eels is not clear, but it may be attributed to genetics or environmental adaptation (Nordeng, 1983; Gross, 1985; Secor, 1999). The Japanese eel is considered to be a panmictic population (Sang et al., 1994). If the divergent migratory contingents of the eel having different genotypic structure needs to be evaluated. However, there is a widely hold view that life histories in animals are selected for and adapted to maximizing the production of progeny (Schaffer and Elson, 1975; Stearns, 1977; Dingle, 1980; Miller and Brannon, 1982; Gross, 1985). In evolutionary terms, the persistence of migration needs to be seen in relation to the balance of advantages obtained from migration and the costs incurred by the population/species. Advantages include such aspects as increased food supply, avoidance of potentially harmful environmental conditions and/or a movement to more favorable ones, the occupation of habitats that have specific or specialized habitat requirement, and the availability of more living space. Costs of migration include mortalities resulting from migration itself, changed environmental conditions that may be intolerable (in diadromous fishes, specifically osmoregulatory stress) (McDowll, 1988). Gross (1987) proposed that diadromy occurs when the gain in fitness from using a second habitat minus the migration costs of moving between habitats exceeds the fitness from staying in only one habitat. When the elvers migrate from offshore seawater to upstream freshwater for living habitat and feeding, they have to overcome the osmotic pressure of salinity gradient environment. If they stayed in estuary their osmoregulatory cost would be lower than those in both freshwater and sea water. Meanwhile, the stability of water volume, living space and food abundance for the eel is generally superior in the estuary than the upper stream of the river in the island countries, such as Japan and Taiwan. The estuary functions as a nursery and feeding ground for the juveniles of many inshore fish and offers commercially important fish (Wallace et al., 1984; Blaber and Blaber, 1980; Day et al., 1981; Lenanton, 1982; Bell et al., 1984). This is because the estuary provides suitable food resources as well as shelter, absence of turbulence, and a reduction of predation (Blaber et al., 1985). These conditions may confine the eel to the estuarine waters, and subsequently the estuarine contingents were predominant in the Japanese eel population in both Japan and Taiwan.

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