

**A study to investigate the persistence of polydimethylsiloxane
and to determine the detection of polydimethylsiloxane in a
body fluid matrix following Cellmark's Sperm Elution[®].**

By

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A thesis submitted in partial fulfilment for the requirements for
the degree of MSc (by Research) at the University of Central
Lancashire.

May 2018

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Abstract

Lubricant trace evidence stands as associative evidence in sexual assault cases when there is an absence of semen. Polydimethylsiloxane (PDMS) is a substance applied as a lubricant by the majority of condom manufacturers worldwide. As the awareness of sexually transmitted infections has amplified immensely over recent years, the number of perpetrators wearing condoms during sexual assaults has risen also.

Accounts of false positive results arising from lubricant analysis in alleged sexual assault cases are becoming progressively common. This led to suggestions that PDMS may be unintentionally transferred onto blank cotton swabs during manufacturing. This project accumulated a selection of blank swabs from vaginal swab kits and sexual assault early evidence kits. All swabs were extracted with isooctane then analysed using an FTIR microscope and u-ATR accessory. No PDMS was detected on any swabs examined.

Recently, the lubricating jelly used in sexual assault kits has been replaced with a more cost-effective option. Aquagel and OptiLube are the lubricants now found in sexual assault kits. However, manufacturers have failed to report the ingredients incorporated within these lubricants. If PDMS was to be present, this could lead to false positive results in sexual assault cases. A sample of Aquagel and OptiLube were examined using the u-ATR, and both substances were revealed to be water-based lubricants containing no PDMS.

Previous research has indicated that 0.125µl is currently the smallest volume of PDMS detected using FTIR analysis. However, the quantity of PDMS expected on intimate swabs following sexual intercourse with a silicone lubricated condom is unknown. Extracting various volumes of PDMS from swab heads, and analysing using FTIR could potentially produce a quantitative curve. This could allow the volume of an unknown sample of PDMS remaining on intimate swabs to be identified. Results indicated that there is a positive correlation between the amount of PDMS detected and the intensity of the peaks on the spectrum. However, examining samples using a u-ATR and FTIR microscope is not a fully quantitative method due to multiple

external factors. For example, the technique is unable to include the whole sample to allow complete analysis.

In current forensic casework, forensic experts don't usually suggest lubricant analysis if the time that the intimate swabs were taken exceeded 24 hours since the alleged sexual assault. Previous research implied that PDMS was able to be detected in the vagina 48 hours after intercourse; however this research is very outdated. This was explored by participants undertaking sexual intercourse with their partner using a PDMS lubricated condom. Volunteers were then asked to take blind vaginal swabs at specific time intervals. Results indicated that PDMS was able to be detected on all swabs after 6 hours, and only 33% of swabs taken at 12 hours. No PDMS was recovered after 12 hours.

Up to this stage in the project, it can be confidently stated that volumes as low as 0.125µl are able to be detected on the swab head extracts using u-ATR after Sperm Elution. However, it is currently unknown whether the addition of multiple body fluids, in addition to semen, affects whether PDMS is able to persist on the swab head. Eight different body fluid combinations of semen, blood, saliva, vaginal material and faecal material were seeded onto blank cotton swabs alongside 2µl of PDMS. Swabs were then sperm eluted and extracted with isooctane. PDMS was able to be detected using the u-ATR on all of the swabs examined. This information will aid in the interpretation of casework results involving scenarios whereby multiple body fluids may be present.

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Chapter 1: Literature Review

1.1 Sexual Assault Statistics

In society today, sexual assaults are regrettably becoming more of a frequent social deviance. Worldwide, approximately 1 in 5 women and around 1 in 71 men will experience a sexual assault during their lifespan [1]. Rape is the foremost major offence within crimes associated with sexual assault. A study compiled by the Home Office conveyed that 23% of women suffered sexual assault; however, only 5% of the women were victims of rape [2]. It is predicted that fewer than 37% of reported rapes resulted in prosecution, and in these cases less than half of the suspects were convicted of the crime [3]. Yearly in the United Kingdom, roughly 500,000 adults suffer a form of sexual assault and approximately 85,000 women and 12,000 men are raped. Furthermore, figures indicate that roughly 90% of rape victims have previously encountered the offender prior to the assault [4]. Roughly £8.5 billion is consumed yearly through examining sexual assault cases; therefore additional tools are required to deliver comprehensive scientific evidence to convict committers of crime [2].

If an alleged sexual assault has occurred, it is crucial that all physical evidence is collected by medical and forensic examiners. It is essential that all intimate swabs are taken rapidly from both the victim and the accused, if feasible, to maximise the likelihood of any forensic evidence being detected. Sexual Assault Early Evidence Kits, provided by Scene Safe, are used by medical examiners to recover forensic evidence. Equipment for the collection of semen, vaginal cells, lubricants, urine and blood is all available within the SAEE kits [5]. However, many victims tend to report the crime long after the alleged assault was committed. As a result, the evidence recovery may be compromised due to too much time having passed, causing the traces of biological material or lubricant to have degraded or been washed away [4].

1.2 What is a Lubricant?

A lubricant is a substance which is applied in order to decrease the measure of friction between two surfaces upon contact. Often a greasy liquid, oil or thick gel, lubricants can be incorporated into many different scales including industrial applications to lubricate heavy machinery, medical roles to enhance the mechanism of artificial joints, to prevent food sticking to cooking appliances and for personal use to assist penetration during sexual intercourse [6].

Focusing specifically on lubricants associated with sexual intercourse, there are three main categories employed to facilitate penetration. These are condom lubricants, personal lubricants and improvised agents including Vaseline (petroleum jelly) and body oils [7]. Condom lubricant formulations can be grouped into four key classes which are silicone based, water based, oil based and organic [1]. The array of various lubricant types allows for differentiation between each compound. The chemical structures of each lubricant type are very different, therefore valuable in directing towards a particular lubricant class. This characteristic can be interpreted to allow the identification of an unknown lubricant sample via investigation using analytical techniques [8].

1.3 DNA Evidence

The first and foremost beneficial evidence type able to be recovered following an alleged sexual assault is biological evidence. To attain a positive DNA match against a significant biological transfer is extremely likely to lead to the successful prosecution of a suspect. DNA evidence is unique to each individual, with the exclusion of identical twins. However, even in this case, they possess different fingerprints to allow individualisation [9]. This distinctive profile is specific to each individual and is very effective at being able to identify a person. This feature can be applied to determine an association between the scene of a crime and an individual. This is principally the circumstance during sexual assault cases whereby biological traces are regularly excreted, hence offering an increased likelihood of DNA transfer. If biological evidence is detected in these situations, reliant upon the quality of the

subsequent profile, there is a strong likelihood the source of its origin may be verified. If this is accomplished, an extremely strong link could be provided between an individual and the scene of crime. As a result, a prosecution can be made and a perpetrator is brought to justice.

Brauner and Gallili [10] investigated a forensic case whereby both a condom and DNA evidence was present. A young girl was taken to an isolated area by the suspect, whom she had met beforehand, whereby she was supposedly raped with the use of a condom. The condom was discarded following the alleged assault and was recovered from the scene of the assault by police two days later. The victim underwent routine medical examinations. Furthermore, the condom was examined to reveal the presence of blood on the outer surface, semen on the inner surface and the presence of 5 pubic hairs. The blood and semen allowed a DNA profile to be determined. All of the pubic hairs attached to the condom matched the samples provided by the victim. The blood on the outer surface of the condom corresponded to the victim, which aided in corroborating her version of events claiming she was a virgin prior to the assault. The recovered semen was found to match that of the suspect, therefore providing confirmatory evidence that the suspect had sexual intercourse with the victim. The suspect was later found guilty of rape.

Although DNA is the most confirmatory result when investigating a sexual assault case, fingerprint evidence can provide answers of valuable significance with the absence of, or alongside biological support. Fingerprint evidence together with lubricant traces can not only place an individual at a scene, but also suggest the use of a condom if the lubricant traces match those from the intimate swabs from the victim. Upon handling a condom, the outer lubricated coating is transferred onto the fingers of the perpetrator. The offender could then potentially leave contaminated finger marks on objects and surfaces at the crime scene [2]. Bradshaw employed Matrix-Assisted Laser Desorption/Ionisation MS Imaging (MALDI MSI) for the analysis of condom lubricant contaminated fingerprints. However, if a fingerprint is recovered from a scene within a lubricant, fingerprint evidence is favoured over lubricant analysis as fingerprints are unique to each individual, even identical twins

as previously noted [11]. To enhance the findings, if a non-destructive fingerprint recovery technique is utilised, lubricant analysis could be conducted consecutively.

1.4 Swab Examination for Lubricants

If DNA evidence is anticipated to be present on a swab recovered from a sexual assault victim, it is highly unlikely a portion of the swab head will be offered for lubricant analysis. This is because DNA evidence can deliver definite confirmation as to who executed the crime as DNA is unique to each individual [12]. Alternatively, lubricant traces only act as associative evidence, determining whether an offence was committed. At present, there is no standard or routine procedure for lubricant examinations within forensic laboratories nationwide. In cases whereby lubricant analysis is required, the swab head is cut into two portions, one for DNA analysis and the other for lubricant analysis. The swab is then extracted in a non-polar solvent such as isooctane or cyclohexane [13]. Unfortunately, cutting the swab can compromise any potential biological material present as each substance may be unevenly distributed on the swab head [14]. The decision as to whether lubricant analysis is required depends upon the likelihood of the presence of DNA evidence and how beneficial the presence of a lubricant would be towards the case.

