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BRIEF COMMUNICATION

The exotic American eel in Taiwan: ecological implications

Y.-S. HAN*, C.-H. YU*, H.-T. YU*, C.-W. CHANG*, I-C. LIAO† AND W.-N. TZENG*‡

**Institute of Zoology, College of Science, National Taiwan University, Taipei, Taiwan, R.O.C. and †Taiwan Fisheries Research Institute, 199 Hou-Ih Road, Keelung, Taiwan, R.O.C.*

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Six American eels *Anguilla rostrata* were identified in the Kaoping River of south-western Taiwan by sequencing the mitochondrial cytochrome b gene (mt*Cyt-b*). This was the first record of an exotic eel species in the natural waters of Taiwan. The exotic eels may have been imported from North America at the elver stage for aquaculture and escaped from culture ponds.

Key words: *Anguilla rostrata*; species identification; mitochondrial cytochrome b gene.

Anguillid eels are catadromous fishes that spawn in the ocean and develop in fresh water (Tesch, 1977). There are 15 described species and 3 subspecies of anguillid eels (Ege, 1939; Castle & Williamson, 1974). All anguillids are Indo-Pacific except the European eel *Anguilla anguilla* (L.) and the American eel *Anguilla rostrata* (LeSueur) inhabiting the North Atlantic Ocean. In Taiwan, there are four native anguillid eels: *Anguilla japonica* Temminck and Schlegel, *Anguilla marmorata* Quoy and Gaimard, *Anguilla bicolor pacifica* Schmidt, and *Anguilla celebesensis* Kaup (Han *et al*., 2001; Tzeng, 1982; Tzeng & Tabeta, 1983). The Japanese eel *A. japonica* is a commercially important species for aquaculture in Taiwan. As the catch of Japanese eel elvers has been deemed insufficient to meet the demand of eel aquaculture (Tzeng, 1996), elvers of exotic eel species, mainly American eel and European eel, have been imported since 1969 and 1977, respectively (Li, 1997). In Japan, European eels have been found in the Shinjiko Lake and Mikawa Bay (Zhang *et al*., 1999), and are abundant in the Uono River (Aoyama *et al*., 2000). To date, no exotic eels have been documented in the natural waters of Taiwan. The external morphologies of native and introduced eels are quite similar, complicating the identification of exotics. In this study, exotic eel species were identified by phylogenetic analysis of the mitochondrial cytochrome b gene (mt*Cyt-b*). In addition, the age, growth and maturity of exotic eels were examined to evaluate the condition of introduced eels in the natural environment of Taiwan.

Among 184 eels collected from the Kaoping River of south-western Taiwan between July 1999 and February 2001, six individuals, four females (AR1 to AR4) and two males (AR5 and AR6), were found that differed morphologically from Japanese eels. These eels had thick snouts, round heads, abundant mesenteric fat and unusually large eyes. Their total length (L_T), body mass, and horizontal and vertical diameters of the left eye were measured and compared with those of Japanese eels. The ocular index was calculated according to Pankhurst (1982). Maturation stages of eels were divided into silver, pre-silver and yellow stages by the colouration of the pectoral fins and the dorsal region

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‡Author to whom correspondence should be addressed. Tel.: +886 2 23639570; fax: +886 2 23636837; email: wnt@ccms.ntu.edu.tw

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TABLE I. Comparison of morphological and gonadal characteristics between the unknown eels and Japanese eels

	Unknown eels	Japanese eels
Sample size	6	178
Sex composition	♀4 ♂2	♀154 ♂24
L_T (mm)	415–700	362–785
Body mass (g)	125–591	55–829
Age (year)	8–12	3–11
No. of vertebrae	105–107	112–119*
Ocular index (OI)	6·71–20·71	1·87–6·90
Oocyte diameter (μm)	90–150	25–225

*Data according to Ege (1939).

