MOLECULAR ECOLOGY

Molecular Ecology (2011) **20**, 4385–4387 doi: 10.1111/j.1365-294X.2011.05263.x

NEWS AND VIEWS

PERSPECTIVE

Small is the new big: assessing the population structure of microorganisms

ALEEZA C. GERSTEIN and JEAN-SÉBASTIEN MOORE

Department of Zoology and Beaty Biodiversity Research Centre, The University of British Columbia, 6270 University Boulevard, Vancouver, BC, Canada V6T 1Z4

Microorganisms are a tremendously large and diverse group spanning multiple kingdoms, yet they have been considerably under-studied by ecologists and evolutionary biologists compared to their larger relatives. Although a few microbial species have become the stars of laboratory experiments, relatively few studies have examined microbial species in their natural habitats. As such, the question of whether microbial diversity parallels that of larger bodied species is contentious (Lachance 2004; Fenchel & Finlay 2004). It has been suggested that large population sizes, high dispersal potential and low extinction rates lead to genetically homogeneous populations of microbial species over large geographical scales—arguments that bring to mind discussions about speciation and population structure in the marine environment. In this issue of *Molecular Ecology*, Herrera *et al.* (2011) add to this debate by examining 91 isolates of the flower-living yeast *Metschnikowia gruessii* from southeastern Spain. Their AFLP results support both spatial structuring of genetic diversity across the region, as well as microsite-dependent diversifying selection within single flowers. This study adds to a growing body of literature suggesting that although microbes have much larger population sizes and many differ in their principal mode of reproduction (primarily clonal rather than sexual), patterns of genetic diversity and phylogenetic structure for some microbial species may be similar to that of larger species. This study highlights the need for vastly more research that specifically examines biogeographic structure in this under-utilized group of organisms.

Received 7 July 2011; revision received 29 July 2011; accepted 4 August 2011

Microorganisms (species with body length less than 1mm; prokaryotes, unicellular eukaryotes and micrometazoans) form a tremendous part of the biota of the planet. In part owing to difficulty in identification and culturing, these species have received less attention from ecologists and evolutionary biologists relative to macroorganisms. A small number of these species (e.g., *Saccharomyces cerevisiae*, *Pseudomonas fluorescens*, *Chlamydomonas*, *Escherichia coli*) have become the model organisms of choice for laboratory evolutionary biologists, who take advantage of features including fast growth, large population sizes and largely clonal growth. A number of the same attributes that make these species so desirable for laboratory study have led some researchers to suggest that eukaryotic microbial species have a ubiquitous distribution around the world and lack population structure. Because of large population sizes, high dispersal rates and low rates of extinction, it is hypothesized that microbial species do not show the same population structure and biogeographic diversification as larger organisms (Fenchel & Finlay 2004). In particular, species that are predominantly or completely asexual are expected to show a lack of population structure, because high rates of clonal reproduction have been shown theoretically to decrease genetic diversity (Balloux *et al.* 2003; but see Arnaud-Haond *et al.* 2005). Scientists on the other side of the debate, however, have argued that microbes are an incredibly diverse group with variation in size, life history, morphology, behaviour, etc. and that it is premature to assume that all small-bodied species should share distribution properties (Caron 2009; Lowe *et al.* 2005). Indeed, many contend that endemism and vicariant distributions are common in microorganisms and that population structure and allopatric speciation are possible (Lachance 2004).

Readers of Molecular Ecology may remember another group of organisms that were long believed to lack intraspecies genetic discontinuities over large geographical scales: marine organisms with planktonic larvae. This group, much like microbes, enjoys large population sizes and high dispersal potential and should therefore display weak population differentiation (Palumbi 1992). Yet, research into these marine organisms routinely uncovers genetic discontinuities. Many evolutionary mechanisms have been invoked to explain such high levels of differentiation (Palumbi 1994). In particular, data show that high dispersal potential does not always translate into high rates of realized dispersal (e.g., Christie *et al.* 2010) and that heterogeneous natural selection in the marine environment could lead to population differentiation at selected loci, and in some cases in neutral loci (e.g., Colbeck *et al.* 2011).