An alternative way allowing both DNA and lubricant analysis to be investigated would be to primarily assess for the presence of semen on the swab, followed by the extraction of lubricants. However, many extraction techniques permitting DNA analysis obliterate lubricant traces which may be present on the swab. The 'Christmas Tree' staining method comprising the use of Nuclear Fast Red and Picroindigocarmine allows for the identification of sperm heads within a sample [15]. Regrettably, this test is of a destructive nature indicating further analysis for lubricant traces is prohibited unless only a section of the swab was provided for the analysis of semen [16]. J.P. Allery [17] directed a study which disclosed that only 25% of smears from women who had unprotected sexual intercourse within the past 24 hours displayed spermatozoa. This indicates the importance of providing a portion of a swab for lubricant analysis as the confirmation of sexual intercourse could be

achieved even with the absence of semen, subject to false positives or alternative explanations for the ingress of components of lubricants.

1.5 The Use of Condoms in Sexual Assaults

The presence of seminal fluid on high vaginal swabs can act as incriminating evidence in sexual assault cases demonstrating that penetration occurred [18]. While DNA based evidence possesses the utmost value in sexual assault cases, it is not always present, therefore other forms of evidence should be considered. Condom lubricant residues act as associative evidence when no seminal fluid is able to be recovered. It is therefore essential any lubricant traces are identified as they could contribute as circumstantial evidence towards a sexual assault case. Furthermore, it may be feasible to establish whether an offence was committed despite the absence of DNA confirmation [19].

Statistical evidence has indicated that the quantity of sexual assault crimes involving the use of a condom has dramatically amplified over recent years, and is projected to continue increasing [7]. In America, it has been estimated that approximately 15% of perpetrators resort to wearing a condom when committing a sexual assault [20]. In addition to the exceptionally reduced likelihood of depositing any biological material at a scene, multiple justifications are available as to why many offenders opt to wear a condom during sexual assaults [21]. A plausible explanation for this may be the recent advances in DNA analysis and the increased awareness of sexually transmitted infections through the media, especially human immunodeficiency virus whereby an inclusive cure has yet to be discovered [10]. Furthermore, condoms offer protection against other common sexually transmitted diseases including chlamydia, gonorrhoea, vaginitis and syphilis [21].

Criminals, repeat offenders and individuals with a DNA profile previously documented on a national database, in particular, may choose the use of a condom in order to prevent their identity being detected and being connected to manifold crime scenes [20]. Despite possessing awareness regarding the prevention of leaving behind biological evidence, offenders often overlook the transmission of other

substances which can act as valuable evidence for securing a conviction, including fibres, fingerprints and lubricant traces when a condom is involved [22]. Criminals with greater intelligence may vary the type and brand of the condom used whilst committing the offence with the attempt to avoid associations linking their crimes.

Locard's exchange principle states that 'every contact leaves a trace' and this is especially the case in sexual assault crimes. The physical contact concerning the assailant, victim and the scene potentially instigates the exchange of traces of evidence including condom lubricants [23]. These are chemical substances applied to the outer surface of condoms to minimise the friction experienced during penetration. The presence of condom lubricants, particularly PDMS, can aid in determining the outcome of a sexual assault case. Confirmation can be contributed as to whether a condom was used or can be entirely omitted from the case, furthermore corroborate either the victims or suspects account of the incident [24].

The absence of genetic material being transferred from the offender stemming from the dramatic rise in condom use has instigated a negative effect on the ability of forensic scientists to learn the identity of the criminal. Fortunately, it has amplified the number of assignments whereby a condom lubricant is left as prospective trace evidence. If biological material is unavailable, lubricant evidence may perhaps be the most valuable piece of evidence available for the case [25]. This prompted research with respect to the identification and examination of condom lubricants and their proficiency to be used as trace evidence in sexual assault cases.

Lubricant traces act as circumstantial evidence whereby a suspect can be linked to a crime scene if the PDMS recovered from the suspect matches that found on the victim or at the scene. Furthermore, if the recovered evidence is obtained from the victim internally, the probability of a conviction rises significantly as it establishes sexual intercourse must have occurred in order for the PDMS to enter the body [26]. Alternatively, if the victim undertook voluntary sexual intercourse with a condom up to a few days prior to the alleged assault, any detected lubricant traces could have resulted from the earlier intercourse. This information signifies the importance of evaluating each case independently and assembling together as many facts as

possible prior to the alleged assault to inhibit the occurrence of any false prosecutions [7].

1.6 Components of a Condom

Research conducted by Blackledge [27] in 1999 revealed that 6-9 billion condoms were used worldwide. The quantity of condoms mass-produced in Germany between 1978 and present has practically doubled. The augmented marketing and improved availability may have additionally contributed towards the £21.6 million growth in sales between 1990 and 1995. It can be assumed from this study that the sum of sexual crimes involving the use of a condom will similarly rise based on the progressively increasing use of condoms worldwide.

The majority of condoms available internationally constitute of a polymer of isoprene, a material forming the basic structure of both natural and synthetic rubbers. This is commonly referred to as latex rubber. The natural elastomer latex is a very cost-effective and successful material in preventing the transfer of semen to the vagina. Alternatively, materials including lambskin, polyurethane and synthetic elastomers are often incorporated as alternative materials for individuals with latex allergies. However, research discovered that non-latex condoms express a lower performance and are more likely to result in breakages during intercourse.

Latex condoms hold numerous varying structural features including shape, size, colour, texture, sensitivity, thickness and lubricant type. Furthermore, a variety of flavours including strawberry, chocolate and cherry are sometimes applied to the condom surface to enhance customer appeal, and different flavouring agents, including maltol, can be incorporated to create specific tastes and aromas of condoms [24]. In addition, substances containing menthol produce a change in sensation [1]. However, the lubricant remains the foremost characteristic which can benefit a sexual assault case where the condom is absent. Condoms are coated with substances which possess suitable lubricating properties. Polydimethylsiloxane (PDMS) is an ideal constituent for a condom lubricant as the latex rubber will not absorb the silicone, resulting in the rubber remaining undamaged. Approximately

100-500ml of lubricant is applied to each condom during manufacturing, hence it is likely to be detected after use [25]. Previous research identified that PDMS acts as the lubricant on 90% of latex condoms in the United Kingdom [8]. Furthermore, in New Zealand, a sample of 25 different types of condoms were obtained and 88% were found to be lubricated with PDMS [28]. Cho & Huang extended this international research by conducting a study in Taiwan which involved accumulating a total of 35 different condoms. The outcome disclosed 91% of the samples were lubricated with PDMS [29]. This suggests that PDMS is likely to be the most common lubricant applied to condoms worldwide, not just in the United Kingdom. Detection of trace samples as small as 0.5 μ l of PDMS have been published in literature, and 0.125 μ l have been recorded at Cellmark using Fourier Transform Infra-Red Spectroscopy (FTIR) [30].

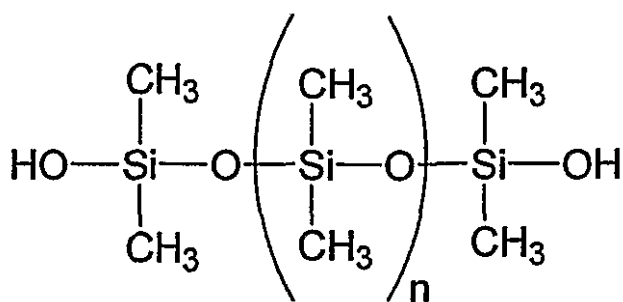
Condom lubricants contain a selection of additives including spermicides and starch particles. Spermicides are chemicals which digest the cell membranes of spermatozoa to minimise the likelihood of conception [31]. Nonoxynol-9 is a clear, colourless liquid and is the most common spermicide incorporated into lubricants [26]. Being an alkyl phenol ethoxylate attached to a benzene ring with a 9 carbon alkyl chain, Nonoxynol-9 is easily identified and the source is more accurately determined due to few alternative uses in commercial products [32]. However in recent studies, a minimal percentage of condoms on the market contain this substance due to reports suggesting spermicides don't protect against sexually transmitted disease and can increase the risk of infections [7]. From a sample of 65 condoms, the only incorporated spermicide appeared to be nonoxynol-9 [7]. Holloenbeck [33] interpreted spectra using electrospray ionisation and MALDI to discover nonoxynol-9 is easily identifiable in lubricants. Maynard [7] extracted intimate swabs seeded with a lubricant containing a silicone-based substance combined with nonoxynol-9 using a nonpolar solvent. Results determined that spermicides combined with dry lubricants as an alternative to water-based persisted longer after intercourse due to not being absorbed by the body. Therefore, lubricant analysis for the presence of both PDMS and nonoxynol-9 is likely to produce a more valid outcome in a sexual assault case.

Starch particles, including corn starch are integrated into lubricants. Through applying these substances it can be ensured the assembly of the condom is preserved in the packaging and an extended use by date is maintained. Polarised Light Microscopy (PLM) is a technique often applied to identify starch components [27]. This method involves illuminating a sample using polarised light to examine the optical features of the specimen. The particles are stained using Haematoxylin and Eosin dye modifying the appearance to dark marks visualised through the microscope. Upon further magnification, starch particles exhibit a maltese cross characteristic which is unique to this specific polymer, exposing the beneficial features of PLM [34]. Polarised light enhances the specificity of this technique, as in standard light the particles would exhibit a spherical appearance which is a typical characteristic of many particles [35]. Unfortunately, starch particles alone are unable to confirm the use of a condom, therefore the analysis of other components, particularly the lubricant, is required to support this proposal.

