



Forensic science and the right to access to justice: Testing the efficacy of self-examination intimate DNA swabs to enhance victim-centred responses to sexual violence in low-resource environments



Lisa L. Smith ^{a,*}, Jon H. Wetton ^b, Gurdeep K.M. Lall ^b, Heather D. Flowe ^c, Mark A. Jobling ^b

^a University of Leicester, Department of Criminology, 154 Upper New Walk, Leicester LE1 7QA, UK

^b University of Leicester, Department of Genetics, Adrian Building, Leicester LE1 7RH, UK

^c Loughborough University, School of Sport, Exercise, and Health Sciences, Epinal Way, Loughborough LE11 3TU, UK

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ABSTRACT

In developed countries, DNA profiling routinely forms part of the forensic strategy in the investigation of sexual violence. Medical examinations provide opportunities for recovering DNA evidence from intimate swabs, which can be particularly probative in cases where the identity of the perpetrator is unknown and proof of intercourse between two people is required. In low-resource environments, such as developing countries, remote geographic locations, conflict (and post-conflict) affected regions and displaced communities where access to medical examinations is lacking, DNA evidence is not available to support prosecutions and perpetrators are rarely identified and held accountable for crimes of sexual violence. This paper reports the results of a proof-of-concept study testing the efficacy of a novel self-examination intimate swab designed for recovering DNA following unprotected sexual intercourse. The results of this study corroborate previous research which has demonstrated that male DNA profiles can be successfully recovered by post-coital, self-examination methods, and discusses how this novel approach could enable the integration of DNA evidence into victim-centred approaches to investigating and prosecuting sexual violence in low-resource environments. The results and discussion challenge the prevailing assumption that intimate DNA swabs must be collected by trained medical professionals in order to be of evidential value.

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1. Background

The development of DNA profiling revolutionised the investigation and prosecution of violent crime, and sexual violence in particular, in developed countries. Sexual violence is notoriously difficult to investigate and prosecute, and although DNA evidence is not helpful in every case, it can be very powerful evidence in cases where the perpetrator is a stranger and cannot be identified by the victim. In addition to identifying perpetrators and exonerating the innocent, DNA is also extremely valuable as a source of intelligence about serial assaults committed by the same perpetrator. In regions with legislation and technology to support a DNA database, comparing DNA from a crime scene with stored criminal justice profiles can identify a previously unknown suspect and in some cases significantly influence the successful detection of the crime.

In the majority of domestic (UK) sexual violence cases, the legal issue argued at trial is not the identity of the alleged perpetrator, or a question of whether sexual intercourse has occurred, but the determination of consent [1]. In such cases, DNA evidence is rarely considered probative, as it cannot be used to prove presence, or lack of, consent. However, in cases where the core issue is identity and whether or not sexual intercourse took place between two parties, DNA from semen recovered from an intimate swab of the alleged victim is often a valuable indicator of recent sexual activity with a particular individual [2,3]. DNA evidence has had a profound impact on the progression of sexual violence cases through the criminal justice system, and research has shown that the collection and examination of DNA increases the chances of a case successfully progressing to trial, and that the greatest impact is seen on cases with traditionally low detection rates and those in which the investigation did not identify a suspect [4]. One of the few cost-benefit analyses of DNA in criminal investigations noted that the most significant contribution of DNA is aiding in solving cases which would otherwise remain unsolved and also its role in identifying prolific offenders [5].

* Corresponding author at: University of Leicester, Department of Criminology, 154 Upper New Walk, Leicester LE1 1QH, UK.

E-mail address: ls149@le.ac.uk (L.L. Smith).

1.1. Domestic sexual violence investigations

In the UK, and similarly in other developed countries, there are well established protocols and guidelines for the collection, storage, and analysis of evidence obtained during investigations of sexual violence. For example, the Faculty of Forensic and Legal Medicine (UK) outlines the collection of various sources of forensic material from both victims and suspects which includes mouth swabs and rinse, fingernail clippings, urine, and external and intimate swabs [6]. These forensic examinations are conducted by a trained medical examiner, and 'early evidence kits' can also be administered by investigating police officers, which maximise the chances of recovering forensic evidence that may be lost before the victim can undergo a full medical examination [7,8]. Research has demonstrated the effectiveness of early evidence kits for recovery of spermatozoa from urine samples and vulval gauze wipes, and highlighted the value of these kits in cases where remote locations delay access to a full medical examination [8]. Although best-practice guidelines emphasise that early evidence kits should ideally be used alongside full medical examinations, these kits provide an alternative for alleged victims who refuse an invasive medical exam on cultural or religious grounds. As a result of these available methods, DNA is routinely included in the forensic strategy of sexual violence investigations in developed countries.

