



Genetic and morphometric differences between yellowtail snapper (*Ocyurus chrysurus*, Lutjanidae) populations of the tropical West Atlantic

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Abstract

Populations of *Ocyurus chrysurus* were compared genetically and morphometrically along the West Atlantic coast to test the null hypothesis of population homogeneity in the area. Brazilian populations were found to be differentiated in shape (canonical variates analysis; $F_{[48,515]} = 10.84$, $p < 0.0001$). Analyses of mitochondrial DNA sequences (663 bp of the control region) did not show any differences between Brazilian populations but could detect differences between Brazilian and Caribbean (Belize) populations. The samples from Pernambuco differed significantly from the other Brazilian populations in allozyme frequencies (11 loci; $F_{ST} = 0.167$; $p < 0.05$), but this may have resulted from the small number of samples analysed for that population. Sequence variation of Belize samples departed from neutral expectations (F_u 's $FS = -8.88$; $p < 0.001$). A mismatch distribution analysis points to an ancient population expansion in that area. We conclude that the genetic data do not allow the rejection of the null hypothesis of panmixia for Brazilian yellowtail snapper populations which should be treated as a single genetic stock, with a latitudinal gradient on their morphology which probably results from phenotypic plasticity. On the other hand, there is a severe restriction to gene flow between *O. chrysurus* populations from the Caribbean and from the southwestern Atlantic.

Key words: mtDNA, allozymes, morphometry, fisheries, phylogeography.

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Introduction

The yellowtail snapper *Ocyurus chrysurus* (Bloch, 1791) is a Lutjanid reef fish found on the east coast of the Americas from New England (USA) to southeastern Brazil (Menezes and Figueiredo, 1980; Allen, 1985). It is dioecious and its spawning times vary between geographic regions (Thompson and Munro, 1983; Claro, 1983; Grimes, 1987). *O. chrysurus* has opportunist and generalist alimentary habits and, hence, it is an important predator in some reef zones (Fallows, 1984; Parrish, 1987). They are long-lived (up to 17 years Allman *et al.*, 2005) and their larvae are pelagic (Riley *et al.*, 1995).

Yellowtail snappers are important fisheries resource where they occur. The production of the species in Northeast Brazil is high (Costa *et al.*, 2003) by 2001 averaging 1683 tons.year⁻¹ (Resende *et al.*, 2003). In Southeast Brazil

yellowtail snappers accounted for 16% of the total fish biomass captured between 1986-1989 (Paiva and Andrade-Tubino, 1998). The formation of spawning aggregations in this species (Claro, 1983) makes it particularly vulnerable to overfishing (Costa *et al.*, 2003), so that it is important that the stock structure of this species be known in order to better preserve it. This is also important because of the reported post-recruitment site fidelity of this species (Watson *et al.*, 2002) which might lead to large stock differentiation and fragility.

In recent years there has been a worldwide decrease in fishing stocks (Garcia and Grainger, 2005; Pauly *et al.*, 2005): the overexploited, fully exploited and exhausted fisheries that were 69% in 1995 increased to 75% in 2002 and only 1% of the stocks are in a state of recuperation (FAO, 2002; Ormerod, 2003). Similar problems are faced by the Brazilian fisheries stocks (Vasconcellos and Gasalla, 2001). For this reason efficient management policies based on unambiguous scientific data are necessary both to protect the fishing stocks and to maximize their exploitation without compromising their integrity. The correct stock as-

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assessment and identification of the species is fundamental to establish the maximum sustainable effort of a given marine resource (Ryman and Utter, 1987; Ryman, 1991). Many stock concepts can be found in the literature (Booke, 1981; Ovenden, 1990; Smith *et al.*, 1990; Carvalho and Hauser, 1995), but one of the most accepted and used is that “a stock is an intraspecific group of randomly mating individuals with temporal and spatial integrity” (Ihssen *et al.*, 1981) which covers most of the definitions given by other authors.

The identification of fish stocks can be made efficiently by the use of highly polymorphic molecular markers (Blaber *et al.*, 2005; Caddy and Seijo, 2005; Carmen and Ablan, 2006). In this study nuclear (allozymes) and mitochondrial (control region sequences) markers are used for the first time to analyse the stock structure of populations of *O. chrysurus* along 2380 km of Brazilian coast. A significant latitudinal gradient was observed for geometric morphometric data. The lack of observable heterogeneity in allozymes or in the highly variable mitochondrial DNA sequences indicates that this variation results from phenotypic plasticity. Additionally, samples from the Caribbean differed genetically from the Brazilian ones indicating restriction in gene flow between the two areas for this fish species.

Material and Methods

Sampling

Specimens of *Ocyurus chrysurus* were collected between August and October 2001 by fishing vessels from four locations along the Brazilian coast (Table 1; Figure 1). Samples of muscle and liver tissues from each fish were transported frozen to the laboratory where they were stored in liquid nitrogen until analyses. Muscle samples from *O. chrysurus* were also collected in Belize (17° 29' 34" N, 88° 06' 00" W; N = 20) and transported in 90% ethanol to the laboratory where they were used in DNA sequencing.

Morphometric analyses

Morphometric variation among populations was assessed through geometric methods (Zelditch *et al.*, 2004). For this purpose all 192 individuals collected at the four localities were measured. Measurements were taken with a Vernier manual calliper (0.05 mm of precision) between 10 homologous landmarks using a truss network protocol (Strauss and Bookstein, 1982) which provided 21 inter-landmark distances (Figure 2).

