The effect of climatic simulations on DNA persistence on glass, cotton and polyester

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Abstract

It is important to understand the variables impacting DNA persistence when considering the recovery, and evaluative interpretation, of DNA evidence from crime scenes. Whilst it is known that temperature, humidity and UV affect DNA persistence, little research has been conducted to explore these effects in a combined and controlled manner. This study includes two experiments in which a climate chamber was used to simulate climatic conditions over a repeating 24-hr period. Aliquots of ~50ng DNA were added to each substrate and DNA recovered at 0, 1, 3 and 7 days after deposition. Samples were run in triplicate, extracted and quantified. The first experiment investigated the effect of typical Southern English winter and summer days on DNA persistence on glass and cotton, with DNA being recovered by wet and dry swabs from glass and mini-tapes from cotton. The second experiment investigated the effect of typical Northern Italian winter and summer days on DNA persistence on cotton and polyester, with DNA being recovered by wet and moist swabs from both fabrics. Quantities of DNA on all substrates significantly declined over 7 days under summer conditions ($p<0.05$), and more DNA tended to persist on the fabric substrates in both studies under conditions of winter than summer. These results contribute to our understanding of DNA persistence under different climatic conditions and will help inform investigators' DNA recovery strategies.

Keywords

Trace DNA; DNA persistence; Climate; DNA-TPPR

1. Introduction

It is important to understand the variables impacting DNA persistence when considering the recovery, and evaluative interpretation, of DNA evidence from crime scenes. These variables include various climatic conditions, such as temperature and humidity. It has been observed that DNA can persist on surfaces for longer in the dark at ambient temperature (in the laboratory) than when left outside with an average temperature and relative humidity of 24.1°C, 63% (day) and 18.8°C, 71% (night) [1]. Similarly, Lee *et al.* [2] observed better DNA persistence on a range of surfaces under controlled conditions of temperatures of 19-25°C and relative humidity of 50-77%, than uncontrolled conditions of higher temperatures (22-34°C) and relative humidity (50-99%). Whilst it is accepted that temperature, humidity and UV will impact DNA persistence [3], there is a paucity of research exploring these effects and further research has been recommended [4]. Here, we investigate the effect of simulated climatic conditions on the persistence of acellular DNA on glass, cotton and polyester.

2. Materials and Methods

Fabric substrates (100% cotton swatches, 100% polyester swatches) were soaked in 25% bleach, rinsed three times with DNA-free water and UV-irradiated after drying to remove any extraneous DNA present. Glass substrates (microscope slides) were also cleaned with 25% bleach, rinsed with DNA-free water and UV-irradiated. Negative controls taken from each substrate type confirmed the absence of DNA. Stock solutions of acellular human DNA were prepared from buccal swabs of a consenting volunteer.

Ten µl aliquots of ~50 ng acellular DNA were deposited on the substrates, left to dry and then sampled after 0, 1, 3 or 7 days in a climate chamber (Memmert ICH260L), which was programmed with fluctuating temperature, humidity and daylight hours to simulate specific climatic conditions. Two experiments were conducted in triplicate; one to simulate a Southern English climate and the other to simulate a Northern Italian climate. In the first experiment, DNA was added to cotton (n=24) and glass substrates (n=24), subjected to winter (1-6°C; 80% av. humidity; 8hr sunlight) or summer

conditions (15-26°C; 64% av. humidity; 14hr sunlight), and sampled using wet and dry cotton swabs from glass and SceneSafe FAST™ tapes from cotton. In the second experiment, DNA was added to cotton (n=24) and polyester substrates (n=24), subjected to winter (0-18°C; 70% av. humidity; 12hr sunlight) or summer conditions (13-35°C; 60% av. humidity; 16hr sunlight), and sampled using wet and moist cotton swabs from both fabrics. Whilst it is routine to use mini-tapes to recover DNA from fabrics within UK casework, wet and moist swabs have been used by Australian casework laboratories for DNA recovery from fabrics. Here, wet and moist swabbing appeared to have a better recovery efficiency from cotton than mini-taping, with an average of 24% of the DNA deposited being recovered, as opposed to an average of 2% with the mini-tapes.

DNA was extracted from swabs and mini-tapes using the swab protocol of the QIAamp® DNA Investigator Kit, with an overnight incubation and elution into 35 µl. Extracts were then quantified using the Quantifiler® Human DNA Quantification Kit. Results are presented as percentages of DNA recovered at Day 0 (median of three replicates), and DNA persistence over time under each climatic simulation was analysed using Spearman's rank correlations.

3. Results

3.1 Simulated Southern England conditions

When exposed to the simulated Southern England conditions, the quantity of DNA recovered declined over the 7-day period (Fig. 1A & B). Under summer conditions, statistically significant strong negative correlations were found between time and the DNA quantity recovered from both glass (rho = -0.950; $p = 0.000$) and cotton (rho = -0.799; $p = 0.002$). Under winter conditions, a statistically significant strong negative correlation was found between time and the DNA quantity recovered from cotton (rho = -0.756; $p = 0.004$), but only a moderate negative correlation that was not statistically significant was observed for glass (rho = -0.432; $p = 0.161$). It was also observed that more DNA persisted on cotton under winter conditions than summer (Fig. 1B), but this effect was not seen with glass (Fig. 1A).

