GENETIC STRUCTURE OF *BRACHIDONTES PUNICEUS* POPULATIONS IN CAPE VERDE ARCHIPELAGO SHOWS SIGNATURE OF EXPANSION DURING THE LAST GLACIAL MAXIMUM

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ABSTRACT

Quaternary climatic fluctuations had major impacts on species' distributions over the last 2.5 Myr. Expansions and contractions of the polar ice caps throughout glacial cycles strongly affected terrestrial fauna and flora whereas eustatic sea level variations had major consequences on rocky-shore communities. The effect of these glacial episodes on marine organisms inhabiting oceanic islands is still poorly understood. We analysed the genetic structure of the marine mussel *Brachidontes puniceus* from the Cape Verde Islands using mitochondrial sequence data. The apparent absence of physical oceanographic barriers or ecological filters in the geographical range of *B. puniceus* is reflected in the lack of genetic structure found among populations from the entire archipelago. Larval dispersal in *B. puniceus* likely played a critical role in the demographic connectivity of populations across the Cape Verde archipelago. Results from demographic analysis were consistent with a population expansion promoted by an increase in the habitat available for larval settlement resulting from a low sea-level stand during the last glacial maximum.

INTRODUCTION

Climatic fluctuations of the Quaternary over the last 2.5 Myr had a major impact on species' distributions due to the expansion and contraction of the ice sheets (Webb & Bartlein, 1992; Hewitt, 2000). The effect of the glacial cycles on the geographical range of the European terrestrial fauna and flora has been extensively studied (Hewitt, 1996; Stewart & Lister, 2001; Hewitt, 2004). The 'expansion–contraction' model of Pleistocene biogeography (Provan & Bennett, 2008) is often invoked to explain geographic isolation or response of species to extreme climatic conditions. This model proposes a reduction of population sizes during the last glacial maximum (23,000–18,000 years ago) followed by northward movement of the species from southern refugia as the climate warmed (population expansion). However, more complex histories associated with different modes and rates of dispersal or distinct environmental niches might obscure demographic inference during glacial cycles. Studies on the impact of eustatic sea-level changes in the phylogeography and demographic history of marine communities have significantly increased in the last few years (Provan, Wattier & Maggs, 2005; Wares & Cunningham, 2005; Marko *et al*., 2010). Yet the effect of these glacial episodes on marine organisms inhabiting oceanic islands, especially those in tropical regions, is still poorly understood and only few examples are available (Paulay, 1990; Park & Ó Foighil, 2000; Cunha *et al*., 2005). Late Cenozoic sea-level fluctuations affected oceanic-island shallow-water communities differently depending on whether they inhabited soft or hard-bottom substrates (Paulay, 1990). During Plio-Pleistocene low sea-level stands (sea level dropped *c*. 100–150 m; Kukla, 1977; Haq, Hardenbol & Vail, 1988), soft-bottom bivalves experienced local extinctions through habitat loss. On the contrary, hard-bottom organisms exhibited higher speciation rates due to an increase of niche opportunities.

The origin of the worldwide intertidal mussel genus *Brachidontes* Swainson, 1840 (Mytilidae) dates back to the Jurassic (Coan, Scott & Bernard, 2000). The early life history involves feeding larvae drifting in the plankton for up to 40 days (Campos & Ramorino, 1980; Fields & Moore, 1983). The genus is represented in the Cape Verde Islands by a single species, *Brachidontes puniceus* Gmelin, 1791. This intertidal hard-bottom species is distributed throughout the archipelago and along the West African coast from Mauritania to Angola. The Cape Verde Islands are under the influence of the strong North Equatorial Countercurrent and the southern limit of the North Equatorial Current, which may prevent larval-mediated gene flow and increase genetic structure among populations (Fig. 1). Thus far, the only genetic study performed on *B. puniceus* included specimens from a single locality in Cape Verde (Lee & Ó Foighil, 2005) and established a phylogenetic relationship with the ecologically dominant member of a western Atlantic cryptic species complex present in the Florida Keys, Bahamas and Bermuda.