To retain the relative geometry of landmarks, inter-landmark distances were converted to Cartesian landmark coordinates for each specimen using the import truss feature of the Morphueus program (Slice, 1998). Landmark coordinates were then adjusted by the generalized least squares Procrustes superimposition (GLS) method (Rohlf and Slice, 1990), centering all specimens, and scaling them to an equal centroid size. The GLS-adjusted landmark coordinates



Figure 1 - Collection localities of *Ocyurus chrysurus*.

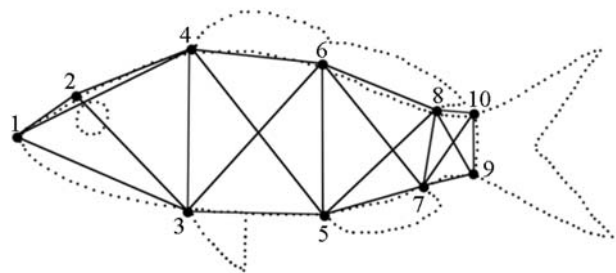


Figure 2 - Outline of *Ocyurus chrysurus* showing the 10 landmarks and the truss network of measured distances.

Table 1 - Sampling localities and number of specimens used for each analysis.

Locality	Latitude	Longitude	Morphometry	Allozymes	mtDNA
CE - Ceará	3° 43' 02" S	38° 32' 34" W	57	45	17
PE - Pernambuco	8° 45' 35" S	35° 06' 17" W	17	5	12
BA - Bahia	13° 22' 13" S	39° 04' 23" W	60	55	12
ES - Espírito Santo	20° 19' 10" S	40° 20' 16" W	58	58	16

dinates were used for thin-plate spline (TPS) analysis (Bookstein, 1989) which provided shape parameters for subsequent multivariate analyses. Both uniform and non-uniform (= partial warps) components of shape were submitted to a canonical variate analysis (CVA) in order to assess shape variation among populations (Cavalcanti *et al.*, 1999; Zelditch *et al.*, 2004). The PAST program (Hammer *et al.*, 2001) was used for thin-plate spline analysis and partial warps computations. Multivariate statistical analysis (CVA) was performed with SYSTAT® package, v. 10.

Allozyme electrophoresis

Muscle and liver samples were tested for thirty allozymes and three buffer systems, using 12.5% starch gel electrophoresis. Based on preliminary results, liver tissue and twelve allozyme systems were chosen for the analyses (Table 2).

Gel slices were stained following standard conditions (Manchenko, 1994; Batista and Solé-Cava, 2005; Lima *et al.*, 2005). Samples that could not be reliably scored for at least half of the allozyme loci were excluded from the analyses. This was the case for most samples from Pernambuco, leading to a severe reduction in allozymes sample size for that locality. Allozyme genotypes were used to estimate allele frequencies, from which heterozygosity levels and inbreeding indices (Global and pairwise F_{ST}) were calculated, using the Genetix 4.01 program (Belkhir *et al.*, 2002). An exploratory factorial correspondence analysis (Excoffier *et al.*, 1992) was also used, to check for hidden partitions in the allelic hyperspace.

DNA extraction

Total genomic DNA was extracted from muscle tissues of 77 individuals, using a CTAB extraction protocol (CTAB 2%, EDTA 20 mM, 2-mercaptoethanol 0.2% v/v,

NaCl 1.4 M, Proteinase K 30 micrograms in 500 μ L of Tris 100 mM). The extracted DNA was precipitated with ethanol and 3 M sodium acetate, re-suspended in 20 μ L of ultrapure water, and stored at -20 °C.

PCR amplification and sequencing

The PCR reactions of the control-region of mitochondrial DNA (D-loop) were done using the external primers THR (5'AGCTCAGCGCCAGAGCGCCGGTCTTGTA AA3') (Lee *et al.*, 1995) and 12S (5'ATAGTGGGGTAT CTAATCCCAGTT3') (Palumbi *et al.*, 1991). PCR reactions were set up using 1 unit of Taq polymerase, 0.2 mM of each dNTP, 0.5 μ M of each primer, 1.5 mM of MgCl₂, 30 μ g of BSA (bovine serum albumin) and 1 μ L of DNA (approx. 50 ng) as a template, in a final volume of 30 μ L of PCR buffer (Promega). Thermocycling conditions were: one initial cycle of 5 min at 94 °C, followed by 35 cycles of 1 min each at 94 °C, 48 °C and 72 °C, and one final extension step of 5 min at 72 °C. Negative controls (without DNA template) were used in all PCR reactions to check for contamination. PCR products were purified, and both strands were sequenced in an ABI 3730 XL automatic sequencer, using the internal PRO (5'CCCAAAGCTAAAA TTCTAA3') (Kocher *et al.*, 1989) and PHE (5'GCTTTAG TTAAGCTACG3') (Hedgecock and Strong, 1994) primers. The different haplotypes obtained were deposited in GenBank under accession numbers DQ666423-DQ666447 and EF624354-EF624386.

Sequence analyses

Sequences were edited using the Seqman™ II program (DNASTAR Inc.) and aligned using ClustalW (Thompson *et al.*, 1994). For phylogeographic nested clade analysis (NCA), a haplotype network was generated using the TCS 1.21 program (Clement *et al.*, 2000) using the 95% parsimony

Table 2 - Enzyme systems used in allozyme electrophoresis; buffer systems: TC8 = 0.25 M Tris, 0.06 M citrate, pH 8.0 (Ward and Beardmore, 1977); TC7 = 0.135 M Tris, 0.043 M citrate, pH 7.0 (Shaw and Prasad, 1970); Poulik = 0.03 M Tris, 0.005 M citrate, pH 8.5 (gel), 0.06 M LiOH, 0.30 Borate, pH 8.1 (buffer tank) (Poulik, 1957). E.C.: Enzyme Commission numbers.

