*Bulletin of Insectology* ***69*** *(1): 117-126, 2016*
ISSN 1721-8861

# Distribution and genetic variability of Staphylinidae across a gradient of anthropogenically influenced insular landscapes

**José A. P. Marcelino[1,2], Rosanna Giordano[3], Paulo A. V. Borges[1], Patricia V. Garcia[1,2], Felipe N. Soto-Adames[3,4], António O. Soares[1,2]**
[1]*CE$_3$C, Centre for Ecology, Evolution and Environmental Changes, Azorean Biodiversity Group, Department of Biology, University of the Azores, Portugal*
[2]*CIRN, Department of Biology, University of the Azores, Portugal*
[3]*Department of Biology, University of Puerto Rico, San Juan, USA*
[4]*Department of Entomology, University of Illinois, Urbana, USA*

## Abstract

This paper describes the distribution and genetic variability of rove beetles (Coleoptera Staphylinidae) in anthropogenically influenced insular landscapes. The study was conducted in the Azores archipelago, characterized by high anthropogenic influence and landscape fragmentation. Collections were made in five islands, from eight habitats, along a gradient of anthropogenic influence. The species of Staphylinidae from the Azores collected for this study were widely distributed and showed low habitat fidelity. Rove beetle richness was associated with anthropogenic influence and habitat type, increasing from less to more anthropogenic impacted habitats. However, genetic diversity of profiled species (i.e. with three or more specimens per species/habitat) does not seem affected by anthropogenic influence in the different habitat types, isolation or landscape fragmentation. COI haplotypes were, as a rule, not exclusive to a given island or habitat. High level of genetic divergence and nucleotide saturation was found in closely related morphological designated species, demonstrating possible disparities between currently defined taxonomic units based on morphology and molecular phylogenies of Staphylinidae. This study found evidence of cryptic speciation in the *Atheta fungi* (Gravenhorst) species complex which had thus far remained undetected. Similar trends were found for *Oligota parva* Kraatz, *Oxytelus sculptus* Gravenhorst, *Oligota pumilio* Kiesenwetter. Previous studies with lower taxonomical resolution may have underestimated the biotic diversity reported in the Azores in comparison to other Macaronesian archipelagos.

**Key words:** habitat type, landscape fragmentation, geographic isolation, genetic variability, Staphylinidae, rove beetles.

## Introduction

Remote volcanic oceanic archipelagos are usually characterized by high endemism and represent a significant portion of terrestrial biodiversity (Holland and Hadfield, 2002; Whittaker and Fernández-Palacios, 2007; Martín *et al*., 2008). The archipelago of the Azores possesses a nutrient rich volcanic soil that supports an abundant flora. As a result, the Azores are recognized worldwide as important conservation hotspots (Myers, 2000; Dias, 2007), despite the high level of landscape fragmentation and anthropogenic impact experienced in its islands (Cardoso *et al*., 2009; 2010; Meijer *et al*., 2011).

In comparison to other Macaronesian archipelagos, the Azores have comparatively lower endemic diversity (Borges *et al*., 2008; 2010a; 2010b; Arechavaleta *et al*., 2010). They are characterized by the ubiquitous presence of shared endemics across islands and few single island endemics (Borges *et al*., 2010b). Recent studies have reported a paucity of species radiation for the flora (Carine and Schaefer, 2010) and fauna (Amorim *et al*., 2012) of the Azores. Reasons for this phenomenon have been debated, and they range from: 1) Lack of past climatic fluctuations (Carine and Schaefer, 2010); 2) Low taxonomical species resolution and species underestimates (Schaefer *et al*., 2011a); 3) Relatively recent geological origin and thus in the first stages of their biological evolution (Borges and Hortal, 2009; Triantis *et al*., 2012) and 4) Low habitat heterogeneity (Triantis *et al*., 2012). The interplay between these factors may have dictated the comparative lower level of taxonomic diversity in the Azores, and the lower level of endemics may have facilitated dispersal within the archipelago.

Landscape genetic studies could help to further clarify the patterns observed in the distribution, speciation and radiation of species in the Azores. Landscape genetics has been used as a tool in strategies to manage biodiversity, with the aim of discerning the impact of ecological, anthropogenic and geographic constrains on the distribution of genetic variability, as well as evolutionary processes in complex landscape matrices (Manel *et al*., 2010; Segelbacher *et al*., 2010; Sommer *et al*., 2013). Molecular data have only recently been incorporated into biodiversity studies in the Azores (Carine and Schaefer, 2010; Schaefer *et al*., 2011a; 2011b; Amorim *et al*., 2012), and they have been focused mainly on Azorean plants (Rumeu *et al*., 2011; Schaefer *et al*., 2011a; 2011b; Silva *et al*., 2011; Martins *et al*., 2013) and more recently on *Tarphius* beetles (Amorim *et al*., 2012), gastropods (Jordaens *et al*., 2009, Dayrat *et al*., 2011) and birds (Rodrigues *et al*., 2013). A common conclusion from these works is that to better understand patterns of distribution of genetic variation in the Azores, studies with more robust genetic data (e.g., number and length of genes profiled) are necessary. More than half of the endemic species found in the Azores are arthropods (Borges *et al*., 2010b), rendering this group an optimal choice for the study of the distribution of genetic variability in populations isolated on oceanic islands and fragmented habitats. Arthropods have been used in landscape conservation studies, to test for the correlation between genetic diversity and anthropogenic influence (Heidinger *et al*., 2013). One of the reasons species on oceanic islands are of particular interest is due to their isolation from mainland species pools (Gillespie and Roderick, 2002; Whittaker and Fernández-Palacios, 2007). The Azores have been inhabited by humans for the past 600 years. Anthropogenic effects on the archipelago have given rise to a variable landscape consisting of mainly pastures interspaced with commercial production forests of *Cryptomeria japonica* (L.f.) D.Don and few remnants of pristine forests. The cultural and natural history of the Azores makes these islands an ideal environment to study how landscape fragmentation, and anthropogenic influence across a large range of habitats, influences the genetic distribution of arthropod diversity.

In order to study the distribution and genetic variability of the arthropod fauna of the Azores, we have chosen rove beetles (Coleoptera Staphylinidae) as a model group, since they occupy a wide diversity of habitats in the Azores. Staphylinids represent one of the most diverse lineages of arthropods, inhabiting practically all terrestrial niches (Thayer, 2005). To date 47,744 species of Staphylinidae have been reported worldwide (Thayer, 2005) with 117 described from the Azores (Borges *et al*., 2010a). Rove beetles are a common and ecologically important component of soil fauna and known as potential bioindicators of environmental quality (Bohac, 1999; Rainio and Niemela 2003; Basset *et al*., 2004; Hodkinson and Jackson 2005; Pohl *et al*., 2007). Despite their abundance and widespread occurrence, information as to their ecological role is scarce (Chatzimanolis, 2004) and limited to few species (Dennis *et al*., 1997; Krooss and Schaefer, 1998; Andersen and Eltun, 1999; Byers *et al*., 2000; Caballero *et al*., 2009). Studies of staphylinids on insular ecosystems are limited to species lists (Assing, 2002; Klimaszewski *et al*., 2002; Borges *et al*., 2010a; Théry and Leschen, 2013). Moreover, the taxonomic treatment of members of this group has been difficult due to the paucity of diagnostic morphological characters. As a result, the taxonomic and phylogenetic relationship among tribes has been extremely challenging (Anderson and Ashe, 2000; Ashe, 2005; Beutel and Molenda, 1997; Caterino and Vogler, 2005; Chatzimanolis *et al*., 2010; Elven *et al*., 2010; 2012; Gutiérrez-Chacón *et al*., 2009; Hansen, 1997; Thayer, 2005; Thomas, 2009; Osswald *et al*., 2013).