Recent research literature, and current practical guidelines endorsed by advocacy organisations, focus on the importance of a victim-centred approach to the investigation of sexual violence [9]. This shift has highlighted the need to ensure that victims can access justice and that the criminal justice system takes steps to treat victims with respect, sensitivity and dignity in order to reduce the negative impact of secondary victimisation. As a result, the availability of resources such as Sexual Assault Referral Centres [10] and organisations such as Rape Crisis England and Wales [11] aim to support victims of sexual violence at every stage of the criminal justice process. However, even with these measures in place to support victims, reporting rates for sexual violence remain very low [12] and the criminal justice system is regularly criticised for its treatment of victims during the investigation and prosecution of cases.

1.2. Sexual violence investigations in low-resource environments

Despite the routine use of forensic science, particularly DNA evidence, in developed countries to support sexual violence prosecutions there are millions of victims worldwide who's right to access to justice is denied by a lack of access to evidence. Many of these victims are located in what this paper refers to as 'low-resource' environments, which includes developing countries, remote geographic locations, conflict (and post conflict) affected regions, and displaced communities. Under these circumstances what has been referred to as a 'culture of impunity' exists, where prosecutions are so rare that there is no legal deterrent for perpetrators and sexual violence has become a way of life for women and girls [13].

There are complex cultural, political, and social factors which play a role in the perpetration, reporting, investigation, and prosecution of sexual violence in various regions and deeply embedded gender inequality is at the root of the issue [13]. Although national courts are largely responsible for the prosecution of cases of sexual violence, where these crimes are perpetrated in the context of armed conflict they can fall under the jurisdiction of international law and be prosecuted as war crimes, crimes against humanity, and even genocide [14]. Criminal tribunals which preceded the establishment of the International Criminal Court (ICC) in 2002, such as the International Criminal Tribunal for the former Yugoslavia (ICTY) and the International Criminal Tribunal for Rwanda (ICTR), have had some success in prosecuting sexual violence cases. However, the lack of forensic evidence to support these prosecutions has remained a key barrier in many cases [15].

Access to DNA evidence in low-resource environments is a key issue for discussion, particularly as courts (e.g. in Kenya, Uganda, and India) have been setting dangerous legal precedents by requiring DNA evidence to support prosecutions. Therefore in these circumstances victims face an impossible situation where DNA evidence is required to prosecute cases and promote justice, but the facilities and trained professionals to collect DNA evidence are often not available. In addition to the lack of availability of medical staff and facilities, other barriers to justice exist in these regions, such as cultural norms that deter women from seeking invasive medical exams (particularly where it is not possible to accommodate a victim's expressed preference for a female medical examiner), as well as a lack of security and legal safeguards for victims. These circumstances create a 'culture of impunity' for perpetrators of sexual violence, and support continued cycles of violence.

There are a number of victim-centred programmes operating in low-resource environments that aim to support victims and tackle the culture of impunity. Many of these are implemented by human rights organisations and NGOs, and provide guidelines for supporting victims, as well as clinics where victims can access psycho-social support as well as advocacy services and awareness campaigns. Whilst these approaches are invaluable for supporting victims, they do not directly address the lack of prosecutions or provide access to forensic evidence collection. A recent example is the International Protocol on the Documentation and Investigation of Sexual Violence in Conflict published by the UK Foreign and Commonwealth Office [16] which provides valuable guidance for the interviewing of victims and witnesses but neglects the possibility of collecting forensic evidence to support prosecutions.

