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Comparison of morphological traits and SSR markers to analyze genetic diversity of alfalfa cultivars

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Detailed knowledge of the level of genetic diversity and patterns of population structure in cultivated alfalfa is needed for further progress in traditional and molecular breeding as well as for the management of genetic resources in this species. In this study, ten European cultivars were investigated : three cultivars (two varieties and one landrace) of Provence type, i.e. low dormancy cultivars mainly based on the subspecies *M. sativa sativa*, and seven cultivars (six varieties and one landrace of Flamande (or Flemish) type, i.e. more dormant cultivars introgressed with different parts of the subspecies *M. sativa falcata*. For each variety, 40 genotypes were examined using morphological traits and SSR markers.

Both morphological traits and molecular markers revealed a large within-cultivar diversity and a small but significant variation among cultivars. For morphological traits, a clear separation of cultivars of Flamande and Provence types was observed. Luzelle, a cultivar related to Flamande type with a high part of *Falcata* genome, was clearly separated from the rest of the cultivars. Among the other Flamande cultivars, we found no differentiation. Contrastingly, the three Provence cultivars were significantly differentiated for morphological traits. For molecular markers, we found significant differentiation between Luzelle and the other cultivars, as for morphological traits. Barmed, a non dormant cultivar bred from American and North-African genetic resources, was also separated from the other cultivars. No structure between Flamande and Provence types was detected using molecular markers. Finally, even if the structure among cultivars was different for morphological traits and molecular markers, a large within-cultivar was observed in both cases and Luzelle was separated from the other cultivars.

The history of alfalfa introduction in Europe (17th Century), the reproductive mode of this species (allogamy), its genetic characteristics (autotetraploidy), the patterns of seed exchanges between farmers contribute to explain the large within-cultivar variation and the low differentiation among populations. Breeding and maintenance of genetic resources should exploit this situation for more efficiency.

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