INTRODUCTION

Introgressive hybridization is an important factor of biological evolution, as evident from molecular, ecological, and phylogeographical data [1, 2]. Introgressive events play an important part in increasing genetic and phenotypic variation of various taxa. In addition, introgressive hybridization is a potential source of adaptive evolution of species. Eventually, hybridization and an increase in genetic variation may provide for unusually rapid speciation.

Natural interspecific hybridization and mitochondrial DNA transfer is known for many vertebrates, including lizards, frogs, mice, rats, birds, and fishes [3, 4]. In particular, in studies of mtDNA variation in species of the genus \textit{Salvelinus} have detected hybridization of \textit{S. fontinalis} and Arctic char \textit{S. alpinus} [5], lake trout \textit{S. namaycush} and \textit{S. alpinus} [6], and dolly varden \textit{S. malma} and bull trout \textit{S. confluentus} [7, 8].

In this work, mtDNA introgression in several char species of the Russian Far East was inferred from the nucleotide sequences of the mitochondrial cytochrome b gene.

MATERIALS AND METHODS

The material (muscle tissue fixed with 70% ethanol) was obtained from northern dolly varden \textit{S. malma malma} (\(N = 13\)) of the Yana, Taiui, Yama, Arman’ (northern coast of the Sea of Okhotsk), Getlyangen, Marich, and Utaatap (Chukotka) rivers and the Vilyuchinskii Stream (southeastern coast of Kamchatka); southern dolly varden \textit{S. malma krascheninnikovi} (\(N = 1\)) of the Shikotan Island (Kurils); white-spotted char \textit{S. leucomaenis} (\(N = 1\)) of the Taiui River (northern coast of the Sea of Okhotsk), and Arctic char \textit{S. alpinus} (\(N = 1\)) of Finland. The sites of char collection are shown in Fig. 1. Ichthyological specific identification was performed by P.K. Gudkov.

Total DNA was isolated as in [9]. The nucleotide sequence of a 405-bp fragment of the mitochondrial cytochrome b gene was established in the School of Biological Sciences, Washington State University (Vancouver, United States). The mtDNA fragment was sequenced at both strands by fluorescence labeling of double-stranded PCR products and their separation on a 373A Perkin Elmer automated sequencer (Applied Biosystems, United States).

Phylogenetic analysis of the nucleotide sequences of the cytochrome b mtDNA gene was carried out with the PHYLIP 3.2 and MEGA 1.01 packages [10, 11]. Phylogenetic trees were reconstructed using four methods: unweighted pair-group method with arithmetic average (UPGMA), neighbor joining (NJ), maximum likelihood (ML), and maximum parsimony (MP). The divergence between nucleotide sequences of the cytochrome b mtDNA gene was estimated as percent nucleotide substitutions per site (\(p\)-distance), using the MEGA 1.01 package. Atlantic salmon \textit{Salmo salar} was used as an outgroup in construction of phylogenetic trees. The validity of clustering was checked by bootstrap analysis with 1000 iterations, using the MEGA 1.01 package.

RESULTS

The nucleotide sequence was established for the 405-bp mtDNA fragment (15 152–15 556) of the cytochrome b mtDNA gene. Nucleotide coordinates corresponded to the mammalian mitochondrial cytochrome b mtDNA gene [12]. Variable nucleotide positions found in the 405-bp fragment for the char species examined are shown in Fig. 2.

Abstract—Nucleotide sequence analysis of the mitochondrial cytochrome b gene in the genus \textit{Salvelinus} revealed hybrids with \textit{S. leucomaenis} in \textit{S. malma malma} populations of the Northern Okhotsk Sea basin. Hybrids of \textit{S. m. malma} and \textit{S. m. krascheninnikovi} were found in \textit{S. m. malma} populations from the Northern Okhotsk Sea basin and Kamchatka. The findings testify to a secondary contact and mtDNA transfer between these species and forms. It was assumed that introgressive hybridization took place both long ago and relatively recently and occurred in one direction: from \textit{S. leucomaenis} or \textit{S. m. krascheninnikovi} to \textit{S. m. malma}.

O. A. Radchenko

Institute of Biological Problems of the North, Russian Academy of Sciences, Magadan, 685000 Russia; fax: (41322)344-63; e-mail: mradchenko@mail.ru

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The numbers of different nucleotides established in pairwise comparisons of the char species are shown in Table 1.

Comparisons with other species revealed the maximum difference in sequence of the 405-bp mtDNA fragment for *S. m. malma* from the Yama River (Table 1, fish 10, 12–23 substitutions), the Yana River (fish 11, 13–24 substitutions; no. 14, 22–25 substitutions), Kamchatka (no. 12, 12–22 substitutions), and the Arman’ River (fish 15, 21–24 substitutions); *S. m. kraschenin-