1.3. Expanding the use of forensic science to address human rights violations

Gender inequality continues to be one of the world's most enduring violations of human rights, and sexual violence is one of the most damaging manifestations of gender inequality. Violence against women and girls not only has devastating effects on half of the population, but also adversely impacts all aspects of the economic, social, and political realm [13]. Men and boys are also frequently victims of sexual violence, and should not be overlooked in these discussions; however, women and girls are disproportionately at risk of victimisation.

Forensic science has traditionally been utilised in investigations of war crimes and crimes against humanity, but this has focused almost exclusively on the use of pathologists, archaeologists, and anthropologists who address questions of manner of death, assessment of injuries resulting from torture, and identification of the remains of deceased individuals [17]. However, what is lacking is an attempt by the forensic science community to improve access to evidence that can support prosecutions for sexual violence in low-resource environments. Research and innovation in this area could be vital for closing the justice gap for victims, ensuring that individuals' right to access to justice is upheld, and tackling the culture of impunity by providing high-quality evidence to support prosecutions in these regions.

Although the domestic forensic science strategy in the UK is largely focused on expanding digital forensic capabilities and implementing a more coherent and cost-effective national approach to forensic science delivery [18], the UK Aid strategy outlines the Government's priority areas internationally [19]. The international strategy identifies investment in tackling crime and security issues, as well as prioritising the rights of girls and women, in striving to address extreme poverty and assisting the world's most vulnerable people [19]. This paper argues that research and innovation that can make DNA evidence accessible to legal systems in low-resource environments in an effort to support sexual violence prosecutions, is one way that the forensic science community can contribute to the UK Government's global initiatives.

In order to challenge some of the existing barriers to DNA evidence outlined in Section 1.3, the following sections of this paper present a novel DNA recovery method and the results of a small-scale proof-of-

concept study which tested the effectiveness of self-examination, intimate swabs for the collection of DNA following sexual intercourse.

1.4. A novel approach to DNA evidence recovery for low-resource environments

As outlined in Section 1.2, domestic investigations of sexual violence can benefit from DNA evidence due to the accessibility of trained police officers, medical facilities, and support services. Indeed, when these services are available to victims these are the ideal conditions under which to collect forensic evidence [8]. However, in situations where medical facilities and trained staff do not exist or are not accessible, recovery of DNA evidence is currently not available to support investigations and prosecutions.

The aim of this paper is to question the prevailing assumption that intimate DNA recovery swabs can only be administered by medical professionals during sexual assault examinations. The legal precedent for admissibility of DNA evidence collected from alleged victims using early evidence kits demonstrates that it is not a legal requirement for evidence to be recovered by a medical professional. However, there are limitations to the probative value of evidence collected using current early evidence kits, including the fact that urine samples and vulval gauze wipes cannot provide evidence of penetration (unlike intimate forensic specimens collected during full medical examinations) [8].

The approach taken in the current study was to extend the capabilities of current early evidence kits, in order to enable the recovery of DNA evidence from a self-examination intimate swab. There are numerous examples of validated self-examination intimate swabs which are routinely used to diagnose sexually transmitted pathogens such as gonorrhoea [20] and chlamydia [21]. Typically, users are instructed to insert these swabs 2–4 cm into the vagina - similar to the low vaginal swab administered during a sexual assault examination [22]. High vaginal swabs conducted during forensic medical examinations can be obtained with the use of a speculum in conjunction with an endocervical swab; however, 'blind' high vaginal swabs can be obtained without a speculum. These 'blind' high vaginal swabs are typically inserted 4–6 cm into the vagina and are effective for the collection of semen, contact DNA, lubricant, and saliva [23].

The goal of the current study was to test the effectiveness of self-examination, high vaginal swabs for collecting DNA after unprotected sexual intercourse using two swab techniques. The first technique involved self-examination using the same procedure as existing 'blind' high vaginal swabs, and the second method utilised a tampon applicator to assist the user in guiding the swab into the correct position. The following section describes the methods and results of this proof-of-concept study.

2. Materials and methods

Participants who volunteered for this study were required to be heterosexual couples engaged in consensual, unprotected intercourse. For the purposes of this study, 'unprotected' intercourse referred to an

absence of the use of condoms; the use of oral contraceptives and IUDs was permitted. Informed consent was required from both the male and female participant, and couples were given a participant kit which included: a reference buccal swab for each participant (male and female), two versions of the self-examination intimate swab (swab A included a tampon applicator, and swab B was applicator free - see Fig. 1a and b), a participant information sheet, consent forms (signed by both parties), full instructions describing how to use each version of the swab, and a brief questionnaire about ease of use.

