Isolating Plant DNA Without Standard Laboratory Tools

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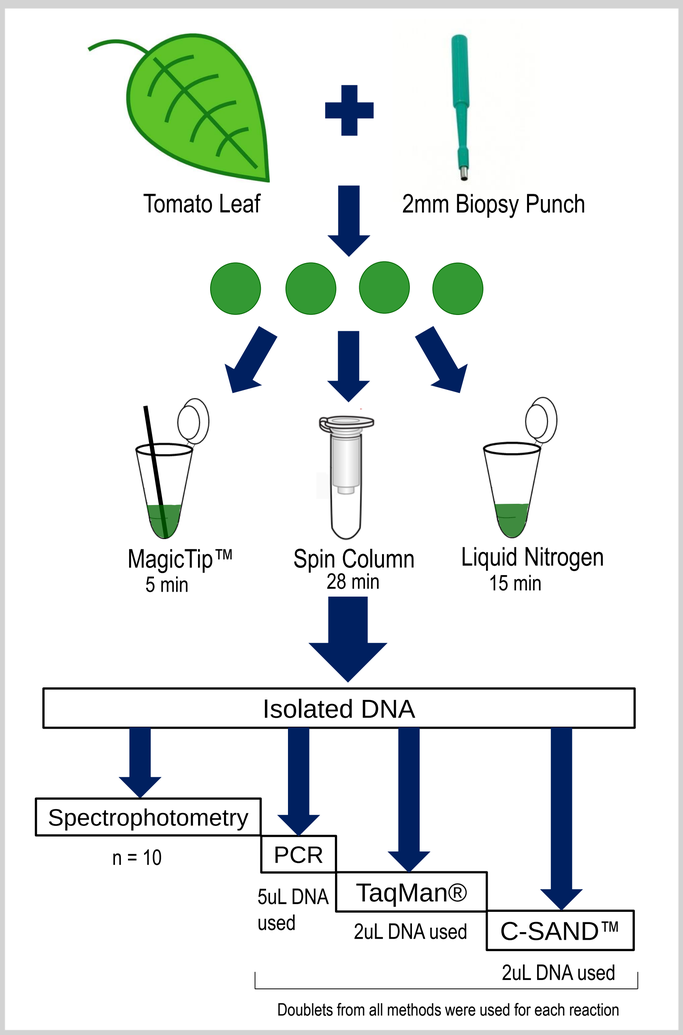
Abstract

Current methods of plant DNA isolation can be time consuming and labor intensive, requiring laboratory equipment and technical skills. Mechanical methods of plant DNA isolation often require liquid nitrogen or the use of a bead and shaker system to lyse the plant cells. Column based kits require a centrifuge or vacuum manifold to isolate the DNA from the cell debris. Some of the above methods require lengthy incubations, multiple centrifugation steps, and pipetting steps. All these methods also require laboratory equipment or hazardous materials that are not easy to acquire making field use impractical or dangerous. A recently developed method uses a substrate (MagicTip™) simply placed into the cell lysate to bind DNA before being removed into a buffer where the DNA is then eluted all within 5 minutes. DNA isolation kit buffers are supplied in dropper bottles eliminating the need for pipettes allowing the protocol to be performed in a field setting. To test the efficacy of DNA template eluted using the MagicTip™ DNA isolation method, we compared the performance of this template alongside template produced using other isolation methods in a polymerase chain reaction (PCR), TaqMan® assay, and C-SAND™ assay. Tomato leaf samples (four, 2mm punches) had DNA isolated using a MagicTip™ DNA isolation kit, a commercially available spin column kit, and a second commercially available kit requiring liquid nitrogen. The results suggest that the DNA isolated using the MagicTip™ method is of similar quality to the other methods tested. The ease of use and portability of the MagicTip™ DNA isolation method allows for high quality DNA to be generated quickly in the lab or in the field.

