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Fisheries Research 80 (2006) 333-338

www.elsevier.com/locate/fishres

#### Short communication

# Spatial and temporal mitochondrial DNA genetic homogeneity of dolphinfish populations (*Coryphaena hippurus*) in the eastern central Pacific

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Received 26 January 2006; received in revised form 13 April 2006; accepted 27 April 2006

#### **Abstract**

Dolphinfish (*Coryphaena hippurus*) samples were collected over four consecutive years from four locations in the eastern central Pacific to evaluate the genetic variation in a 751 bp segment of the mitochondrial NADH subunit 1 (ND1) to test for the presence of genetic population structure. Sequence analyses revealed no significant differences among collections from the same location in the different years sampled nor between locations. Mismatch distributions, estimations of population expansion parameters, and neutrality tests revealed significant fluctuations in population size in coincidence with past glacial and interglacial periods during the late Pleistocene. The low levels of nucleotide diversities and shallow coalescence of mtDNA genealogies observed were coincident with the estimated demographic parameters and neutrality tests, in suggesting the presence of important past population size fluctuations or range expansion. The prevention of the accumulation of deep lineages independently on how it was originated, probably delayed the emergence of a population divergence process which might account for the lack of genetic differences detected.

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Keywords: Subpopulations; Historical demography; GENE flow; Population expansion

#### 1. Introduction

The dolphinfish (*Coryphaena hippurus*) is a cosmopolitan, highly migratory pelagic fish found in tropical and subtropical waters (Palko et al., 1982). The dolphinfish is a fast growing fish reaching two meters of fork length and weighting up to 30 kg at 3 years old, and is targeted by both recreational and commercial fisheries. A significant catch is taken along the Pacific coast of Mexico by artisanal fisheries, even though the area within 80 km of the coastline is reserved

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by law for sport fishing. However, as there are plans to open the resource for commercial exploitation, the definition of the population structure in the fishing area is needed before proceeding to conduct population dynamics studies to define management strategies.

Dolphinfish populations are abundant in eastern Pacific tropical and subtropical areas. Off Mexico, it is distributed between the Gulf of Tehuantepec and the Baja California Peninsula, including the Gulf of California.

The seasonality of catch is a feature of dolphinfish populations (Oxenford, 1999; Lasso and Zapata, 1999) and may be related to seasonal migrations to spawning areas or to seasonal changes in sea temperature. Although no information about spawning activity is available, spatial and temporal iso-

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lation of spawning groups may result in genetic divergence of populations. Significant abundance fluctuations in dolphin-fish from the Atlantic and Caribbean have been associated with the presence of divergence between populations, based on differences in allozyme allele frequencies (Oxenford and Hunt, 1986). In the Mexican Pacific, large abundances have been reported around Los Cabos and into the Gulf of California, where important catches are taken from July to September and from May to August, respectively (Zúñiga-Flores, 2004). Another area off southern Mexico has also been recognized in the Gulf of Tehuantepec (off Oaxaca and Chiapas), with maximum abundances during February to May, (Mendizabal et al., 1990).

Some features of large, pelagic, tropical fish species, such as their wide distribution, considerable population sizes, high reproductive potential, dispersal ability, and rapid growth rates are thought to explain the observed levels of the population structure, which range from shallow, but significant, structuring to lack of heterogeneity because of the absence of obvious barriers to dispersal (Graves, 1998; Ward, 2000). Even though the possible presence of population structure between populations in a local rather than wide scale could be remote in the marine realm, clarification of the presence of discrete genetic populations in the area where the fishery is taking place is nevertheless important before opening this fish resource to commercial exploitation.