Enzyme	Abbreviation	Buffer	E.C. number
Acid phosphatase	ACP	TC8	3.1.3.2
Alpha-Esterases	α EST	Poulik	3.1.1.1
Glucose 6-Phosphate dehydrogenase	G6PDH	TC8	1.1.1.49
Glutamate dehydrogenase	GDH	Poulik	1.4.1.2
Glutamic-Oxaloacetic transaminase	GOT	TC7	2.6.1.1
Lactate dehydrogenase	LDH	TC7	1.1.1.27
Malic enzyme	ME	TC8	1.1.1.40
Phosphoglucomutase	PGM	TC7	5.4.2.2
Phosphogluconate dehydrogenase	PGD	TC8	1.1.1.44
Phosphoglucose isomerase	PGI	Poulik	5.3.1.9
Superoxide dismutase	SOD	TC8	1.15.1.1
Xanthine oxidase	XOD	TC8	1.1.3.22

mony criterion (Templeton, 1998). Clades were nested from haplotypes (0-step) to the highest level, each separated by one substitution step (Templeton *et al.*, 1995; Templeton, 1998), and the statistical significance of each group was assessed, jointly with geographical coordinates for each sample, using the program GeoDis 2.2 (Posada *et al.*, 2000). Selective neutrality of the control region sequences was tested with Tajima's (Tajima, 1996) and Fu's (Fu and Li, 1993) tests. Exact tests of population differentiation (Raymond and Rousset, 1995) were performed with 1,000 dememorisation steps and 10,000 Markov Chain steps. Selective neutrality tests, mismatch distribution analyses (Rogers and Harpending, 1992), Analyses of Molecular Variance (AMOVA; Excoffier *et al.*, 1992) and exact tests of population differentiation were done using the Arlequin 3.0 program (Excoffier *et al.*, 2005). Haplotype and nucleotide diversity indices were estimated using the DNAsp 4 program (Rozas *et al.*, 2003).

Results

Morphometry

Morphometric variation among populations was visualized via scatter plot of the scores of the first two canonical variables of the CVA applied to the shape variables of the TPS analysis (Figure 3). The first variable accounted for 76.8% of the among-population relative to within-population variation and discriminated the southern-most populations (Espírito Santo, Bahia) from the northern ones (Pernambuco, Ceará). A geographical gradient was also observed when considering the intermediate position of the Bahia specimens between Espírito Santo and the northern populations. The second variable accounted for only 26% of total variation, being related to differences between Ceará and Pernambuco and, to a lesser extent, between Bahia and Espírito Santo. Results of CVA indicated significantly among-populations differences (Wilks lambda = 0.1255; $F_{[48,515]} = 10.84$; $p < 0.0001$). Percentage of individuals correctly classified to the four original populations ranged from 80% (Bahia) to 84% (Ceará and Espírito Santo).

Allozyme electrophoresis

Eleven *loci* were scored for all samples. Another four *loci* were scored for all populations except that of Pernambuco (Table 3). Observed heterozygosities (average Hardy-Weinberg expected heterozygosity, $H_e = 0.1917$; Table 3) were high (Smith and Fujio, 1982; Ward *et al.*, 1994). Overall, the populations were found to be genetically structured ($F_{ST} = 0.0277$; 95% confidence interval (bootstrapping over *loci*) = 0.00078-0.05648; standard error (jackknifing across *loci*) = 0.0195 reject the null hypothesis of population homogeneity). However, this resulted, mostly, from the large gene frequency differences found in two *loci* (*G6PD-2* and *GOT*, Table 3) of the population

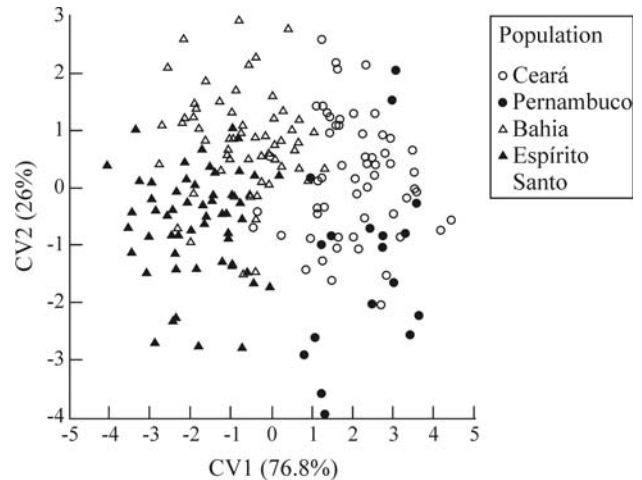


Figure 3 - Scatter plot of individual scores from the first two canonical variates.

from Pernambuco, which was the only one significantly different from the others (Table 4). When the samples from Pernambuco were excluded from the analyses, the F_{ST} became non-significant ($F_{ST} = 0.0098$; $p > 0.30$). The factorial correspondence analysis of unclassified samples showed a wide non-significant scatter of the data with only 27.1% of the total inertia explained by the first two factors (Figure 4). When the gravity centroids of each of the four sampling sites were compared, the two first factors accounted for 84.9% of the total inertia, showing significant differentiation between Pernambuco and the other sites.