as well as by silver colour on the belly and the state of gonadal development. Vertebral counts were determined from soft X-ray photographs. Sex was determined by gonadal histology. Maturation stages of ovary and testis were classified according to Yamamoto & Yamauchi (1974) and Miura *et al.* (1991), respectively. Sagittal otoliths of these eels were extracted and prepared for age determination following Tzeng *et al.* (1994). DNA was extracted from the liver tissues of the six unknown individuals and eleven randomly selected individuals of *A. japonica* (AJ, n=10), and *A. marmorata* (AM, n=1). The two oligonucleotide primers used for PCR amplification of the mitochondrial cytochrome b gene, named *Cyt-b*1 (5′-TGCTAACGATGCCCTAGTGG-3′) and *Cyt-b*2 (5′-CTAGT CAACCTACTAATGGG-3′), were designed based on the conserved regions of cytochrome b sequences of the genus *Anguilla* deposited in GenBank/NCBI. The locations of *Cyt-b*1 and *Cyt-b*2 are at nucleotides 39–58 and 1098–1117, respectively, in the open reading frame of the mt*Cyt-b*. After PCR, both strands (1079 bp length) were sequenced commercially (Mission Biotech Inc., Taipei, Taiwan). DNA sequences of the six unknown individuals (AR1–AR6) and other known eel species were then aligned with those of 15 anguillid species deposited in the GenBank using Clustal W (v1.81) network service. A phylogenetic tree was constructed by the neighbour-joining method with genetic distance calculated by Kimura's two-parameter model using the programme MEGA 2 (Kumar *et al.*, 2001).

The total length and body mass of the six unknown eels were comparable to those of Japanese eels (Table I). Their ocular indexes ranged between 6·71 and 20·71 (Table I), however, markedly larger than those of Japanese eels (1·87–6·90, Table I). Such large eyes are known only in some American eel specimens (Vladykov, 1973). The gonads of the unknown eels were intermingled with adipocytes [Fig. 1(a), (b)], similar to those of American eels (Wenner & Musick, 1974), but different from Japanese eels (Fig. 1(c), (d)]. The numbers of vertebrae of these six eels ranged between 105 and 107, overlapping with the range of American eels (103–109) (Ege, 1939), but markedly less than Japanese eels (112–119) (Ege, 1939). Mesenteric fat was abundant in the six individuals, in contrast to the low level of mesenteric fat in Japanese eels. Topology of the phylogenetic tree showed that the six unknown eels, AR1 to AR6, formed a clade with the American eel with robust bootstrap support (Fig. 2). The mt*Cyt-b* sequences of the six eels showed low nucleotide differences (AR1–AR6, 1,9,6,7,8,5, respectively) compared to that of American eel deposited (AF006716) with mean transition : transvertion ratio of 4·14, and all of these nucleotide differences were synonymous except AR2, which had four nonsynonymous sites. The mt*Cyt-b* sequences of the six eels could be clearly distinguished from the closest species European eel (48–53 nucleotides difference). Hence, the six unknown eels were validated to be American eel *A. rostrata*.

According to the sexual maturation of Japanese eel (Yamamoto & Yamauchi, 1974), one female of the exotic eels was at the pre-silver stage with oocytes in the peri-nucleolus stage and the lipid vesicles apparent at the periphery of the oocyte. The other three