To understand the impact of Pleistocene sea-level changes on the demographic history of a planktotrophic organism inhabiting an oceanic archipelago, we investigated the genetic structure of *B. puniceus* from the Cape Verde Islands using mitochondrial (mt) sequence data. We analysed factors that can affect larval-mediated gene flow and plausible alternative scenarios of the species' response to climatic changes. If the 'expansion–contraction' model (Provan & Bennett, 2008) best explains the biogeographic response of *B. puniceus* to sea-level changes, a population contraction during glacial periods followed by an expansion after the last glacial maximum is expected, *c*. 18,000 years (18 Kyr) ago (Bennett, 1990; Hewitt,

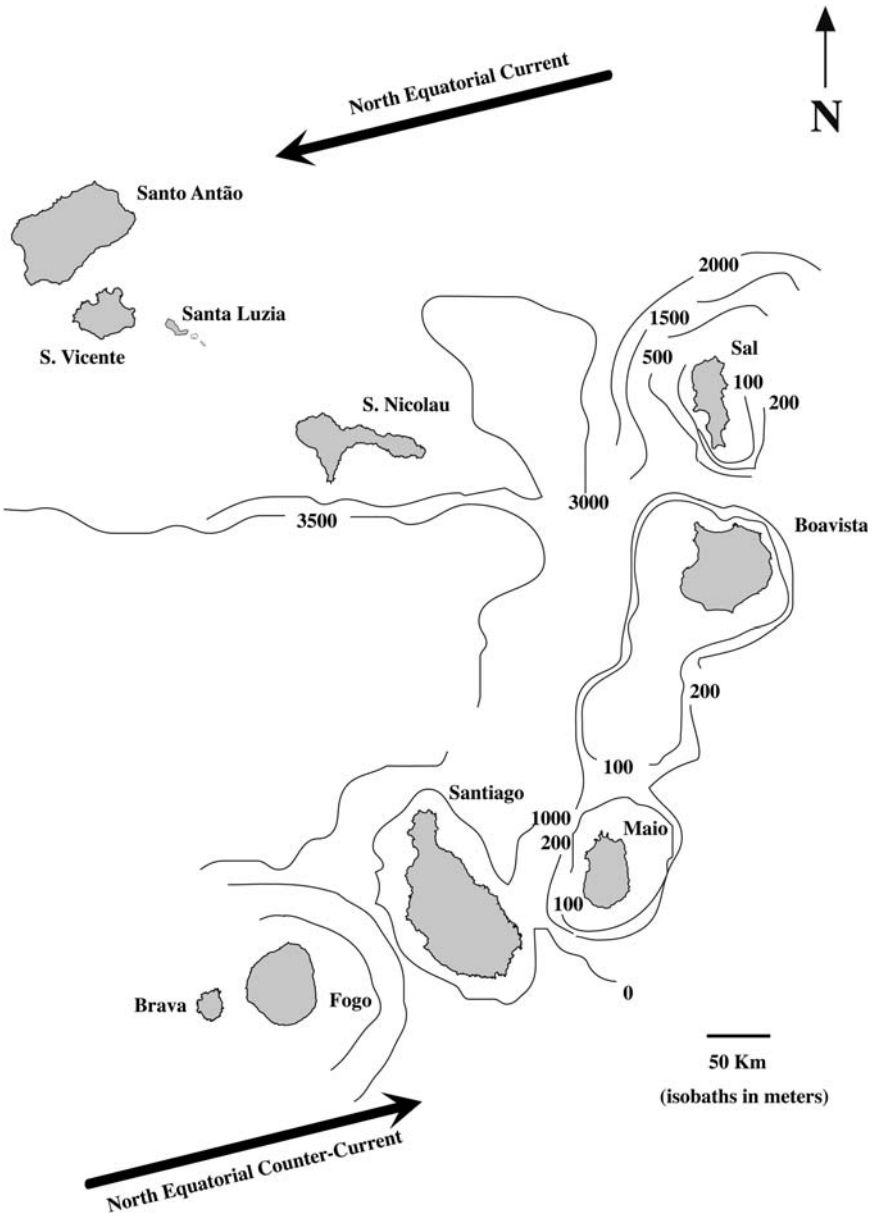

Figure 1. Map and bathymetry of the Cape Verde archipelago. Isobaths (in metres) are indicated, as well as the direction of the prevailing currents.