3.2 Simulated Northern Italy conditions

When exposed to the simulated Northern Italy conditions, the quantity of DNA recovered also declined over the 7-day period (Fig. 1C & D). Under summer conditions, statistically significant strong and moderate negative correlations were found between time and the DNA quantity recovered from cotton (rho = -0.864; $p = 0.000$) and polyester (rho = -0.660; $p = 0.020$), respectively. Under winter conditions, moderate negative correlations were observed between time and the DNA quantity recovered from both cotton (rho = -0.518; $p = 0.084$) and polyester (rho = -0.497; $p = 0.101$), but these were not statistically significant. In addition, more DNA generally persisted on both cotton and polyester under winter conditions than summer conditions (Fig. 1C & D).

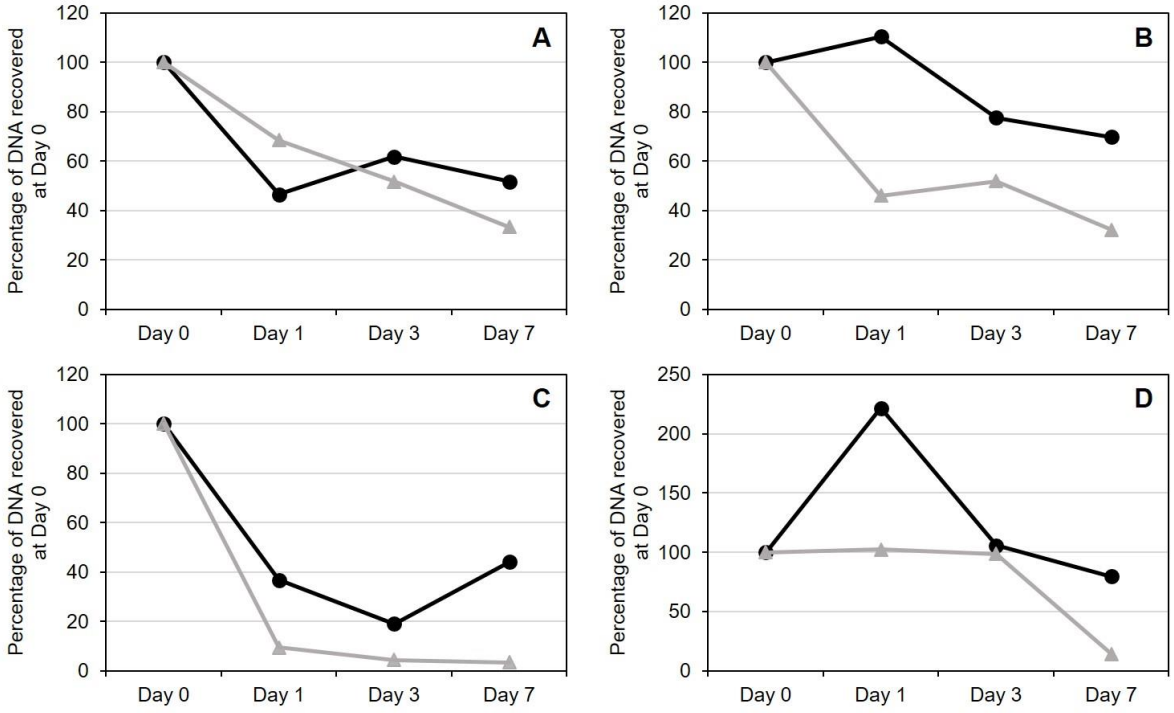

Fig. 1. Recovery of DNA from substrates exposed to simulated Southern England conditions (A & B) and Northern Italy conditions (C & D). Substrates were glass (A), cotton (B & C) and polyester (D) and were exposed to summer conditions (grey triangle and line) or winter conditions (black dot and line).

4. Discussion

Irrespective of the differences in simulated climatic conditions and recovery methods used, quantities of DNA on all substrates tended to decline over 7 days of simulated climatic conditions, in agreement with previous observations of DNA persistence on surfaces exposed to conditions of varying daylight, humidity and temperature [1, 2]. In the experiments herein, this decline in DNA over time was generally only statistically significant under summer conditions. Since the summer conditions in both experiments had lower average humidity than the winter conditions, this suggests that temperature could have a larger impact on DNA persistence than humidity and this should be investigated further.

The observed decline in DNA over time was not always consistent; for example, more than 100% of DNA recovered at Day 0 was observed for cotton under Southern England winter conditions at Day 1 (Fig. 1B) and for polyester at Days 1 and 3 (Fig. 1D). Such a phenomenon has been observed previously [1] and could result from variation in the interaction of the DNA solution with the substrate that might impact its recovery. Considering a study in which DNA was recovered from rubbed cotton and plastic substrates that were sampled either immediately after deposit or 24 hours later [5], it has been previously proposed that substrate type might impact DNA persistence [6], with DNA persisting better on plastic than cotton [5]. When considering results from both experiments herein, it appeared that summer conditions had a greater impact than winter conditions on DNA persistence on porous than non-porous substrates, as more DNA tended to persist on the fabric substrates under conditions of winter than summer. Fig. 1 also shows that DNA tended to persist better on polyester than cotton, further illustrating the potential impact of substrate type on DNA persistence; this requires further investigation.

Overall, these experiments illustrate the value of using a climate chamber to simulate climatic conditions for investigations into the impact of temperature, humidity and daylight exposure on DNA persistence. The results obtained contribute to our

understanding of DNA persistence under different climatic conditions and will help inform investigators' DNA recovery strategies.

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Conflict of interest statement

None.

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