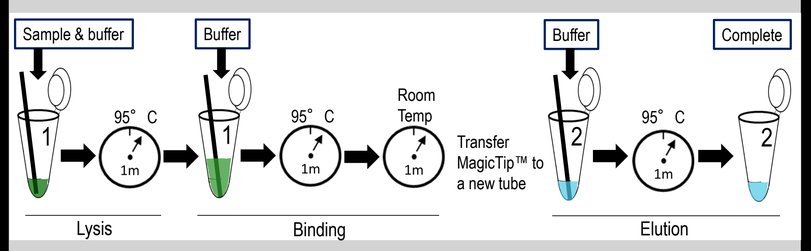
Methods



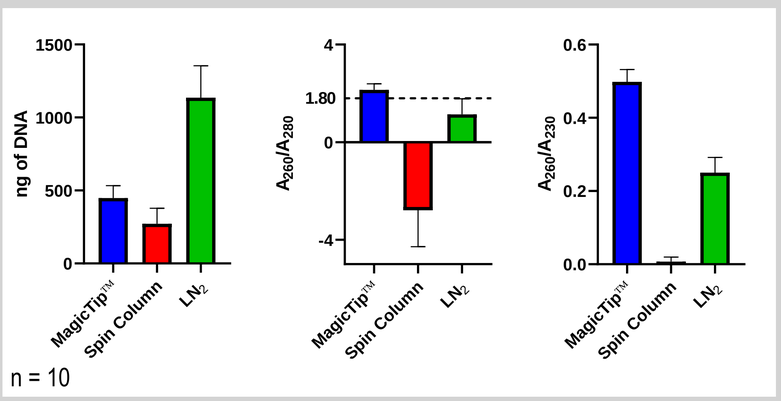
MagicTip™ Kit

Kits include all components* and are stable at room temperature.
Buffers are supplied in dropper bottles and applied dropwise.
Entire process takes about 5 minutes and only requires a heat source.
*Ethanol not included and must be purchased by user

The MagicTip™ Plant DNA Isolation Kit System Overview



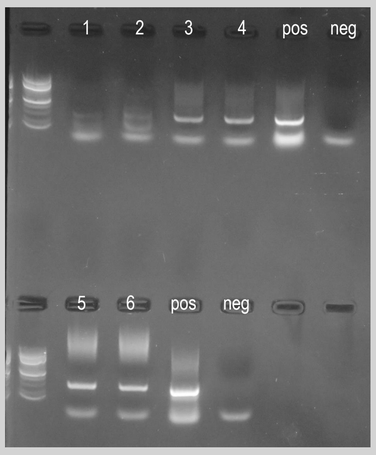
Plant DNA Isolation Yield and Purity Comparison



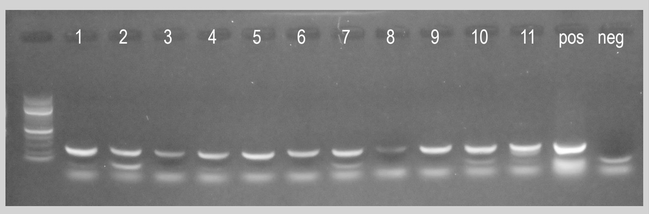
MagicTip™ Method Isolates PCR-Quality DNA

Lanes	Method
1,2	Spin Column
3,4	MagicTip™
5,6	Liquid Nitrogen

Primers designed for tomato actin with an expected product size of 255bp



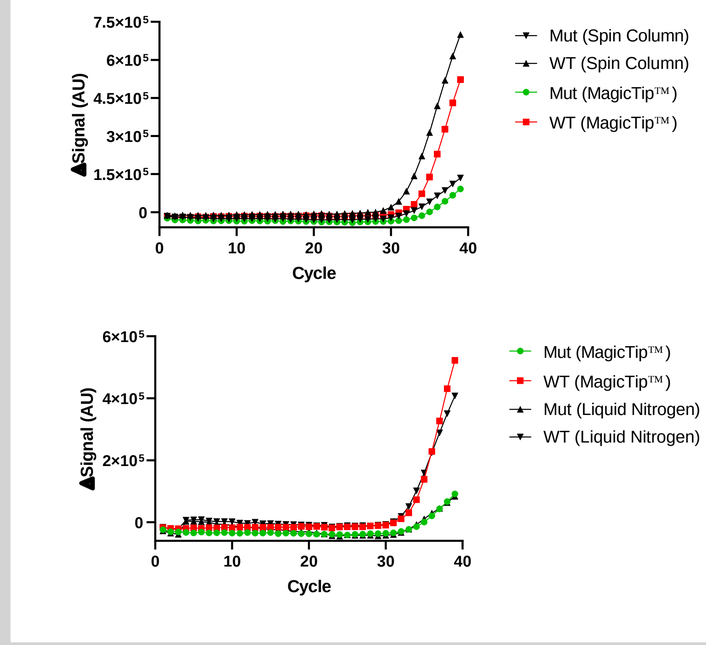
MagicTip™ Method Compatible with Many Plant Species



Primers designed for NAD1 with an expected product size of 230bp.