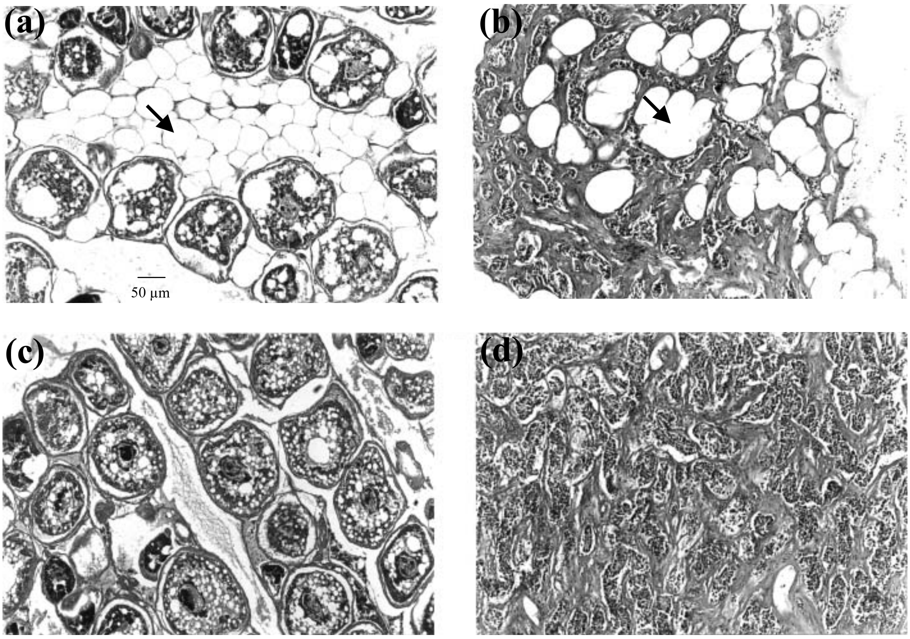

FIG. 1. Microscopic histology of the gonads stained with haematoxylin and eosin (magnification × 200). (a) Female American eel, 612 mm L_T, 519·2 g. (b) Male American eel, 415 mm L_T, 125·1 g. (c) Female Japanese eel, 695 mm L_T, 775·4 g. (d) Male Japanese eel, 494 mm L_T, 177·7 g. ←, adipocytes.

females were already in the silver stage with oocytes in the oil-droplet stage with a mean diameter >140 μm [Table I and Fig. 1(a)].

It seems impossible that American eels had migrated naturally from the North Atlantic to Asia (Tsukamoto & Aoyama, 1998) and the only possible route would seem to be human transplantation. The six exotic American eels may have been discarded by a culturist or escaped from culture ponds and have inhabited the natural waters of Taiwan for years (Table I). They were found to be at the silver stage in July and October, which is somewhat earlier than that of Japanese eels (August to February) in the rivers of Taiwan. The overlap in time of metamorphosis from yellow to silver eel between native Japanese eels and exotic American eels may lead to the risk of interspecific hybridization. Since the spawning ground of the American eel is in the Sargasso Sea, North Atlantic (Schmidt, 1925), and the routes of eel migration are believed to be species-specific (Tsukamoto, 1992), the American eel in Taiwan is unlikely to propagate successfully. As the life cycle and larval transport routes (e.g. North Equatorial Current and Kuroshio Current in Japanese eel; North Equatorial Current and Gulf Stream in American eel) of both eel species are very similar to each other (Cheng & Tzeng, 1996; Wang & Tzeng, 1998, 2000), however, the possibility that American eels may establish new spawning grounds in Asia cannot be excluded. A migrating European eel in the silver phase was captured in the East China Sea (Aoyama *et al*., 2000), indicating that the spawning migration of introduced eels is possible.

The potential ecological impacts of exotic eel species on native eels may be significant: (a) exotic eels may introduce pathogens and parasites to which native eel species were previously unexposed; (b) exotic eels may compete with native eel species for space and food; (c) interspecific hybridization may cause a disturbance of local gene pools. Hence, the policy of stocking non-native eel species to make up the shortage of Japanese eels in Taiwan should be re-evaluated carefully. Instead, management efforts should focus on the protection of native eels to sustain pure populations of this species.