1999; Provan & Bennett, 2008). However, during periods of falling sea level promoted by glacial episodes, islands increase in area and niche opportunities. Therefore, in hard-bottom organisms such as *B. puniceus* a population expansion during these periods might also be expected. An additional hypothetical scenario would be demographic stability throughout the last glacial maximum as observed in planktonic rocky-shore species along the northeast Pacific coast (Marko *et al.*, 2010).

MATERIAL AND METHODS

Taxon sampling, DNA amplification and sequencing

A total of 114 specimens of *Brachidontes puniceus* with an average of 14 specimens per population were collected in eight of the 10 islands of the Cape Verde archipelago (Table 1). Between two to three different sampling points were chosen from each island to evaluate intrasite variation, except in Santa Luzia because of its small size. All specimens were preserved in 96% ethanol and total genomic DNA was extracted from muscle tissue using a salt-extraction method (Sambrook, Fritsch & Maniatis, 1989). Universal primers of Folmer *et al.* (1994) were used in PCRs to amplify about 600 bp of the mt cytochrome oxidase subunit I (COI) gene in a few specimens of *B. puniceus*. Internal *Brachidontes*-specific COI primers (COI – F: CCTTTATTTAA CGTTTGGAA; COI – R: GTCGTATTAAAATTTCGATC) were used to amplify a fragment of 548 bp. All PCR amplifications were conducted in 25-μl reactions containing 1× PCR buffer, 0.2 mM of each dNTP, 0.2 μM of each primer, 2 mM of $MgCl_2$, 1 μl of template DNA and *Taq* DNA polymerase (1 U, GoTaq®, Promega). The PCR programme was as follows: one cycle of 2 min at 94°C, 40 cycles of 1 min at 94°C, 30 s at 42°C and 1 min at 72°C, with a final extension step of 5 min at 72°C. PCR products were purified by ethanol/sodium acetate precipitation and directly sequenced using the corresponding PCR primers. Sequencing was performed in an automated sequencer

Table 1. Sample locations and summary statistics for *Brachidontes puniceus*.

Island	Location	Latitude	Longitude	*N*	*H*	*H*	π
São Vicente	Calhau	16°51′16″N	24°51′47″W	15	6	0.705	0.198
São Nicolau	Tarrafal	16°33′44″N	24°21′33″W	16	5	0.650	0.141
Sal	Murdeira	16°40′36″N	22°56′11″W	15	8	0.758	0.248
Boavista	Ponta de Ervatão	16°02′24″N	22°41′40″W	15	6	0.702	0.312
Santiago	Cidade Velha	14°54′53″N	23°36′23″W	15	10	0.895	0.309
Santo Antão	Ponta do Sol	17°12′11″N	25°05′38″W	15	9	0.876	0.348
Santa Luzia	Ponta Branca	16°46′59″N	24°47′26″W	7	4	0.714	0.209
Maio	Ponta Preta	15°07′34″N	23°12′21″W	16	11	0.875	0.333

Number of sequenced individuals (*N*) per island, number of haplotypes (*H*), haplotype diversity (*h*) and nucleotide diversity (π) calculated in DnaSP.

(ABI PRISM 3700) using the BigDye® Terminator v.3.1 Cycle Sequencing Kit (Applied Biosystems), and following the manufacturer's instructions.

Sequence analysis

The dataset included a 548 bp fragment of the mt COI gene from 119 specimens of *B. puniceus* (114 from this study and five sequences retrieved from GenBank with the accession numbers: AY825105–8). Sequences were aligned with CLUSTALX v.1.83 (Thompson *et al.*, 1997) using the default settings and further verified by eye in MACCLADE v.4.08 OS X (Maddison & Maddison, 2003) in order to maximize positional homology.

Population genetic structure

Pairwise genetic divergences between sampling sites were estimated with the fixation indexes F_{ST} and Φ_{ST} (Excoffier, Smouse & Quattro, 1992) using ARLEQUIN v.3.5 (Excoffier & Lischer, 2010). Significance of pairwise population comparisons was tested by 20,000 permutations to construct null distributions and test the significance of variance components for each hierarchical comparison (Guo & Thompson, 1992). The standard Bonferroni correction (Rice, 1989) was applied for multiple tests. Partitioning of genetic variability among sites was further tested using a hierarchical analysis of molecular variance, AMOVA (Excoffier *et al.*, 1992) as implemented in ARLEQUIN.

