

Did Farmed Coho Salmon *Oncorhynchus kisutch* that Escaped during the Earthquake and Tsunami Disaster in 2011 Interbreed with Native Masu Salmon *Oncorhynchus masou*?

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Abstract: During the Great East Japan Earthquake on March 11, 2011, several million individuals of farmed coho salmon (*Oncorhynchus kisutch*) escaped from aquaculture facilities in the Northeast Pacific Ocean, Tohoku, Japan. In fall of the same year, sexually mature coho salmon migrated up rivers in this area. Farmed coho salmon that migrate up rivers to breed may affect the genetic material of native salmon species and result in weakened populations. Especially, there is a strong concern that coho salmon may cross with the native masu salmon (*Oncorhynchus masou*); it is known that hybrids of these species have survivability. In this study, we surveyed masu salmon landed at a local fish market, using genetic and morphological methods in order to determine whether there are hybrids. As a result, hybrids were not found in this survey. Therefore, at this moment the impact on the genetic resources of masu salmon is considered to be low.

Key words: Farmed coho salmon (*Oncorhynchus kisutch*), escaped, masu salmon (*Oncorhynchus masou*), hybrids, DNA fragment analysis

Introduction

Coho salmon (*Oncorhynchus kisutch*) has been transplanted from North America to Hokkaido since the 1970s (Ishida *et al.*, 1975, 1976; Nara *et al.*, 1979; Umeda *et al.*, 1981), but has not become naturalized in Japan. They migrate to waters off Hokkaido for feeding, and sometimes stray and go up rivers in Hokkaido (Kikuchi *et al.*, 1998); however, they do not regularly spawn in Japan. Sea farming of coho salmon has been conducted around the Tohoku Pacific coast in Japan since 1975. In recent years, production has remained at over 10,000 tons per year.

Due to the effects of the Great East Japan Earthquake and tsunami on March 11, 2011, all farmed coho salmon (more than 5 million fishes) escaped into the Northeast Pacific Ocean off Tohoku,

northeastern Japan. In fall of the same year, sexually mature coho salmon migrated up rivers in this area. These coho salmon may have affected the genetic material of native salmon species and resulted in weakened populations. There is a strong concern that coho salmon may cross with the native masu salmon (*Oncorhynchus masou*) because it is known that hybrids of these species have survivability (Ito *et al.*, 2006). On the other hand, the ability to survive is low for crosses with chum salmon (*Oncorhynchus keta*) (Foerster, 1935). The upriver season is different between masu salmon and coho salmon; however, the spawning season overlaps for these species. The spawning season of masu salmon (Honshu pacific region) is from September to November (Kiso, 1995), while that of coho salmon is from November to January (Koseki, 2013). It is unclear how many years

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hybrids will need to mature. However, based on the maturation age of both species being 3 years old, the possibility that hybrids returned in spring of 2014 was considered to be high. In this study, we surveyed masu salmon landed at a local fish market, using genetic and morphological methods in order to determine the presence of hybrids.

Materials and methods

We conducted visual checks of masu and coho salmon landed at a Miyako fish market from May to September, 2014 (approx. 2,000 individuals) (Fig. 1). In general, it is possible to distinguish both species by checking the gill raker and the radial silver stripes on the caudal fin (Fig. 2). We surveyed presence of individuals with mixed characteristics. The reasons we choose this market were as follows: (1) large number of escaped coho salmon were landed at this market during summer to autumn season in 2011 and (2) upriver coho salmon were detected in several rivers near the market. We selected 39 masu, and 5

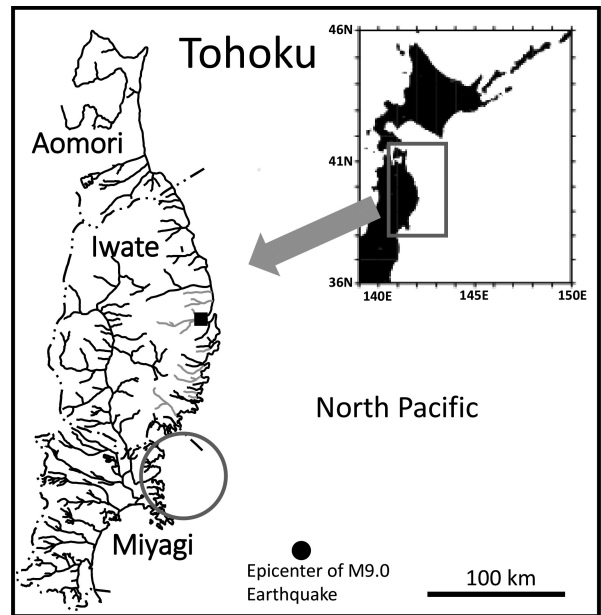


Fig. 1. Map of the Tohoku Pacific Coast. Rivers marked in gray indicate that adult coho salmon were detected in 2011. Open circle indicates the main farming area. "■" indicate study site.