This study aims to determine whether anthropogenically fragmented insular landscape matrices influence the spatial distribution and genetic variability of staphylinids on the Azores, as well as to determine the taxonomic relationship of Azorean taxa to the four informal groups of Staphylinidae *sensu* Lawrence and Newton (1995), i.e. Omaliine, Tachyporine, Oxyteline and Staphylinine), vis-à-vis published COI data from other species published in Genbank. We hypothesize that the distribution, species richness, abundance and genetic diversity of staphylinids will change with increasing anthropogenic influence. We stress that the genetic analysis in this work aims to determine the taxonomic relationship of the species collected on the Azores in relation to other species for which genetic information is available, not to establish a definitive molecular phylogeny of the family. Such a task is beyond the scope of this work.

## Materials and methods

### Habitat selection and sampling

Habitats were selected to represent the land-use diversity of the Azorean archipelago, located across the Mid-Atlantic ridge (36°35'-39°43'N 24°45'-31°17'W). The five islands sampled were selected on the basis of the proportion of land present in a pristine and fragmented state along a gradient of anthropogenic influence, i.e., São Miguel (SMG) with a large proportion of land allocated to agriculture and low/medium proportion of natural habitats; Terceira (TER) with a large proportion of land dedicated to pastureland and a medium/high proportion of natural habitats; Pico (PIC) with a high proportion of land dedicated to pastureland and medium/high proportion of natural habitats, classified as UNESCO World Heritage cultural landscapes; Flores (FLO) with a small proportion of land dedicated to pastureland and a high proportion of natural communities, classified as UNESCO Biosphere Reserve; Santa Maria (SMR) with a high proportion of land allocated to agriculture, although not all presently used, and a small proportion of natural habitats, albeit of unique conservation value. The habitats sampled for this study, depicting gradients of increasing anthropogenically influenced habitats were as follows; Herbaceous gradient: meadows, semi-natural pastures, intensive pasture land and corn fields. Arborescent gradient: pristine forest of *Laurus azorica* (Seub.) Franco, invasive forests of *Pittosporum undulatum* Vent., production forests of *C. japonica*, and orchards of *Citrus sinensis* L. (see Marcelino *et al*., 2013 for further description of the habitats and supplemental material (figure S1, supplement 1) for further description of the islands and distribution of biodiversity).

To obtain a representative sampling of Staphylinidae species in each habitat, pitfall traps (PF) and Berlese-Tullgren traps (BT) were used. A total of sixteen sites/island were sampled for Staphylinidae diversity, these spanned eight habitats (2 replicates per habitat type and island sampled, with two transects of 100 m each). Habitats were ca. 5 km-20 km apart. PF consisted of plastic cylinder cups, 78 mm deep and 42 mm in diameter, filled with ca. 80 ml of 96% ethanol and 0.04% commercial liquid dishwashing detergent to break the surface tension of the ethanol. PF were placed in the soil with the rim flushed with the surface and covered with a plastic plate at ca. 3 cm above the trap to avoid desiccation, flooding or insectivore predation. Traps remained in the soil for 7 days prior to removal. Thirteen PF were set up in each transect. The number and distance of BT traps was equal to PF traps. Soil and litter samples, as well as sweeping and vacuum samples, were collected at each PF trap area. Soil and litter samples were stored at 10 °C prior to being placed in BT traps. Sweeping and vacuum samples were also stored at 10 °C and subsequently sorted manually. BT consisted of two plastic darkened containers assembled together to provide an upper vented area (14 cm diameter × 11.5 cm high) with four openings (1 cm diameter covered with a 0.3 × 0.3 mm diameter mesh), coupled to a 15 W lamp on top, and a collecting area at the bottom (13 cm diameter × 10 cm high) partially filled with ca. 80 ml of the same liquid mixture used in PF. Litter samples were processed in the same device as described above but with a 1.8 × 1.8 mm mesh attached to a plastic funnel in the center of the device that filtered small invertebrates. The number and distance of BT traps was equal to the PF traps. Litter samples remained in BT traps for 72 h before removal, storage and taxonomic identifications. All sampling was conducted in June 2009 to ensure similar climatic and vegetation conditions. Species inventory completeness was determined by plotting species rarefaction curves using EstimateS (Colwell, 2011). Sampling representativeness was determined as:

$$S_{\text{Observed species}}/S_{\text{Jackknife1 species estimator}}$$

We selected the Jackknife estimator since it is one of the most robust when using incidence data (Hortal *et al.*, 2006).

### Taxonomic identifications

Specimens were sorted under a stereomicroscope and assigned to morpho-species using reference image galleries generated for the study (www.eden-azores.com). Reference voucher specimen collections were generated for each habitat type and islands surveyed and were stored at the Entomology Laboratory, Department of Biology of the University of the Azores. They are part of the Arthropoteca of the University of the Azores (ARZ), at Ponta Delgada, Portugal. All specimens were preserved in 96% ethanol and stored in a chamber at 4 °C, prior to higher taxonomical identifications (genus and species).

All specimens of staphylinids were identified to species based on morphological characters by a member in the group (PAVB), using as a reference the Entomoteca Dalberto Teixeira Pombo, at University of the Azores, Terceira, Portugal. Subsequently these were reconfirmed by a Staphylinidae systematics expert (Volker Assing), using dissecting techniques. A list of species can be found in table S1 (supplement 1).

### DNA extraction

Single specimens preserved in ethanol were allowed to air dry prior to DNA extraction using the QIAamp® DNA microkit (QIAGEN Inc., Chatsworth, CA). The nearly complete Cytochrome Oxidase (COI) gene (1265 bp) was amplified using primers indicated in table S2 (supplement 1) and Illustra PuReTaq™ Ready-To-Go PCR™ beads (GE, Fairfield, CT) using the following protocol: 95 °C 2 min followed by (95 °C 30 sec, 50 °C 30 sec, 72 °C 2 min) for 40 cycles. PCR amplifications were verified by gel electrophoresis using 1% agarose gel with 1 µl GelGreen™ Nucleic Acid Gel Stain, 10,000X (Biotium Inc., Hayward, CA). If multiple non-specific bands were present, PCR products were cleaned by excising the target length band from the gel using a sterile scalpel and cleaned with the Zymoclean™ Gel DNA recovery kit (Zymo Research, Irvine, CA). If only a single band was present, PCR reactions were cleaned using QIAquick PCR purification Kit (QIAGEN, Valencia, CA) according to the manufacturer's protocol. PCR reactions were carried out in a DNA Engine Peltier thermal cycler (Bio-Rad, Hercules, CA). PCR products were purified using the QIAquick PCR purification kit (QIAGEN, Valencia, CA) according to the manufacturer's protocol. PCR products were measured using a Nanodrop (Thermo Scientific, Hudson, NH). Five ng of DNA per 100 bp was used in each 20 µl sequencing reaction, as well as 1 µl of primer and a mixture of 2:1:1 BigDye Terminator v3.1; dGTP BigDye Terminator v3.0; Big Dye Terminator v1.1, and v3.1 5X sequencing buffer (Life Technologies, Carlsbad, CA). The following sequencing protocol was used: 95º C 2 min followed by (98º C for 10 s, 50º C for 5 s, 60º C for 4 min) for 30 cycles. Sequencing products were cleaned using PERFORMA Ultra 96-well Plate (Edge Bio, Gaithersburg, MD) and run on a ABI 3730 automated capillary Sequencer (Life Technologies, Carlsbad, CA) at the Keck Center for Comparative and Functional Genomics, University of Illinois at Urbana-Champaign, USA. Raw sequence data were edited using Sequencher™ 4.7 (Gene Codes Corporation, Ann Arbor, MI).