Haplotype and nucleotide diversities were calculated with DnaSP v.5 (Librado & Rozas, 2009). When haplotype diversity is high and most haplotypes are unique, it is not feasible to assess the genetic structure solely from the haplotype frequency data. The statistic *Snn* is a measure of how often the 'nearest neighbours' of sequences are from the same locality or geographic area. It was proposed as a more powerful method for handling loci with high haplotype diversity (Hudson, 2000). Values of *Snn* are expected to be close to 0.5 if individuals from different locations are panmictic and close to 1 if they are highly differentiated (Hudson, 2000). To account for the effect of multiple comparisons and small samples sizes, *P*-values of the *Snn* statistics were adjusted using the sequential Bonferroni correction (Rice, 1989) in DNASP. One thousand simulations evaluated the significance of the statistic. The location of samples was assigned randomly but in the same proportions as in our original dataset.

To assess the potential impact of gene flow on genetic structure, we tested for a positive correlation between genetic distance $[\Phi_{ST}/(1-\Phi_{ST})]$ and geographic distance using the Isolation by Distance Web Service v.3.16 (Jensen, Bohonak & Kelley, 2005). Following Rousset (1997), we tested for isolation by distance estimating Φ_{ST} based on the Kimura two-parameter distance. Distances were regressed against the natural logarithm of geographic distance using the shortest (straight line) distance. Under the assumption of isolation by distance, a linear relationship between the genetic distance and logarithmic distance is expected (Rousset, 1997). Therefore, log-transformed distances were used because the sampled area represents a two-dimensional habitat for *B. puniceus* larvae. The significance of the associations was tested with a Mantel test using 20,000 permutations.

Haplotype genealogies using parsimony (Templeton, Crandall & Sing, 1992) were reconstructed with TCS v.3.14 (Clement, Posada & Crandall, 2000). Pairwise distance values for all haplotypes were estimated above the 95% level to create the network. In addition, nonmetric multidimensional scaling (MDS) (Kruskal, 1964) was used to assess group structure with the ALSCAL program included in the SPSS v.18.0 package (SPSS Inc., Chicago, IL, USA). The MDS was based on intersite Euclidean distances calculated from the matrix of estimated TN93+I genetic distances between locations.

Mutation rates in Brachidontes puniceus

The available mutation rates (18.29 and 24.38% per Myr/per generation) determined by Lee & Ó Foighil (2005) for *Brachidontes* were calculated using the divergence of geminate pairs of species separated by the Isthmus of Panama. However, these estimates were based on third codon positions of COI and, according to the authors, there was evidence of saturation. Therefore, to estimate mutation rates we used COI sequence data from Lee & Ó Foighil (2005) excluding third codon positions. Nucleotide substitutions among the geminates *Brachidontes semilaevis* (Pacific) *vs. B. exustus (*from Gulf+Atlantic clades) (see Lee & Ó Foighil, 2004, 2005, for sample locations) were calculated based on Tamura–Nei corrected distances with MEGA v.4 (Tamura *et al.*, 2007). These estimates were calibrated using the same calibrations as in Lee & Ó Foighil (2005) at 2.7 and 3.6 Ma. The first calibration is based on the intercontinental exchange of land mammals (Marshall, 1988) and the latter on the differentiation of nearshore molluscan palaeofaunas (Coates *et al.*, 1992). We used the formula $G\ [(1/2)d/(D \times 10^6)]$ to calculate the mutation rate per site and per generation, where G is the generation time, d represents nucleotide substitutions per site and D corresponds to the estimated divergence date for the geminate species (Wares & Cunningham, 2001). The estimated generation time for *Brachidontes* is 3 years (Morton, 1988).

Demographic history of Brachidontes puniceus

To determine whether the individuals were in mutation–genetic drift equilibrium, the following neutrality tests were performed in DNASP v.5 (Librado & Rozas, 2009): Tajima's *D* (Tajima, 1989), Fu's F_S (Fu, 1997) and R_2 (Ramos-Onsins & Rozas, 2002). Significant negative values of *D* and F_S and positive R_2 values can be interpreted as signatures of population expansion. The R_2 test has greater statistical power when sample sizes are small (Ramos-Onsins & Rozas, 2002).