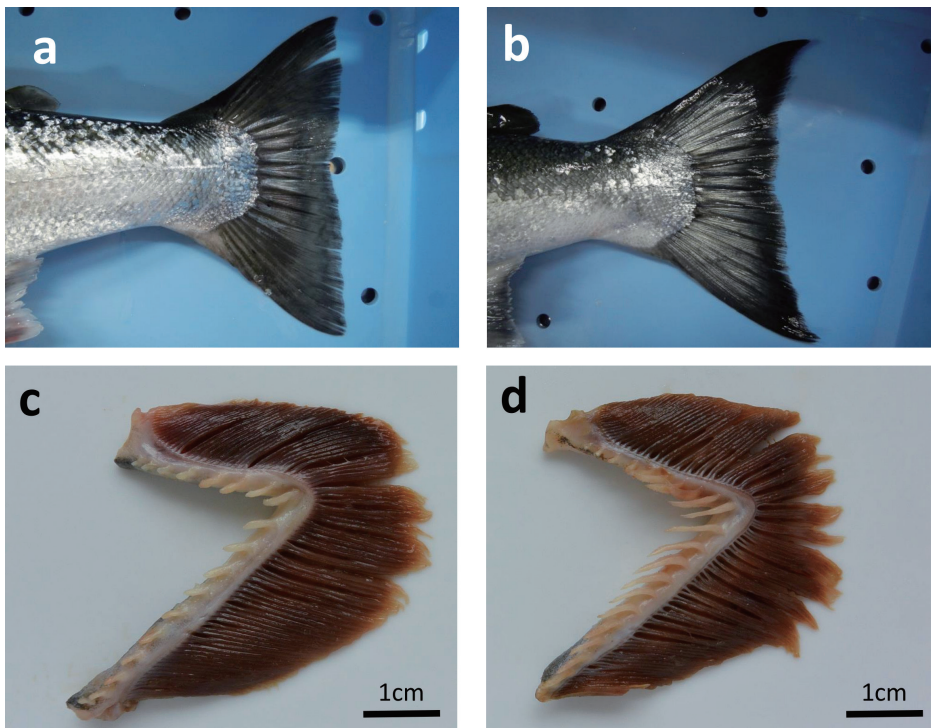


Fig. 2. Comparison of the tail and the first gill of adult coho and masu salmon. Upper panels shows tail of masu (a: fork length 57.0cm) and coho (b: fork length 60.6cm), lower panels shows the first gill of masu (c) and coho (d). Radial silver stripes on the caudal fin are obvious in coho, and the gill rakers longer and slender in coho.

coho salmon (all individuals were sampled during the survey) for more detailed analysis. For the morphological comparison, we compared the number of rays of each fin, and the number and length of the gill rakers. We conducted sequence analysis of the

intron C of the growth hormone 1 (GH-1) using a primer set with 17 bp adapter sequence for the fluorescent label. PCR amplified product of masu salmon (256bp) was 34 bp less than that of coho salmon (290 bp) (Fig. 3), thus making it possible to

