

MINING MICROSATELLITES IN THE *TUBER MELANOSPORUM* GENOME FOR POPULATION GENETIC ANALYSES

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The level of genetic diversity and genetic structure in *T. melanosporum*, the most appreciated black truffle worldwide, has been debated for many years. Bertault *et al.* (1998) claimed that *T. melanosporum*, also known as the Perigord truffle, has a low genetic diversity and its populations lack any genetic structure. As a consequence, the different bouquet exhibited by different truffle populations has been primarily attributed by these authors to ecological rather than genetic determinants. The absence of genetic structure within *T. melanosporum* populations has been however questioned by Murat *et al.* (2004) and, more recently, by Riccioni *et al.* (2008) who identified a significant level of genetic differentiation among populations of this species. However, all these studies relied on a limited set of molecular markers that also showed a low level of polymorphism. Thanks to the availability of the whole genome assembly of *T. melanosporum* (Martin *et al.*, 2010) it is now possible to identify a plethora of markers for population genetic analyses.

To this end, this study aimed at annotating and characterizing SSR (simple sequence repeats) loci in the genome of *T. melanosporum*. The *T. melanosporum* genome is rich in microsatellite loci with 22,425 SSRs. Mono-nucleotide SSRs are the most frequent motifs. SSRs were found in all genomic regions although they are more frequent in non-coding regions. Sixty out of 135 PCR-amplified mono-, di-, tri-, tetra, penta, and hexanucleotides were polymorphic (44%) within black truffle populations and 27 were randomly selected to analyse 139 *T. melanosporum* specimen collected in France, Italy and Spain. The number of alleles per locus varied from 2 to 18 and the expected heterozygosity from 0.124 to 0.815. One hundred and thirty-two different multilocus genotypes out of the 139 *T. melanosporum* isolates were identified and the genotypic diversity was high (0.999).

In conclusion, the SSRs characterized in this study were highly polymorphic and our results showed that *T. melanosporum* is a species with an important genetic diversity, which is in agreement with its recently uncovered heterothallic mating system (Rubini *et al.*, 2011).

The availability of highly polymorphic molecular markers coupled to a large-scale truffle collection, representative of the species distributional range, present in our labs will allow us to gain insight into the extent and geographical distribution of genetic variability among black truffle populations. In turn, robust population genetic analyses will allow us to test the hypothesis that not only environmental but also genetic factors play a critical role in shaping the aroma of the black truffle.

REFERENCES

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