Sequence variation

From the 663 base pairs of the control region of mitochondrial DNA sequenced, 105 were polymorphic, producing 58 haplotypes in the 77 individuals analysed. Genetic variation over all sampling sites was high (haplotype diversity, $h = 0.9614$; nucleotide diversity, $\pi = 0.0185$). The highest levels of variation were observed in Belize ($h = 1.000$; $\pi = 0.02378$). The null hypothesis of selective neutrality was not rejected for the studied sequences (Tajima's D test; $p > 0.05$; Fu's F_s test; $p > 0.05$) from most sampling sites. An exception was the Belize population, where D was not significantly different from zero, but Fu's F_s was significantly negative ($F_s = -8.88$; $p < 0.001$). A negative F_s results from an excess of rare alleles (as can be observed in the haplotype network in Figure 5), and it is usually interpreted as an indication of population expansion (Harpending, 1994). Hence, a mismatch distribution analysis (Rogers and Harpending, 1992) was performed on that population (Figure 6), and the observed distribution did not deviate significantly from the null hypothesis of population expansion ($SSD = 0.004$; $p > 0.95$). The Harpending's Raggedness index was low ($r = 0.010$), indicating a smooth distribution, also consistent with the hypothesis of population expansion (Harpending, 1994).

Table 3 - Allele frequencies for the 14 *loci* analysed. For simplicity, the slowest migrating allele for each *locus* is not shown: its frequency can be calculated as one *minus* the frequency of the other alleles at that *locus* in the population. N = number of individuals analysed; He = mean Hardy-Weinberg expected heterozygosity; Ho = mean observed heterozygosity (values shown as average \pm standard error)

Locus/allele	Ceará (N = 45)	Pernambuco (N = 5)	Bahia (N = 55)	Espírito Santo (N = 58)
ACP/A	0.97	1	1	1
α -EST/A	0.28	0.10	0.15	0.13
GDH/A	0.37	0.30	0.41	0.41
G6PD-1/A	0.53	0.30	0.44	0.41
G6PD-2/A	0.58	1	0.31	0.41
GOT/A	0.23	0	0.39	0.41
/B	0.74	1	0.61	0.59
LDH-1/A	0	0	0.01	0.05
LDH-2/A	1	1	1	1
ME/A	1	1	1	1
PGD-1/A	1	1	1	1
PGI-1/A	0.59	ND	0.54	0.64
PGI-2/A	0.50	ND	0.51	0.59
PGM/A	0.50	ND	0.63	0.42
/B	0.46	ND	0.37	0.58
SOD/A	0.97	ND	1	0.98
XOD/A	1	1	1	1
He	0.234 \pm 0.240	0.076 \pm 0.533	0.223 \pm 0.236	0.234 \pm 0.232
Ho	0.140 \pm 0.180	0.094 \pm 0.182	0.192 \pm 0.244	0.185 \pm 0.219

Table 4 - Pairwise F_{ST} values among the four Brazilian populations for allozyme data. Probability values (H_0 : $F_{ST} = 0$) were calculated by 1000 bootstrap permutations. NS = not significant; * = $p < 0.05$; ** = $p < 0.01$

Population 1	Population 2	Allozyme F_{ST}
Espírito Santo	Ceará	0.0113 ^{NS}
Espírito Santo	Pernambuco	0.1726*
Espírito Santo	Bahia	0.0005 ^{NS}
Ceará	Pernambuco	0.1010 ^{NS}
Ceará	Bahia	0.0202 ^{NS}
Pernambuco	Bahia	0.2256**

Levels of population differentiation, as indicated by pairwise F_{ST} values, were significant only between the population from Belize and the others (Tables 4 and 5). No differentiation was observed among populations from Brazil with the exact tests of population differentiation (Markov chain test; exact P value = 0.72255 ± 0.02941). This was confirmed by AMOVA, where none of the Φ_{ST} values among Brazilian populations was significant, and less than 7% of the total variance was explained by differences between geographical locations (Table 6). The nested clade analysis did not reveal significant ($p > 0.05$ after Bonferroni correction) groupings at any hierarchical level. Seven haplotypes, five of which from Belize, were excluded from the nested-clade analysis because they did not meet the 95% confidence level on the TCS haplotype network. This

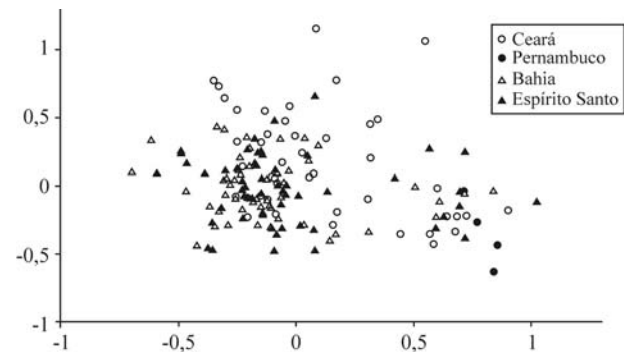


Figure 4 - Correspondence factorial analysis of allozyme data.

was caused by the very large divergence of those sequences in relation to the others, as can be seen in the neighbor-joining tree of all haplotypes (Figure 7).

Discussion

The analysis of both nuclear and mitochondrial genetic data of populations of yellowtail snappers, *Ocyurus chrysurus*, did not reveal genetic differences that could justify rejecting the null hypothesis of panmixia along the 2380 km of Brazilian coast, but significant differences were observed between Brazilian and Caribbean populations. In contrast, we observed significant differences in the mor-

phology of the Brazilian samples ($F_{[48,515]} = 10.84$; $p < 0.0001$).

