

DNAgard Tissues & Cells preserve plant tissue at room temperature

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INTRODUCTION

DNAgard Tissue is a liquid storage reagent that rapidly permeates cell membranes to stabilize and protect genomic DNA. It was developed to assist scientists with the storage and shipping of tissue samples at room temperature. It is ideal for field sample collections. Simply collect your sample and add just enough DNAgard Tissue to cover your sample. There is no need for dry ice collection or freezer storage. Storage of plant material and subsequent DNA isolations can be challenging. DNAgard Tissue protects DNA in the presence of plant inhibitors. For example, plant species such as banana (*Musa* spp.) have high amounts of polyphenolic metabolites, Wheat (*Triticum* spp.) has high amounts of nucleases, while *Chlorophytum* species contain high amounts of mucous compounds. All these conditions make it difficult to obtain high quality DNA.

MATERIALS AND METHODS

DNA Isolation

Five different plant species were collected and stored in DNAgard Tissue for 30 days at room temperature. The plant species included representatives of five genera: *Musa*, *Ensete*, *Hordeum*, *Triticum*, and *Chlorophytum*. 75mg of leaf segment was added to a 2mL screw cap tube containing 500µl of DNAgard. After 30 days the leaf segments were removed and washed in distilled water (3x) at room temperature. Leaf segments were placed into liquid nitrogen and homogenized. DNA was isolated using a plant genomic DNA isolation kit (Invisorb Spin Plant Mini Kit). Approximately 100ng of plant genomic DNA was viewed on a 1.2% agarose gel.

RESULTS AND DISCUSSION

For plant genomic analyses, it is essential to obtain intact, high molecular weight DNA. This can be challenging as many plant species accumulate secondary metabolites that can directly damage DNA and/or interfere with downstream DNA analysis. Results from this study show complete protection of DNA in DNAgard Tissue at room temperature after 30 days preservation. Figure 1 shows that intact, high molecular weight genomic DNA is recovered from plant tissue of five species stored in DNAgard Tissue formulation. Figure 3 demonstrates that DNA recovered from these samples is free from inhibitors that interfere with restriction digestion.

1 2 3 M 4 5 6

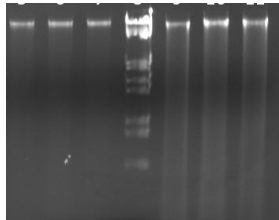


Figure 1. Lyophilization of plant genomic DNA compared to room temperature storage in DNAgard Tissue. Lanes 1, 2, and 3 are purified banana (*Musa balbisiana*) genomic DNA stored in DNAgard Tissue: Lanes 4, 5, and 6 are purified banana genomic DNA that was stored lyophilized (positive controls).

Figure 2. Stabilization of plant genomic DNA.

Lanes 1 and 14 represent λ DNA at 100ng. The remaining lanes represent DNA extracted from samples stored in DNAgard Tissue: Lanes 2-5 are purified banana DNA (*Musa balbisiana*). Lanes 6-7 represent DNA of *Ensete gillettii* (related to banana). Lanes 8-9 are barley DNA (*Hordeum vulgare*). Lanes 10-11 are *Chlorophytum* DNA. Lanes 12-13 represent wheat DNA (*Triticum aestivum*). M = λ DNA/PstI ladder.

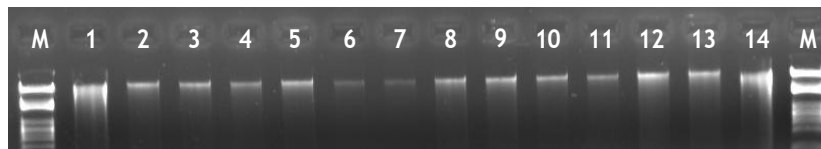


Figure 3. DNA compatibility with restriction digestion. This gel is a typical example of restriction pattern of banana (*Musa balbisiana*) DNA recovered from plant samples stored in DNAgard Tissue. Lane 1 is λ DNA/PstI. Lane 2 is banana DNA digested with *Alu*I. Lane 3 banana DNA digested with *Fok*I and Lane 4 is banana DNA digested with *Mse*I.

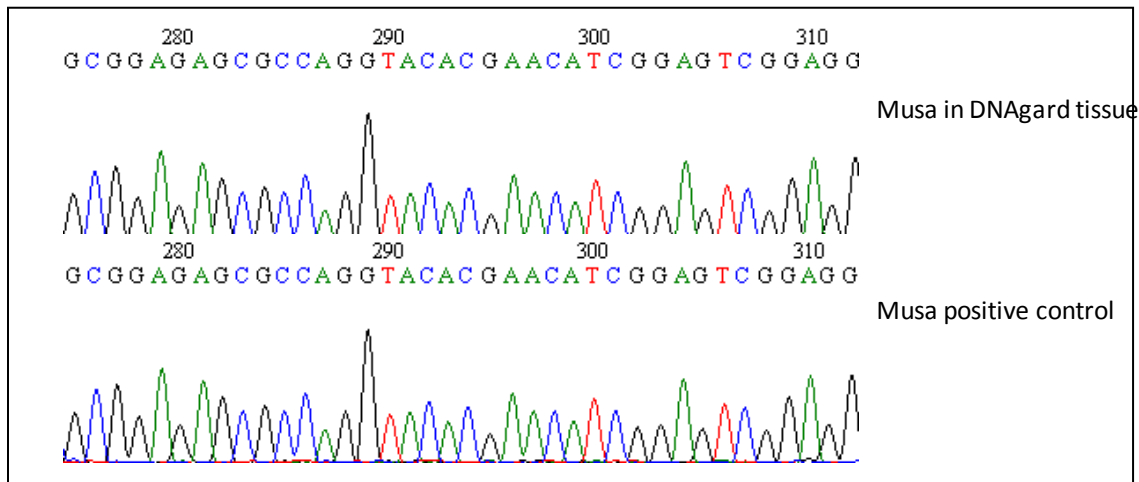
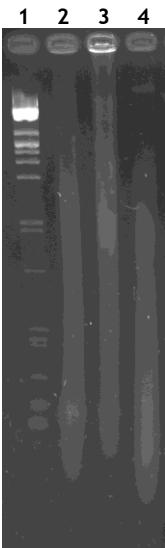


Figure 4. Previous data based on sequenced clones of the ITS region from banana (*Musa balbisiana*) show that DNAgard Tissue retains sequence integrity compared to previous sequence data.

CONCLUSION

The results clearly show that DNAgard Tissue preserves DNA in plant tissues at room temperature. This is a convenient product for collecting and preserving field samples. Samples can be processed at a later time for DNA isolation yielding high quality DNA for any downstream application. There is no need for dry ice/ice box (wet ice) at collection site. DNAgard Tissue is convenient to use and allows for flexibility in the choice of field collection format.