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No sex for Daphnia in the Italian Alps...



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Introduction

Differential distribution of asexual species at high altitudes and latitudes has long been noted and coined geographical parthenogenesis. This pattern is thought to arise from the better colonization abilities of asexuals.

Daphnia appear to comply with this as subarctic and arctic species typically reproduce by obligate parthenogenesis (Beaton & Hebert 1988, Dufresne & Hebert 1994). Some *Daphnia* populations from high altitudes are also known to reproduce asexually i.e. those in the Bolivian Andes (Mergeay et al. 2008) and also those in the High Tatra mountains (Dufresne et al. 2011). By contrast, although also inhabiting alpine environments, *D. pulicaria* from the Pyrenees reproduce primarily by cyclic parthenogenesis (Dufresne et al. 2011), suggesting complex glaciation history may play a role in transitions to asexuality.

This study aimed to determine the reproductive mode and clonal diversity patterns of *Daphnia pulicaria* from four lakes in the Italian Alps.

Materials and methods

Four alpine lakes were sampled in Gran Paradiso, National Park (GPNP, Western Alps) including Lake Nivolet, Lake Trebecchi Inferiore, Lake Trebecchi Superiore, and Lake Lillet. The lakes are spatially very close to each other but belong to different river catchments: lake Lillet is situated in the Orco river basin, whereas the others three lakes are located within the Dora di Savarenche river catchment. Four alpine lakes



were sampled in Gran Paradiso, National Park (GPNP, Western Alps) including Lake Nivolet, Lake Trebecchi Inferiore, Lake Trebecchi Superiore, and Lake Lillet. The lakes are spatially very close to each other but belong to different river catchments: lake Lillet is situated in the Orco river basin, whereas the others three lakes are located within the Dora di Savarenche river catchment. 24 individuals per lake genotyped at six microsatellite loci (Dp183, Dp502, Dp512, Dp513, Dp514, Dp514alt). These genoypes were added to a previous set of genotypes from other alpine and lowland European sites (Dufresne et al. 2011). Alleles were transformed into a presence/absence matrix and Cavalli-Sforza chord distances were obtained and a neighbor-joining tree constructed using MEGA7.

Discussion

It is most likely that Daphnia inhabiting the four lakes in the Western Alps reproduce by obligate parthenogenesis as there is no clonal diversity and no males were observed in laboratory cultures.

Daphnia from GPNP had microsatellite genotypes that clustered in two different clades in the NJ tree. These results are congruent with a previous mtDNA study that showed that *Daphnia* from Lake Nivolet, Lake Trebecchi inferiore and Lake Trebecchi superiore belong to the boreal clade of European *D. pulicaria* whereas those from Lake Lillet were genetically distinct and belong to the alpine clade of European *D. pulicaria*.

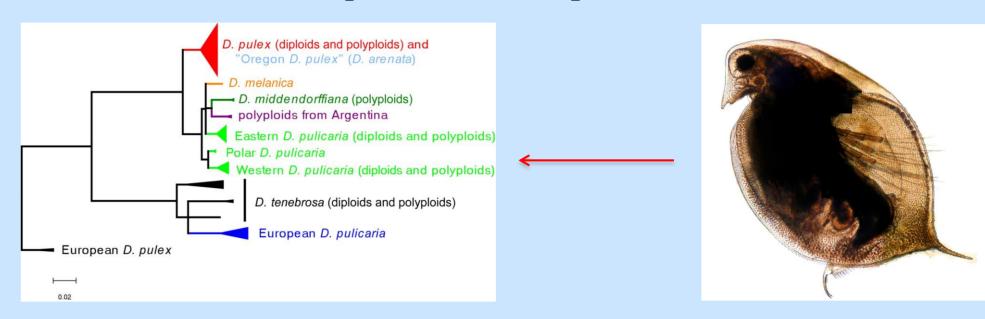
The two clones were diploids as the other alpine clones of the High Tatra mountains.

How did transitions to obligate parthenogenesis occurred in these populations?

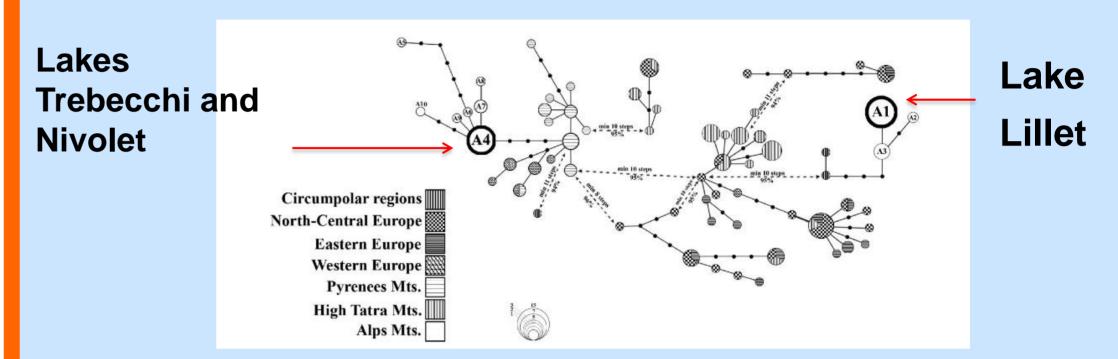
Preliminary results indicate that alleles that suppress meiosis in females but not in males as in North American Daphnia likely involved (as in Lynch et al. 2008).

