Genetic divergence and ecotype formation in *Epipactis* - Implications for conservation

Hans JACQUEMYN*, Hanne DE KORT, An VANDEN BROECK, Rein BRYS

KU Leuven, Dept of Biology, Plant Conservation and Population Biology, BE-3001 Leuven, Belgium

* **hans.jacquemyn@kuleuven.be**

Ecological speciation is the process by which one species diverges into two distinct phylogenetic lineages that gradually become reproductively isolated from each other after they have colonized a new habitat. Because ecological speciation typically occurs across a continuum of time and several intermediate stages, often called ecological races or ecotypes, can be discerned during the speciation process, it may result in a complex of taxa among which species limits are difficult to define. A typical example of a species group among which species limits are difficult to define is the genus *Epipactis*. It contains a complex of autogamous and non-autogamous taxa that may have arisen after the colonization of new habitats, followed by rapid adaptation and evolutionary changes in key traits that allow establishment and survival in these newly colonized habitats. However, the taxonomic status of these species is problematic and different authors have treated the taxonomy of *Epipactis* in different ways, some recognizing the different taxa as distinct species, others considering them only as minor intraspecific variants or ecological races. Here we present the results of genomic, meta-genomic and morphological analyses aimed at investigating the taxonomic status of coastal dune populations of the widespread terrestrial orchid *Epipactis helleborine*. Investigations of the mycorrhizal fungi associating with coastal dune populations and typical forest populations has shown that they associate with significantly different fungal communities. Crossing experiments show that both taxa easily cross and produce viable seeds. However, germination of seeds of dune populations in forest habitat and vice versa was always lower than that of seeds of coastal populations in dune habitats or of forest populations in forest habitat, leading to strong reproductive isolation as a result of immigrant inviability. Genomic analyses using SNP markers further revealed that coastal dune populations diverged only about 50 generations ago from inland populations, went through a significant bottleneck and were most likely the result of a single colonization event. Current levels of population genetic diversity in 27 populations along the Dutch and French coast were low and not related to population size or spatial isolation. The sampled dune populations also showed very little genetic differentiation and no apparent spatial genetic structure was observed. Overall, these results are consistent with a process of genetic divergence after a single, very recent colonization event followed by extensive gene flow among populations. From a taxonomic point of view, coastal dune populations of *E. helleborine* should not be treated as a separate species, but rather as an ecotype.