1.7 Polydimethylsiloxane (PDMS)

PDMS, a form of siloxane, is composed of only silicone, carbon, hydrogen and oxygen atoms [28]. Consisting of a flexible silicone-oxygen-silicone backbone, the silicone atoms are further bonded to two methyl groups causing a high level of viscoelasticity. The functional groups present at the end of the polymer chain differ between alcohol, methyl and ethyl groups dependent upon the application of the PDMS.

Polydimethylsiloxane, frequently referred to as dimethicone or simethicone, is a silicone based organic compound regularly applied to most condoms, gels and personal lubricants [36]. PDMS is typically synthesised via the hydrolysis and polycondensation reaction of dichlorodimethylsiloxane and chloromethylsiloxane [37]. Possessing the general formula $(\text{CH}_3)_3\text{Si}[\text{OSi}(\text{CH}_3)_2]_n(\text{CH}_3)_3$, PDMS can exhibit a viscosity ranging between 5 and 20000 centistokes dependent upon the requirement of the role [37]. The exact viscosity can be determined by multiplying the molecular weight by the number of times each single unit is repeated.



n = The number of repeat units.
 PDMS as a lubricant often has a
 value of approximately n=200.

Figure 1.7: A diagram to show the structure of polydimethylsiloxane.

PDMS is a non-water based lubricant which is colourless, odourless, viscous and non-staining [27]. Being a hydrophobic substance, PDMS is an ideal component within a dry lubricant [38]. Furthermore, PDMS isn't absorbed by the body meaning it is unable to accumulate and cause harm, implying it can be applied both internally and externally and will persist for a length of time unless disturbed or wiped away [27]. Being a non-toxic, particularly robust substance exhibiting a high thermal stability, PDMS displays multiple positive characteristics being an adaptable chemical for an array of diverse applications.

In order for a positive confirmation to be concluded to verify the use of a condom, the viscosity of the PDMS requires to be measured to allow the exclusion of alternative sources [39]. PDMS used as a condom lubricant frequently possesses a viscosity of approximately 200 centistokes. Although, a reported sample of condoms displayed viscosities between the range of 100 centistokes and 350 centistokes [40]. Comparing the viscosity of a recovered sample of PDMS from a case to known viscosities from popular branded condoms will increase the likelihood of the PDMS stemming from that specific source in the case of a match [28]. If the viscosity identified is far from the standard range for condom lubricants, it implies the PDMS has arisen from alternative sources. The same viscosity of PDMS is likely to be incorporated into different styles of condoms from the same brand, meaning the exact condom may not be accurately identified from the PDMS alone. By identifying the presence of additives in the sample of PDMS which correlate with those present in the lubricant, the probability of the match is enhanced.

1.8 Polyethylene Glycol (PEG)

PEG is the second most common substance incorporated as a condom lubricant. Being a water-based substance with an average molecular weight of 300 Da, PEG can be absorbed into the body therefore is unlikely to be recovered from a victim long after a sexual assault [31]. Cho & Huang conducted a study in Taiwan revealing that only 2 condoms from a sample of 35 were lubricated with PEG [29]. Conversely, the sample size of this research is relatively small; hence the results aren't representative worldwide. Furthermore, Coyle & Anwar supplemented this investigation by discovering fewer than 10% of condoms sold on the UK market are lubricated with PEG [8].

In conclusion, PDMS is the favoured lubricant in comparison to PEG mainly due to having a greater shelf life and reducing the risk of the user contracting human immunodeficiency virus. Furthermore, PEG is a highly water-based substance resulting in a reduced peak feeling during sexual intercourse when compared to condoms lubricated with PDMS [3].

1.9 The Importance of Lubricant Evidence

When investigating rape cases, despite victims insisting the offender wore a condom whilst committing the assault, condoms are rarely recovered as evidence. If the account from the victim is revealed to be factual, the odds of seminal fluid being present at the scene of the crime are extremely low. The analysis of lubricant traces is the subsequent stage in resolving a sexual assault case. The significance of the investigation of lubricants varies dependent upon the situation of each case. The case studies explained below illustrate the importance of lubricant evidence in aiding the conviction of a perpetrator in some circumstances.

Spencer [3] identified the importance of the need to undertake lubricant analysis as routine during the investigation of alleged sexual assaults. A woman stated she had been raped by the defendant without the use of a condom. DNA evidence was recovered and found to match that of the defendant. However, the suspect claims

that the sexual intercourse was consensual with the use of a condom. The suspect claimed that the sample of evidence collected from the victim, a mixture of saliva and semen, could only have been recovered from the condom used during the consensual act. This case was resolved outside of court resulting in no prosecutions. However, the examination of the biological samples for the detection of condom lubricants might have allowed the corroboration of either the victim's or the suspect's account of the version of events.

1.9.1 PEG Case Study

Shen [32] investigated a situation, where PEG was the lubricant present, which aided the outcome of a sexual assault case. It was alleged an underage girl was sexually assaulted by her stepfather. Prior to the occurrence of the assault, the suspect provided the victim a vaginal insert to prevent the transfer of biological evidence. Intimate swabs were taken from both the victim and suspect to search for lubricant traces from the vaginal insert as the victim implied the suspect did not wear a condom. Reports indicate that 43% of the insert consists of PEG, indicating analysis is likely to detect traces of the lubricant during examination. The suspect denied all accounts of the assault, stating no vaginal insert was provided to the victim [32]. The swabs taken from both the suspect and victim contained traces of PEG which matched that from the vaginal insert. However, once these findings had been recovered, the suspect revealed he often used a personal lubricant which may account for the traces of PEG. Upon analysis of the personal lubricant, PEG was stated as not being a component within the ingredients; however, traces had been detected in the personal lubricant when analysed using Desorption/Ionisation on Silicon and Time of Flight Mass Spectrometry. In conclusion, the case ended with no prosecutions due to the lack of DNA evidence and weak lubricant evidence.

1.9.2 PDMS Case Study

Blackledge and Vincenti [26] investigated two cases from The Naval Criminal Investigation Service in America to express the importance of lubricant traces in alleged sexual assault cases. Both cases supposedly involved the use of a lubricated condom. Firstly, a female student within the armed forces stated she had been out

drinking with some friends and became unconscious. She stated that she had been raped whilst comatose by one of her classmates as her underwear had been removed. It is believed the suspect kept the condom following the assault to boast to his friends. The condom was disposed of prior to the suspect's interview, resulting in his clothing being recovered for examination [26]. It was revealed during the interview that the suspect obtained a 'Sheik Elite' condom, lubricated with PDMS, from a peer. While, the suspect had confessed to committing the crime, lubricant analysis had identified that the traces found on the suspects clothing and the victim matched the PDMS from the condom.

The second case involved a teenage girl being allegedly assaulted by a member of the Armed Forces who had previously been diagnosed with AIDs. The suspect claimed the sexual intercourse was consensual and he wore a PDMS lubricated 'Prime' condom which broke during intercourse. However, the victim denied the intercourse was consensual and insisted the suspect did not wear a condom. The victim was examined shortly after the time of intercourse before showering, in a time frame whereby PDMS would have persisted in the vagina. No PDMS traces were recovered from the intimate swabs from the victim meaning inadequate evidence is available to corroborate the suspect's account [36].

1.9.3 Determination of Specific Condom

Proficiency tests regarding lubricant examinations have been simulated to assess the accuracy of the techniques applied in sexual assault cases. A scenario was imitated where a female college student claimed that she had been sexually assaulted and insisted the suspect had worn a condom. The victim had been medically examined within 2 hours of the alleged assault. A suspect had been identified and upon examiners searching his dormitory, three different brands of condoms were recovered. All three condoms produced FTIR spectra indicating the presence of PDMS. However, both the intimate swab and the swab from the first condom deposited a white substance during the solvent evaporation, producing an extra peak at 1481cm^{-1} which was not present in the spectra from the other two condoms. Scientists reported that the test did not simulate a real case scenario as the

examined condom was in an unused condition and the amount of lubricant present on the 'intimate' swab was extremely high and would not be expected in such quantities in a real case. The swabs were too heavily seeded when setting up the scenario and more care is required if additional proficiency tests are to be implemented. Furthermore, no SEM analysis of control swabs was provided to eliminate any potential traces of PDMS on blank cotton swabs [41]. In combination with EDX, this would have allowed the elemental composition of the swab to be determined, allowing the presence or absence of silicone to be confirmed.

1.10 External Sources of Polydimethylsiloxane

Although it would be expected that intimate swabs would be collected from both the accused and the victim in an alleged sexual assault case and despite the constant appeals from national police forces, the procedure is implemented infrequently by the majority of forensic examiners. As identified previously, PDMS is a versatile substance occupying many uses. Several of these roles mistakenly contribute in traces of PDMS being recovered in sexual assault cases and can be unintentionally interpreted as a condom lubricant. As a result, false positives resulting from PDMS surfacing from alternate sources are possible in the examination of intimate swabs [8]

Very little research has been conducted considering the ability to differentiate between different samples of PDMS, and no successful technique has been adapted which is able to completely individualise a sample. Lipp [42] effectively analysed diverse samples of PDMS using FTIR and was able to distinguish between different chain lengths of the substance. However, this technique was incapable of accurately identifying each sample specifically when compared to other samples possessing the same chain length.