Sequences were aligned using a combination of algorithmic (Clustal W software) and manual alignment, and subsequently analyzed using PAUP 4.0b10 (Swofford, 2002). Start codon positions were determined using the mitochondrial genome of *Drosophila yakuba* Burla (Genbank accession number NC001322) as a reference (Clary and Wolstenholme, 1985). Differences in the genotype of individuals were determined with pairwise distances in PAUP (table S3, supplement 1). Haplotypes were determined for each species between islands and between habitats within island (table S4, supplement 1).

### Genetic analysis

The COI gene was sequenced for a total of 142 individuals in subfamilies and tribes of the four informal groups of Staphylinidae *sensu* Lawrence and Newton (1995), found in the Azores. The COI sequences obtained from Azorean specimens were aligned with those of other staphylinid species in Genbank. The final data set of 275 individuals is comprised of: 54 species in 13 tribes of the Tachyporine group; 57 species in 7 tribes of the Staphylinine group; 23 species in 2 tribes of the Omaliine group; and 14 species in 2 tribes of the Oxyteline group. The complete list of the 275 individuals used in the genetic analysis is given in table S5 (supplement 1).

We evaluated the saturation level of the nucleotide sequences for the four Staphylinidae groups individually using the Akaike information criterion implemented in MrBayes. Graphic representations were obtained by plotting nucleotide corrected pairwise distances, obtained in PAUP (Swofford, 2003), using the best substitution model identified in jModeltest v0.1.1 (Posada 2008; Guindon and Gascuel, 2003). Maximum Likelihood was then used to measure pairwise genetic distances between nucleotide sequences and plotted against uncorrected pairwise distances. Saturation was assessed as deviation from a linear unsaturated data set as established by Jeffroy *et al.* (2006). In addition, the nucleotide data sets for each Staphylinidae group *sensu* Lawrence and Newton (1995) was partitioned by codon position and saturation estimated independently for each codon position during the analyses. Four Markov chains with a temperature of 0.5 °C were run for 30 million generations. Samples were taken every 1000 generations. The first 15000 trees produced were discarded during burn in.

We inferred the taxonomic relationship of taxa in the four groups of Staphylinidae found in the Azores, based on their COI genetic profile, and published data from species found across the globe, using a Neighbor Joining (NJ) analysis as implemented in PAUP version 4.0a147. The outgroup taxa was selected based on the taxonomical proximity to all tribes included in the analysis. We chose a species in the superfamily Staphylinoidea (Coleoptera, infraorder Staphyliniformia), in which the family Staphylinidae is included, (see table S5, supplement 1). The output tree was visualized using FigTree v1.3.1 (Rambaut, 2009).

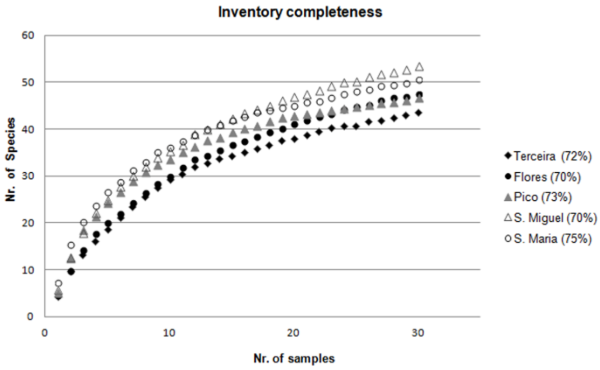

**Figure 1.** Species rarefaction curves and inventory completeness (%) for each island based on the number of species collected (Jackknife richness estimator).

## Results

Inventory completeness for Staphylinidae collected during this project in the Azores archipelago reached 70-75%, depending on the island sampled (figure 1). A total of 38 species were found (table S1, supplement 1). The distribution of species richness across islands, and within habitat types sampled, reflect anthropogenic influence, ranging from three species in the less anthropogenic impacted habitats to 18 species in the more influenced habitats (figure S1, supplement 1). Disparities were more evident in the degree of abundance, ranging from eight specimens in less anthropogenic influenced habitats, to 260 in the most anthropogenic influenced habitats (figure S1, supplement 1). Species turnover was more evident in highly anthropogenically influenced habitats such as corn is all the islands sampled except S. Miguel and invasive forest in Flores Island. With few exceptions, habitat types with high anthropogenic influence, i.e. corn, intensive pasture and orchards, presented higher abundance when compared with less anthropogenically influenced habitats such as pristine forests, semi natural pastures and meadows. Santa Maria and Pico had the highest abundance of staphylinids (figure S1, supplement 1). Geologically, these are the oldest and youngest islands of the archipelago, respectively.

In order to discriminate species diversity at a finer resolution as well as to test the level of genetic diversity at the population level we obtained COI mitochondrial DNA sequences for all morphospecies represented by more than three specimens per locality. In total we sequenced the COI gene for 142 staphylinids. Specimens sequenced belonged to the four informal Staphylinidae groups *sensu* Lawrence and Newton (1995) i.e., Tachyporine, Staphylinine, Omaliine and Oxyteline groups. The nucleotide dataset showed high levels of saturation (figures S2 and S3, supplement 1), therefore, to avoid biased inferences, all analyses were conducted using the corresponding amino acid sequences. Within the taxonomically assigned putative species, amino acid sequence divergence (table S3, supplement 1) for Tachyporine, ranged between 0% [*Atheta fungi* (Gravenhorst)] and 10.3% (*Oligota pumilio* Kiesenwetter) while across species it was 17.8%; for Staphylinine we observed 0-0.25% within species [*Quedius simplicifrons* Fairmaire, *Rugilus orbiculatus orbiculatus* (Paykull)] and 13.06% across species; Oxyteline species divergences varied from 0.8% [*Anotylus nitidifrons* (Wollaston)] and 15.57% (*Oxytelus sculptus* Gravenhorst) within species, and 17.9% across species; Omaliine divergence within species was 3.6% (*Phloeonomus punctipennis* Thomson) and 14.2% across species. The invasive *A. fungi* was the most widespread and abundant species across all islands (figure S4, supplement 1). Analysis of COI sequences for this species shows no geographic differentiation, as none of the haplotypes were found to be exclusive to a given island (figure 2 and table S4, supplement 1). The majority of haplotypes were disseminated across habitats and islands sampled, showing amino acid level pair-wise divergences ranging from 0.2%-5.7%. The second most widespread species, the invasive *A. nitidifrons*, was found in five islands and six of the eight habitats, and had four haplotypes differing from 0.16%-0.87% at the amino acid level. This species was followed by the native *R. orbiculatus orbiculatus* (five islands, four habitats, two haplotypes, 0.24% divergence) and the native *O. pumilio* (four islands, three habitats, two haplotypes, 10.3% divergence).

