Droplet Vitrification - A Viable Method of Cryopreservation for Deeringothamnus rugelii, Deeringothamnus pulchellus, and Asimina tetramera

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Genetic diversity is crucial to the survival of species. To preserve plants threatened by natural and human causes, ex situ conservation has been implemented with seed banking and cryopreservation. Since not all plants produce seeds, or have seeds that bank successfully, cryopreservation is an alternative method to produce reserves of these "exceptional species." Pawpaws are a group of these exceptional species that cannot be preserved through traditional methods. The purpose of this study was to determine if a newer cryopreservation protocol, droplet vitrification (DV), could be a better method of preservation for Deeringothamnus rugelii, Deeringothamnus pulchellus, and Asimina tetramera, compared to encapsulation vitrification (EV). Shoot tips and nodes (n=10) were dissected and cryopreserved for one hour using DV, followed by recovery on gel plates for 6 weeks, and a transfer to culture tubes that was monitored for 12 additional weeks. The species were viable post DV, since 54% of A. tetramera shoot tips demonstrated growth at 18 weeks, followed by 30% of D. rugelii nodes and tips, and 9% of D. pulchellus tips. While the cultured D. rugelii nodes and tips had the same percentage of tissues demonstrating growth, phenotypical differences were observed between the recovered node and tip cultures. The culture tubes containing the most growth for each species and tissue group were subcultured and monitored for 18 weeks to compare the growth between the two generations of cultures. While further testing is required, droplet vitrification may be an alternative method to effectively conserve these essential species.

Awards Won: Fourth Award of \$500