A potential external source of PDMS is cosmetic and pharmaceutical products. Examples include body lotions, deodorants, vaginal suppositories, shampoo, sun cream and makeup [43]. Siloxane containing substances are frequently

supplemented into cosmetic items, in minimal concentrations to provide a glow to the skin, maintain high quality and an assured enhanced radiance. PDMS viscosities of approximately 5 centistokes are integrated in a cosmetic product which is considerably lower than the viscosity of PDMS used as a lubricant. However, this value can fluctuate substantially dependent upon the specific article [44]. If the swab analysis undertaken in a sexual assault case is unable to correctly ascertain the viscosity, the direct source of the PDMS may not be accurately identified.

Coulson & Fabian [45] gathered 16 different branded high street personal care products. Exhibits included shower gels, lotions and cleansers. 6 of the 16 samples were analysed to discover they contained PDMS, despite only 4 of the 6 stating on the ingredients that they contained a silicone-based compound. Following this, Cellmark Forensic Services conducted a study whereby 21 different branded cosmetic items consisting of body lotions, oils and creams were analysed for traces of PDMS using FTIR and Liquid Cell. 9 of the 21 products, 43%, were found to contain PDMS. The report does not state whether the list of ingredients specified silicone substances were present within the product [46]. Therefore, if identified, the source of PDMS is unable to be directly associated with a condom as the traces could have originated from environmental contamination [7]. In addition to this, 5 sanitary wear products were examined using the same equipment stated above. None of the items tested positive for PDMS. Therefore no PDMS would be transferred to the gusset of knickers from female hygiene products. This suggests that if a victim is wearing a sanitary item during the time of an alleged assault, the product could be examined for lubricant traces to aid in determining whether an assault was committed. However, a larger sample size is required to be tested, consisting of a wider range of brands and repeats to assist in validating these findings [46].

Silicone based components are incorporated into fabrics within many platforms of the textile industry. PDMS exhibits multiple properties which are desirable in the textiles manufacturing process. Features including heat stability, reduction of static coefficient and acting as an anti-foaming agent, all of which contribute towards enhancing the performance and appearance of materials [47]. Furthermore, many fabric softeners contain PDMS as it prevents the creasing and tearing of materials

[48]. Softeners are often applied as a finish to clothing items also, to provide a waterproof coating due to the hydrophobic nature of the material [49]. Similar viscosities of PDMS to those displayed in condom lubricants are applied in the textile industry. Therefore if PDMS is recovered from garments, particularly underwear, the originating source is unable to be verified. However, if recovered internally, the likelihood of the substance sourcing from the undergarment is slightly reduced. If a control section of an item of clothing is examined also delivering a negative result for PDMS, a more precise conclusion is able to be established [27].

A study compiled by Cellmark Forensic Services investigated the effect of washing on the persistence of silicone fabric finishes on garments. Research indicated that traces of PDMS are detectable on upper garments for approximately 6 washes and roughly 14 washes on the gusset from a pair of knickers. This characteristic should be considered in evaluating significance based upon the age and condition of the clothing [46]. For example, if a garment is well worn and not recently purchased, traces of PDMS are unlikely to have originated from the initial fabric finish.

1.11 Presence of Polydimethylsiloxane on Blank Cotton Swabs

In 2003, Hutchinson [25] analysed sealed blank cotton swabs extracted in cyclohexane using an FTIR. The spectra observed were typical of polydimethylsiloxane. Two different brands of swabs were examined (both with a cotton tip, one with a plastic shaft and one with a wooden shaft) and exhibited the same characteristics. However, the findings were revealed to be inconsistent with only a small quantity exhibiting traces of PDMS.

In recent years, it has been questioned by scientists that polydimethylsiloxane could have been inadvertently deposited on blank cotton swabs during the manufacturing process. This is because PDMS has been detected in Forensic Casework whereby both the victim and suspect claimed a condom was not used during an alleged sexual assault. Forensic equipment, including an FTIR has detected volumes as low as 0.125µl trace samples of PDMS via a swab extraction using isooctane. It is therefore

essential that any contaminants are identified for the prevention of false positive results [50]. Subsequently, negligible research has been conducted into this area as a result of the decline in funding within the forensic science community. Insufficient staff, resources and time have prevented the development of answers to this allegation.

Coyle & Anwar [8] used Infra-red analysis to examine 24 blank cotton sterile swabs and discovered no peaks correlating to PDMS. This suggests that it is doubtful PDMS contaminates the swabs during manufacturing. However, an insignificant quantity of swabs were analysed during this study, hence it is not representative. Moreover, only one brand of swabs was examined which isn't typical of all swabs used during sexual assault cases, therefore accurate conclusions cannot be formulated. Secondly, Campbell & Gordon [28] inspected 38 blank cotton swabs for residues of PDMS. Mass Spectrometry was the system operated with the spectra indicating no presence of PDMS. Likewise, this too is an insignificant measure of swabs, providing no signal to the brand(s) of swabs tested. Cellmark provided a study in 2013 whereby a total of 45 blank cotton swabs were analysed with 15 swabs each, from the following brands, using FTIR: MWE, Copan and SceneSafe [46]. All of the swabs analysed displayed spectra negative for PDMS.

In support of this research, Cellmark Forensic Services fulfilled a study in 2016 whereby 523 blank cotton sterile swabs were analysed to test for the presence of PDMS. A total of 4 different universal brands were donated from 12 different Police Forces nationwide with a range of round and mini point tips. The extraction process involved the use of isooctane as the non-polar solvent with all samples being analysed using an FTIR with microscope attachment. All of the swabs analysed displayed spectra negative for PDMS [51]. The only spectra resulting were approximately 95% matches with Dibutyl phthalate and Bis(3,5,5-trimethylhexyl) phthalate on comparison. Reports suggest that these traces are components within the adhesive used to glue the swab head to the wooden shaft [52]. In conclusion, the manufacturing process is classed as being an improbable source for PDMS contamination.

Following the previous research, Cellmark Forensic Services completed an additional study focusing on the presence of PDMS on blank cotton swabs. A total of 15 blank cotton swabs were retrieved from 3 SceneSafe swab collection kits and were prepared for analysis using the isooctane extraction as used in the previous study. Results indicated that 5 swabs from one SceneSafe kit tested positive for PDMS, whereas all swabs in the second and third kit tested negative. The intensity of the peaks on the spectra for the positive samples was low, however consistent with the peaks characteristic of PDMS. Analysis concluded that in the case of the 5 swabs containing PDMS, none was present on the plastic shaft; suggesting the PDMS originated on the swab head [53]. In conclusion, further research is required with regards to the contamination of blank cotton swabs, especially focusing on the swabs present in sexual assault kits with a plastic shaft.

1.12 Persistence of Polydimethylsiloxane

The persistence of spermatozoa or a lubricant is the measurement of time that the substance is recoverable and identifiable following the transfer from the penis or condom to the vagina, penis, anus or external genitalia [54]. Research has indicated that spermatozoa exhibited following ejaculation only have an approximate lifespan of 20 minutes unless an egg is fertilised initiating pregnancy. It is therefore crucial that recovery and analysis is undertaken promptly to allow a greater chance of detection. If no spermatozoa are identified, condom lubricants display a lengthier persistence time in comparison. Hence, if recovered, the presence of PDMS may also confirm that sexual intercourse had occurred in addition to delivering a justified explanation for the absence of DNA.

Water-based lubricants including PEG display a considerably poorer persistence time in comparison to silicone-based lubricants. This substantial difference is the outcome of polyethylene glycol being absorbed by the vaginal membranes, consequently causing a reduction in persistence. Initial research regarding the persistence of condom lubricants suggested a 1-2 hour recovery time of water-based lubricants following sexual intercourse [33]. Following this, Dalton [55] revealed that both

vaginal and penile swabs acquired greater than 5 hours post coitus will contain no traces of PEG. Most currently, an increased estimate of approximately 6 hours on vaginal swabs and 8 hours on penile swabs was suggested [13]. Overall, unless the victim of sexual assault reports the incident promptly, the likelihood for detecting lubricant trace evidence, if a water-based condom was employed, is minimal.

Silicone based lubricants, including polydimethylsiloxane, retain non-polar properties indicating the substance is unable to pass through the vaginal walls unlike polyethylene glycol. Attributable to this feature, Cain & Nguyen [57] discovered PDMS in the vagina at least 48 hours after sexual intercourse. Following this, Campbell & Gordon [28] revealed traces of PDMS were detectable on vaginal swabs of participants who denied being involved in any sexual activity involving the use of a condom or personal lubricant within three days prior. This evidence indicates that PDMS may persist in the vagina for more than 72 hours post coitus. PDMS has been detected on penile swabs more than 50 hours after intercourse. A second source of research discovered that PDMS was detected on 93% of penile swabs up to 20 hours after wearing a condom. In addition to this, Thomson [13] learned PDMS was detected on anal swabs, using FTIR, 7-8 hours after sexual intercourse.

PDMS is of significant interest compared to other non-biological evidence due to the low degradation rate and the difficulty in being able to destroy the evidence. However, multiple factors can reduce the persistence time of lubricants. If a breakage occurs in the condom during sexual intercourse and the offender ejaculates, the semen may dilute the PDMS until it is no longer detectable [56]. Furthermore, menstruation, washing and the activity the individual has undertaken during the day may cause traces of PDMS to be excreted from the body [57]. The greater the elapsed time since intercourse, the lower the likelihood of recovering lubricants.