The NJ analysis of the COI gene, for the complete Staphylinidae dataset, shows one large core group of genera for three informal Staphylinidae groups *sensu* Lawrence and Newton (1995), i.e., Oxyteline, Staphylininae and Tachyporinae (figure 2) (figure S5, supplement 2 for high resolution imaging). Clades of these major core groups of genera seldom included genera from other groups. The Omaliine group does not show a large core group of taxa, but several smaller groups dispersed across the middle and base of the tree.

## Discussion

Staphylinidae in the Azores presented an evenly distributed pattern of species richness across islands, indicative of low habitat fidelity and strong dispersal capabilities. This was not the case for the different habitats in the gradients of anthropogenic influence, where species richness and abundance increased as human impact rose. This pattern could be a consequence of sampling

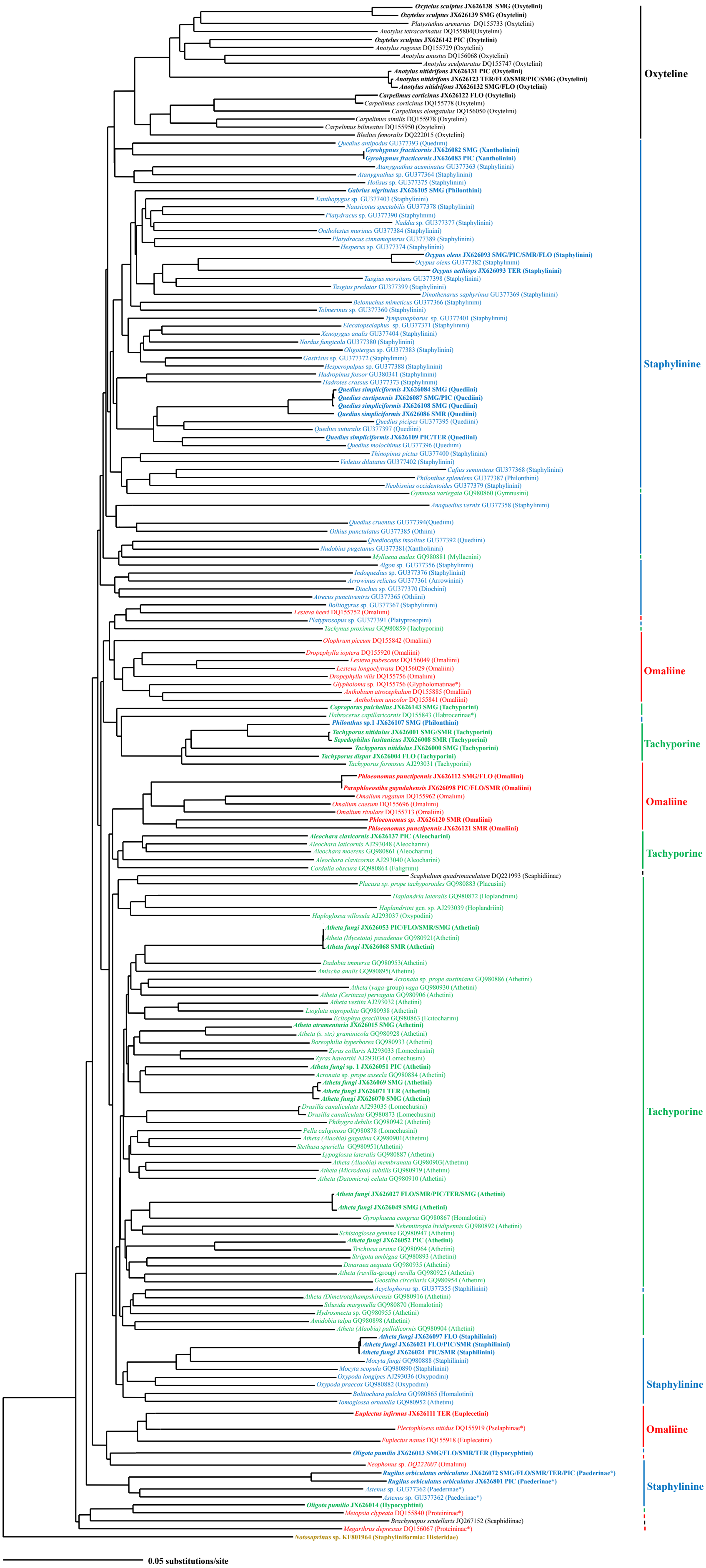

**Figure 2.** Neighbor Joining tree for Staphylinidae species in subfamilies and tribes of the four informal groups of Staphylinidae *sensu* Lawrence and Newton (1995), found in the Azores and worldwide, for which COI sequences were generated (1265 bp) or available in Genbank, respectively. Staphylinidae groups coloured as follows: Tachyporine (green), Omaliine (red), Oxyteline (black) and Staphylinine (brown). Outgroup was coloured in brown. Taxa are followed by their Genbank accession number, tribe or subfamily (*) when tribe was not available in Genbank. COI sequences for taxa in bold were generated in this study. Island of origin for each species is specified as follows: SMG - Sao Miguel; TER - Terceira; SMR - Santa Maria; FLO - Flores; PIC - Pico.

A high-resolution figure is available in the supplemental material (figure S5 supplement 2).

in artificially nutrient enriched habitats composed of depauperate communities (i.e., low β diversity and high species turnover) such as intensive pasture land, corn fields and orchards which were dominated by the most tolerant and competitive introduced arthropod species (data not shown). Although natural and anthropogenically impacted habitats seemed close in species richness, they were in fact very distinct since introduced cosmopolitan species did not contribute to the unique species pool characteristic of a given community assemblage, such as the one in pristine forests, meadows and semi natural pastures. These findings concur with previous reports that also found a broad dissemination of generalist species across the Azorean islands and the existence of few single island endemics (Borges *et al*., 2010a). Similar results have also been shown for populations of highly vagile insects in central Europe (Černá *et al*., 2013). Landscape fragmentation may be of crucial importance for the success of some species and of less consequence to highly vagile insects that can easily move across a fragmented matrix. Different habitats may act as stepping stones for dispersal across a highly fragmented landscape (Finn *et al*., 2007; Louy *et al*., 2007; Prevedello and Vieira, 2010; Bossart and Antwi, 2013). This may be the case for staphylinids in the Azores, which do not seem to have specific macrohabitat requirements and are ubiquitously distributed across the fragmented matrices, particularly in more anthropogenic impacted habitats (both in abundance and number of species). Moreover, the strong influence of human activity in the region suggests that passive dispersal may be a common bridge for inter-island colonization in anthropophilic staphylinids. This has been the case for the Japanese beetle *Popillia japonica* Newman in the Azores (Martins and Simões, 1985). The low habitat fidelity and strong dispersal capabilities of Staphylinidae in the Azores was also supported by genetic data. Human induced landscape fragmentation, as well as geographic distance between habitat patches and islands, was not correlated with genetic differentiation and isolation. Genetic diversity among Staphylinidae populations, subjected to geographic and anthropogenically influenced barriers, does not appear affected by isolation (in opposition to species richness and abundance), as haplotypes represented with multiple specimens were, as a rule, not exclusive to a given island. This contradicts the expected decrease in genetic connectivity with increasing geographic distance (Wright, 1943; Slatkin, 1993). The absence of an effect of landscape fragmentation on genetic isolation has been reported in other animals (Sumner *et al*., 2004; Banks *et al*., 2005; Richmond *et al*., 2009; Dixo *et al*., 2009).