One reference buccal swab was requested from each of the male and female donors as well as two post-coital intimate swabs, one collected with each version of the swab following separate occasions of intercourse. Participants were asked to use the self-examination swab 12 to 36 h after intercourse, in order to mimic realistic reporting delays experienced in cases of sexual violence. Participants were asked to indicate the time since intercourse for each of the samples submitted on the paperwork returned with the samples.

2.1. DNA extraction

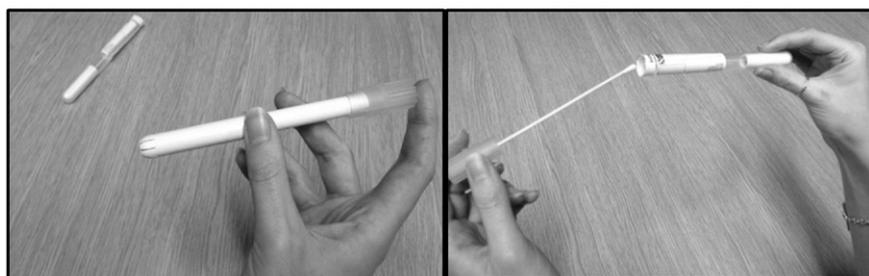
The buccal and intimate swab shafts were broken near the swab head and placed in 1.5 ml micro-centrifuge tubes with 400 µl sterile water. After brief vortexing of the tubes, 200 µl water was transferred for DNA extraction. The robotic QIAGEN QIAcube workstation was used for automated extraction of DNA from both buccal and intimate swab samples using a non-differential protocol. Extracted DNA was eluted in 100 µl AE buffer.

2.2. DNA quantification

DNA was quantified using the Quantifiler Trio DNA Quantification Kit (Applied Biosystems, ThermoFisher Scientific) on an Applied Biosystems 7500 Fast Real-Time PCR System. Quantifiler Trio uses TaqMan real-time PCR technology to simultaneously quantify the total amount of amplifiable human DNA and the proportion of human male DNA in a sample (Table 1) by targeting both Y-chromosomal and autosomal regions.

2.3. Yfiler Plus STR profiling

Y-STR rather than autosomal STR profiling was applied as it can yield a male specific profile even in the presence of a large excess of female DNA and is therefore more appropriate for samples collected following prolonged time since intercourse (TSI). Whilst autosomal profiling has the potential to identify assailants from national DNA databases these are unlikely to exist in low-resource countries and we envisage the most likely use of DNA to be linking serial assaults and comparisons against specific individuals suspected of sexual offences. The high frequency of multi-male rapes in low-resource settings also favours the adoption of a Y-STR approach and we intend to explore application of rapidly mutating Y-STR (RM Y-STR) multiplexes combined with Next



a) Swab A, with tampon applicator

b) Swab B, standard swab with no applicator

Fig. 1. a) Swab A, with tampon applicator. b) Swab B, standard swab with no applicator.

Table 1
Summary of results for eight participant couples.

Donor couple	Swab	TSI (hrs)	M:F ratio	Total PCR input (pg)	Male PCR input (pg)	Profile quality
1	A	23	1:16	614	38	FP
	B	32	1:20	2079	104	FP
2	A	23	1:11	689	63	FP
	B	23	1:15	961	64	PP (–1)
3	A	0.5	1:2	858	429	FP
	B	0.5	1:3	2689	896	FP
4	A	12	1:43	26,762	622	FP
	B	22	1:19	2781	146	FP
5	A	13	1:250	3741	15	Failed
	B	34	1:1447	2838	2	Failed
6	A	12	1:54	1902	35	PP (–1)
	B	15	1:2383	12,088	5	PP (–12)
7	A	24	1:5	1123	225	FP
	B	24	1:4	606	152	FP
8	A	25	1:8	1215	152	FP
	B	N/A ^a	–	–	–	–

^a Couple did not use swab B, so no results recorded.

