Genetic Differences and Variation in Black Rockfish (Sebastes schlegeli) and Hwanghae Rockfish (S. koreanus) from the Yellow Sea

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ABSTRACT

Black rockfish (Sebastes schlegeli; BR) and Hwanghae rockfish (S. koreanus; HHR) were obtained from the Yellow Sea of Gunsan in Korea. In this study, seven oligonucleotide primers generated a total of 884 bands in the BR species, and 632 in the HHR species, with a DNA fragment size ranging from 150 to 2,200 bp. On average, a decamer primer generated 126.3 amplified products in the BR species. A RAPD primer generated an average of 11.5 amplified bands per sample, ranging from 7.3 to 15.5 bands in this rockfish species. In the HHR species, shared unique bands, corresponding to bands of 230 bp, 750 bp, 800 bp and 1,600 bp, were generated by the decamer primer, OPA-16. Seven primers generated 75 polymorphic bands (75/884 bands, 8.48%) in the BR species and 62 (62/632 bands, 9.81%) in the HHR species. The percentage of polymorphic bands obtained from the BR species varied from 7.2% to 9.9%. Seven primers were the highest levels of polymorphism, which was 12.9%, in the HHR species. The 180 bp and 200 bp bands produced by the primer OPB-14 were identified as being unique to individual BR species. Individuals of the BR species exhibited comparatively higher bandsharing values than did fish of the HHR. The average bandsharing values were approximately 0.838 ± 0.004 and was 0.690 ± 0.012 in the BR and HHR species, respectively. The average bandsharing value between the two rockfish species was approximately 0.527 ± 0.005. The dendrogram, generated by seven reliable primers, indicates three genetic clusters. To distinguish the species of rockfish, our data suggested that the molecular marker based differentiation method could be a better than the traditional morphological character based one.

Key words: black rockfish, dendrogram, genetic distance, Hwanghae rockfish, Sebastes koreanus, Sebastes schlegeli, Yellow Sea.

INTRODUCTION

Rockfish are a very diverse group in term of size, habitat, and distribution. The scorpionfish of Sebastes are
the largest genus with about 100 species throughout the world in the family Scorpaenidae of the order Perciformes (Chen, 1975, 1986; Nelson, 1984). Among the scorpionfish of Sebastes, black rockfish (Sebastes schlegeli; BR) are an economically important marine fish species. BR lives in the Far East Asia, the East and South China Sea, the Philippines, Canada and USA. Especially, BR is widely distributed in the West Sea and the South Sea of the Korean Peninsula, and in the Gulf of Pohai of China, as well as in several areas around Japan. Generally, the ovoviviparous season for the fish starts in May and continues through to June in the South and the West Sea of the Korean Peninsula. The rockfish inhabit in a reef area of the shore. This fish grow to a maximum length of 40 cm. BR is ranked the highest among marine fish in Korea as a game fish, and attracts millions of anglers all through the year. The size and type of the rockfish vary according to habitat which includes such factors as the temperature and the depth of the water, and the availability of nutrients. They also possess black-fray transverse string at the flanks, and two black bands below the eyes. BR is predatory fish and will eat just about anything.

The Hwanghae rockfish (Sebastes koreanus; HHR) are a new scorpion fish species (Kim and Lee, 1994). The HHR inhabit in the West Sea of Korea at depths ranging from two to thirty meters. The HHR is also indigenous to some regions of the West Sea (Kim et al., 2005). This fish grow to a length of about 15 cm. The HHR has 14 dorsal spines, 30 ~ 31 lateral line pores and have no dark spots at the isthmus and at base of pectoral fins. The colors of the dorsal, anal, ventral, pectoral and caudal fins are pale grayish with small black spots. They also possess the dorsal and the lateral brown or grayish bodies mottled with black, and the pale or light pink isthmus without spots. They possess black-fray transverse string at the flanks, and three black stripes on the cheek.

BR and HHR are highly desirable species from the sea and are available year round. These species of fish are also one of very tasty marine fish species available throughout the year. Especially, BR has become Korean popular fish in a variety of restaurants for a long while. The consumption of these species has increased considerably, as restaurants that specialize in serving rockfish in various forms, such as raw with lemon, boiled with vegetables, or fried, have recently become established. As the necessity of rockfish increase, the understanding of the genetics of this teleost species becomes necessary. However, in spite of their economic and scientific importance, little information currently exists regarding the genetics of the BR and HHR species in Korea.

The maturation, fatty acid requirement, feeding habit, early embryonic development and spawning of the various rockfish species has been reported by researchers (Lee and Kim, 1992; Kim and Han, 1993; Bai and Lee, 1996; Kang et al., 2003). Especially, a new scorpionfish, S. koreanus has been described by morphological traits such as dorsal spines and lateral line pores (Kim and Lee, 1994). The genetic differences and DNA polymorphism were analyzed by RAPD method within and between black rockfish (S. schlegeli) populations from the Yellow Sea and the Southern Sea in Korea (Yoon and Kim, 2001b). However, little information currently exists regarding the genetic difference between BR and HHR.

In the present study, we performed a clustering analysis to elucidate the genetic differences between individuals of BR and HHR collected in Gunsan of the Yellow Sea by RAPD analysis.

MATERIALS AND METHODS

Sample collection and isolation of genomic DNA

Black rockfish (S. schlegeli) and Hwanghae rockfish (S. koreanus) were obtained from the same site of the Yellow Sea of Gunsan in Korea. The muscle tissues of the rockfish were collected in sterile test tubes, which were immediately placed in liquid nitrogen and stored until needed. The RAPD-PCR analysis was performed on the muscle extracts of 22 rockfish (11 individuals for black rockfish; 11 for Hwanghae rockfish) using seven arbitrarily selected primers. DNA extraction was performed as described previously (Yoon and Kim, 2003). After several washings, lysis buffer I [155 mM NH₄Cl, 10 mM KHCO₃, 1 mM EDTA] was added to the samples, and the tubes were gently inverted. The precipitates obtained were then centrifuged and resuspended in lysis buffer II [10 mM Tris-HCl (pH 8.0), 10 mM EDTA, and 100 mM NaCl, 0.5% SDS], and 15 μl of proteinase K solution (10 mg/ml) was added. DNA samples were extracted from the lysates by the addition of ice-cold 70% ethanol and centrifugation at 6,289 × g for 5 min. The DNA pellets were dried for 2 hrs and maintained at -40°C until analysis. The concentrations of the extracted genomic DNA samples were calculated