Future studies using genomic tools will help to better assess genetic relationships among alpine and lowland populations of European *D pulicaria*. As well, gene expression studies comparing lowland and alpine populations are needed to decipher local adaptations to life in high altitude environments. Since impacts of human-related activities and climate changes on mountain species are known to be dramatic, we pose strong issues for the conservation of these extremely localized taxa.

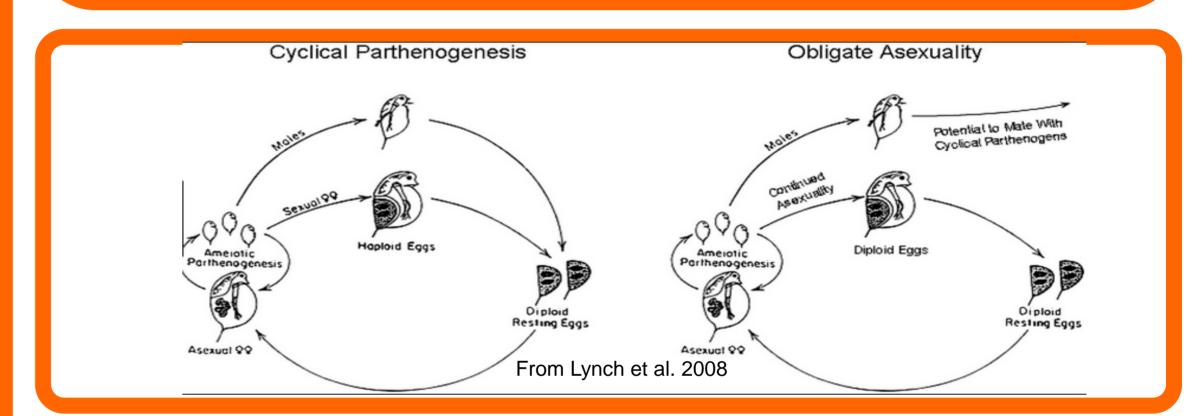
European *D. pulicaria*



ND5 tree of members of the *Daphnia pulex* complex

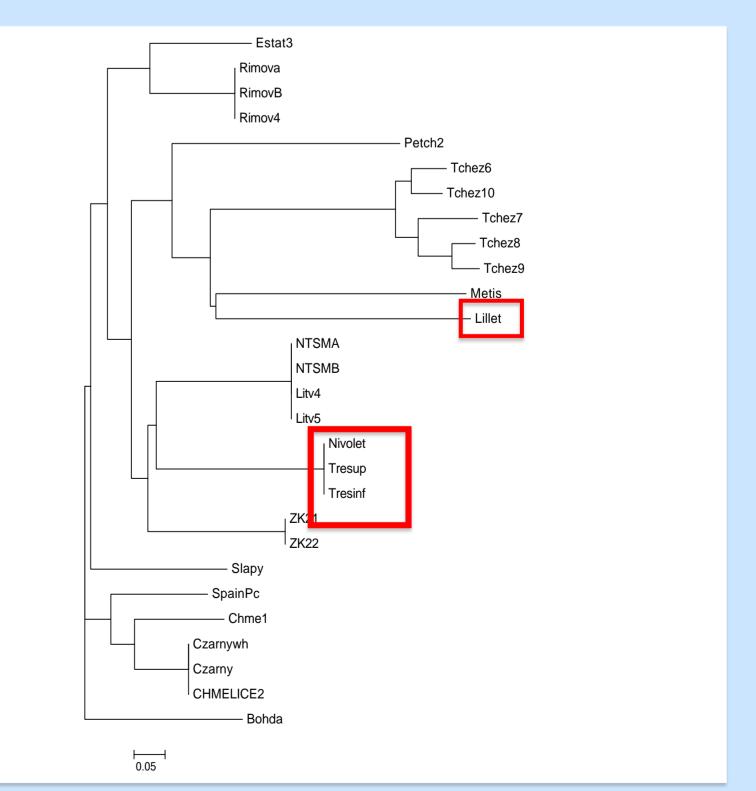


Haplotype network based on ND5 sequences (Bellati et al. 2014)



Results

A single multilocus genotype among the 72 individuals genotyped in Lakes Nivolet, Trebecchi Inferiore and Trebecchi Superiore! Another multilocus genotype among the 24 individuals from Lillet



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