We compared the Staphylinidae taxa obtained from the Azores archipelago to taxa present in Genbank based on a large dataset of complete and partial mtDNA COI (1265 bp) sequences, using Neighbor Joining analysis. As previous studies have attested with analysis at the group or tribe level (Maus *et al*., 2001; Thomas, 2009; Chatzimanolis *et al*., 2010; Elven *et al*., 2010; 2012; McKenna *et al*., 2015), our results suggest that the relationships for species and tribes of Staphylinidae are unresolved. The Tachyporine and Oxyteline are the Staphylinidae groups *sensu* Lawrence and Newton (1995) in which taxonomical affinities seem more robust. Only seldom we found genera interspersed among other groups. Conversely, the Omaliine and to a smaller extent the Staphylinine, showed minor groups and isolated taxa distributed among other Staphylinidae groups. This is not surprising since taxonomic classifications of rove beetles are largely based on poorly understood anatomical and morphological characters (Maus *et al*., 2001). The taxonomic and phylogenetic relationships among tribes have been reported as extremely challenging (Anderson and Ashe, 2000; Ashe, 2005; Beutel and Molenda, 1997; Caterino and Vogler, 2005; Chatzimanolis *et al*., 2010; Elven *et al*., 2010; Gutiérrez-Chacón *et al*., 2009; Hansen, 1997; Thayer, 2005). The recognition of non-monophyletic tribes and phylogenetic arrangements has been extensively reported in molecular analyses focusing on particular subfamilies or tribes of Staphylinidae, namely the Tachyporine and Staphylinine groups (Ashe, 2005; Elven *et al*., 2010; 2012; Osswald *et al*., 2013; Maus *et al*., 2001; Thomas, 2009; Chatzimanolis *et al*., 2010), as well as the Staphyliniformia (Caterino *et al*., 2005). Moreover, difficulties arising from the high level of genetic divergence and nucleotide saturation in closely related species of this study are documented here. These latter findings corroborate the known disparities between currently defined taxonomic units and the existing molecular phylogenies of Staphylinidae.

The *A. fungi* complex of species suggests that cryptic speciation in Staphylinidae is extensive in the Azores, and probably beyond this region (Volker Assing personal communication), and has remained undetected due to the lack of taxonomic studies. Cryptic speciation has been documented in many biological groups (Bickford *et al*., 2006). Low species identification resolution and underestimates of species number in the Azores have been previously described as a probable cause for the presumably lower biota diversity found in the Azores when compared with other Macaronesian archipelagos (Borges *et al*., 2008a; 2010a; Arechavaleta *et al*., 2010; Schaefer *et al*., 2011a; Amorim *et al.,* 2012), Lack of detailed taxonomic work also can explain the disparities between current assigned taxonomic units and the genetic profiles of Staphylinidae taxa. A work integrating morphological and molecular data, across the four Staphylinidae lineages, would help to further resolve the intricate taxonomic relationships of the family, most probably rearranging tribes and species into a more cohesive classification.

In this study we also determined the effect of anthropogenic influence, landscape fragmentation and isolation, in the distribution of species richness and the genetic variability of rove beetles, collected from an isolated archipelago, the Azores, which has lost more than 95% of its native vegetation in less than 600 years. Using a traditional measure of biodiversity, i.e. number of species, coupled with genetic diversity, a measure not commonly used in community ecology studies, we observed that Staphylinidae in the Azores present low habitat fidelity and strong dissemination capabilities, independent of landscape fragmentation. It is likely that the unresolved taxonomic status of a significant proportion of staphylinid taxa has contributed to underestimating the diversity of members of this group in the Azores. This study gives evidence for the existence of cryptic speciation among some taxa indicating the need for further morphological and molecular studies to be able to elucidate the biodiversity of rove beetles in the Azores.

Our study indicates that anthropogenic influence in different habitats can contribute to the increase in species richness and abundance of a few opportunistic species, with limited genetic variability, possibly compromising the viability of less competitive and unique species. Retaining genetic variability at the population level is crucial for species resilience in habitats that are continually and drastically altered due to anthropogenic and/or climatic impacts. This research underscores that more attention should be given to the long-term effects of these impacts on species richness. Oceanic islands continue, since Darwin and Wallace, to provide insights into the mechanisms that promote biodiversity, as well as the effects of anthropogenic action.

## Acknowledgements

We would like to thank Roberto Resendes and João Moniz for field assistance, and Betty Ujhelyi, Massimo Pessino and Therese Anne Catanach for technical support. We also thank Volker Assing for taxonomic identification of some specimens. This work was supported by a grant from the Fundo Regional Ciência e Tecnologia, Regional Government of the Azores and PROEMPREGO and the research project "PROBIO, Profiling Reliable Organisms as Bioindicators: an integrated approach for island systems" financed by FLAD (Fundação Luso-Americana para o Desenvolvimento). This study was also financed by Portuguese National Funds, through FCT – Fundação para a Ciência e a Tecnologia, within the project UID/BIA/00329/2013.

## References

Amorim I. R., Emerson B. C., Borges P. A. V., Wayne R. K., 2012.- Phylogeography and molecular phylogeny of Macaronesian *Tarphius* (Coleoptera: Zopheridae): why so few species in the Azores?.- *Journal of Biogeography*, 39: 1583-1595.

Anderson R. S., Ashe J. S., 2000.- Leaf litter inhabiting beetles as surrogates for establishing priorities for conservation of selected tropical montane cloud forests in Honduras, Central America (Coleoptera; Staphylinidae, Curculionidae).- *Biodiversity and Conservation*, 9: 617-653.

Andersen A., Eltun R., 1999.- Long-term developments in the carabid and staphylinid (Col., Carabidae and Staphylinidae) fauna during conversion from conventional to biological farming.- *Journal of Applied Entomology*, 124: 51-56.

Arechavaleta M., Rodríguez S., Zurita N., Garcia A., 2010.- *Lista de especies silvestres de Canarias. Hongos, plantas y animales terrestres*.- Gobierno de Canarias, Spain.

Ashe J. S., 2005.- Phylogeny of the tachyporine group subfamilies and 'basal' lineages of the Aleocharinae (Coleoptera: Staphylinidae) based on larval and adult characteristics.- *Systematic Entomology*, 30: 3-37.

Assing V., 2002.- On the Staphylinidae of the Canary Islands. IX. New synonyms and records, and asystematic rearrangement of some endogean and cavernicolous Aleocharinae (Coleoptera).- *Vieraea*, 30: 45-66.

Banks S. C., Lindenmayer D. B., Ward S. J., Taylor A. C., 2005.- The effects of habitat fragmentation via forestry plantation establishment on spatial genotypic structure in the small marsupial carnivore, *Antechinus agilis*.- *Molecular Ecology*, 14: 1667-1680.

Basset Y., Mavoungou J. F., Mikissa J. B., Missa O., Miller S. E., Kitching R. L., Alonso A., 2004.- Discriminatory power of different arthropod data sets for the biological monitoring of anthropogenic disturbance in tropical forests.- *Biodiversity and Conservation*,13: 709-732.

Beutel R. G., Molenda R., 1997.- Comparative morphology of larvae of Staphylinoidea (Coleoptera, Polyphaga) with phylogenetic implications.- *Zoologischer Anzeiger*, 236: 37-67.

Bickford D., Lohman D. J., Sodhi N. S., Ng P. K., Meier R., Winker K., Ingram K. K., Das I., 2006.- Cryptic species as a window on diversity and conservation.- *Trends in Ecology and Evolution*, 22: 148-155.