We characterized past changes in the effective population size (N_e) of *B. puniceus* by generating Bayesian skyline plots (BSPs) (Drummond *et al.*, 2005) with BEAST v.1.5.4 (Drummond &

Rambaut, 2007). We used only third codon positions from our COI dataset because they are less likely to cause amino acid substitutions and therefore more likely to follow assumptions of neutrality (Wares & Cunningham, 2001), and there was no evidence of saturation in our dataset (data not shown). The mutation rates were estimated at 3.5 and 2.65% per Myr/site/generation, determined as explained above. BSPs generate a posterior distribution of N_e through time using Markov chain Monte Carlo (MCMC) sampling. The reconstructed BSPs were obtained using the TN93 model as selected by the AIC criterion implemented in MODELTEST. A piecewise-constant model with 10 groups was selected. All analyses ran for 10 million iterations with the first 10% discarded as burn-in. Genealogies and model parameters were sampled every 2,000 iterations. Bayesian skyline reconstructions were conducted in TRACER v.1.5 (Drummond & Rambaut, 2007) to determine the effective population size over time (N_eT), the median and the corresponding credibility intervals. Demographic analyses were carried out using the resources of Bioportal from University of Oslo (https://www.bioportal.uio.no).

RESULTS

Population genetic structure of Brachidontes puniceus

The 114 sequences (GenBank accession numbers HM999676–HM999789) yielded 39 haplotypes. Of these, 33 haplotypes were unique. Haplotype diversity (Table 1) was relatively high, varying between 0.650 and 0.895 (mean = 0.769 ± 0.037). Nucleotide diversity (Table 1) varied between 0.141 and 0.348% (mean = 0.266 ± 0.032%). The hypothesis of panmixia, tested with AMOVA with all localities as one-gene pool, was not rejected ($F_{ST} = -0.012$, $P = 0.784$; $\Phi_{ST} = -0.011$, $P = 0.881$). The nearest-neighbour statistic did not detect any genetic differentiation among neighbouring localities ($Snn = 0.106$, $P = 0.956$). Moreover, single pairwise site comparisons were all nonsignificant for these three statistics even after Bonferroni corrections.

The mtDNA haplotype network (Fig. 2) showed a star-like shape with a central most common ancestral haplotype connected to many rare haplotypes by one to eight mutational steps. Neither the regression of the pairwise $\Phi_{ST}/(1 - \Phi_{ST})$ values against log distance ($R_2 = 0.014$; $P = 0.972$) nor the MDS analysis showed any evidence of phylogeographic or genetic structuring (not shown).

Demographic history of Brachidontes puniceus

Both Tajima's and Fu's neutrality tests indicated significant departures from equilibrium ($D = -2.54$; $P < 0.001$ and $F_S = -53.97$; $P < 0.001$, respectively). The R_2 test resulted in a significant positive value (0.0169; $P = 0.002$).

The resulting output of the MCMC analysis of *B. puniceus* using BSP models is summarized in Figure 3. Figure 3-A1 and -A2 represents the same demographic analysis with different time scales using a mutation rate of 2.65%. Figure 3-A1 indicates the variation in N_e during the last 28 Kyr, whereas Figure 3-A2 shows its variation over the last 264 Kyr. Figure 3-A2 shows an increase in N_e starting at about 30 Kyr, and Figure 3-A1 indicates that population size remained constant during the last 20 Kyr. Figure 3-B1 and -B2 shows the variation of the effective population size (N_e) of *B. puniceus* using a mutation rate of 3.5%. Figure 3-B2 shows an increase in N_e starting at about 20 Kyr, and Figure 3-B1 indicates that population size remained constant during the last 10 Kyr.