1.13 Semen Extraction Techniques

In sexual offence cases, intimate body swabs and fabrics are frequently submitted for analysis by forensic casework examiners. The discovery of seminal fluid on recovered exhibits and the successive incorporation of DNA analysis are of essential significance, combined with an evaluation as to the importance of the findings [58]. The most common technique for extracting semen from both swabs and materials in forensic laboratories is via a water-based extraction. However, Cellmark Forensic Services have developed a technique named the 'Sperm Elution' process which is greatly favoured over the traditional water-based extraction. Although Cellmark occasionally perform the water-based extraction dependent upon case circumstances, the 'Sperm Elution' method possesses numerous positive features contributing to an extraction with greater success.

Both spermatozoa and epithelial cells are expected to be present on intimate swabs containing semen obtained from sexual assault victims. The presence of nucleated epithelial cells can interfere with the identification of spermatozoa during microscopic examinations [59]. Through the Sperm Elution process, the epithelial cells are separated from the spermatozoa allowing sperm heads to be more easily identifiable whilst slide searching. This stage is feasible because the procedure facilitates a two buffer system whereby two end products remain. These are the epithelial pellet and the seminal pellet [60]. The process is initiated via the addition of Mo Lite only, which acts as a weak buffer to extract only a small portion of the sperm heads from the swab to allow the formation of the epithelial pellet. The second stage involves the addition of Proteinase K and sodium dodecyl sulphate, in which all cellular material, excluding sperm cells, is digested, allowing an effective isolation of spermatozoa [61]. Furthermore, sperm heads are expected to be detectable in a larger quantity following the removal of the epithelial cells [58]. Cell separation prior to microscopic examination develops the examiners sureness in observing and recording spermatozoa which could possibly be present. An enhanced likelihood of obtaining an accurate DNA profile from an intimate swab is an outcome of successful cell separation [59].

Initially, the estimated recovery rate of spermatozoa from intimate swabs following a water-based extraction is less than 20% from the cell pellet. The Sperm Elution process is reported to recover greater than double the quantity of spermatozoa compared to the water-based extraction method [59]. This vast increase in detection is imperative within forensic science as often, only trace amounts of spermatozoa are recovered following sexual assaults, therefore the likelihood of detection is rapidly increased with the improved method.

A study conducted by Hulme, Lewis & Davidson [59] involved 4 different simulations on intimate swabs. These include a high volume of approximately 4700 sperm cells being seeded onto 2 swabs and a low volume of approximately 470 sperm cells being seeded onto 2 additional swabs. The first set of swabs was analysed using the Sperm Elution method, and the second set using the water-based extraction. On average across all 4 validation experiments, the Sperm Elution method recovered 71% of spermatozoa compared to only 25% from the water eluted swabs. As the expected number of spermatozoa recovered is likely to increase following the Sperm Elution procedure, if a victim didn't report the assault immediately, the increase could affect the interpretation of time since intercourse if the examiner is not aware of the efficiency of the process [59].

One of the foremost variances between the Sperm Elution and the water elution is the type of reagents used. The water elution only uses water as the material to extract the spermatozoa, whereas the Sperm Elution method used 4 different reagents which are: Mo Lite, Mo Classic A, Proteinase K and Mo Wash. The exact composition of these chemicals is confidential to Cellmark Forensic Services; therefore the exact reason as to why the Sperm Elution has such a high recovery rate when compared to other techniques is currently not published.

Extraction of whole swab heads for semen extraction, without providing half a swab head for lubricant analysis, provides the most sensitive detection of spermatozoa [58]. However, if semen is unknowingly absent from the intimate swabs in an alleged sexual assault case as a result of the use of a condom, traces of PDMS could be present. Minimal research has been conducted into the effects of the Sperm Elution

process and whether PDMS can be detected following the extraction. A study conducted by Cellmark Forensic Services revealed that some PDMS remains on the swab head following the Sperm Elution; however the results obtained weren't quantitative and the effects of the further addition of other bodily fluids including blood, saliva, vaginal material and faeces, which are expected on intimate swabs in forensic casework, is currently unknown [53].

1.14 Analytical Techniques for the Analysis of Polydimethylsiloxane

Limited research has been conducted into the analysis of condom lubricants in relation to sexual assault cases. However, a wide variety of techniques have been applied to determine the identification of lubricants, especially PDMS. A list of techniques examined is Fourier Transform Infra-red (FTIR), Attenuated Total Reflectance (ATR), Raman Spectroscopy, Gas Chromatography Mass Spectrometry (GCMS), Desorption Chemical Ionisation Mass Spectrometry (DCI/MS), Nuclear Magnetic Resonance (NMR), Pyrolysis Mass Spectrometry (Py-GCMS) and Capillary Electrophoresis. Previous research has indicated that Raman Spectroscopy requires no sample preparation and is a non-destructive technique for lubricant analysis [62]. However, this technique alone would be unsuitable for the examination of PDMS as contamination and fluorescence within the sample could interfere with the peaks forming an inaccurate spectrum. The fluorescence is caused by particles absorbing photons, resulting in the electronic state of the particles reaching an excited state followed by emitting a new photon by relaxation [8].

GCMS was deemed to be an incompatible technique as silicone oils aren't sufficiently volatile enough to pass through the gas chromatography column. PDMS is known to possess a molecular weight from 3000Da up to approximately 15000Da, whereas the detection limit for GCMS is 1000Da. Wachholz [63] coupled GCMS with GC-FTIR for the identification of siloxane compounds. Both methods are supplementary to each other and provide more accurate results. Py-GCMS can be used as an alternative technique. This application is beneficial as it breaks down long chained polymers allowing them to be identified by GCMS. Kleinert [64] successfully analysed PDMS with viscosities of up to 30000 centistokes. Therefore, if PDMS undergoes pyrolysis

at high temperatures, around 600°C, to break down the compound into smaller polymers before GCMS analysis, PDMS as a lubricant is able to be identified. Py-GCMS is a straight forward process with the equipment being available in most forensic laboratories.

Polydimethylsiloxane possesses a differing range of viscosities due to being incorporated for numerous different applications. The mean chain length of PDMS can be established using a technique named Diffuse Reflectance FTIR (DRIFTS). This method discloses whether the examined sample of PDMS is likely to have been sourced from a condom or an alternate use. Research conducted by Maynard [7] involved the analysis of 27 cotton swabs with an applied condom lubricant using DRIFTS. Each sample was seeded 90 days prior to the analysis. The standard identifying peaks for each Infra-red spectrum persisted as predicted for all samples demonstrating that the base component of each lubricant, including PDMS maintains the same structure for a minimum of 3 months with no apparent decomposition [7]. Blackledge assessed Fourier Self-Deconvolution (FSD) together with FTIR to interpret characteristics of the viscosities of diverse samples of PDMS. This is a mathematical term for decreasing bandwidths, allowing overlapped peaks to be distinguished from one another. The results identified viscosities between 5 and 500 centistokes could be distinguished. This allowed for comparisons between alternate sources of PDMS and to differentiate between multiple brands of condoms [42].

Desorption Chemical Ionisation Mass Spectrometry (DCI/MS) is an analytical technique whereby the sample is dissolved in a volatile solvent then heated prior to ionisation. It is occasionally applied for lubricant analysis for the reason that it acquires exceptionally high sensitivity by being able to detect nanolitre traces of PDMS in comparison to other sensitive methods such as FTIR which have only been perceived to detect microlitre volumes [26]. In addition to being competent in identifying PDMS, this apparatus would allow for lesser volumes to be observed and contrasts in PDMS applied by varying condom manufactures. Like DRIFTS, DCI/MS is capable of differentiating between different chain lengths, though only great differences are recognised, which is detrimental as many producers incorporate

PDMS with a similar viscosity of approximately 200 centistokes to their condoms [30]. Furthermore, Py-GCMS displays greater sensitivity in comparison to DCI/MS meaning this technique is seen as a more appropriate option. Unfortunately, the equipment to perform this method is very costly, therefore isn't generally offered in most forensic laboratories. Nevertheless, if accessible this technique is very beneficial towards lubricant identification and an extremely powerful confirmatory method.

^1H , ^{13}C and ^{29}Si are examples of different NMR techniques often applied for analysis. Each lubricant possesses a unique structure whereby the hydrogen atoms are situated in different environments allowing a specific spectrum to be produced, hence allowing each lubricant to be distinct in the spectra. ^1H is the primary NMR selected for lubricant analysis and allows the different proton environments within the lubricant to be characterised. Furthermore, proton NMR is a quantitative method where only one experiment is required to be undertaken for multiple samples [31]. Lee [31] conducted research which shows the ability of NMR to effectively verify the presence of condom lubricants in addition to identifying spermicides. In all experiments, PDMS was detected; however, only three quarters of samples were able to be distinguished between. As previously stated, most condom manufacturers apply formulations of PDMS with a viscosity of approximately 200 centistokes, the differentiation between condoms on the UK market is very unlikely using any analytical tool. NMR is a robust piece of equipment, but expensive and, therefore, isn't available in many laboratories employed for forensic casework. Furthermore, the sensitivity of NMR is much weaker in comparison to techniques applying Mass Spectrometry.

Capillary Electrophoresis is an analytical technique rarely performed in forensic laboratories. Although a technique which is very quick and simple, and little sample preparation is required, the process would only be beneficial if combined with additional analytical techniques [65]. Burger applied the technique to the detection of PDMS on fabrics. 30 minutes after the application of the lubricant, PDMS was no longer able to be detected on the fabric using Capillary Electrophoresis [36].