Some heterogeneity was observed in allozyme frequencies among Brazilian populations ($F_{ST} = 0.0277$; $p < 0.05$). This heterogeneity was caused by the large differences observed between Pernambuco and other Brazil-

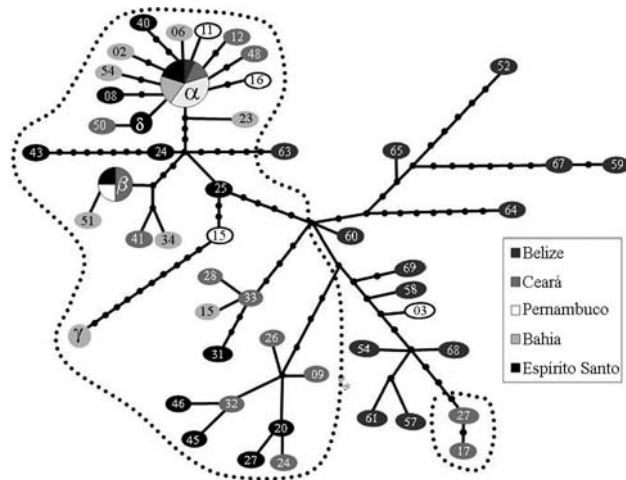


Figure 5 - mtDNA control-region haplotype network. Different shades of grey are used to indicate sample locations. Hypothetical non-sampled haplotypes are indicated by dots. Lines connecting haplotypes correspond to single nucleotide changes. Dotted line surrounds haplotype clusters from the southwestern Atlantic. Haplotype α : samples CE22, CE06, PE01, PE06, PE10, PE13, PE14, PE17, BA10, BA11, BA47, ES04, ES07, ES41, BE62; Haplotype β : CE05, CE53, PE4, ES42; Haplotype γ : BA43, BA46; Haplotype δ : ES28, ES52.

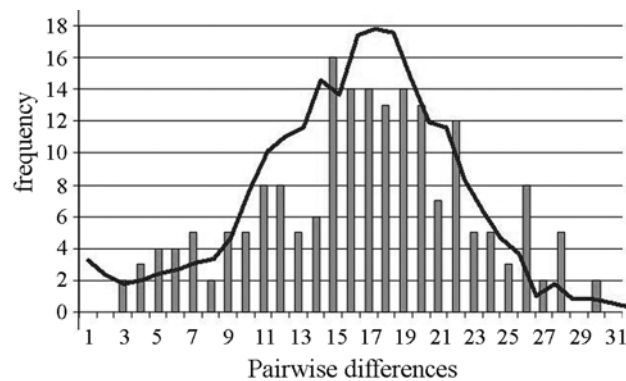


Figure 6 - Mismatch distribution analysis for the Belize population. The line shows the expected distribution under the null hypothesis of population expansion.

ian populations (Table 4, Figure 4). However, since only 5 individuals were analysed from that population, it is likely that the differences encountered simply result from sampling error. Indeed, when we excluded Pernambuco from the analysis, the mean F_{ST} values became non-significant ($F_{ST} = 0.0098$; $p > 0.30$). Therefore, we cautiously conclude that there is no support from the allozyme data to reject the null hypothesis of panmixia in this species along the studied area on the Brazilian coast.

The control region of *O. chrysurus* is extremely variable (haplotype diversity = 0.9614). However, this variability is much higher within than between sampling locations in Brazil (Tables 5 and 6), and both the haplotype network generated by nested-clade analysis (Figure 5) and the complete haplotype tree (Figure 7) show that the haplotypes from the Brazilian samples are distributed haphazardly along the coast. The Caribbean samples, on the other hand, differ significantly from the southwestern Atlantic ones ($F_{ST} = 0.17$, $p < 0.05$; Table 5). This can also be seen in the haplotype network and the haplotype tree (Figures 5 and 7), where most samples from Belize cluster outside of the southwestern Atlantic ones. The haplotype network has a

Table 6 - Results of the analysis of molecular variance (AMOVA) from mtDNA sequence data of the Brazilian populations. O.B.P. = Other Brazilian Populations. * $p < 0.05$.

Groups	Among groups	Within populations
Ceará X O.B.P.	-3.95	100.00
Pernambuco X O.B.P.	1.51	97.66
Bahia X O.B.P.	-2.01	99.39
Espírito Santo X O.B.P.	4.25	96.31
Ceará and Pernambuco X O.B.P.	-1.50	98.90
Ceará and Bahia X O.B.P.	-2.30	99.16
Ceará and Espírito Santo X O.B.P.	3.74	97.18
Ceará X Pernambuco X O.B.P.	-3.33	98.95
Pernambuco X Bahia X O.B.P.	2.59	97.98
Pernambuco X Espírito Santo X O.B.P.	3.43	97.84
Bahia X Ceará X O.B.P.	-7.19	99.58
Bahia X Espírito Santo X O.B.P.	0.91	98.25
Ceará X Espírito Santo X O.B.P.	3.43	97.84
Belize X Brazilian populations	17.57	81.63*

Table 5 - Pairwise mtDNA F_{ST} values between *O. chrysurus* populations (below the diagonal). The significance of each value ($H_0: F_{ST} = 0$) is shown above the diagonal. * $p < 0,05$.

	Belize	Ceará	Pernambuco	Bahia	Espírito Santo
Belize		0.000+-0.00*	0.000+-0.00*	0.000+-0.00*	0.000+-0.00*
Ceará	0.07763		0.054+-0.03	0.117+-0.03	0.468+-0.04
Pernambuco	0.21298	0.07341		0.657+-0.07	0.216+-0.02
Bahia	0.17277	0.04022	-0.02348		0.315+-0.03
Espírito Santo	0.17159	-0.00254	0.01821	0.01320	