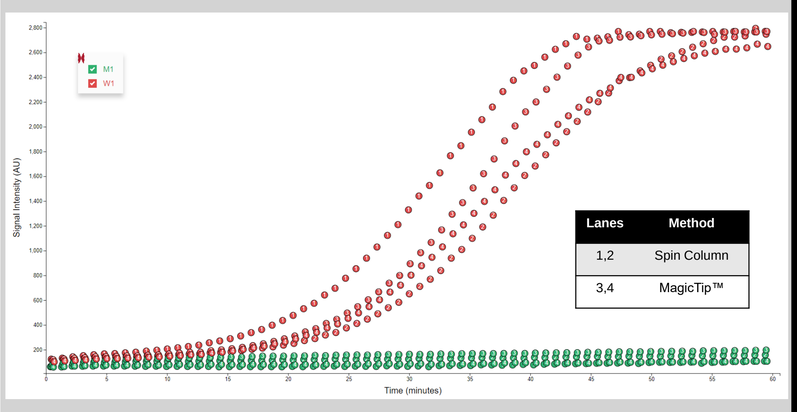
Lane	Sample
1	Clover
2	Tomato
3	Arabidopsis
4	Camelina
5	Sorghum
6	Tobacco
7	Soy
8	Cotton
9	Hemp
10	Corn
11	Wheat

MagicTip™ Plant DNA Template Performance in TaqMan® Assays

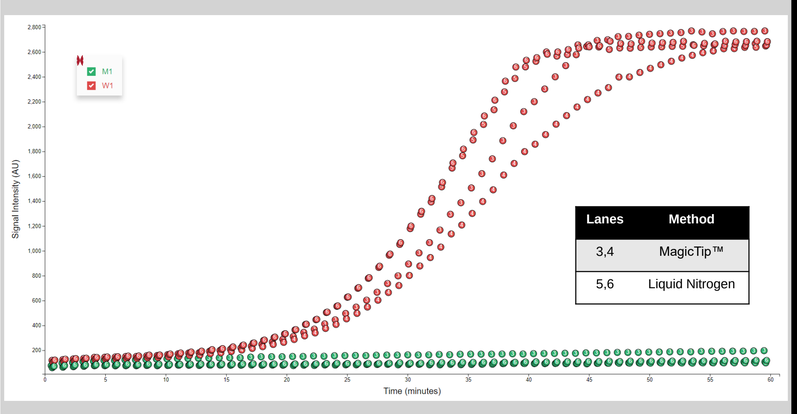


TaqMan® assay designed for the Tomato Locule SNP. Tomato leaf used for sample was homozygous wild-type.

MagicTip™ Plant DNA Comparable to other DNA extraction methods in C-SAND™ Assays



C-SAND™ assay developed for the SNP detection of mutant (green) or wild-type (red) alleles in the gene that controls the number of tomato locules. Individual sampled is homozygous wild-type.



Conclusion

In this study, it was shown that the MagicTip™ plant DNA isolation kit produces DNA of similar quality to other commercially available kits utilizing spin column or liquid nitrogen extraction methods. The DNA isolated from the MagicTip™ was shown to be comparable and functional in many different downstream applications such as:

- PCR
- TaqMan®
- C-SAND™ (**C**ombined **S**equence **A**mplification and **N**ucleotide **D**etection)

The MagicTip™ method requires no technical skill or training, no additional lab equipment, and all necessary components are included in the kit. Compared to other DNA isolation methods, it is easy to use and takes only 5 minutes to produce quality DNA. This assures the user that they will be able to isolate DNA in a short time period in any setting, allowing for high quality DNA to be generated quickly in the lab or in the field.

Acknowledgements

Development and design of the Tomato Locule C-SAND™ assay was performed by **Heather Piscatelli** and **Alyssa Hangman.**

Design of the Tomato Locule TaqMan® assay was performed by **Heather Piscatelli.**

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