Finally, SEM-EDX is a technique often used for the identification of silicone. The Scanning Electron Microscope allows the production of signals used to obtain information regarding the surface and structure of the material. In combination with EDX, the chemical composition is determined allowing the presence of Silicone to be confirmed or excluded.

1.14.1 Infra-red Spectroscopy

Infra-red spectroscopy is a class of vibrational spectroscopy which measures the absorbance of photons of both organic and inorganic compounds [66]. The main aim of IR is to identify functional groups present in a molecule. When a molecule absorbs a photon, the molecule gains energy and moves to a higher vibrational energy state. This is known as the excited vibrational state. This increases the average bond length of the molecule [67]. The process functions by analysing the number of IR photons and the amount of energy in the IR photons absorbed by the molecule [66]. The IR light passes through a beam splitter which separates the different wavelengths of light. This passes through to a moving mirror which only allows one frequency of IR light to pass through the sample at a time. Once the light has passed through the sample, any photons which aren't absorbed reach the detector. The reference detector receives all IR light. This reveals how much IR light is present in total. By comparing the amount of unabsorbed photons with the amount of photons which reached the detector allows the quantity of absorbed photons to be calculated. This information forms an interferogram, which is converted into an IR spectrum using a logarithmic equation [66].

Fourier Transform Infra-red Spectroscopy (FTIR) is a specialised form of Infrared Spectroscopy that records the effect on a sample when struck by a light beam within the IR range, 1mm-750nm of the electromagnetic spectrum, by measuring the interactions between the molecules. The technique is applied to establish the molecular structure of a material by labelling and categorising the functional groups present in a substance [67]. FTIR is a procedure fulfilling the same process, however, it displays amplified sensitivity in comparison to a traditional Infra-red. Furthermore, FTIR displays a higher signal to noise ratio, greater wavenumber accuracy and a

shorter scan time. Conversely, not all groups within a molecule are able to be recognised using Infra-red. Bonds present within the structure must display a changeable dipole moment in order to be Infra-red active and be detected [67].

An Infra-red spectrum of transmittance (%) on the y axis against wavenumber (cm^{-1}) on the x axis is then usually calculated allowing the functional group of each peak to be identified and compared to a suspected compound [13]. Peaks present appear between the approximate range of $600\text{-}4000\text{cm}^{-1}$. The wavenumber of a substance is the volume of wave cycles per centimetre. The quantity of energy travelling through the sample is the transmission [49]. When a beam of energy reaches a functional group within the sample, the transmission levels drop forming a peak. This is as a result of the bonds reaching the excitation state through absorption and vibrating [68]. Every peak within the spectrum possesses a wavenumber which relates to particular bonds associating with a functional group within the molecule. The Infra-red spectrum is exclusive to each substance. Common peaks often occur in compounds possessing very similar characteristics. However, the distinct fingerprint region of the spectrum is unique to each molecule [67].

FTIR is an analytical technique able to allow the identification of polar molecules within the solid or liquid state. In order to further confirm the identity of an unknown sample, a known standard as a reference should be analysed following the examination. Also, if possible, qualitatively compare the unknown spectrum to a recognised database to assist in determining the identity of the sample [69]. Though, reference libraries rarely contain vast amounts of spectra, and aren't updated frequently, therefore it is advised to compare against multiple sources.

1.14.2 Infra-red Spectroscopy in Relation to Lubricants

Fourier Transform Infra-red Spectrometry is an analytical instrument extensively incorporated for use of multiple types of forensic evidence including the examination of fibres, paint and condom lubricants in alleged sexual assault cases [29]. Even though volumes of research regarding lubricants have substantially increased over recent years, FTIR is still the favoured process for the analysis despite competition

from alternate techniques [62]. Extensive research has been conducted by implying a diverse array of analytical techniques as described in the previous sub-chapter. However, due to the FTIR being widely available, cost effective, fast, simple and reliable, the method has been the principal choice by forensic research scientists for the utmost degree of study carried out into determining the most suitable routine for the extraction and analysis of lubricants.

Individual studies have been performed aiming to establish a further successful route for examinations, proficient at identifying unique characteristics between each individual condom available nationwide. To this date, although ineffective at distinguishing between all condoms (due to the great similarity of the chemical composition of condoms on the market), combining FTIR with supplementary techniques could ease the interpretation of evidence within the case.

1.14.3 Fourier Transform Infra-red Spectroscopy of Polydimethylsiloxane

PDMS displays a basic, yet distinctive structure to allow for a straight forward identification. The lubricant is a substance suitable for analysis by FTIR due to being a liquid susceptible to undergoing a dipole moment, allowing the material to be IR active. Each dipole produces a bond vibration which forms a characteristic peak on the Infra-red spectrum. PDMS presents specific characteristic peaks at approximately 1260cm^{-1} , 1090cm^{-1} , 1020cm^{-1} and 800cm^{-1} [70]. A symmetric CH_3 deformation is formed at 1260cm^{-1} , anti-symmetric Si-O-Si stretching at 1090cm^{-1} and 1020cm^{-1} and CH_3 rocking at 800cm^{-1} [63]. The C-H bonds at approximately 3000cm^{-1} are very common in many materials and therefore aren't indicative of PDMS. Furthermore, all four of the characteristic peaks are required to be present for the positive identification of PDMS as other similar substances may contain peaks at the same wavenumber. These peaks are explained in Table 1.14.3.

Fourier Transform Infra-red Spectrometry is the routine non-destructive method used for the identification of PDMS and personal lubricants as it's a fast, uncomplicated and cost-effective procedure and the equipment is present in the majority of forensic laboratories [40]. However, the technique lacks the power to identify specific condom brands due to being unable to distinguish between varying

viscosities of polydimethylsiloxane. This raises a problem for Reporting Scientists when endeavouring to decipher the origin of the lubricant.

Table 1.14.3: A table to show the bonds which represent the peaks of PDMS on an Infra-red spectrum.

Bond	Vibration	Peak Appearance	Wavenumber
Si-CH ₃	Rocking	Singlet	Approx. 800cm ⁻¹
Si-O-Si	Asymmetrical Stretch	Doublet	Approx. 1020cm ⁻¹ & 1090cm ⁻¹
Si-CH ₃	Symmetric Stretch	Singlet	Approx. 1260cm ⁻¹
C-H	Asymmetric Stretch	Singlet	Approx. 3000cm ⁻¹

The Si-O-Si asymmetric stretch is a doublet because the oxygen atom which is bonded to the silicone closest to the end of the polymer chain will be a stronger bond compared to the silicone closer to the centre of the polymer chain. This is because the O-H functional group at the end of the chain is a very strong bond, hence an increased wavenumber on this Si-O bond.

FTIR can normally detect marginal volumes of a substance, hence it is an extremely sensitive piece of equipment [68]. Research conducted by Cellmark Forensic Services revealed PDMS was able to be identified on extracted cotton swabs initially seeded with 0.125µl of PDMS [53]. Blackledge & Vincenti [26] explored sexual assault cases; allegedly including the use of a condom lubricated with PDMS using FTIR. The technique was established and presented to the forensic industry by Blackledge whilst incorporating microscopic analysis of a material, followed by FTIR to examine PDMS samples from a range of manufacturers. Maynard [7] discovered FTIR is eligible to detect PDMS on vaginal swabs for approximately 48 hours following sexual intercourse.

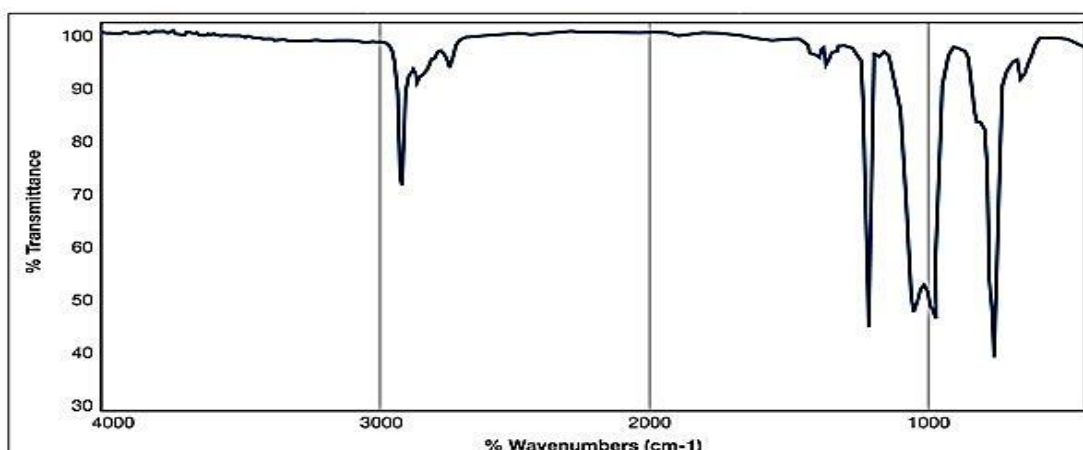


Figure 1.14.3: An Infra-red Spectrum of polydimethylsiloxane.

1.14.4 Fourier Transform Infra-red Microscopy

Fourier Transform Infra-red microscopy follows the exact same procedure as described in Section 1.14.1, but in addition a microscope is incorporated to enhance the view of the sample. This technique is applied for materials visually too small for the naked eye, or when substances are present in volumes too low for alternative analysis.