Generation Sequencing (NGS) as a means of deconvoluting complex mixtures from groups of potentially related males.

The AmpFISTR® Yfiler® Plus Kit (Thermo Fisher Scientific) is a 6-dye multiplex assay for short tandem repeats (STRs) that allows amplification of 27 Y-STR markers from samples. Yfiler Plus profiling was carried out according to manufacturer's instructions on all male buccal DNA samples and on DNA extracts from all intimate swabs. Following PCR, products were separated by capillary electrophoresis on an ABI 3130xl Genetic Analyzer. The resulting peaks were then inspected and analysed using GeneMapper software version 4.0. Y-STR profiles generated from the intimate swabs were compared to the reference genotypes obtained from male buccal swabs.

3. Results

Seven donor couples returned both swab A and B, with and without tampon applicator respectively. One participant returned only intimate swab A. Three women indicated on the questionnaire that they preferred swab A and four preferred swab B. The time since intercourse (TSI) for intimate swabs A and B were reported and these varied among the couples (Table 1). The swab with the applicator was not significantly better in recovering DNA or more comfortable than the applicator-free swab.

The total amount of DNA recovered ranged from 2.6 µg to 57 ng with male:female (M:F) DNA ratios between 1:2 and 1:2383. Neither total DNA recovered (ng) nor the quantity of male DNA differed significantly between swab types or donors whereas M:F ratio did vary between couples ($P = 0.001$, OneWay ANOVA). Other than swabs collected by the donor couple who had both the lowest mean M:F ratio and lowest mean male DNA recovery which gave no profile, the least complete profile (15 out of 27 alleles) was derived from a 5 pg male input, roughly equal to that from a single cell, and a M:F ratio of 1:2383. Full or near complete profiles lacking a single allele were derived from all other intimate swabs from other donor couples, including Couple 2 in which the male partner was reported to be vasectomised. Yfiler Plus profiles obtained from male reference buccal swabs were fully concordant with the male profiles from the intimate swabs in all cases (Table 1).

4. Discussion

This paper has considered the inherent difficulties with the recovery of DNA evidence in cases of sexual violence encountered in a variety of low-resource environments, including developing countries, remote geographic locations, conflict (and post-conflict) affected regions and

displaced communities. In the absence of high-quality evidence to support prosecutions, sexual violence perpetrators in these regions are rarely identified, which provides no deterrence for these crimes, and victims continue to be denied their right to access to justice.

To date, there has been no research and development focusing on innovative methods to make DNA evidence available in low-resource regions, particularly in situations where a trained medical professional is not accessible to carry out a forensic medical examination. This paper has presented results from a proof-of-concept study demonstrating that innovative, self-examination intimate (high vaginal) DNA swabs can be a viable source of DNA up to 32 h after unprotected sexual intercourse. These self-examination swabs could therefore potentially be added to the components currently provided in early evidence kits, thereby improving the probative value of DNA recovered using these kits.

The concept of self-examination intimate swabs is not new (e.g. low vaginal swabs for diagnosing sexually transmitted infections); however, the design and testing of a prototype self-examination high vaginal swab for DNA collection is novel (patent pending). Although the results of this small-scale initial study seem promising, further work on the design and validation of the swab is required and currently underway by the authors. In particular, the applicator feature of swab A in the current study is considered to be of particular importance in the context of forensic self-examination swabs for two reasons. Firstly, the use of a tampon-style applicator assists the user during the insertion of the swab both protecting the user from injury and ensuring that the swab is positioned correctly to maximise the likelihood of recovering DNA. Secondly, the applicator protects the end of the swab during insertion and removal, which minimises the chances of contamination due to the swab accidentally coming into contact with other surfaces. This is an important consideration when interpreting the results in the context of a case of alleged sexual violence involving penetration, particularly regarding the probative value of the DNA recovered.

The impact of this novel approach to the collection of DNA evidence would ultimately be measured by the contribution of the recovered evidence to improving the reporting and prosecution rates for sexual violence in low-resource environments. There is still substantial work to be done on the validation of this technique, but equally important is consideration of whether the legal and cultural framework exists to support such forensic innovation. This includes the existence of robust sexual violence legislation, and consideration of regional variations in laws governing the admissibility of DNA evidence at trial, legislation enabling DNA samples to be obtained from suspects, and the establishment of DNA databases.