Interestingly, the study by Herrera *et al.* (2011) suggests that similar mechanisms may be responsible for the observed population structure in *Metschnikowia gruessii*. Clustering analyses performed using STRUCTURE and a nonmodel-based equivalent (K-means) both uncover genetic clusters that appear congruent with environmental differences between sampling sites (Fig. 1a); isolates sam-

Correspondence: Aleeza C. Gerstein, Fax: (604) 822 2416; E-mail: gerstein@zoology.ubc.ca

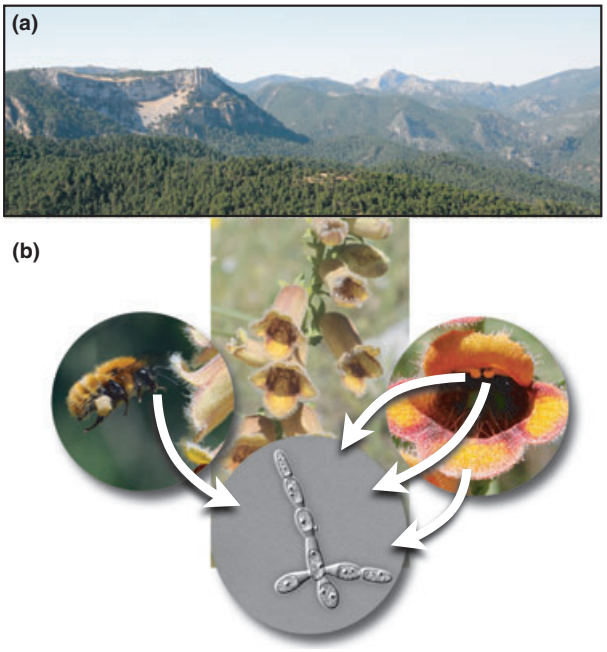

Fig. 1 Population structure of the wild flower-living yeast *Metschnikowia gruessii* at two different spatial scales. (a) Picture showing the area of southeastern Spain where inoculates were collected. The study by Herrera et al. (2011) showed that samples from similar elevations cluster together genetically. Lack of isolation by distance, however, suggests that gene flow is not limited by dispersal potential per se, but by features of the environment. (b) The study also uncovered genetic differences between inoculates of *M. gruessii* (bottom center) collected on the glossae of bumble bee vectors (bottom left), and the corolla, anthers and nectar (bottom right) of the flower *Digitalis obscura* (center). Those differences in allele frequencies support the hypothesis of diversifying selection operating on the scale of floral microsites. Photo credits: C.M. Herrera.

pled from lower elevation sites appear to cluster together, while isolates from higher elevations were found to belong to two different clusters. In contrast, there does not seem to be any relationship between genetic differentiation and geographical distance between sampling sites. Together, these findings suggest that it is not dispersal potential per se that limits population connectivity in *M. gruessii*, but that environmental characteristics determine how the dispersal potential of the species is realized.

Differences in the strength and direction of natural selection on very small scales also appear to contribute to the structuring of *M. gruessii* populations. Herrera *et al.* (2011) demonstrate this very elegantly by showing differences in genetic composition between isolates of *M. gruessii* sampled from their bumblebee vector and isolates sampled from various floral parts (Fig. 1b). Specifically, there appears to be a reduction in genotype diversity from the bumblebee isolates to those found on the corolla of flowers. Furthermore, the frequency of five putatively selected loci differs between different floral microsites, suggesting diversifying selection. This microsite-specific filtering of genotypes could then maintain high levels of genetic polymorphisms on which natural selection can act. This type of microsite-dependent natural selection could not only favour allopatric speciation, but could also help maintain intraspecific genetic diversity.

Herrera *et al.* (2011) thus find strong evidence for population structure at two different geographical scales in *M. gruessii*, a species with both large population sizes and high dispersal potential. Herrera *et al.* (2011) also present results that suggest *M. gruessii* has a largely or completely asexual life cycle, a factor that has been predicted to reduce genotypic diversity. Their findings are consistent with the results of another recent paper that examined population structure in *S. cerevisiae*, also a primarily clonal species (with an estimated one outcrossing event every 50 000 cell divisions, Ruderfer *et al.* 2006). Goddard *et al.* (2010) assayed 172 strains of *S. cerevisiae* from multiple locations in New Zealand (including grape juice ferments, soil, flowers, apiaries and bark) with nine variable microsatellite loci and found their isolates to be distinct from previously assayed international isolates (Liti *et al.* 2009). Together, these results suggest that clonality does not necessarily preclude intraspecific diversity.