This technique requires substantial preparation time prior to analysis. Dependent upon the physical state of the sample, a section or extract is applied to a glass window. Barium Fluoride, BaF_2 , is the favoured material for the window as it contains water resistant properties, and absorbs no moisture from any of the samples [71]. Previous research used Sodium Chloride and Potassium Bromide windows, but these can be easily broken, and affect the quality of the spectra produced. The microscope slide, containing the BaF_2 window, is then positioned onto the stage of the microscope. Using an appropriate application, the sample can be visualised on the computer at a greater magnification. By modifying the position of the stage, different areas of the sample are able to be viewed, and the required area is indicated for examination.

Although a highly sensitive piece of equipment, the quality of the spectra observed is highly reliant upon the accuracy of the application of the sample onto the glass window. For example, if a liquid material is spotted on to the window too heavily,

forming a thick layer of material, the spectra produced will be sharp and precise. But, visualising the position of the sample with the microscope is difficult because the individual spherical globules of the lubricant don't separate. On the other hand, if the spotting of the matter onto the window isn't precise, the sample can spread to the outer edges of the disc and result in a thinner layer of material and provide difficulties in locating the circumference, hence producing a poorer spectrum.

Infra-red microscopy uses reflective optics as an alternative to glass which is present in standard optical microscopes. This makes certain that the entire Infra-red region of the electromagnetic spectrum is observed with only a negligible effect on the signal. Reflective, condensing objectives are situated centrally in the microscope. These feature a Cassegrain design which controls the focusing of the light beam both towards and from the sample to allow transmission [72]. A Mercury Cadmium Telluride detector, cooled with liquid nitrogen, is where the light source is redirected once passed through the sample [73].

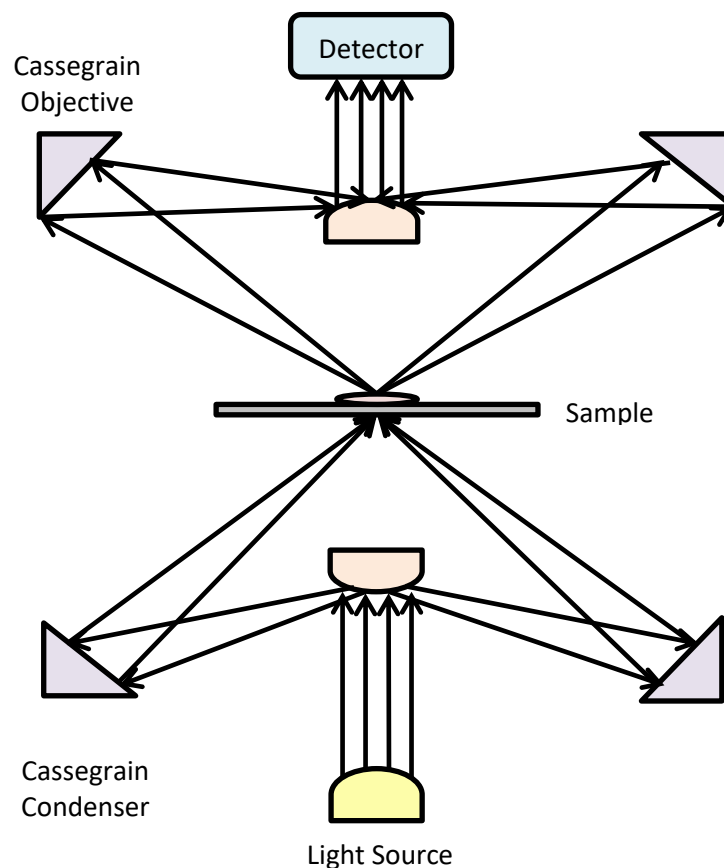


Figure 1.14.4: A diagram to show the direction of the Infra-red light source during microscopy.

1.14.5 Universal Attenuated Total Reflectance Fourier Transform Infra-red Spectroscopy

Universal Attenuated Total Reflectance (u-ATR) is an alternate sampling technique used in conjunction with FTIR. The process initiates by introducing a material to the optically compact crystal, often comprising of Zinc and Selenium, on the stage. A single beam of Infra-red light is then addressed onto the crystal, which has a high refractive index, at a designated angle [74]. This interior reflectance generates a transient wave that ranges further than the plane of the crystal, passing through the sample. This wave will be attenuated in regions of the spectrum where the sample is absorbing the Infra-red energy. Upon completion, the beam departs via the opposite end of the crystal and is aimed towards the detector within the Infra-red spectrometer. The beam of energy is recorded by the detector producing an interferogram, leading to the creation of the Infra-red spectrum. u-ATR is an ideal method for generating qualitative results [74]. Minimal sample preparation is required; the equipment is simple to operate and spectra are produced effectively. As one beam of energy is transferred through the sample, a single-bounce reflectance is produced.

Maynard [7] applied u-ATR as an analytical technique in his research based on condoms and personal lubricants often involved in sexual assault cases. The project aimed to consider the effectiveness of u-ATR and whether the process could be deemed a more beneficial alternative in comparison to DRIFTS. Results disclosed the quality of the spectra attained possessed indistinguishable peaks where it was incapable of differentiating between two different samples of PDMS. However, u-ATR was considered to be the more successful technique due to being a more cost effective, quick, simple and highly sensitive process.

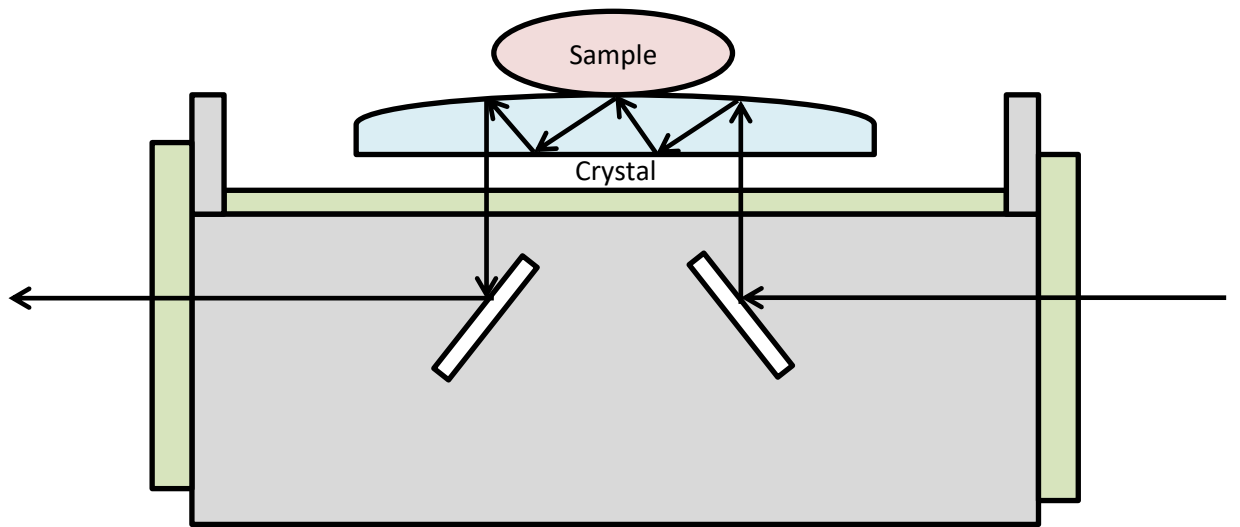


Figure 1.14.5: A diagram to show the direction of the Infra-red light source during u-ATR.

Chapter 2: Aims & Objectives

2.1 Aims

The predominant aims of conducting this research are to assess the validity of Cellmark's Sperm Elution procedure applied to a body fluid matrix, and to determine the persistence of PDMS on the penis and in the vagina following sexual intercourse.

2.2 Objectives

- To determine whether blank cotton swabs are unintentionally contaminated with PDMS during the manufacturing process, by extracting the swab head from blank cotton swabs using isooctane and analysing the extracts using u-ATR FTIR and FTIR Microscopy for the presence of PDMS.
- To determine the smallest detectable volume of PDMS on cotton swabs following an isooctane extraction using u-ATR FTIR.
- Produce a quantitative curve for the detection of PDMS based up on the volume of the substance.
- To determine the smallest detectable volume of PDMS on cotton swabs following the Sperm Elution process and an isooctane extraction using u-ATR FTIR.
- Complete the Sperm Elution process using a body fluid matrix to determine the effects of the process on the detection of PDMS.
- Determine the persistence of PDMS on the vagina and penis following sexual intercourse between partners using a lubricated condom.
- To discover whether PDMS is transferred onto undergarments after sexual intercourse with a silicone-based condom.
- Compare and contrast the sensitivity of analytical techniques for the detection of PDMS.
- Identify the lubricant present in Sexual Assault Kits to rule out PDMS contamination in alleged sexual assault cases.

- Conduct a survey to determine the most popular shower gels and body lotions on the UK market. Identify whether PDMS is present within the products and if this is likely to be a contaminant in sexual assault cases.
- Collect a variety of condoms sold on the UK market and analyse for the presence of PDMS.
- Determine whether further extractions of swabs containing PDMS using Direct Semen Extraction and Fast Differential affect the detection of PDMS.
- Identify whether PDMS is present on the fabric of underwear, and, if so, how many washes are required to eliminate the contamination.

Chapter 3 – Methods & Materials

3.1 List of Equipment and Reagents

The two main processes undertaken in this project are an isooctane extraction and Sperm Elution. A range of different equipment is incorporated into each technique to ensure accuracy and prevent contamination. Below are two tables listing all the equipment, samples and reagents used to complete these two sections of the project.

Table 3.1: A table to show the samples, reagents and equipment used in the Sperm Elution process.