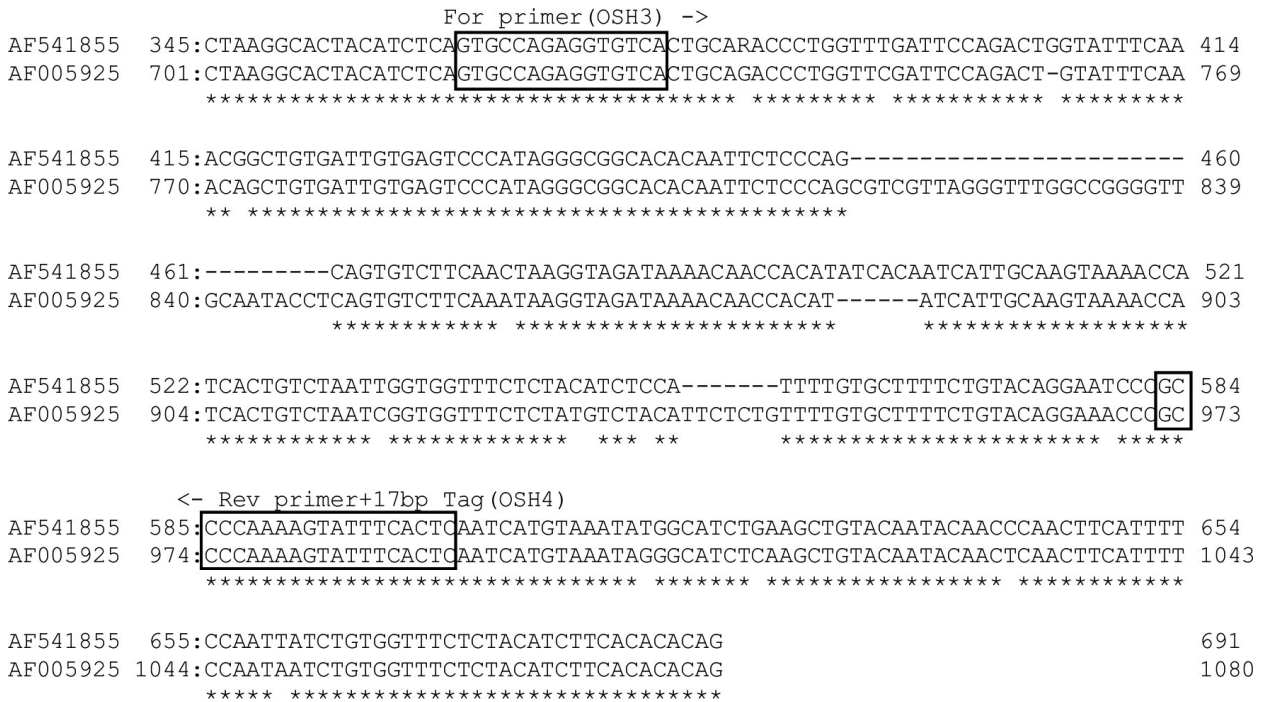


Fig. 3. Alignment of the intron C of the growth hormone 1 (GH-1) between masu salmon (GenBank: AF541855) and coho salmon (AF005925). PCR product amplified using primer set OSH3 and OSH4 of masu salmon is 34 bp less than that of coho salmon.

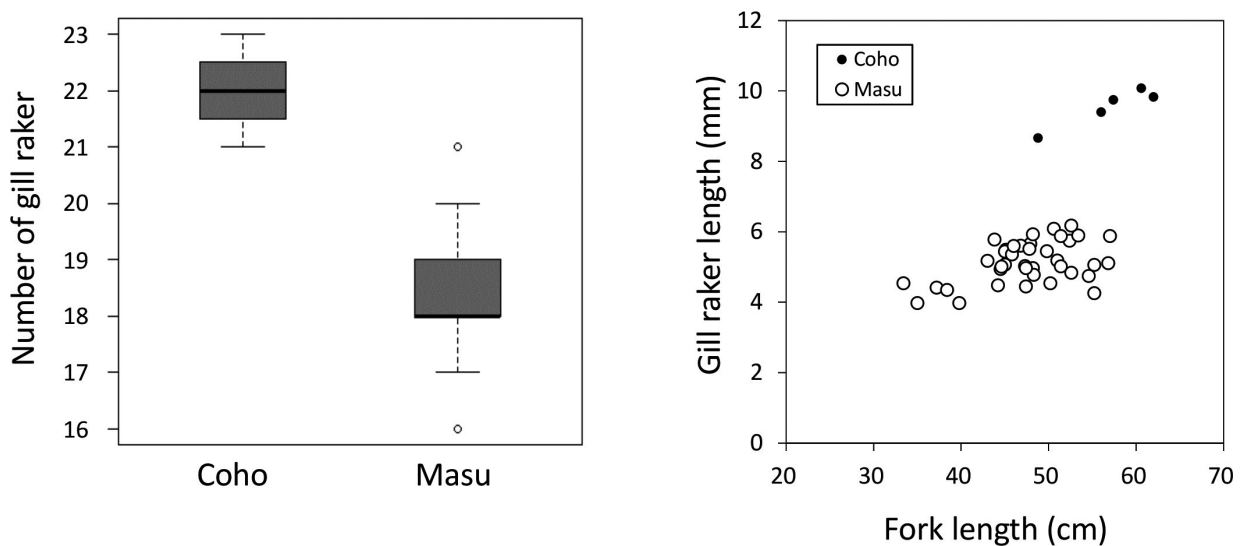


Fig. 4. Box plot of the number of gill rakers (left panel), and relationship between the longest gill raker length in the first gill and the fork length (right panel).

distinguish the two species. The amplicon was examined in both to see if there was a hybrid.