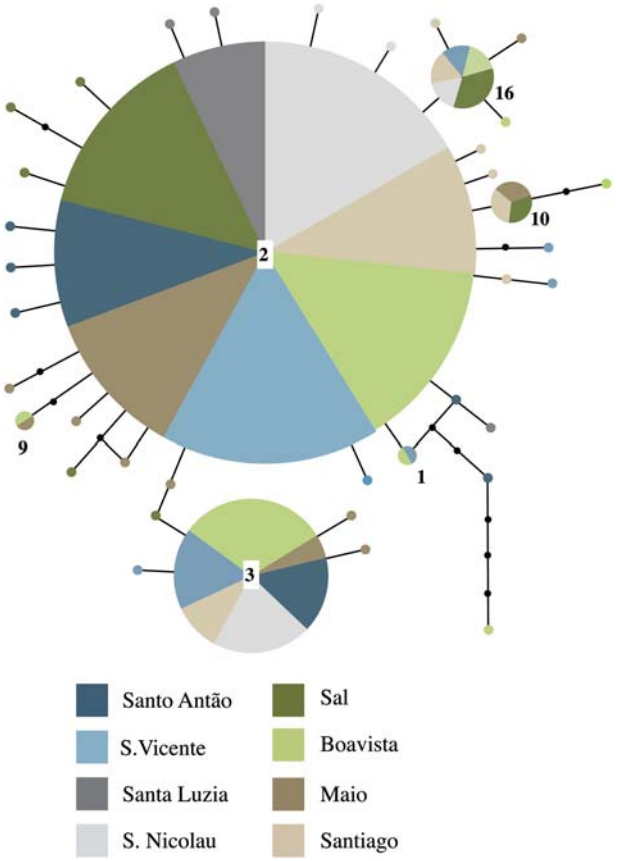

Figure 2. TCS parsimony network of *Brachidontes puniceus*. Pairwise distance values for all haplotypes were estimated defining the limits of parsimony above the 95% level. Numbers represent the most common haplotypes.

DISCUSSION

Genetic structure and larval development of Brachidontes puniceus *from the Cape Verde archipelago*

It is often assumed that benthic organisms with a long pelagic early life stage will display wide geographic ranges and reduced genetic structure compared to those lacking a free-drifting stage (Jablonski, 1986; Scheltema, 1986). On the other hand, species with lecithotrophic larval development reach metamorphosis without feeding on plankton and usually exhibit reduced dispersal capabilities and more restricted geographic distributions. However, connectivity between rocky intertidal invertebrate populations exhibiting direct development was not affected by the existence of a strong marine biogeographic barrier in southeastern Australia (Ayre, Minchinton & Perrin, 2009). Hence, prediction of gene flow among populations based on their larval development should be carefully evaluated.

Members of the genus *Brachidontes* exhibit a planktotrophic veliger lasting up to 40 days before becoming competent to settle (Campos & Ramorino, 1980; Fields & Moore, 1983). Accordingly, wide dispersal abilities and reduced genetic population structures are expected. However, recent studies on the Caribbean *Brachidontes exustus* (Lee & Ó Foighil, 2004, 2005) showed the existence of distinct geographic areas and a clear signature of genetic structure, which questions this prior assumption. These genetic discontinuities seem to be maintained by postrecruitment ecological filters rather than oceanographic barriers to larval dispersal (Lee & Ó Foighil, 2005). The nature