Bohac J., 1999.- Staphylinid beetles as bioindicators.- *Agriculture, Ecosystems and Environment*, 74: 357-372.

Borges P. A. V., Hortal J., 2009.- Time, area and isolation: Factors driving the diversification of Azorean arthropods.- *Journal of Biogeography*, 36: 178-191.

Borges P. A. V., Abreu C., Aguiar A. M. F., Carvalho P., Jardim R., Melo I., Oliveira P., Sérgio C., Vieira P., Serrano A. R. M., 2008.- *A list of the terrestrial fungi, flora and fauna of Madeira and Selvagens archipelagos*.- Direcção Regional do Ambiente da Madeira and Universidade dos Açores, Funchal and Angra do Heroísmo, Portugal.

Borges P. A. V., Costa A., Cunha R., Gabriel R., Gonçalves V., Martins A. F., Melo I., Parente M., Raposeiro P., Rodrigues P., Santos R. S., Silva L., Vieira V., 2010a.- *A list of the terrestrial and marine biota from the Azores*.- Princípia, Cascais, Portugal.

Borges P. A. V., Costa A., Cunha R., Gabriel R., Gonçalves V., Martins A. F., Melo I., Parente M., Raposeiro P., Rodrigues P., Santos R. S., Silva L., Vieira V., Vieira P., Vieira V., Mendonça E., Boeiro M., 2010b.- Description of the terrestrial and marine biodiversity of the Azores, pp. 9-33. In: *A list of the terrestrial and marine biota from the Azores* (Borges P. A. V., Costa A., Cunha R., Gabriel R., Gonçalves V., Martins A. F., Melo I., Parente M., Raposeiro P., Rodrigues P., Santos R. S., Silva L., Vieira V., Eds).- Princípia, Cascais, Portugal.

Bossart J. L., Antwi J. B., 2013.- Species-specific traits predict genetic structure but not genetic diversity of three fragmented Afrotropical forest butterfly species.- *Conservation Genetics*, 14: 511-528.

Brook B. W., Sodhi N. S., Bradshaw C. J., 2008.- Synergies among extinction drivers under global change.- *Trends in Ecology and Evolution*, 23: 453-60.

Bush J. W. M., Prakash M., Hu D. L., 2007.- The integument of water- walking arthropods: form and function.- *Advances in Insect Physiology*, 34: 117-192.

Byers R. A., Barker G. M., Davidson R. L., Hoebeke E. R., Sanderson M. A., 2000.- Richness and abundance of Carabidae and Staphylinidae (Coleoptera) in northeastern dairy pastures under intensive grazing.- *Great Lakes Entomologist*, 33: 81-105.

Caballero U., León-Cortés J. L., Morón-Ríos A., 2009.- Response of rove beetles (Staphylinidae) to various habitat types and change in Southern Mexico.- *Journal of Insect Conservation*, 13: 67-75.

Cardoso P., Lobo J. M., Aranda S. C., Dinis F., Gaspar C., Borges P. A. V., 2009.- A spatial scale assessment of habitat effects on arthropod communities of an oceanic island.- *Acta Oecologica*, 35: 590-597.

Cardoso P., Borges P. A. V., Dinis F., Gaspar C., 2010.- Patterns of alpha and beta diversity of epigean arthropods at contrasting land-uses of an oceanic island (Terceira, Azores), pp.73-88. In: *Terrestrial arthropods of Macaronesia - biodiversity, ecology and evolution* (Serrano A. R. M., Borges P. A. V., Boieiro M., Oromí P., Eds).- Sociedade Portuguesa de Entomologia, Lisboa, Portugal.

Carine M., Schaefer H., 2010.- The Azorean diversity enigma: why are there so few Azorean endemic flowering plants and why are they so widespread?- *Journal of Biogeography*, 37: 77-89.

Carlson C. S., Thomas D. J., Eberle M. A., Swanson J. E., Livingston R. J., Rieder M. J., 2005.- Genomic regions exhibiting positive selection identified from dense genotype data.- *Genome Research*, 15: 1553-1565.

Caterino M. S., Hunt T., Vogler A. P., 2005.- On the constitution and phylogeny of Staphyliniformia.- *Molecular Phylogenetics and Evolution*, 34: 655-672.

Černá K., Straka J., Munclinger P., 2013.- Population structure of pioneer specialist solitary bee *Andrena vaga* (Hymenoptera: Andrenidae) in central Europe: the effect of habitat fragmentation or evolutionary history?.- *Conservation Genetics*, 14: 875-883.

Chatzimanolis S., Cohen I. M., Schomann A., Solodovnikov A., 2010.- Molecular phylogeny of the mega-diverse rove beetle tribe Staphylinini (Insecta, Coleoptera, Staphylinidae).- *Zoologica Scripta*, 39: 436-449.

Clary D. O., Wolstenholme D. R., 1985.- The mitochondrial DNA molecule of *Drosophila yakuba*: nucleotide sequence, gene organization, and genetic code.- *Journal of Molecular Evolution*, 22: 252-271.

Clement M., Posada D., Crandall K. A., 2000.- TCS: a computer program to estimate gene genealogies.- *Molecular Ecology*, 9: 1657-1660.

Colwell R. K., 2011.- *EstimateS, Version 8.2: statistical estimation of species richness and shared species from samples*.- Software and user's guide.

Dayrat B., Conrad M., Balayan S., White T. R., Albrecht C., Golding R., Gomes S. R., Harasewych M. G., Martins A. M. F., 2011.- Phylogenetic relationships and evolution of pulmonate gastropods (Mollusca): new insights from increased taxon sampling.- *Molecular Phylogenetics and Evolution*, 59: 425-437.

Dennis P., Young M. R., Howard C. L., Gordon I. J., 1997.- The response of epigeal beetles (Col.: Carabidae, Staphylinidae) to varied grazing regimes on upland *Nardus stricta* grasslands.- *Journal of Applied Ecology*, 34: 433-443.

Dias E., Elias R. B., Melo C., Mendes C., 2007.- Biology and ecology of the forests of the islands. Azores. Trees and forests of Portugal, Azores and Madeira, pp. 51-80. In: *A floresta das ilhas* (Silva L., Ed.).- Público, Comunicação Social, S.A. and Fundação Luso-Americana, Lisbon, Portugal. (In Portuguese).

Dixo M., Metzger J. P., Morgante J. S., Zamudio K. R., 2009.- Habitat fragmentation reduces genetic diversity and connectivity among toad populations in the Brazilian Atlantic Coastal Forest.- *Biological Conservation*, 142: 1560-1569.

Elven H., Bachmann L., Gusarov V. I., 2010.- Phylogeny of the tribe Athetini (Coleoptera: Staphylinidae) inferred from mitochondrial and nuclear sequence data.- *Molecular Phylogenetics and Evolution*, 57: 84-100.

Elven H., Bachmann L., Gusarov V. I., 2012.- Molecular phylogeny of the Athetini-Lomechusini-Ecitocharini clade of Aleocharine rove beetles (Insecta).- *Zoologica Scripta*, 41: 617-636.

Finn D. S., Blouin M. S., Lytle D. A., 2007.- Population genetic structure reveals terrestrial affinities for a headwater stream insect.- *Freshwater Biology*, 52: 1881-1897.

