

## Abstract

Walnuts are highly valued for their rich nutritional profile and wide medicinal applications. This demand has led to the intensification of breeding activities in major walnut production areas such as southwest China, in order to develop more superior cultivars. With the increasing number of cultivars, accurate identification becomes fundamental to selecting the right cultivar for grafting, industrial processing or development of new cultivars. To ensure proper identification of cultivars and understand the genetic structure of wild and cultivated material, we genotyped 362 cultivated and wild individuals of walnut trees from southwest China (with two additional populations from Xinjiang, plus three cultivars from Canada, France and Belgium) using 36 polymorphic microsatellite loci. We found relatively low indices of genetic diversity ( $H_o = 0.570$ ,  $H_E = 0.404$ ,  $N_A = 2.345$ ) as well as a high level of clonality (>85% of cultivars), indicating reliance on genetically narrow sources of parental material for breeding. Our STRUCTURE and PCoA analyses generally delineated the two species, though considerable levels of introgression were also evident. More significantly, we detected a distinct genetic group of cultivated *Juglans sigillata*, which mainly comprised individuals of the popular ‘Yangbidapao’ landrace. Finally, a core set of 18 SSR loci was selected, which was capable of identifying 32 cultivars. In a nutshell, our results call for more utilization of genetically disparate material, including wild walnut trees, as parental sources to breed for more cultivars. The data reported herein will significantly contribute towards the genetic improvement and conservation of the walnut germplasm in southwest China.

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