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Possible use of SSR markers to optimise DUS testing in *Capsicum annuum* L.

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BACKGROUND Chili pepper and sweet pepper (*Capsicum annuum* L.) are important vegetable crops with a significant economic value. The French Group for the study and evaluation of Varieties and Seeds (GEVES) is in charge of Distinctness, Uniformity and Stability (DUS) tests for varieties registration and/or protection. This examination process is exclusively based on phenotypic traits and mainly involves field observations. For pepper, a large number of morphological characters are recorded as well as some biochemical properties such as pungency. According to the size of the pepper reference collection and varieties of common knowledge (more than 2500 varieties in the reference collection), field trials may be time and space consuming. In order to enhance field assays and manage reference collection, the use of molecular markers have already been assessed and validated on different crop species, for example maize and barley [1]. The present study aimed to examine the potential use of DNA based techniques as a tool to optimise DUS test in *Capsicum*.

MATERIALS & METHODS DNA extraction was carried out on a sample of 30 seeds with 57 cultivars. These different varieties represented a diverse sample in terms of morphological characters specially fruit shape and pungent characteristic. A set of 38 SSR were first selected according to previous studies ([2] and [3]) on different *Capsicum* species. Currently, 13 SSR markers have been assessed. First analysis on molecular diversity were carried out in R (R core Team). Genetic indices were used to select the most informative markers and Rogers genetic distances were calculated to examine genetic structure via UPGMA tree and PCOA analysis.

RESULTS All 13 primers tested amplified. However, one primer showed inconsistency and was difficult to score and therefore discarded for further analysis in this study. The 12 SSR primers generated a total of 42 polymorphic fragments with an average of 3.5 alleles per locus. The number of alleles per locus ranged from 2 to 9. PIC (Polymorphism Index Content) was estimated from 0.018 to 0.476. Few markers had a low PIC value. The polymorphic SSR markers were not able to distinguish the 57 pepper varieties tested. Rogers genetic distance values ranged from 0 to 0.88. To analyse the genetic relationships among the varieties, a cluster analysis was performed. PCOA and UPGMA dendrogram gave similar results, one major cluster containing 41 non-pungent varieties was observed. For the 16 remaining varieties a grouping pattern according to the morphological traits tested, i.e., pungent and fruit shape was not observed.

DISCUSSION & CONCLUSION To improve molecular descriptions of varieties, clustering analysis results from the 25 remaining SSR will be analysed. If results are conclusive, the marker set selected will be used to calculate genetic distances among candidate varieties and varieties from the collection. These genetic distances could be used as complementary information for field comparisons. Moreover, UPOV models now consider positively the use of molecular markers directly linked to a phenotypic trait. Diagnostic markers should then be examined to assess potential benefits in the framework of DUS testing. In *Capsicum*, the pungent or non-pungent character is a good candidate to evaluate this approach.

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