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Minimizing hand-to-glove DNA contamination

M. van den Berge*, S. Wagner, E. Meijers, B. Kokshoorn, A. Kloosterman, M. van der Scheer, T. Sijen

Division of Biological Traces, Netherlands Forensic Institute, the Netherlands

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ABSTRACT

In this study, the Netherlands Forensic Institute's (NFI) contamination prevention recommendations to prevent hand-to-outside-glove DNA transfer were assessed for effectiveness and subsequently optimized. The results of this study support the choice of using a 0.3% sodium hypochlorite solution or commercially available RNase AWAY as decontamination reagents to clean and subsequently dry the exterior of donned gloves prior to entering the lab and/or handling items of evidence.

1. Introduction

With the increasing sensitivity of forensic DNA profiling methodologies, DNA contamination prevention procedures may need to be revisited and revised to increase stringency. One of the potential contamination risks is the hand-to-outside-glove DNA transfer induced by laboratory personnel when putting on gloves. In this study, the extent to which cellular material can be transferred to the outside of a laboratory glove upon donning was examined. Secondly, the effect of a cleaning protocol in removal of contaminant DNA of the outside of donned gloves was assessed using various types of decontamination reagents. Lastly, the potential removal of trace material after applying glove cleaning and contact with an evidentiary trace was assessed.

2. Material and methods

First, the degree of contact of bare hands to the outside of gloves in the process of donning gloves was inventoried. A fluorescent solution (GlowTec UV germ powder in mineral oil) was applied to the hands of six volunteers (forensic professionals trained in DNA contamination prevention), who were then asked to put on laboratory gloves. A fluorescent light source was used to visualize dye that had transferred to the outside of the gloves. Next, a second pair of laboratory gloves was put on (standard procedure for NFI trace recovery experts), that was again illuminated. Each donor participated three times.

Secondly, the extent of DNA transfer to the outside of the glove was examined. Six volunteers put on a glove starting with the left hand. The little finger and thumb areas of the glove were sampled using a H_2O-moistened cotton swab ("uncleaned blank controls"). Next, cellular material was applied to the remaining three fingers by rubbing on the forehead for ten seconds. The ring finger was sampled to serve as uncleaned positive control. The middle and index finger were individually sampled after cleaning using RNase AWAY™ (Thermo Scientific) and drying with a paper tissue. The same set-up was repeated for the right hand, but now 0.3% sodium hypochlorite (Actisan) was used to clean the middle and index finger (total $n = 60$).

Thirdly, it was assessed whether use of cleaning reagents could have a negative effect when cleaned gloves come into contact with evidentiary traces. A cotton cloth was divided in 45 squares (± 3*4 cm each) after which the cloth was irradiated (CL-1000 UV-crosslinker, 900 mJ/cm^2 for 60 min) and confirmed cleared from contamination nucleic acids based on DNA quantification results. Cellular material was applied to the cloth by rubbing on the forehead. Three "positive control" samples were collected by tape lifting (25x tapping). A second volunteer put on a laboratory glove, cleaned the glove with sodium hypochlorite and dried it with a paper towel. The index finger was placed on the cloth and the touch location of the cloth was sampled by tape lifting. This was repeated ten times (new glove, cleaned, press finger) using sodium hypochlorite for cleaning. The same set-up was repeated using RNase AWAY or no cleaning solution ($n = 10$ each).

Samples were subjected to DNA extraction using the QIAamp DNA Mini Kit (QIAGEN, manufacturer's instructions), DNA quantification as described in [1] and half-volume PPF6C STR-profiling (Promega, manufacturer's instructions).

3. Results and discussion

Hand-to-glove contact became visible after the first test as

* Corresponding author at: Laan van Ypenburg 6, The Hague, 2497 GB, the Netherlands.
E-mail address: m.van.den.berge@nfi.nl (M. van den Berge).

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Table 1
Average DNA concentrations and standard deviations (SD) of samples taken after donning laboratory gloves and purposely adding cellular material. Finger areas of the gloves were sampled that were either uncleaned, or cleaned with one of two decontamination reagents.

	Uncleaned (n = 24)	Sodium hypochlorite (n = 12)	RNase AWAY (n = 12)
Average DNA concentration (ng/μL)	0.031	0.002	0.003
SD DNA concentration (ng/μL)	0.023	0.002	0.006

Table 2
Average DNA concentrations and standard deviations of samples taken from a mimicked evidentiary trace after the trace came into contact with a glove that was either not cleaned, or cleaned with either of two decontamination reagents.

	Positive control (n = 3)	No decontamination reagent (n = 10)	Sodium hypochlorite (n = 8)*	RNase AWAY (n = 8)*
Average DNA concentration (ng/μL)	0.25	0.26	0.24	0.20
SD DNA concentration (ng/μL)	0.08	0.24	0.12	0.04

* Two samples appeared outliers (high DNA yield) and were excluded from the dataset as uneven distribution of cellular material to the cloth is possible.

fluorescence was observed on various parts of the donned laboratory gloves. The majority of the fluorescence was observed on the wrist area of the glove (likely from tightening the glove). Sporadic illuminations were visible on various parts of the middle hand area of the glove (by pulling the glove from the glovebox) and the finger area of the glove (by donning the glove). Occasional fluorescence was detected on the second layer of gloves indicating very limited transfer from gloved hands to the outside of a second set of gloves (data not shown).

One of the 24 “uncleaned blank control” samples taken directly after donning a glove resulted in a partial DNA profile corresponding to the wearer of the glove (41% detected alleles). This emphasizes the importance of a contamination prevention protocol. Table 1 gives an overview of the results obtained when donned gloves, to which cellular material was intentionally applied, were sampled with and without cleaning using two types of decontamination reagents. Compared to the uncleaned samples, the average DNA concentration was reduced by 94% when a glove was cleaned with sodium hypochlorite. RNase AWAY resulted in a 90% reduction of the average DNA concentration. Six of the 12 sodium hypochlorite-cleaned samples resulted in partial, degraded DNA profiles (avg. 18% detected alleles). Out of the 12 RNase AWAY-cleaned samples, five samples resulted in partially, degraded profiles (avg. 19% detected alleles).

Sodium hypochlorite and RNase AWAY are thus similarly effective in removing DNA contaminations. The third test was performed to examine that the use of these decontamination detergents does not have a negative effect when gloves subsequently come into contact with evidentiary traces. Regardless of the decontamination reagent used, all samples resulted in full STR profiles. Quantification results are presented in Table 2 which shows that the obtained average DNA concentrations are similar for all samples, regardless of touching by a cleaned or uncleaned glove. The variations observed are likely caused by the suboptimal distribution of cellular material on the cloth, resulting in relatively large standard deviations. Overall, use of decontamination reagents showed no negative effect on the DNA quantity or quality of the evidentiary trace.

4. Conclusion

We confirm [2] that hand-to-glove DNA contamination is a realistic scenario to be considered when working in a forensic laboratory. Results of this study led to an update of the contamination prevention strategies at our laboratory to reduce the potential risk of transferring self-DNA, which includes the cleaning of the exterior of donned gloves (both first and second layers in case two layers are used) with a chloride-based reagent prior to entering the laboratory.

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N/A.

Declaration of Competing Interest

None.

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