insects

Review

Phylogeography of Saproxylic and Forest Floor Invertebrates from Tallaganda, South-eastern Australia

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Abstract: The interaction between physiogeographic landscape context and certain life history characteristics, particularly dispersal ability, can generate predictable outcomes for how species responded to Pleistocene (and earlier) climatic changes. Furthermore, the extent to which impacts of past landscape-level changes 'scale-up' to whole communities has begun to be addressed via comparative phylogeographic analyses of co-distributed species. Here we present an overview of a body of research on flightless low-mobility forest invertebrates, focusing on two springtails and two terrestrial flatworms, from Tallaganda on the Great Dividing Range of south-eastern Australia. These species are distantly-related, and represent contrasting trophic levels (*i.e.*, slime-mold-grazers vs. higher-level predators). However, they share an association with the dead wood (saproxylic) habitat. Spatial patterns of intraspecific genetic diversity partly conform to topography-based divisions that circumscribe five 'microgeographic regions' at Tallaganda. In synthesizing population processes and past events that generated contemporary spatial patterns of genetic diversity in these forest floor invertebrates, we highlight cases of phylogeographic congruence, pseudo-congruence, and incongruence. Finally, we propose conservation-oriented recommendations for the prioritisation of areas for protection.

Supplementary Material

1. Methods

1.1. Data Availability

As part of our review paper, we performed some re-analyses of published datasets. GenBank accession numbers for mitochondrial DNA (mtDNA) sequences from Collembola are given in Garrick *et al.* [1–3], and accessions for flatworm mtDNA sequences can be found in Sunnucks *et al.* [4]. Collembola nuclear genotypes used here in clustering analyses are available on request.
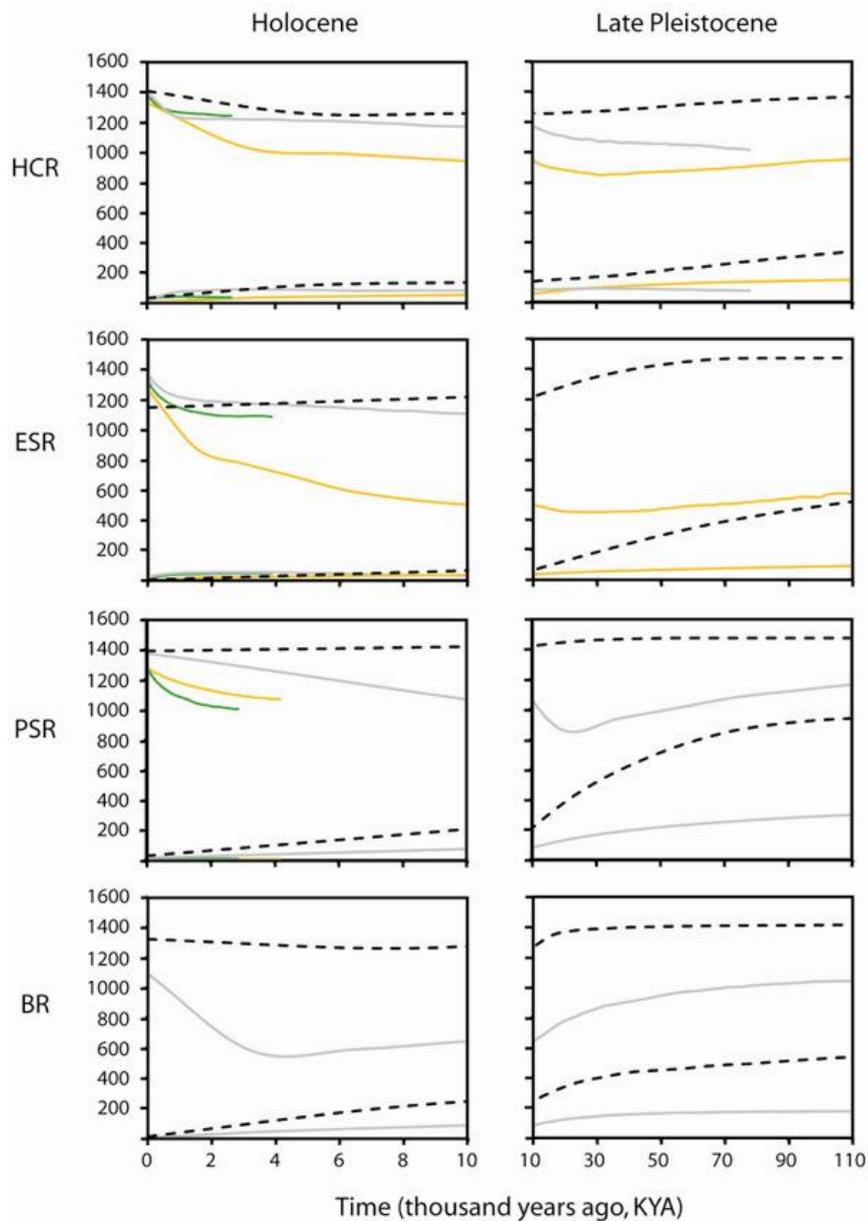
1.2. Genetic Diversity Summary Statistics