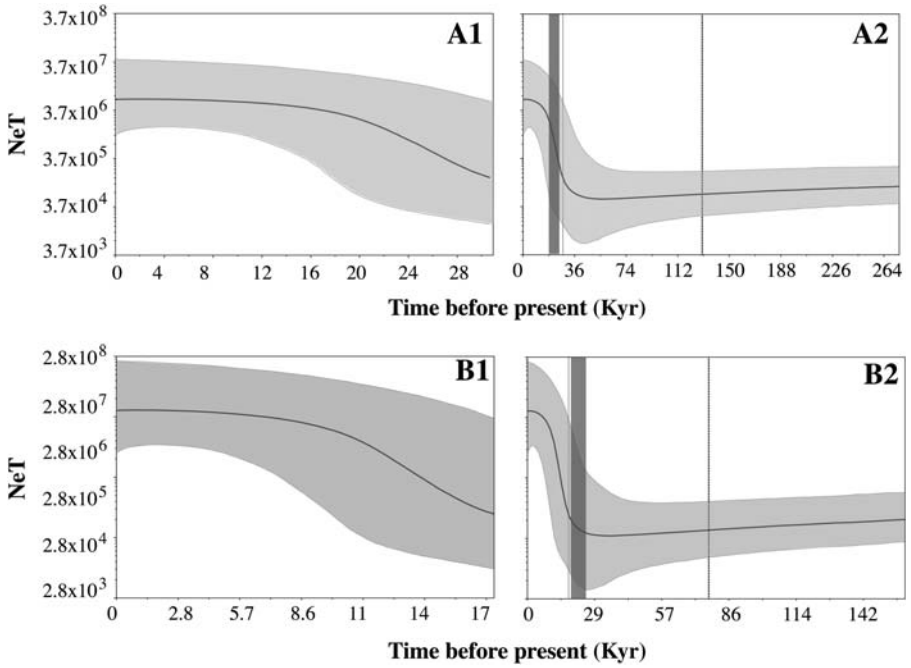

Figure 3. BSPs of changes in effective population size over time based on mitochondrial COI sequence data of *Brachidontes puniceus* analysed with BEAST. BSPs from A1 and A2 represent the same demographic analysis (as well as B1 and B2) shown with different time scales. BSPs represented in A1 and A2 were based on a mutation rate of 2.65%, whereas BSPs from B1 and B2 were based on a mutation rate of 3.5%. On the *y*-axes N_e represents effective population size and T = generation time; *x*-axes represent time in thousands of years (Kyr). The thick solid line represents the median and the grey area the 95% highest posterior density of the N_eT estimates. Vertical dark-grey bars represent the duration of the last glacial maximum.

of the environmental exclusion mechanisms is related to transitions between two distinct habitats. Continental rocky-shores present lower salinity and higher nutrient conditions whereas oceanic islands habitats exhibit higher salinities and lower productivity (Lee & Ó Foighil, 2005).

Here, we analysed the genetic structure of the marine mussel *B. puniceus* from eight of the 10 islands of the Cape Verde archipelago, but found no evidence of any genetic differentiation. All F_{ST} and Φ_{ST} pairwise comparisons, as well as *Snn* statistics, were nonsignificant. The hypothesis of panmixia was not rejected. Neither the regression between genetic and geographic distances nor the MDS revealed the existence of any phylogeographic groups. Niche differences (oceanic islands *vs.* continental rocky-shores) caused environmental exclusion promoting genetic discontinuities in the Caribbean *Brachidontes*. Regardless of the absence of such habitat transitions in the Cape Verde Islands, a recent study on demersal fish communities with a sedentary lifestyle on the rocky bottoms around the islands showed the existence of an ecological structure associated with geographic distance and habitat suitability (Medina *et al.*, 2007). Nevertheless, given the absence of genetic structure in all analysed locations there is no evidence for the existence of ecological transitions in the habitat occupied by *B. puniceus*. Moreover, increasing geographic distance or depth among islands is not sufficient to promote any genetic structuring. The Cape Verde Islands are under the strong influence of large-scale oceanic circulation and dominant currents are likely to promote a physical barrier to larval drifting across the archipelago, increasing the genetic structure. However, according to our results the influence of the prevailing currents (see Fig. 1) seems insufficient to create a physical barrier and larval-mediated gene flow maintains population connectivity along the archipelago. This outcome is not completely unexpected considering that Bermudan populations showed no evidence of genetic isolation from Bahamian/Floridian populations separated by an oceanic barrier greater than the Cape Verde – Africa geographic break (Lee & Ó Foighil, 2005).

Sea-level changes and demographic history of Brachidontes *in Cape Verde archipelago*

Several possible scenarios can be hypothesized regarding the response of marine communities to sea-level changes. According to the biogeographic 'expansion–contraction' model (Provan & Bennett, 2008), a contraction in size and a shift towards lower latitudes during periods of cooling followed by population growth and recolonization during postglacial warming is expected. Another plausible scenario would be population expansion determined by an increase of niche opportunities due to the lowered sea level during glacial episodes. Alternatively, demographic stability over the last glacial maximum (18–23 Kyr ago) has been observed in several rocky-shore populations along the northeast Pacific coast (Marko *et al.*, 2010).

