



# Room Temperature DNA Storage

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## Abstract

Successful long term storage of extracted DNA is of critical importance to the field of forensic science. DNA evidence may require future retesting; therefore, it must be preserved in order to obtain accurate results over time. While current frozen storage methods are effective at maintaining DNA samples over long periods of time, they are limited by increased cost and potential sample loss due to power failure or unsuccessful transport. Consequently, new technology has been developed that allows for room temperature DNA storage. In this study, GenVault GenTegra™, Biomatrixa DNAsable® LD and microbiology grade trehalose dihydrate were evaluated for their ability to preserve DNA in various storage conditions over a period of four weeks. Results indicated that Biomatrixa DNAsable® LD was the most effective stabilizer at room temperature. However, extra precaution should be taken when utilizing Biomatrixa DNAsable® LD in particularly humid conditions. In addition, further studies are suggested to evaluate the continued performance of Biomatrixa DNAsable® LD for up to one year.

## Studies Performed

### Concentration Study

Stock dilutions: 40, 7, 1, 0.25, 0.1, 0.05, 0.00625, 0.003125ng/μL  
Analysis interval: 4 weeks  
Storage conditions: Room temperature

### Accelerated Aging Study

Stock dilution: 0.05ng/μL  
Analysis interval: 4 weeks  
Storage conditions: 56°C and 70°C

### Uncontrolled Humidity Study

Stock dilution: 0.05ng/μL  
Analysis intervals: 2 weeks, 4 weeks  
Storage conditions: 50°C water bath & incubation container

### Time Study

Stock dilution: 0.05ng/μL  
Analysis intervals: 1 day, 1 week, 2 weeks, 3 weeks, 4 weeks  
Storage conditions: Room temperature

### Extraction Method Study

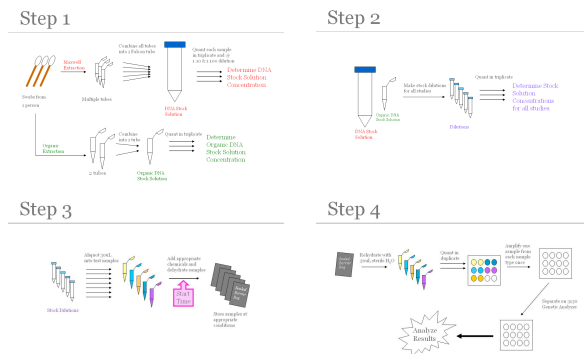
Stock dilution: 0.05ng/μL (Maxwell® and phenol/chloroform)  
Analysis interval: 4 weeks  
Storage conditions: Room temperature

### Contamination Study

No DNA added  
Analysis interval: 4 weeks  
Storage conditions: Room temperature

## Experimental Procedure

Below is a visual representation of how samples were prepared for each study:



## Results

### Concentration Study

Although most stabilizers were effective at preserving DNA in samples with lower concentrations, they appeared to be less effective in samples with higher concentrations. In particular, trehalose performed the worst at higher DNA concentrations. In addition, the untreated frozen sample showed a decrease in DNA concentration; however this was probably due to variability in quantification procedures since previous research has proven frozen storage effective.

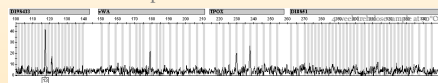
Concentration	Treatment				
	Untreated	GenTegra™	DNAsable® LD	Trehalose	Frozen
40ng/μL	Yes	No	Yes	Yes	Yes
7ng/μL	No	No	No	Yes	Yes
1ng/μL	Yes	Yes	No	Yes	Yes
0.25ng/μL	No	No	No	No	No
0.1ng/μL	No	Yes	No	No	No
0.05ng/μL	No	No	No	No	No
0.00625ng/μL	No	No	No	Yes	No
0.003125ng/μL	No	No	No	No	No

Statistical significance of stabilizer performance at various concentrations

### Accelerated Aging Study

Time acceleration projection equates 4 weeks at 56°C and 70°C to 42 weeks and 111 weeks at room temperature, respectively. All samples at 56°C produced full profiles with several peaks below stochastic threshold. At 70°C, some samples produced full profiles while others produced partial profiles; however, all samples contained peaks below the stochastic threshold.

The trehalose sample exhibited almost complete profile dropout with only one peak present which was below the stochastic threshold. Therefore, it was determined that trehalose should not be used to preserve DNA in samples at storage temperatures above 56°C or in samples stored for longer than 42 weeks at room temperature.



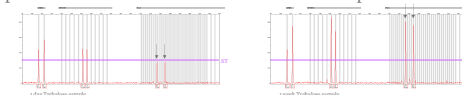
### Uncontrolled Humidity Study

All 50°C water bath samples yielded full profiles with all peaks above the stochastic threshold.

Four week incubation container samples produced full profiles with several peaks below the stochastic threshold. However, the DNAsable® LD sample produced undetermined results for quantification; therefore the sample was concentrated. Consequently, allele and peak height data for this sample was not applied to subsequent sample comparison

### Time Study

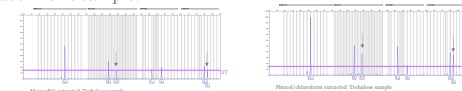
All samples at all time intervals produced full profiles; however, the 1-day trehalose sample contained a few peaks below the stochastic threshold. Subsequent analysis of the 1-week trehalose sample showed that all peaks were above the stochastic threshold. This result was likely due to differences in amplification or DNA concentration between samples.



### Extraction Method Study

All phenol/chloroform extracted samples produced full profiles with all peaks above the stochastic threshold.

All Maxwell® extracted samples produced full profiles with a few peaks below stochastic threshold. In particular, the trehalose sample showed an overall decrease in peak height and two peaks below the stochastic threshold. This variation in peak height was perhaps due to the presence of residual magnetic beads in this sample.



### Contamination Study

All samples yielded no quantifiable DNA during quantification. In addition, all samples produced no allele or peak height data. Thus, stabilizer contamination was not a factor in any of the corresponding studies.

## Conclusion

Untreated samples produced a decrease in DNA concentration at all analysis intervals. Observed decreases were most likely due to DNA oxidation as stabilizers were not present to protect the samples from degradation. GenTegra™ samples yielded variable results over the course of each study. While GenTegra™ was effective at preserving DNA at elevated temperatures and high/low concentrations, it was less effective over time and in humid conditions. In addition, it had an extensive dehydration time in comparison to the other products tested. DNAsable® LD samples showed the best results in a majority of the studies. However, while DNAsable® LD effectively preserved DNA in most samples, it showed decreases in DNA concentration at elevated temperatures of 70°C and humid conditions. Trehalose samples yielded significant decreases in DNA concentrations at elevated temperatures and high DNA concentrations. However, trehalose was most effective in humid conditions. Frozen samples produced a considerable decrease in DNA yield at high concentrations; however, these results were most likely due to variability in quantification procedures.