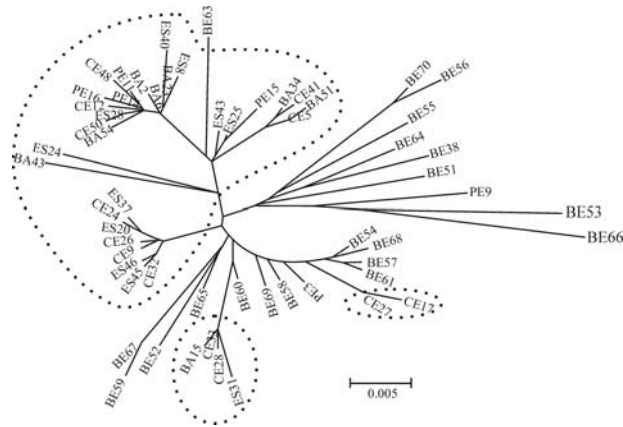


Figure 7 - mtDNA control-region haplotype neighbor-joining tree (Kimura 2-parameter distance). Dotted line surrounds haplotype clusters from the southwestern Atlantic.

signal of recent population expansion (star-shaped topology around haplotype α in Figure 5), but that grouping was not significant in the nested-clade analysis, and none of the Brazilian samples presented deviations from neutrality in both Fu's and Tajima's tests. On the other hand, the sequences of the population from Belize diverged from neutral expectation (Fu's $F_s = -8.88$; $p < 0.001$), and the mismatch distribution analysis (Figure 6) had a mode of 15 nucleotide differences, which is a clear signal of an old population expansion. The analysis of mtDNA thus indicates that the population from the Caribbean is old and currently stable. It shows also that the Caribbean population is different from the Brazilian one which is homogeneous along its distribution.

The results of the morphometric geometric analyses show what seems to be a north-south gradient of body shape, ranging from fusiform individuals, with a long caudal fin in specimens from the north to more rounded individuals, with shorter caudal fin in the south. Gradients in morphometric characters are often associated to phenotypic plasticity (O'Reilly and Horn, 2004). The morphometric differences observed are not incompatible with the existence of discrete fish stocks, since patterns of reef fish community change along the Brazilian coast often show a north-south differentiation (Floeter *et al.*, 2001), but this result contrasts with the homogeneity observed with both nuclear and mitochondrial data. It has been argued that morphometric analyses can sometimes detect subtle differences in stock structure which are undetectable by genetic data, particularly when stock structure is the result of very recent population subdivision (Cadrin, 2000). However, the differences in morphometric data on a background of genetic homogeneity could also indicate phenotypic plasticity. This phenomenon may occur as response to gradients in environmental cues (Via *et al.*, 1995). Such environmental gradient is likely to occur along a geographic range of more than 15° of latitude and ca. 2,400 km of coastline as

the one inhabited by the studied Brazilian populations of *Ocyurus chrysurus*. Differences in oceanographic conditions, mainly sea-water temperature, were reported to this area (Castro and Miranda, 1998) being regarded as an important factor in the genetic differentiation of *Macrodon ancylodon* populations along the Brazilian coast (Santos *et al.*, 2003). Cases where morphometric and genetic data indicate different scenarios of population structuring are not uncommon (Salini *et al.*, 2004; Levi *et al.*, 2004). In the case of *Ocyurus chrysurus*, the fact that the morphological variation observed formed a gradient is a clear indication that it has likely resulted from phenotypic plasticity. We thus conclude that the genetic data do not allow the rejection of the null hypothesis of panmixia for Brazilian yellowtail snapper populations which could be treated as a single genetic stock, with a latitudinal gradient on their morphology. On the other hand, there is a severe restriction to gene flow between *O. chrysurus* populations from the Caribbean and from the southwestern Atlantic.

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References

- Allen GR (1985) Snappers of the world. An annotated and illustrated catalogue of lutjanid species known to date. FAO Fish Synop 125:1-208.
- Allman JA, Barbieri LR and Bartels CT (2005) Regional and fishery-specific patterns of age and growth of yellowtail snapper, *Ocyurus chrysurus*. Gulf Mex Sci 2005:211-223.
- Batista RP and Solé-Cava A (2005) Baixa diferenciação genética entre populações do peixe-batata (*Lopholatilus villarii* Miranda-Ribeiro, 1915) ao norte e sul do banco dos Abrolhos, Brasil. In: PAS Costa, AS Martins and G Olavo (ed.) Pesca e Potenciais de Exploração de Recursos Vivos na Região Central da Zona Econômica Exclusiva Brasileira. Editora da UFRJ, Rio de Janeiro, pp 241-247.
- Belkhir K, Borsa P, Chikhi L, Raufaste N and Bonhomme F (2002) GENETIX 4.04, logiciel sous Windows TM pour la génétique des populations. Montpellier (France). Laboratoire Génome, Populations, Interactions, CNRS UMR 5000, Université de Montpellier II.
- Blaber SJM, Dichmont CM, Buckworth RC, Badrudin, Sumiono B, Nurhakim S, Iskandar B, Fegan B, Ramm DC and Salini JP (2005) Shared stocks of snappers (Lutjanidae) in Australia and Indonesia: Integrating biology, population dynamics and socio-economics to examine management scenarios. Rev Fish Biol Fish 15:111-127.

