APPLICATION OF LASER CAPTURE MICRO-DISSECTION (LCM) MICROSCOPY IN FOREST TREE GENETICS

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Laser Capture Microdissection (LCM) has been widely used in molecular biology since the early 1990s to study individual cells (Emmert-Buck et al., 1996). Until recently, LCM has not been widely applied to plants due to the complex plant cell wall structures (Kehr et al., 2001). However, LCM can be a very efficient tool in plants as well as animals for precisely harvesting targeted tissues, cells or chromosomes. When applied to loblolly pine (Pinus taeda. L), a very important and widely distributed forest tree in the southern United States, it may be possible to reduce genome complexity for further analysis such as genome sequencing. Loblolly pine has a haploid genome size of 23 Gbp (Neale et al, 2014) with a chromosome number of 2n = 2x = 24 (Islam-Faridi, 2007). Specific loblolly pine chromosomes (or portions of chromosomes) have been successfully captured with LCM and have yielded large amounts of DNA following PCR using whole genome amplification (WGA) methods. Fifteen copies of a targeted chromosome (~15 picograms of DNA) yielded 7 Âµg of DNA from PCR amplification (Islam et al, 2014 unpublished). DNA from such WGA can potentially be used for downstream analysis such as making chromosome-specific probes for fluorescence in situ hybridization (FISH) or for chromosome-by-chromosome genome sequencing. LCM promises to be a very powerful tool for studying conifer genomes. We will provide results and discussion of our recent experiments with LCM and WGA of pine chromosomes.

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