#### 2. Materials and methods

Samples of dolphinfish tissue were collected at four locations over four consecutive years from small (artisanal) fishing boats operating in a local scale in the eastern central Pacific (Fig. 1), including Baja California Sur in 2002

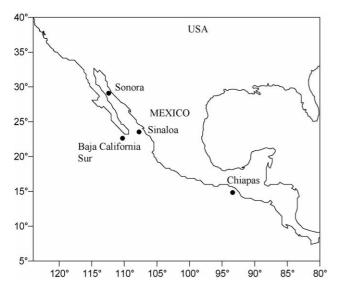


Fig. 1. Locations of four samples of dolphinfish (*Coryphaena hippurus*) from the eastern central Pacific. Sonora (SON), Sinaloa (SIN), Baja California Sur (BCS) and Chiapas (CH).

(BCS02; N= 16) and 2003 (BCS03; N= 23), Sonora in 2003 (SON03; N= 25), Sinaloa in 2003 (SIN03; N= 21) and 2004 (SIN04; N= 33), and Chiapas in 2003 (CH03; 22), 2004 (CH04; N= 22), and 2005 (CH05; N= 15). Total genomic DNA was isolated using the proteinase K — lysis-buffer extraction (Laird et al., 1991) and resuspended in 50–100 μL of TE buffer.

The complete sequence of the mitochondrial NADH dehydrogenase subunit 1 (ND1) gene of dolphinfish (GenBank accession number AF272056) was used to design the internal primers, NADH163 (5'-TAATCCTGCCGCAATTATCC-3') and NADH1128 (5'-AGGCCTTCCAGGTTAGGT GT-3') to amplify a 751 pb segment with the polymerase chain reaction (PCR) in a total of 177 fishes. Reactions for sequencing were made in total volumes of 100 μL containing 10–100 ng DNA, in amplification buffer, 10 mM Tris-HCl (pH 8.4), 50 mM KCl, 1.5 mM MgCl<sub>2</sub>, 0.2 mM of each dNTP, 0.1 mM of each primer and 2.5 U of platinum Taq DNA polymerase (Invitrogen, Cat. 10966-030). PCR amplifications consisted of 35 cycles of 1 min at 95 °C for denaturation, 1 min at 58 °C for annealing, and a final extension at 65 °C for 3 min. Amplicons were purified with a QIAquick purification kit (QIAgen No. Cat. 28104) and sequenced on an ABI 3730xl automated sequencer (Applied Biosystems) by Macrogen Inc., Korea. Sequences were aligned with ClustalX ver. 1.8 (Thompson et al., 1997). Identification of an appropriate substitution model for the mtDNA ND1 gene was made by hierarchical likelihood ratio test implemented in MODELTEST 3.06 (Posada and Crandall, 1998). Haplotype, h, and nucleotide,  $\pi$ , diversities and a minimum spanning network of haplotypes were estimated with Arlequin (Schneider et al., 1997).

F-statistics (Fst), (Weir and Cockerham, 1984) and  $\Phi_{ST}$  (Excoffier et al., 1992), and their respective significance values were calculated using the Arlequin software with the Tamura-Nei distances corrected by the gamma-shape parameter. Arlequin also produced estimates of demographic parameters  $\tau$ ,  $\theta_0$ , and  $\theta_1$  from nucleotide mismatch distributions. Fu's F, as implemented in Arlequin was used to test for departures from neutrality due to recent population expansions or to selection (Fu, 1997). Finally, Harpending raggedness index (Harpending, 1994) and the sum of squared deviations (S.S.D.) were used to test for fit to a unimodal mismatch distribution (Rogers and Harpending, 1992) as implemented in the Arlequin in order to evaluate the Roger's sudden expansion model (Rogers, 1995).

#### 3. Results

A 751 base pairs segment of mtDNA ND1 gene was sequenced for 177 dolphinfishes. A total of 93 variable sites produced 87 haplotypes. Haplotype diversity averaged h = 0.926, and nucleotide diversity averaged  $\pi = 0.0052$  between samples (Table 1).

The minimum spanning network (MSN) revealed two clades, each showing a star-like phylogeny centered on two

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