One legal challenge that may arise is whether false rape allegations may be facilitated by the use of a self-examination intimate swab by allowing a complainant to fabricate evidence. In view of this concern, it is important to note that while the prevalence of false rape allegations is difficult to establish [24], studies that have used the most reliable methodologies suggest that false rape allegations are relatively rare, with an estimated 3% to 6% of cases being false reports [25,26]. These very few false allegations of rape are rarely (if ever) made against an unknown perpetrator, and in cases where the accused is a stranger to the victim it does not seem logical to maintain such scepticism of the complainant. Indeed, in these sorts of stranger rape cases evidence such as semen stains on bedding or underwear would be considered of probative value, which is more susceptible to contamination and tampering than a self-examination intimate swab. Finally, it is important to remember that DNA evidence constitutes one element of a case against a defendant. In all cases in which DNA evidence is introduced, the totality of evidence interpreted in the context of the allegation must be considered when investigating and prosecuting cases.

Despite the complexities of multi-sectoral progress towards improving the use of forensic DNA in cases of sexual violence, there has been substantial progress in regions such as Kenya where intense lobbying and a shift in political will has created opportunities for improvements

in recent years. This has included the implementation of the Sexual Offences Act in 2006 [27] which reconceptualises sexual violence as crimes of *violence* rather than crimes against *morality*, which places increased emphasis on prosecutions and justice [28]. This Act also enables the collection of DNA samples from suspects as well as the storage of DNA profiles from convicted perpetrators on a database. Despite providing a legal environment which enables the use of forensic DNA to support prosecutions, a lack of access to trained police and medical practitioners continues to limit the collection of DNA evidence from victims [29]. Therefore innovative collection techniques, such as the one outlined in this paper, have the potential to help victims overcome these barriers to accessing justice.

The legal admissibility of DNA evidence obtained from the novel, self-examination intimate swabs described in this paper is an important consideration and admissibility criteria and relevant legal precedents in both national and international criminal courts will provide guidance for judicial decisions. The Kenyan legal context provides a useful case study for this discussion, due to the endemic sexual violence issue in Kenya and the resulting changes to the legal framework which have provided a mechanism for DNA evidence to feature more effectively in investigative and prosecutorial strategies [28]. Rules of evidence in Kenyan criminal courts allow for qualified experts to provide testimony about forensic analyses, including DNA which is widely accepted in criminal proceedings in cases of both crimes against individuals and wildlife [30].

Therefore challenges are unlikely to be focused on the admissibility of the DNA evidence itself, or the expert testimony associated with it, but rather may be argued on the grounds of the novel collection method proposed by this paper. Although no case law exists for this evidence collection technique on which to draw conclusions, the legal admissibility of DNA evidence collected by early evidence kits (e.g. in the UK) provides support for the use of evidence collected by untrained persons in criminal prosecutions [31]. The ability of agencies to ensure robust chain of custody for swabs will also be of concern during the implementation of this new technique, and the researchers will continue to work closely with stakeholders to develop guidelines outlining acceptable principles of collection and storage of evidence. The importance of considering the relevance and probative value of any DNA evidence obtained using the technique proposed in this paper (or indeed any available technique), in relation to other evidence provided at trial cannot be overstated. Therefore the usefulness of this novel technique in criminal courts, regardless of the geographic region, will rely on the successful integration of high-quality forensic evidence collected, with reliable and detailed victim testimony, combined with contextual information about the alleged offence.

This paper has argued that in order to overcome complex barriers to the use of forensic DNA in cases of sexual violence in low-resource environments, forensic science innovation which challenges domestic assumptions about the collection of evidence may provide access to justice for victims globally. Although the concept of self-examination high vaginal swabs outlined in this paper is new, the legal admissibility of early evidence kits and diagnostic value of self-examination low vaginal swabs in other contexts provides a starting point for discussion about the utility of this approach in a forensic context. In the vast majority of cases a full forensic medical examination should be considered a priority; however, where such an examination is not available or cannot be safely accessed by victims, this novel method may provide an alternative means of recovering valuable DNA evidence to support prosecutions for sexual violence.

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