- Booke HE (1981) The conundrum of the stock concept - Are nature and nurture definable in fisheries science? *Can J Fish Aquat Sci* 38:1479-1480.
- Bookstein FL (1989) Principal warps: Thin-plate splines and the decomposition of deformations. *IEEE Trans Pat Anal Mach Intel* 11:567-585.
- Caddy JF and Seijo JC (2005) This is more difficult than we thought! The responsibility of scientists, managers and stakeholders to mitigate the unsustainability of marine fisheries. *Phil Trans Royal Soc B* 360:59-75.
- Cadrin SX (2000) Advances in morphometric identification of fishery stocks. *Rev Fish Biol and Fish* 10:91-112.
- Carmen MA and Ablan A (2006) Genetics and the study of fisheries connectivity in Asian developing countries. *Fish Res* 78:158-168.
- Carvalho GR and Hauser L (1995) Molecular genetics and the stock concept in fisheries. In: Carvalho GR and Pitcher TJ (eds) *Molecular Genetics in Fisheries*. Chapman & Hall, London, pp 55-79.
- Castro BM and Miranda LBd (1998) Physical oceanography of the western Atlantic Continental Shelf located between 4° N and 34° S coastal segment (4, W). In: Robinson AR and Brink KH (eds) *The Sea VII*. John Wiley and Sons Inc, London, pp 209-251.
- Cavalcanti MJ, Monteiro LR and Lopes PRD (1999) Landmark-based morphometric analysis in selected species of serranid fishes (Perciformes, Teleostei). *Zool Stud* 38:287-294.
- Claro R (1983) Ecología y ciclo de vida de la rabirrubia, *O. chrysurus* (Bloch), en la plataforma cubana. II. Edad y crecimiento, estructura de poblaciones y pesquerías. *Rep Invest Inst Oceanol Acad Cien Cuba* 19:1-33.
- Clement M, Posada M and Crandall KA (2000) TCS: A computer program to estimate gene genealogies. *Mol Ecol* 9:1657-1660.
- Costa PAS, Braga AD and da Rocha LOF (2003) Reef fisheries in Porto Seguro, Eastern Brazilian Coast. *Fish Res* 60:577-583.
- Excoffier L, Laval G and Schneider S (2005) Arlequin ver. 3.0: An integrated software package for population genetics data analysis. *Evol Bioinf Online* 1:47-50.
- Excoffier L, Smouse PE and Quattro JM (1992) Analysis of molecular variance inferred from metric distances among DNA haplotypes-application to human mitochondrial-DNA restriction data. *Genetics* 131:479-491.
- Fallows JA (1984) The behavioural ecology of feeding in the yellowtail snapper, *Ocyurus chrysurus* (Family Lutjanidae). PhD Thesis, University of Newcastle Upon Tyne, Newcastle.
- FAO (2002) *El Estado Mundial de la Pesca y la Acuicultura*. Food and Agriculture Organization of the United Nations, Rome, 150 pp.
- Floeter SR, Guimarães RZP, Rocha LA, Ferreira CEL, Rangel CA and Gasparini JL (2001) Geographic variation in reef-fish assemblages along the Brazilian coast. *Global Ecol Biogeogr* 10:423-431.
- Fu YX and Li WH (1993) Statistical tests of neutrality of mutations. *Genetics* 133:693-709.
- Garcia SM and Grainger RJR (2005) Gloom and doom? The future of marine capture fisheries. *Phil Trans Royal Soc B* 360:21-46.
- Grimes CB (1987) Reproductive biology of the Lutjanidae: A review. In: Polovina JJ and Ralston S (eds) *Tropical Snappers and Groupers: Biology and Fisheries Management*. Westview Press, Boulder, pp 239-294.
- Hammer O, Harper DAT and Ryan PD (2001) PAST: Paleontological Statistics Software Package for Education and Data Analysis. *Palaeontol Elect* 4(1).
- Harpending HC (1994) Signature of ancient population growth in a low-resolution mitochondrial DNA mismatch distribution. *Hum Biol* 66:591-600.
- Hedgecock D and Strong DR (1994) Conservation biology of endangered Pacific salmonids: Introductory remarks. *Cons Biol* 8:863-894.
- Ihssen PE, Booke HE, Casselman JM, McGlade JM, Payne NR and Utter FM (1981) Stock identification: Materials and methods. *Can J Fish Aquat Sci* 38:1838-1855.
- Kocher TD, Thomas WK, Meyer A, Edwards SV, Paabo S, Villablanca FX and Wilson AC (1989) Dynamics of mitochondrial DNA evolution in animals: Amplification and sequencing with conserved primers. *Proc Nat Acad Sci USA* 86:6196-6200.
- Lee W-J, Conroy J, Howell WH and Kocher TD (1995) Structure and evolution of teleost mitochondrial control regions. *J Mol Evol* 41:54-66.
- Levi D, Patti B, Rizzo P, Lo Brutto S, Parrinello N and Arculeo M (2004) Genetic and morphometric variations of Mediterranean hake, *Merluccius merluccius*, in the Strait of Sicily (central Mediterranean): Implications for stock assessment of shared resources. *Ital J Zool* 71:165-170.
- Lima D, Freitas JE, Araujo ME and Solé-Cava AM (2005) Genetic detection of cryptic species in the frillfin goby *Bathygobius soporator*. *J Exp Mar Biol Ecol* 320:211-223.
- Manchenko GP (1994) *Handbook of Detection of Enzymes on Electrophoretic Gels*. CRC Press Inc, Ann Arbor, 341 pp.
- Menezes NA and Figueiredo JL (1980) *Manual de Peixes Marinhos do Sudeste do Brasil. IV Teleostei (3)*. Museu de Zoologia, São Paulo, 96 pp.
- O'Reilly K and Horn MH (2004) Phenotypic variation among populations of *Atherinops affinis* (Atherinopsidae) with insights from a geometric morphometric analysis. *J Fish Biol* 64:1117-1135.
- Ormerod SJ (2003) Current issues with fish and fisheries: Editor's overview and introduction. *J Appl Ecol* 40:204-213.
- Ovenden JR (1990) Mitochondrial DNA and marine stock assessment: A review. *Austr J Mar Freshwat Res* 41:835-853.
- Paiva MP and Andrade-Tubino MF (1998) Distribuição e abundância de peixes bentônicos explotados pelos linheiros ao largo do sudeste do Brasil (1986-1995). *Rev Bras Biol* 58:619-632.
- Palumbi SR, Martin A, Romano S, McMillan WO, Stice L and Grabowski G (1991) *The Simple Fools Guide to PCR, Version 2.0*. Univ. Hawaii, Honolulu, 47 pp.
- Parrish JD (1987). The trophic biology of snappers and groupers. In: Polovina JJ and Ralston S (eds) *Tropical Snappers and Groupers: Biology and Fisheries Management*. Westview Press, Boulder, pp 405-463.
- Pauly D, Watson R and Alder J (2005) Global trends in world fisheries: Impacts on marine ecosystems and food security. *Phil Trans R Soc B* 360:5-12.
- Posada D, Crandall KA and Templeton AR (2000) GeoDis: A program for the cladistic nested analysis of the geographical distribution of genetic haplotypes. *Mol Ecol* 9:487-488.