While the study by Herrera *et al.* (2011) indicates that examining population structure in wild microbes may indeed be a fruitful area of research, much remains to be performed before any general conclusions about the prevalence of such population structure can be reached. A recent paper by Muller *et al.* (2011) examined tiling-array-based genotypes of 88 worldwide clinical and nonclinical isolates of *S. cerevisiae* and found no evidence of clear population structure. It is yet to be determined whether the population structure uncovered by Herrera *et al.* (2011) is temporally stable, as it seems likely that some microbial species may exhibit chaotic population structure similar to marine organisms. Whether the contrasting patterns in microbial structure are the result of interesting biological phenomena, or are simply attributed to a different choice of molecular markers, remains to be seen. As additional molecular tools become available, and as these tools are applied to increasing numbers of microbial species in a variety of contexts, we have no doubt that fundamental questions about the genetic structure of microbial species will be answered. It is high time for some microbial biologists to shed their laboratory coats and don their field hats.

References

Arnaud-Haond S, Alberto F, Teixeira S, Serrão EA, Duarte CM (2005) Assessing genetic diversity in clonal organisms: low diversity or low resolution? Combining power and cost efficiency in selecting markers. *Journal of Heredity*, **96**, 434–440.

Balloux F, Lehmann L, Meeûs T (2003) The population genetics of clonal and partially clonal diploids. *Genetics*, **164**, 1635–1644.

Caron D (2009) Past President's address: protistan biogeography: why all the fuss? *Journal of Eukaryotic Microbiology*, **56**, 105–112.

Christie MR, Stallings CD, Johnson DW, Hixon MA (2010) Self-recruitment and sweepstakes reproduction amid extensive gene flow in a coral-reef fish. *Molecular Ecology*, **19**, 1042–1057.

Colbeck GJ, Turgeon J, Sirois P, Dodson JJ (2011) Historical introgression and the role of selective vs. neutral processes in structuring nuclear genetic variation (AFLP) in a circumpolar marine fish, the capelin (*Mallotus villosus*). *Molecular Ecology*, **20**, 1976–1987.

Fenchel T, Finlay B (2004) The ubiquity of small species: patterns of local and global diversity. *BioScience*, **54**, 777–784.

Goddard M, Anfang N, Tang R, Gardner R, Jun C (2010) A distinct population of *Saccharomyces cerevisiae* in New Zealand: evidence for local dispersal by insects and human-aided global dispersal in oak barrels. *Environmental Microbiology*, **12**, 63–73.

Herrera CM, Pozo MI, Bazaga P (2011) Clonality, genetic diversity and support for the diversifying selection hypothesis in natural populations of a flower-living yeast. *Molecular Ecology*, **20**, 4395–4407.

Lachance M (2004) Here and there or everywhere? *BioScience*, **54**, 884–884.

Liti G, Carter D, Moses A *et al.* (2009) Population genomics of domestic and wild yeasts. *Nature*, **458**, 337–341.

Lowe C, Kemp S, Montagnes D (2005) An interdisciplinary approach to assess the functional diversity of free-living microscopic eukaryotes. *Aquatic Microbial Ecology*, **41**, 67–77.

Palumbi SR (1992) Marine speciation on a small planet. *Trends in Ecology and Evolution*, **7**, 114–118.

Palumbi SR (1994) Genetic divergence, reproductive isolation, and marine speciation. *Annual Review of Ecology and Systematics*, **25**, 547–572.

Ruderfer D, Pratt S, Seidel H, Kruglyak L (2006) Population genomic analysis of outcrossing and recombination in yeast. *Nature Genetics*, **38**, 1077–1081.

A.C.G. and J.S.M. are both finishing their PhDs at The University of British Columbia. Sharing an office, they have had many heated discussions about the potential for integration of their divergent approaches to the study of ecology and evolution (experimental evolution of *Saccharomyces cerevisiae* in a controlled laboratory environment—A.C.G., and fish population genetics in the wild—J.S.M.).

---

doi: 10.1111/j.1365-294X.2011.05263.x