Gillespie R. G., Roderick G. K., 2002.- Arthropods on islands: colonization, speciation, and conservation.- *Annual Review of Entomology*, 47: 595-632.

Grimaldi D., Engel M. S., 2005.- *Evolution of the insects*.- Cambridge University Press, Cambridge, UK.

Guindon S., Gascuel O., 2003.- A simple, fast, and accurate algorith to estimate large phylogenies by maximum likelihood.- *Systematic Biology*, 52: 696-704.

Gutiérrez-Chacón C., Zúñiga M. C., Van Bodegom P. M., Chará J., Giraldo L., 2009.- Rove beetles (Coleoptera: Staphylinidae) in Neotropical riverine landscapes: characterising their distribution.- *Insect Conservation and Diversity*, 2: 106-115.

Hansen M., 1997.- *Phylogeny and classification of the staphyliniform beetle families. (Coleoptera)*.- Biologiske Skrifter 48, Kongelige Danske Videnskabernes Selskab, Copenhagen, Denmark.

Heidinger I. M. M., Hein S., Feldhaar H., Poethke H.-J., 2013.- The genetic structure of populations of *Metrioptera bicolor* in a spatially structured landscape: effects of dispersal barriers and geographic distance.- *Conservation Genetics*, 14: 299-311.

Hillis D. M., Dixon M. T., 1991.- Ribosomal DNA: molecular evolution and phylogenetic inference.- *The Quaterly Review of Biology*, 66: 411-453.

Hodkinson I. D., Jackson J. K., 2005.- Terrestrial and aquatic invertebrates as bioindicators for environmental monitoring, with particular preference to mountain ecosystems.- *Environmental Management*, 35: 649-666.

Holland B. S., Hadfield M. G., 2002.- Islands within an island: phylogeography and conservation genetics of the endangered Hawaiian tree snail *Achatinella mustelina*.- *Molecular Ecology*, 11: 365-375.

Hortal J., Borges P. A. V., Gaspar C., 2006.- Evaluating the performance of species richness estimators: Sensitivity to sample grain size.- *Journal of Animal Ecology*, 75: 274-287.

Howland D. E., Hewitt G. M., 1995.- Phylogeny of the Coleoptera based on mitochondrial cytochrome oxidase I sequence data.- *Insect Molecular Biology*, 3: 203-215.

Jeffroy O., Brinkmann H., Delsuc F., Philippe H., 2006.- Phylogenomics: the beginning of incongruence?.- *Trends in Genetics*, 22: 225-231.

Jordaens K., van Riel P., Martins A. M. F., Backeljau T., 2009.- Speciation on the Azores islands: congruent patterns in shell morphology, genital anatomy, and molecular markers in endemic land snails (Gastropoda, Leptaxinae).- *Biological Journal of the Linnean Society*, 97: 166-176.

Klimaszewski J., Winchester N. N., Bolte K. B., 2002.- Aleocharine Rove Beetles (Coleoptera, Staphylinidae) of the Ancient Sitka Spruce Forest on Vancouver Island, British Columbia, Canada.- *Canadian Entomologist*, 135: 867-868.

Krooss S., Schaefer M., 1998.- The effect of different farming systems on epigeic arthropods: a five-year study on the rove beetle fauna (Coleoptera: Staphylinidae) of winter wheat.- *Agriculture, Ecosystems and Environment*, 69: 121-133.

Laliberté E., Wells J. A., DeClerck F., Metcalfe D. J., Catterall C. P., Queiroz C., Aubin I., Bonser S. P., Ding Y., Fraterrigo J. M., McNamara S., Morgan J. W., Merlos D. S., Vesk P. A., Mayfield M. M., 2010.- Land-use intensification reduces functional redundancy and response diversity in plant communities.- *Ecological Letters*, 13: 76-86.

Lawrence J. F., Newton A. F., 1995.- Families and subfamilies of Coleoptera (with selected genera, notes, references and data on family-group names), pp 779-1006. In: *Biology, phylogeny and classification of Coleoptera: Papers celebrating the 80th birthday of Roy A. Crowson* (Pakaluk J., Slipinski S. A., Eds).- Muzeum i Instytut Zoologii PAN, Warszawa, Poland.

LIBRADO P., ROZAS J., 2009.- DnaSP v. 5: a software for comprehensive analysis of DNA polymorphism data.- *Bioinformatics*, 25: 1451-1452.

LOUY D., HABEL J. C., SCHMITT T., ASSMANN T., MEYER M., MULLER P., 2007.- Strongly diverging population genetic patterns of three skipper species: the role of habitat fragmentation and dispersal ability.- *Conservation Genetics*, 8: 671-681.

MANEL S., JOOST S., EPPERSON B. K., HOLDEREGGER R., STORFER A., ROSENBERG M. S., SCRIBNER K. T., BONIN A., FORTIN M. J., 2010.- Perspectives on the use of landscape genetics to detect genetic adaptive variation in the field.- *Molecular Ecology*, 19: 3760-3772.

MARCELINO J. A. P. , SILVA L., GARCIA P. V., WEBER E., SOARES A. O., 2013.- Using species spectra to evaluate plant community conservation value along a gradient of anthropogenic disturbance.- *Environmental Monitoring and Assessment*, 185: 6221-6233.

MARTÍN J. L., BORGES P. A.V., ARECHAVALETA M., FARIA B., 2008.- The top 100 list, pp. 421-449. In: *The 100 threatened species prioritary for management in Macaronesia* (MARTÍN J. L., ARECHAVALETA M., BORGES P. A.V., FARIA B., Eds).- Consejería de Medio Ambiente y Ordenación Territorial, Canary Islands, Spain.

MARTINS A., SIMÕES N., 1985.- Population dynamics of the Japanese beetle (Coleoptera - Scarabaeidae) in Terceira island - Azores.- *Arquipélago*, 6: 57-62.

MARTINS J. M., MOREIRA O. C. B., RAINHA N. F. P., BAPTISTA J. A. B., SILVA L., MOURA M. M. T., 2013.- Population genetics and conservation of the Azorean tree *Picconia azorica*.- *Biochemical Systematics and Ecology*, 49: 135-143.

MAUS C., PESCHKE K., DOBLER S., 2001.- Phylogeny of the genus Aleochara inferred from mitochondrial cytochrome oxidase sequences (Coleoptera: Staphylinidae).- *Molecular Phylogenetics and Evolution*, 18: 202-216.

MCKENNA D. D., FARRELL B. D., CATERINO M. S., FARNUM C. W., HAWKS D. C., MADDISON D. R., SEAGO A. E., SHORT A. E. Z., NEWTON A. F., THAYER M. K., 2015.- Phylogeny and evolution of the Staphyliniformia and Scarabaeiformia: Forest litter as a stepping-stone for diversification of non-phytophagous beetles.- *Systematic Entomology*, 40: 35-60.

MEIJER S. S., WHITTAKER R. J., BORGES P. A. V., 2011.- The effects of land-use change on arthropod richness and abundance on Santa Maria Island (Azores): unmanaged plantations favour endemic beetles.- *Journal of Insect Conservation*, 15: 505-522.

MYERS N., MITTERMEIER R. A., MITTERMEIER C. G., DA FONSECA G. A. B., KENT J., 2000.- Biodiversity hotspots for conservation priorities.- *Nature*, 403: 853-858.

OSSWALD J., BACHMANN L., GUSAROV V. I., 2013.- Molecular phylogeny of the beetle tribe Oxypodini (Coleoptera: Staphylinidae: Aleocharinae).- *Systematic Entomology*, 38: 507-522.

