Rare allele detection by next-generation sequencing in large scale modern agriculture

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Abstract

Next-generation sequencing (NGS) is becoming a ubiquitous commodity for molecular analysis, and sequencing of new varieties of crop plants is nearing routine. For many applications in modern agriculture R&D programs, there is a need to detect very low abundance sequence variants. Although sequencing costs have reduced significantly in recent years, for such applications a targeted, rather than whole-genome, sequencing method will be the most efficient approach, especially for large volumes of samples. Depending upon the nature of targets and the requirements per specific application, either amplicon sequencing or enrichment sequencing can be suitable options for detecting low levels of multiple targets at production scale. Here, we focus on three distinct example applications: 1) GMO detection by enrichment sequencing, with a comprehensive target panel for detecting known and unauthorized GMOs down to 0.1% in bulk samples; 2) Gene editing variants detection by amplicon sequencing, for multiple targets and a range of variant frequencies; 3) Monitoring for known target genes/alleles associated with resistance to ‘cidal compounds by amplicon sequencing or enrichment sequencing for reliable and large-scale mutation detection. We will briefly discuss some technical challenges and recent improvements.