- Poulik MD (1957) Starch gel electrophoresis in a discontinuous system of buffers. *Nature* 180:1477-1479.
- Raymond M and Rousset F (1995) An exact test for population differentiation. *Evolution* 49:1280-1283.
- Resende SM, Ferreira BP and Fredou T (2003) A pesca de Lutjanídeos no nordeste do Brasil: Histórico das pescarias, características das espécies e relevância para o manejo. *Bol Tecn Cient CEPENE* 11:257-270.
- Riley CM, Holt GJ and Arnold CR (1995) Growth and morphology of larval and juvenile captive bred Yellowtail Snapper, *Ocyurus chrysurus*. *Fish Bull* 93:179-185.
- Rogers AR and Harpending H (1992) Population growth makes waves in the distribution of pairwise genetic differences. *Mol Biol Evol* 9:552-569.
- Rohlf FJ and Slice DE (1990) Extensions of the Procrustes method for the optimal superimposition of landmarks. *Syst Zool* 39:40-59.
- Rozas J, Sánchez-DelBairro JC, Messeguer X and Rozas R (2003) DNASP: DNA polymorphism analyses by the coalescent and other methods. *Bioinformatics* 19:2496-2497.
- Ryman N (1991) Conservation genetics considerations in fishery management. *J Fish Biol* 39:211.
- Ryman N and Utter F (1987) Population genetics and fishery management. Washington University Press, London, 420 pp.
- Salini JP, Milton DA, Rahman MJ and Hussain MG (2004) Allozyme and morphological variation throughout the geographic range of the tropical shad, hilsa *Tenualosa ilisha*. *Fish Res* 66:53-69.
- Santos S, Schneider H and Sampaio MI (2003) Genetic differentiation of *Macrodon ancylodon* (Sciaenidae, Perciformes) populations in Atlantic coastal waters of South America as revealed by mtDNA analysis. *Genet Mol Biol* 26:151-161.
- Shaw PR and Prasad R (1970) Starch gel eletroforesis of enzymes - A compilation of recipes. *Biochem Genet* 4:297-320.
- Slice DE (1998) *Morpheus et al.*: Software for morphometric research. Department of Ecology and Evolution, State University of New York, New York.
- Smith PJ and Fujio Y (1982) Genetic variation in marine teleosts: High variability in habitat specialists and low variability in habitat generalists. *Mar Biol* 69:7-20.
- Smith PJ, Jamieson A and Birley AJ (1990) Electrophoretic studies and stock concept in marine teleosts. *J Cons Int Explor Mer* 47:231-245.
- Strauss RE and Bookstein FL (1982) The truss: Body form reconstruction in morphometrics. *Syst Zool* 31:113-135.
- Tajima F (1996) The amount of DNA polymorphism maintained in a finite population when the neutral mutation rate varies among sites. *Genetics* 143:1457-1465.
- Templeton AR (1998) Nested clade analyses of phylogeographic data: Testing hypotheses about gene flow and population history. *Mol Ecol* 7:381-397.
- Templeton AR, Routman E and Phillips CA (1995) Separating population structure from population history: A cladistic analyses of the geographical distribution of mitochondrial DNA haplotypes in the tiger salamander, *Ambystona tigrinum*. *Genetics* 140:767-782.
- Thompson JD, Higgins DG and Gibson TJ (1994) CLUSTAL W: Improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. *Nucleic Acids Res* 22:4673-4680.
- Thompson R and Munro JL (1983) The biology, ecology and bionomics of Caribbean reef fishes: Lutjanidae (snappers). In: Munro JL (ed) *Caribbean Coral Reef Fishery Resources ICLARM Stud Rev* 7. Manila, Philippines, pp 94-109.
- Vasconcellos M and Gasalla MA (2001) Fisheries catches and the carrying capacity of marine ecosystems in southern Brazil. *Fish Res* 50:279-295.
- Via S, Gomulkiewicz R, de Jong G, Scheiner SM, Schlichting CD and Van Tienderen PH (1995) Adaptive phenotypic plasticity: Consensus and controversy. *Trends Ecol Evol* 10:212-217.
- Ward RD and Beardmore JA (1977) Protein variation in the plaice (*Pleuronectes platessa*). *Genet Res* 30:45-62.
- Ward RD, Woodwark M and Skibinski DOF (1994) A comparison of genetic diversity levels in marine, fresh-water, and anadromous fishes. *J Fish Biol* 44:213-232.
- Watson M, Munro JL and Gell FR (2002) Settlement, movement and early juvenile mortality of the yellowtail snapper *Ocyurus chrysurus*. *Mar Ecol Prog Ser* 237:247-256.
- Zelditch ML, Swiderski DL, Sheets HD and Fink WL (2004) *Geometric morphometrics for biologists: A primer*. Elsevier Academic Press, San Diego, 443 pp.

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