

DNAgard® Tissue preserves DNA from 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 is n useful tool for storage and shipping of tissue samples at room temperature. In addition to stabilizing tissues in the laboratory, it is ideal for field sample collections such as plant tissue. Storage of plant materials and subsequent DNA isolation can be challenging because of various existing 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, *Chlorophytum* species contain high amounts of mucous compounds. These conditions make it difficult to obtain high quality DNA. However, DNAgard Tissue protects DNA in the presence of plant inhibitors including the inhibitors mentioned above.

Materials and Methods

<u>DNA Isolation</u>: 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 were added to a 2mL screw cap tube containing 500µL of DNAgard Tissue. After 30 days, the leaf segments were removed and washed in distilled water three times at room temperature. Leaf segments were placed into liquid nitrogen and homogenized. Subsequently, 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 analysis, 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 and 2 show that intact, high molecular weight genomic DNA is recovered from plant tissue of five species stored in the DNAgard Tissue formulation. Figure 3 demonstrates that DNA recovered from these samples is free from inhibitors that interfere with restriction digestion. Figure 4 shows that DNAgard Tissue retains the sequence integrity of the ITS region from banana (*Musa balbisiana*) compared with previous sequence data.

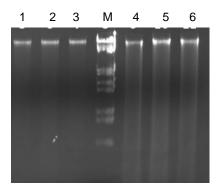


Figure 1 Lyophilizaton of plant genomic DNA compared to room temperature storage in DNAgard Tissue. Lanes 1, 2 and 3 are purified banana (*Musa balbisiana*) genomic DNA stored in DNAgard Tissue: Lanes 4, 5 and 6 are purified banana genomic DNA that was stored lyophilized (positive controls). M= λDNA/Pstl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



Figure 2. Stabilization of plant genomic DNA. Lanes 1 and 14 represent λDNA 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 gilletii* (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= λDNA/Pstl ladder.



Figure 3. DNA from DNAgard Tissue preserved samples shows compatibility with restriction digestion. This gel is a typical example of the restriction pattern of banana (*Musa balbisiana*) DNA recovered from plant samples stored in DNAgard Tissue. Lane 1 is λDNA/Pstl. 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.

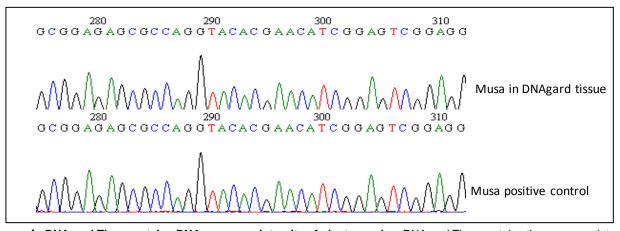


Figure 4. **DNAgard Tissue retains DNA sequence integrity of plant samples.** DNAgard Tissue retains the sequence integrity of the ITS region from banana (*Musa balbisiana*) compared to positive control sequence data.

Conclusion

Our results demonstrate 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. The high quality DNA purified from the samples preserved by DNAgard Tissue can be used for any downstream application. There is no need for dry or wet at the collection sites. DNAgard Tissue is convenient to use and allows for flexibility in the choice of field collection format.

Note: For Research Use Only. Not for use in diagnostic procedures. Please read all instructions for <u>DNAgard Tissue</u> prior to using this protocol. DNAgard® is the registered trademark of Biomatrica. <u>Order DNAgard Tissue</u>