According to our results, both significant negative D and F_S values and the positive R_2 support a signature of population expansion. The mtDNA haplotype network shows a star-like shape with a central most common ancestral haplotype connected to many rare haplotypes by few mutational steps. This result is also consistent with a demographic expansion or past selective sweeps. Regardless of the calibration of the mutation rate used, our Bayesian reconstructions point to a population expansion during the last glacial maximum. *Brachidontes puniceus* expanded in the Cape Verde archipelago at about 30 Kyr (Fig. 3-A2) or 20 Kyr (Fig. 3-B2), depending on whether the calibration of the mutation rate is set at 3.6 or 2.7 Myr, respectively. The available mutation rates for *Brachidontes* estimated by Lee & Ó Foighil (2005) were based on saturated COI third codon positions, which could affect the

estimates. As in any estimation of mutation rate, our estimates should be considered cautiously. We used Lee & Ó Foighil (2005) COI sequences including only first and second positions, which as expected produced lower rates between 3.5×10^{-2} (calibrated at 2.7 Myr) and 2.65×10^{-2} (calibrated at 3.6 Myr) nucleotide substitutions/site/generation per Myr. Our estimates were also smaller when compared to mutation rates provided by Wares & Cunningham (2001) for *Mytilus edulis* (9.51×10^{-2} nucleotide substitutions/site/generation per Myr, again based on third codon positions).

Eustatic sea-level fluctuations can greatly affect shallow-water communities, causing major extinctions or promoting population expansion. All major mass extinctions have been associated with marine regressions, whereas high sea-level stages are often considered to provide opportunities for diversification (Jablonski, 1986). Nevertheless, the scenario of population expansion coinciding with a low sea-level stand has been observed in bivalve faunas of tropical oceanic islands (Paulay, 1990). The ability of bivalves to resist to extinction during a marine regression depends on substratum availability (Paulay, 1990). Hard-bottom species seem to be less affected by regressions when compared with soft-bottom organisms.

The two studies performed thus far on the impact of sea-level changes in marine communities from the Cape Verde archipelago used *Conus* species as subjects for analysis (Cunha *et al*., 2005, 2008). During periods of falling sea-level, increase of niche opportunities promoted speciation in these gastropods (Cunha *et al*., 2005) and the reconstructed phylogeographic patterns were consistent with fluctuations in the sea level (Cunha *et al*., 2008). During high sea-level stands, populations remained isolated on islands separated by deeper channels due to the suspected reduced dispersal abilities of *Conus* species with lecithotrophic larvae. The demographic history of *B. puniceus* revealed a population expansion during the last glacial maximum. During periods of falling sea level, coastline geomorphology is modified and islands increase in area (Graham, Dayton & Erlandson, 2003) providing extra habitats for hard-bottom species such as *Brachidontes*.

According to the 'expansion–contraction' model, we would expect a population contraction during the last glacial maximum between 18 and 23 Kyr, which was not corroborated by our data. It is also important to remember that this model has mostly been applied to terrestrial systems, in which other factors (e.g. expansion of the polar ice caps or temperature) rather than sea level have played a crucial role in species' distributions.

There is growing evidence for demographic stability in marine species with planktonic larvae living in low-shore habitats since the last glacial maximum (Marko, 2004; Wares & Cunningham, 2005). A recent study on rocky-shore species along the Pacific coast of North America showed that demographic histories consistent with regional persistence during the last glacial maximum appears to be a common biogeographic pattern in this area (Marko *et al*., 2010). Our results are not in agreement with this demographic stability throughout the last glacial maximum. The intrinsic particularities of *B. puniceus* with sessile adults and an extended pelagic larval phase, combined with an increase in habitat area during glacial episodes, determined a different response of this rocky-shore species to eustatic sea-level changes.

Conclusions

The apparent absence of physical oceanographic barriers or ecological filters in the geographical range of *B. puniceus* is reflected in the lack of genetic structure found among populations from the entire archipelago. Larval dispersal in *B. puniceus* likely played a critical role in the demographic connectivity of populations throughout the Cape Verde archipelago. A population expansion occurred during the last glacial maximum, most likely determined by an increase in habitat area during low sea-level stages. The 'expansion–contraction' model is not supported by our data and neither is demographic stability during the last glacial maximum, as observed in other planktonic species from the northeastern Atlantic (Jolly *et al*., 2006) and Pacific oceans (Marko *et al*., 2010).

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