An approach to evaluate likelihood of gene flow between *Saccharum* hybrids and wild relatives: A case study from South Africa

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South African biosafety legislation requires an assessment of gene flow before approving genetically modified (GM) crops. Proof is required that hybridization cannot occur between GM crop plants and wild relatives. This study presents an approach to assess the likelihood of gene flow using a case study done in South Africa to investigate the possibility of gene flow from sugarcane to wild relatives in the sugarcane production regions. Spatial and temporal assessments of *Saccharum* and wild relatives was conducted. Eleven *Saccharum* relatives which are common in the study area were selected from the worldwide phylogeny of grasses confirming their close relatedness. Relevant data for these species were sourced from the literature, herbaria and field surveys. Co-occurrence (spatial overlap) of all target species was confirmed in sugarcane cultivation areas. *Imperata cylindrica*, *Sorghum* species and *Miscanthidium* species scored the highest likelihood for prevalence, flowering and spatial overlap, and seed distribution potential. Although these species were flagged by the approach, the crossing of spatial and temporal barriers is no guarantee for gene flow to occur and needs to be considered together with relatedness. Phylogenetic analyses of the extended Internal Transcribed Sequence (ITS) cassette showed that the closest wild relatives to commercial sugarcane were *Miscanthidium capense* and *Miscanthidium junceum* as they are only 3 million years divergent. *Sorghum* was found to be more distantly related. Gene flow likelihood was therefore narrowed down to two species using this approach which involves spatial, temporal and molecular techniques. Future work will include aspects influencing gene flow such as cytological compatibility and introgression between sugarcane and *Miscanthidium* species.

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