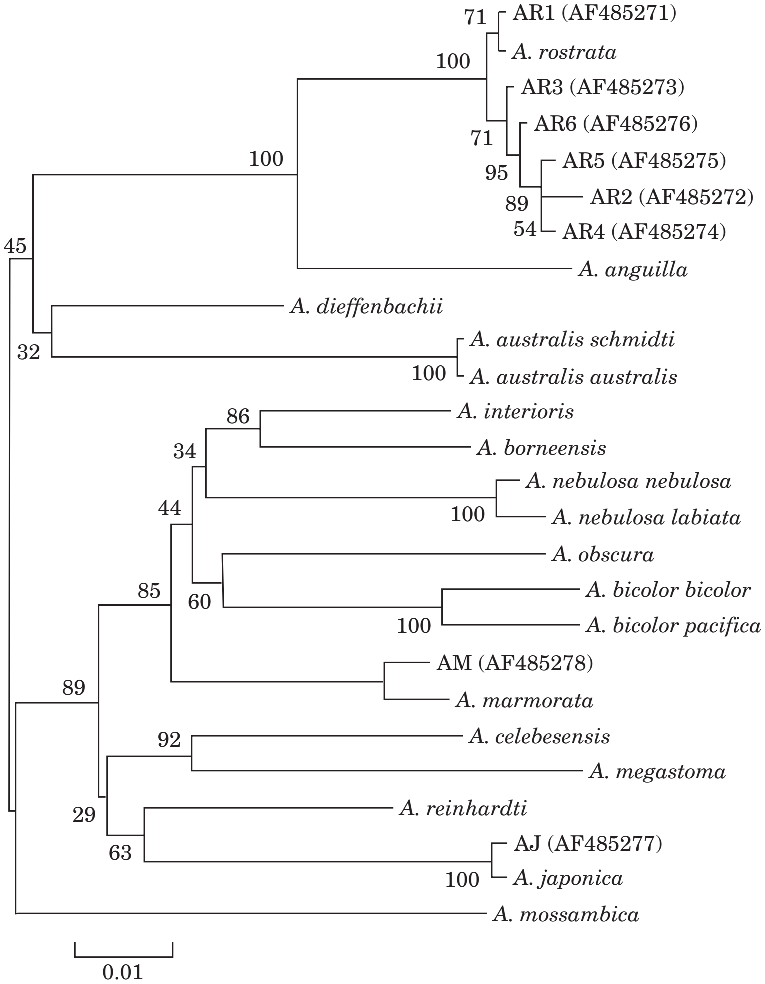

FIG. 2. Phylogenetic tree constructed with the neighbour-joining method. The coding region sequences of mt*Cyt-b* of six exotic American eels (AR1–AR6), one selected *A. japonica* (AJ), one *A. marmorata* (AM), and 18 known eel species and subspecies obtained from GenBank/NCBI were used for analysis. The accession numbers of this study were shown as indicated. The numbers represent bootstrap values of 1000 replicates. Accession numbers: *A. rostrata*, AF006716; *A. anguilla*, AF006714; *A. dieffenbachii*, AF006711; *A. australis schmidti*, AB021769; *A. australis australis*, AF006712; *A. interioris*, AB021773; *A. borneensis* (synonym of *A. malgumora*), AF006718; *A. nebulosa nebulosa*, AB021783; *A. nebulosa labiata* (synonym of *A. bengalensis labiata*), AF074866; *A. obscura*, AB021781; *A. bicolor bicolor*, AF006710; *A. bicolor pacifica*, AF006708; *A. marmorata*, AF074863; *A. celebesensis*, AB021777; *A. megastoma*, AB021771; *A. reinhardti*, AF006706; *A. japonica*, AF006702; *A. mossambica*, AF074864. The scale bar indicates the genetic distance calculated by Kimura's two-parameter model.

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References

Aoyama, J., Watanabe, S., Miyai, T., Sasai, S. & Nishida, M. (2000). The European eel, *Anguilla anguilla* (L.), in Japanese waters. *Dana* **12,** 1–5.

Castle, P. H. J. & Williamson, G. R. (1974). On the validity of the freshwater eel species *Anguilla ancestralis* Ege from Celebes. *Copeia* **1974,** 569–570.

Cheng, P. W. & Tzeng, W. N. (1996). Timing of metamorphosis and estuarine arrival across the dispersal range of the Japanese eel *Anguilla japonica. Marine Ecology Progress Series* **131,** 87–96.

Ege, V. (1939). A revision of the genus *Anguilla* Shaw, a systematic, phylogenetic and geographical study. *Dana Report* **16,** 1–256.

Han, Y. S., Chang, C. W., He, J. T. & Tzeng, W. N. (2001). Validation of the occurrence of short-finned eel *Anguilla bicolor* pacifica in the natural waters of Taiwan. *Acta Zoologica Taiwanica* **12,** 9–19.

