6.9

Fine-scale population genetics of East European *Apis mellifera* reveal differentation and introgression events.

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The East European evolutionary lineage of the honeybee *Apis mellifera* expands its distribution area from the Italian and Balkan Peninsulas to the Carpathian Mountains. Subspecies widely propagated as *A. m. carnica* and *A. m. ligustica*, and others as *A. m. macedonica* are included in this lineage. Our objectives were to test an earlier hypothesis of local *A. m. carnica* morphometric varieties (Alpine and Pannonic) described according to zoogeographic zones, and to evaluate the genetic structure of the subspecies distributed in this vast area. Using a panel of twelve microsatellite loci and honeybees collected from 14 countries, we confirmed the existence of two genetic clusters in *A. m. carnica* corresponding to the described Alpine and Pannonic ecotypes, plus another two within *A. m. macedonica* that we have named as continental and Mediterranean *macedonica* ecotypes. Introgression between neighbor subspecies and ecotypes has been detected in the contact zones and also from the Italian honeybee (*A. m. ligustica*). Our results provide a high resolution analysis of the genetic structure of the eastern honeybee subspecies and argue that successive introductions may be a disruption for the conservation of local varieties.

6.10

Microsatellite analysis in museum samples reveals inbreeding before the regression of *Bombus veteranus*.

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Essential pollinators in natural and managed ecosystems, like bumblebees, are undergoing decline. Several hypotheses have been proposed to explain the observed declines. Also genetic factors could play a role. Population genetics try to correlate genetic parameters with bumblebee decline. In order to do so historical data are needed. In this project, we used eight microsatellite loci to genotype pinned museum specimens of the declining bumblebee Bombus veteranus (Fabricius, 1793). This bumblebee is a good example of a species in decline as it was one of the most abundant bumblebees in Belgium one century ago but after starting to decline in 1950 the species is now almost totally vanished. Bumblebee samples were collected spanning a period of three decades (1895-1923) and their genetic parameters were measured. In all time periods we detected low genetic diversity and high inbreeding coefficients (even when corrected for null alleles). Inbreeding was also confirmed by the occurrence of sterile diploid males, and all this while the population remained stable as the actual decline of B. veteranus occurred after 1950. Based on these results, we speculate that low genetic diversity and inbreeding did not directly result in the collapse of *B. veteranus* in Belgium. However, inbreeding still plays an indirect role in the decline of bumblebee populations because of the appearance of diploid males and because a low HE reduces the capacity to react to the drivers of bumblebee decline.

for your remarks: