Uncovering Identity by Descent for Varietal Protection in Synthetic Populations

John Cameron, Forage Genetics International

DNA fingerprinting utilizes physical linkage between adjacent SNP markers to determine unique genetic profiles for individuals which can be used to detect the presence of that individual's genetics. The physical linkage between SNP loci which DNA fingerprints, or haplotypes, exploit provides increased information about the Identity By Descent (IBD) than that of the SNPs alone. DNA fingerprinting is a common practice for protecting breeding intellectual property in inbred and hybrid crops. However, the utility of analytical haplotyping methodologies for obligate outcrossing populations derived from polycrosses is limited due to the requirements of a) biparental crosses b) acquiring both the parental genotypes and those of tens to hundreds of progeny from each cross, and c) the implementation of computationally intensive statistical models. Genome Wide Allelic Frequency Fingerprinting, via genotyping bulks of tissue in aggregate from synthetic varieties then determining the allele-frequency profiles of the varieties has been proposed as an alternative method to haplotype-based fingerprinting for differentiating obligate outcrossing synthetic varieties. High throughput targeted sequencing has enabled what is referred to in the literature as microhaplotyping; the sequencing of two or more SNP loci located within a few hundred base pairs from each other on a single read so the linkage information between the SNP loci is captured. The result is a multiallelic markers composed of phased SNP loci, which is better able to discriminate IBD at that genomic region than the SNPs alone. An intermediary between unphased SNPs and entire linkage-group marker-phasing, microhaplotyping has recently gained prevalence in forensic sciences for its statistical power in accurately determining relationships between individuals. With the increase in resolution of IBD, microhaplotype markers have the potential of increasing statistical power in population and kinship analyses of synthetic populations using allelic frequency fingerprints, as well as to uncover rare linkage patterns that could additionally be used to discriminate and classify synthetic varieties. This presentation will discuss practical considerations of how a varietal protection program can be cost-effectively implemented in an outcrossing synthetic variety breeding program using targeted-sequencing and microhaplotyping, bulked sampling and sequencing, rare microhaplotype classification, and analytical tests of the level of relationship between varieties.