Results and Discussion

As a result of the morphological comparison, the range of number of fin rays overlapped, and there was no significant difference in the two groups (Table 1). The number and length of gill rakers were larger and longer in coho salmon (Fig. 4). Individuals with mixed characteristics were not found.

As a result of DNA fragment analysis, hybrids were not confirmed. Identification of species from DNA analysis of all the individuals agreed with the

species identification judged from the morphological comparison (Fig. 5). Therefore, based on the current survey, the impact of escaped coho salmon on the genetic resources of masu salmon is considered to be low. However, three of five of coho salmon were of the 2011 brood, and their gonads were developed. These individuals may have derived from the escaped coho salmon during the 2011 earthquake. Therefore, it is necessary to carefully monitor the occurrence of hybrids in the near future.

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Table 1. The number of fin rays in masu salmon and coho salmon

| | Masu salmon <i>O. masou</i> | Coho salmon <i>O. kisutch</i> |
|-------------------|--------------------------------|----------------------------------|
| n | 39 | 5 |
| Dorsal fin rays | 12-16 | 11-12 |
| Caudal fin rays | 19-25 | 19-26 |
| Anal fin rays | 12-15 | 13-15 |
| Pelvic fin rays | 9-11 | 10-11 |
| Pectoral fin rays | 12-15 | 14-16 |

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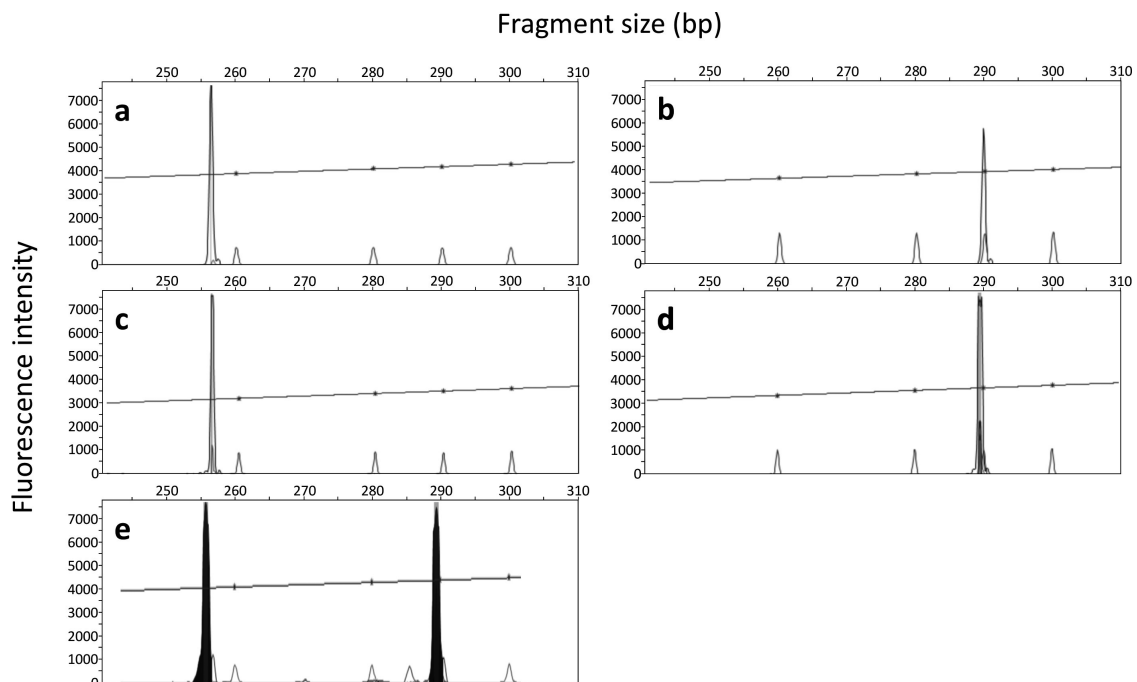


Fig. 5. Fragment size of the PCR amplified products of masu and coho salmon. Upper panel shows control of masu (a) and coho (b) salmon. Middle panel shows present specimens of masu (c) and coho (d). Lower panel shows artificial cross of masu (male) and coho (female) (e), the amplicon is seen in both 256 bp and 290 bp.

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