POHL G. R., LANGOR D. W., SPENCE J. R., 2007.- Rove beetles and ground beetles (Coleoptera: Staphylinidae, Carabidae) as indicators of harvest and regeneration practices in western Canadian foothills forests.- *Biological Conservation*, 137: 294-307.

POSADA D., 2008.- jModelTest: phylogenetic model averaging.- *Molecular Biology and Evolution*, 25: 1253-1256.

PREVEDELLO J. A., VIEIRA M. V., 2010.- Does the type of matrix matter? A quantitative review of the evidence.- *Biodiversity and Conservation*, 19: 1205-1223.

RAINIO J., NIEMELA J., 2003.- Ground beetles (Coleoptera: Carabidae) as bioindicators.- *Biodiversity and Conservation*, 12: 487-506.

RAMBAUT A., 2009.- *FigTree*.- version 1.3.1 [online] URL: http://tree.bio.ed.ac.uk/software/figtree/ (Accessed 16 November 2014).

RICHMOND J. Q., REID D. T., ASHTON K. G., ZAMUDIO K. R., 2009.- Delayed genetic effects of habitat fragmentation on the ecologically specialized Florida Sand Skink (Plestiodon reynoldsi).- *Conservation Genetics*, 10: 1281-1297.

RODRIGUES P., LOPES R. J., DROVETSKI S. V., REIS S., RAMOS J. A., CUNHA R. T., 2013.- Phylogeography and genetic diversity of the robin (*Erithacus rubecula*) in the Azores islands: evidence of a recent colonisation.- *Journal of Ornithology*, 154: 889-900.

ROSENBERG M. S., ANDERSON C. D., 2011.- PASSaGE: Pattern analysis, spatial statistics, and geographic exegesis, version 2.- *Methods in Ecology and Evolution*, 2: 229-232.

RUMEU B., CAUJAPÉ-CASTELLS J., BLANCO-PASTOR J. L., JAÉN-MOLINA R., NOGALES M., ELIAS R. B., VARGAS P., 2011.- The colonization history of *Juniperus brevifolia* (Cupressaceae) in the Azores Islands.- *PLoS ONE*, 6: e27697.

SCHAEFER H., MOURA M., MACIEL M. G. B., SILVA L., RUMSEY F. J., CARINE M., 2011a.- The Linnean shortfall in oceanic island biogeography: a case study in the Azores.- *Journal of Biogeography*, 38: 1345-1355.

SCHAEFER H., HARDY O. J., SILVA L., BARRACLOUG T. G., SAVOLAINEN V., 2011b.- Testing Darwin´s naturalization hypothesis in the Azores.- *Ecological Letters*, 14: 389-396.

SEGELBACHER G., CUSHMAN S. A., EPPERSON B. K., FORTIN M.-J., FRANCOIS O., HARDY O. J., HOLDEREGGER R., MANEL S., 2010.- Applications of landscape genetics in conservation biology: concepts and challenges.- *Conservation Genetics*, 11: 375-385.

SILVA L., ELIAS R. B., MOURA M., MEIMBERG H., DIAS E., 2011.- Genetic variability and differentiation among populations of the Azorean endemic gymnosperm *Juniperus brevifolia*: baseline information for a conservation and restoration perspective.- *Biochemical Genetics*, 49: 715-734.

SIMON C., FRATI F., BECKENBACH A., CRESPI B., LIU H., FLOOK P., 1994.- Evolution, weighting, and phylogenetic utility of mitochondrial gene sequences and a compilation of conserved polymerase chain reaction primers.- *Annals of the Entomological Society of America*, 87: 651-701.

SLATKIN M., 1993.- Isolation by distance in equilibrium and non-equilibrium populations.- *Evolution*, 47: 264-279.

SOMMER S., MCDEVITT A. D., BALKENHOL N., 2013.- Landscape genetic approaches in conservation biology and management.- *Conservation Genetics*, 14: 249-251.

SUMNER J., JESSOP T., PAETKAU D., MORITZ C., 2004.- Limited effect of anthropogenic habitat fragmentation on molecular diversity in a rain forest skink, *Gnypetoscincus queenslandiae*.- *Molecular Ecology*, 13: 254-269.

SWOFFORD D. L., 2003.- *PAUP*. Phylogenetic analysis using parsimony (*and other methods)* version 4.- Sinauer Associates, Sunderland, Massachusets, USA.

TAJIMA F., 1989.- The effect of change in population size on DNA polymorphism.- *Genetics*, 123: 597-601.

TAMURA K., PETERSON D., PETERSON N., STECHER G., NEI M., KUMAR S., 2011.- MEGA5: molecular evolutionary genetics analysis using maximum likelihood, evolutionary distance, and maximum parsimony methods.- *Molecular Biology and Evolution*, 28: 2731-2739.

TAUTZ D., HANCOCK J. M., WEBB D. A., TAUTZ C., DOVER G. A., 1988.- Complete sequences of the rRNA genes of *Drosophila melanogaster*.- *Molecular Biology and Evolution*, 5: 366-376.

THAYER M. K., 2005.- Staphylinoidea, Staphylinidae Latreille, 1802. Coleoptera, Vol I. Morphology and Systematics (Archostemata, Adephaga, Myxophaga, Polyphaga partim), pp. 296-344. In: *Handbook of zoology, arthropoda: insecta*, (BEUTEL R. G., LESCHEN R. A. B., KRISTENSEN N. P., Eds).- De Gruyter, Berlin, Germany.

THÉRY T., LESCHEN R. A. B., 2013.- Pselaphinae (Coleoptera: Staphylinidae) of the Three Kings Islands.- *New Zealand Entomologist*, 36: 37-64.

THOMAS J. C., 2009.- A preliminary molecular investigation of Aleocharine phylogeny (Coleoptera: Staphylinidae).- *Annals of the Entomological Society of America*, 102: 189-195.

TRIANTIS K. A., HORTAL J., AMORIM I., CARDOSO P., SANTOS A. M. C., GABRIEL R., BORGES P. A. V., 2012.- Resolving the Azorean knot: a response to Carine and Schaefer 2010.- *Journal of Biogeography*, 39: 1179-1184.

VOGLER A. P., PEARSON D. L., 1996.- A molecular phylogeny of the tiger beetles: congruence of mitochondrial and nuclear rDNA data sets.- *Molecular Phylogenetics and Evolution*, 6: 321-338.

WESTERN D., 2001.- Human-modified ecosystems and future evolution.- *Proceedings of the National Academy of Sciences of the United States of America*, 98: 5458-5465.

WHITTAKER R. J., FERNÁNDEZ-PALACIOS J. M., 2007.- *Island biogeography - ecology, evolution and conservation*.- Oxford University Press, Oxford, UK.

**Authors' addresses:** José A. P. MARCELINO (corresponding author, jmar06@gmail.com), Paulo A. V. BORGES, Patricia V. GARCIA, António O. SOARES, $CE_3C$, Azorean Biodiversity Group, Department of Biology, University of the Azores, rua da Mae de Deus 13-A, 9501-801 Ponta Delgada, Azores, Portugal; Rosanna GIORDANO, Department of Biology, PO Box 23360, University of Puerto Rico, San Juan, PR 00931, USA; Felipe N. SOTO-ADAMES, Department of Entomology, University of Illinois, Urbana, IL, USA

Received November 25, 2014. Accepted February 29, 2016.