

Sorting Maize Haploids from Hybrids Using Single-Kernel Near-Infrared Spectroscopy

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Abstract : Doubled haploids (DHs) have become an important breeding tool for creating maize inbred lines, although several bottlenecks in the DH production process limit wider development, application, and adoption of the technique. DH kernels are typically sorted manually and represent about 10% of the seeds in a much larger pool where the remaining 90% are hybrid siblings. This introduces time constraints on DH production and manual sorting is often not accurate. Automated sorting based on the chemical composition of the kernel can be effective, but devices, namely NMR, have not achieved the sorting speed to be a cost-effective replacement to manual sorting. This study evaluated a single kernel near-infrared reflectance spectroscopy (skNIR) platform to accurately identify DH kernels based on oil content. The skNIR platform is a higher-throughput device, approximately 3 seeds/s, that uses spectra to predict oil content of each kernel from maize crosses intentionally developed to create larger than normal oil differences, 1.5%-2%, between DH and hybrid kernels. Spectra from the skNIR were used to construct a partial least squares regression (PLS) model for oil and for a categorical reference model of 1 (DH kernel) or 2 (hybrid kernel) and then used to sort several crosses to evaluate performance. Two approaches were used for sorting. The first used a general PLS model developed from all crosses to predict oil content and then used for sorting each induction cross, the second was the development of a specific model from a single induction cross where approximately fifty DH and one hundred hybrid kernels used. This second approach used a categorical reference value of 1 and 2, instead of oil content, for the PLS model and kernels selected for the calibration set were manually referenced based on traditional commercial methods using coloration of the tip cap and germ areas. The generalized PLS oil model statistics were $R^2 = 0.94$ and $RMSE = .93\%$ for kernels spanning an oil content of 2.7% to 19.3%. Sorting by this model resulted in extracting 55% to 85% of haploid kernels from the four induction crosses. Using the second method of generating a model for each cross yielded model statistics ranging from $R^2s = 0.96$ to 0.98 and $RMSEs$ from 0.08 to 0.10 . Sorting in this case resulted in 100% correct classification but required models that were cross. In summary, the first generalized model oil method could be used to sort a significant number of kernels from a kernel pool but was not close to the accuracy of developing a sorting model from a single cross. The penalty for the second method is that a PLS model would need to be developed for each individual cross. In conclusion both methods could find useful application in the sorting of DH from hybrid kernels.

Keywords : NIR, haploids, maize, sorting

Conference Title : ICDHTPBHI 2023 : International Conference on Doubled Haploid Technology in Plant Breeding and Haploid Induction

Conference Location : Barcelona, Spain

Conference Dates : February 16-17, 2023