To quantify mtDNA sequence polymorphism within populations we used DnaSP v5.10 [5] to calculate the number different haplotypes (N_{hap}), haplotypic diversity (H_d; the probability that two randomly drawn sequences are identical), and the mean sequence divergence among haplotypes, as measured using uncorrected *p*-distances (*p*-dist.). The latter two summary statistics are standardized with respect to sample size, thereby facilitating direct comparison across populations and species.

1.3. Bayesian Skyline Plots

To reconstruct population size changes operating over long-term, evolutionary timescales, we used Bayesian skyline plots [6], implemented in BEAST v1.6.1 [7]. For these analyses, the basic underlying unit is assumed to be a single unstructured population. Putatively panmictic groups of springtails (Pseudachorutinae sp. and *Acanthanura* sp.) were inferred from genotypic clustering of nuclear genetic data. However, only mtDNA data were available for flatworms (*Artioposthia lucasi* and *Caenoplana coerulea*). In the latter cases, it was necessary to use phylogenetic lineages on an estimated gene tree (see Figure 4 of the main text) as basic unit of analysis. We recognize that neither of these approaches excludes the possibility of subtle within-group substructure, and thus results should be interpreted with caution. Searches used the GTR+I+G substitution model, empirical base frequencies, a strict-clock with Brower's [8] standard arthropod mtDNA rate ('clock.rate' = 1.15×10^{-8} with the simplifying assumptions of 1 year generation times for each species), a coalescent (Bayesian skyline) tree prior, linear skyline model, UPGMA-generated starting trees with 10 groups, 'skyline.popsize' with a uniform prior and bounded by 0 and 1.5×10^6 (initial value = 10,000), auto-optimized tuning, and other priors as default. Final searches were performed using 1×10^8 MCMC generations sampling parameters every 2,000[th] step (10% discarded as burn-in), and three replicate runs per dataset. One representative replicate run is presented per dataset, and confidence intervals around the median effective population size (N_e) value in these selected replicates are shown in Supplementary Figure S1.

Figure S1. Confidence intervals associated with Bayesian Skyline plots showing changes in N_e over time, based in mtDNA sequence data (Figure 5 of the main text). Curves represent the upper and lower 95% highest posterior density limits for N_e-values (*y*-axis) plotted over time (*x*-axis). Curve colours approximate colours of the species themselves (Figure 2 of the main text) and are as follows: Pseudachorutinae sp., pale gray; *Acanthanura* sp., black (dashed); *Artioposthia lucasi*, yellow; and *Caenoplana coerulea*, dark green (note that *A. lucasi* population PSR corresponds with PSR-1 in Figure 3 of the main text). Note that for a given population, Holocene *vs*. Late Pleistocene represents the same analysis (rescaled).

1.4. Contact Zone Dynamics

Here we focused on two previously identified locations at Tallaganda in which both springtail species show abrupt spatial transitions between genetic populations: the Eastern Slopes / Pikes Saddle

contact zone, and the Pikes Saddle / Badja contact zone [1–3]. For each species, we selected the subset of individuals that were collected at or near these zones. To reduce the impact of kin clustering on inferences about contact zone dynamics, in rotting logs for which ≥5 individuals were sampled, we used KINGROUP v2 [9] to identify putative full siblings. If detected, we retained one randomly selected representative of each family group. We then analysed these reduced datasets using STRUCTURE v2.2.3 [10] with the number of clusters (*K*) fixed at two. Searches used the 'correlated allele frequency' and 'admixture ancestry' models, 1×10^5 MCMC generations burn-in and a run length of 1×10^6 generations. STRUCTURE membership coefficients (*Q*-values) were used to classify an individual's ancestry as purebred (Q ≥ 0.9), or hybrid (Q < 0.9), with hybrids being indicative of ongoing *gene flow* across the contact zone. Similarly, the presence of first-generation migrants—purebred individuals sampled at a spatial location that is characteristic of other genetic population's native range—is indicative of successful *dispersal* across the contact zone.

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