Kumar, S., Tamura, K., Jakobsen, I. B. & Nei, M. (2001). *MEGA 2: Molecular Evolutionary Genetics Analysis Software*. Tempe, Arizona: Arizona State University.

Li, L. S. (1997). Eel aquaculture. In *Aquaculture* (Chang, K. S., ed.), p. 84. Kaohsiung, Taiwan: Chien-Cheng Publishing Company.

Miura, T., Yamauchi, K., Takahashi, H. & Nagahama, Y. (1991). Hormonal induction of all stages of spermatogenesis in vitro in the male Japanese eel (*Anguilla japonica*). *Proceedings of the National Academy of Sciences of the United States of America* **88,** 5774–5778.

Pankhurst, N. W. (1982). Relation of visual changes to the onset of sexual maturation in the European eel *Anguilla anguilla* (L.). *Journal of Fish Biology* **21,** 127–140.

Schmidt, J. (1925). The breeding places of the eel. *Annual Report of the Smithsonian Institute* **1924,** 279–316.

Tesch, F. W. (1977). *The Eel: Biology and Management of Anguillid Eels*. London: Chapman and Hall.

Tsukamoto, K. (1992). Discovery of the spawning area for Japanese eel. *Nature* **356,** 789–791.

Tsukamoto, K. & Aoyama, J. (1998). Evolution of freshwater eels of the genus *Anguilla*: a probable scenario. *Environmental Biology of Fishes* **52,** 139–148.

Tzeng, W. N. (1982). Newly record of the elver, *Anguilla celebesensis* Kaup, from Taiwan. *Bioscience* **19,** 57–66.

Tzeng, W. N. (1996). Short- and long-term fluctuations in catches of elvers of the Japanese eel, *Anguilla japonica*, in Taiwan. In *Developing and Sustaining World Fisheries Resources: The State of Science and Management, Second World Fisheries Congress* (Hancock, D. A., Smith, D. C., Grant, A. & Beumer, J. B., eds), pp. 85–89. Collingwood, Victoria: CSIRO Publishing.

Tzeng, W. N. & Tabeta, O. (1983). First record of the short-finned eel *Anguilla bicolor pacifica* elvers from Taiwan. *Bulletin of the Japanese Society of Scientific Fisheries* **49,** 27–32.

Tzeng, W. N., Wu, H. F. & Wickström, H. (1994). Scanning electron microscopic analysis of annulus microstructure in otolith of European eel *Anguilla anguilla. Journal of Fish Biology* **45,** 479–492.

Vladykov, V. D. (1973). Macrophthalmia in the American eel (*Anguilla rostrata*). *Journal of the Fisheries Research Board of Canada* **30,** 689–693.

Wang, C. H. & Tzeng, W. N. (1998). Interpretation of geographic variation in size of American eel, *Anguilla rostrata* (Le Sueur), elvers on the Atlantic coast of North America by their life history and otolith ageing. *Marine Ecology Progress Series* **168,** 35–43.

Wang, C. H. & Tzeng, W. N. (2000). The timing of metamorphosis and growth rates of American and European eel leptocephali: A mechanism of larval segregative migration. *Fisheries Research* **46,** 191–205.

Wenner, C. A. & Musick, J. A. (1974). Fecundity and gonad observations of the American eel, *Anguilla rostrata*, migrating from Chesapeake Bay, Virginia. *Journal of the Fisheries Research Board of Canada* **31,** 1387–1391.

Yamamoto, K. & Yamauchi, K. (1974). Sexual maturation of Japanese eel and production of eel larvae in the aquarium. *Nature* **251,** 220–222.

Zhang, H., Mikawa, N., Yamada, Y., Horie, N., Okamura, A., Utoh, T., Tanaka, S. & Motonobu, T. (1999). Foreign eel species in the natural waters of Japan detected by polymerase chain reaction of mitochondrial cytochrome b region. *Fisheries Science* **65,** 684–686.