Sample	Reagent	Equipment
Semen	Mo Lite	Heat Block
Saliva	Mo Classic	Centrifuge
Blood	Mo Wash	Vortex
Vaginal Material	Proteinase K	Sonicator
Faecal Material	Nuclear Fast Red	Hotplate
	Picro-Indigo Carmine	Thermometer
	Haematoxylin	2ml flip top tubes
	Eosin	1.5ml screw cap tubes
	Entellan	20, 200, 1000µl pipettes
	Ethanol Absolute	20, 200, 1000µl pipette tips
		Microscope Slides
		Cover Glass
		Plastic Needles
		Spin Baskets

Table 3.11: A table to show the reagents and equipment used for the Isooctane Extraction.

Reagent	Equipment
Isooctane	Micropipette
	Micropipette tips
	Plastic Needles
	Disposable Scalpels
	1.5ml screw neck glass vials
	12ml rimless glass test tubes
	2ml flip top tubes
	Pasteur pipettes
	Black screw cap vial lids
	Acetate sheets

3.1.1 Prevention of Contamination

Prior to beginning the Sperm Elution procedure, all of the equipment and workbench was wiped down using a sodium hypochlorite (presept) disinfectant solution and paper towels. Each 2.5g precept tablet was diluted using 1 litre of water. The workbench was then covered with brown paper and a piece of sugar paper to complete the process.

All worktops were wiped down using isooctane before the extraction of any swabs. Acetate sheets, disposable scalpels and plastic needles were also wiped using isooctane prior to use and discarded after each swab. All test tubes and glass vials were rinsed with isooctane whereby the solvent was drawn through a Pasteur pipette three times to ensure each piece of glassware is thoroughly clean. All glassware washings were transferred to a separate glass vial and analysed prior to extracted samples to exclude any equipment contamination.

The Sperm Elution procedure, the seeding of swabs using PDMS and the extraction of swabs were completed in different laboratories at Cellmark to prevent cross contamination. The PDMS was stored separately whilst securely wrapped in three layers of packaging to prevent being unintentionally transferred to other items within the project. In addition, control samples were completed for each run of analysis to exclude contamination from equipment and reagents.

3.2 Donation of Samples

3.2.1 PDMS

PDMS donated to Cellmark Forensic Services from 'Mates' was utilised to seed the swabs throughout this project. Following Durex, Mates is the second most successful condom manufacturer in the United Kingdom. In order to obtain a reference spectrum for PDMS, a 2 μ l droplet was placed on the u-ATR crystal using a micropipette. The sample was then run producing a spectrum which can be used as a reference.

3.2.2 Lubricants from Sexual Assault Kits

Lubricants are present within sexual assault kits to prevent discomfort to an individual during the recovery of forensic evidence. Manufacturers have recently replaced the lubricant present in sexual assault kits with a more cost-effective alternative. However, professionals failed to report the ingredients incorporated within the lubricants. It is crucial for forensic providers to identify the components in the lubricant because if PDMS is present, this could provide false positive results in sexual assault cases.

The two replacement lubricants within the kits are named 'Aquagel Lubricating Jelly' and 'Optilube Lubricating Jelly'. A sachet of each lubricant was donated to Cellmark Forensic Services for analysis. A 2µl sample of each lubricant was applied to the u-ATR crystal and was left to dry down. Once the sample had fully dried out, the sample was run producing a spectrum for each lubricant. The crystal was thoroughly cleaned using isooctane between each sample, with a blank background control also being completed.



Figure 3.2.2: Aquagel and Optilube Lubricating Jellies.

3.2.3 Body Fluid Donations

In order to complete the Sperm Elution process to determine the limit of detection of PDMS and the detection of PDMS in a body fluid matrix, the donation of body fluids is required. In order to determine the limit of detection of PDMS, only semen is required to be seeded on to the swabs following PDMS. However, to create a body fluid matrix, a range of multiple different body fluids are required.

Cellmark Forensic Services is registered under the Human Tissue Act (HTA); therefore all donated body fluids are able to be stored at Cellmark, Chorley. All biological samples provided for the project were donated by members of staff at Cellmark. An email was sent internally to all Cellmark employees including a participation sheet to inform potential volunteers of how to participate in the research.

The purpose of this research is to determine whether the presence of other body fluids, in addition to semen, including blood, saliva, faecal material and vaginal cells affect the detection of polydimethylsiloxane (PDMS) on intimate swabs following the Sperm Elution process. Furthermore, to identify whether the presence of extra fluids affects whether the PDMS remains on the swab head throughout the process or whether it is extracted into the supernatant. This would be an extremely beneficial technique to have validated for use in forensic casework as scientists are often faced with scenarios where multiple body fluids are present on each intimate swab. For example in sexual offence investigations, especially rape, bleeding may have occurred during the assault.

The donation of body fluids for Sperm Elution gained ethical approval from University of Central Lancashire's Ethics Committee (STEMH). Donation kits, containing a consent form, participant information sheet and swabs/bijoux tubes, were placed in the male and female changing rooms at Cellmark, Chorley. Each donation kit for the required body fluids was labelled with a separate sample reference. A maximum of three to six samples were required of each body fluid, dependent upon the combination of body fluids applied in the project. The statements below clarify the labelling of each sample, and the articles provided for

the recovery of the donations. Details of how to provide each donation is explained on the Participant Information Sheet (Appendix 3).

ST1/01 – ST1/03 – Semen: A plastic universal tube was provided for the participants to donate semen.

ST2/01 – ST2/06 – Saliva: A plastic bijoux tube was provided for the participants to donate saliva.

ST3/01 – ST3/03 – Vaginal material: Three blank cotton SceneSafe swabs were provided for participants to complete blind vaginal swabs to obtain vaginal material.

ST4/01 – ST4/06 – Faecal material: Three blank cotton SceneSafe swabs were provided for participants to complete anal swabs to obtain faecal material.

ST5/01 – ST5/06 – Blood: A plastic bijoux tube was provided for participants to donate blood. It was advised for participants to donate via a prick to the finger with no direct harm being inflicted upon the individual.

Once the donations had been produced, participants were asked to seal all samples in the labelled plastic packaging provided and place in the freezer in sample reception at Cellmark, Chorley.

3.2.4 Persistence

Previous research regarding the persistence of PDMS in the vagina and penis is very outdated and limited. Conducting a new study involving volunteers undertaking consensual sexual intercourse with their partner with the use of a silicone lubricated condom would provide a more accurate time frame for the persistence of PDMS. This information could greatly influence the forensic science market as currently the majority of forensic providers only recommend the analysis of intimate swabs for the presence of lubricants when recovered less than 24 hours after the offence. However, if this time frame could be extended, it may be of use in more forensic cases. This is valuable as many victims of alleged sexual assault don't come forward and notify the police immediately after the assault.

Only Cellmark employees are allowed to participate in this section of the study due to only DNA present on the company's internal database being allowed to enter the laboratories. Participation in the project would require the volunteer to have sexual intercourse with their partner using the provided condoms. All condoms are of the same brand and are lubricated with PDMS. The condom used for this section of the project is 'Durex Pleasure Me'. Male participants will then be required to swab the penis in the following three areas: shaft, glans and coronal sulcus at the stated time intervals. Female participants are required to complete two blind vaginal swabs at the same stated time intervals. Female participants are also required to take a swab outside the vagina up to one hour prior to sexual intercourse to allow any external contaminants to be identified.

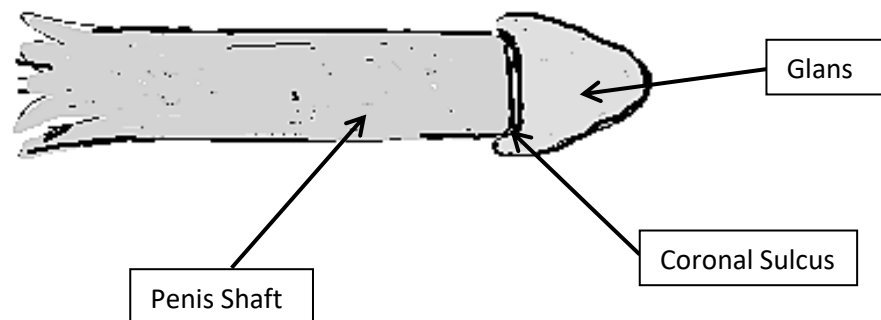


Figure 3.2.4: Labelled diagram of a penis.

Alternatively, to prevent coercion, willing participants who aren't in a relationship and potential participants whose partners feel uncomfortable in participating in the research, may opt to conduct solo sex. Male participants are directed to masturbate whilst wearing the provided condom, and female participants are advised to apply the condom to a sex toy whilst masturbating. The same conditions apply as for intercourse whereby intimate swabs are taken at the stated time intervals and a 7 day abstinence prior to taking part is required to prevent contamination.

As forensic providers only recommend lubricant analysis from sexual assault cases for up to a maximum of 24 hours following the alleged attack, the following time

intervals have been selected for participants to provide the intimate swabs: 0 hours, 6 hours, 12 hours, 18 hours, 24 hours and 36 hours. Participants are asked to fill in a Completion Sheet to ensure the time frames are being accurately adhered to and allow the possibility of any external contaminants to be identified. The following questions are asked on the completion sheet:

Gender: Male Female
(Please tick as appropriate)

I am undertaking: Sexual intercourse Solo sex

Time elapsed since intercourse/solo sex: 12 hours

Date & Time:

Swabs taken: Outer Vaginal Blind Vaginal x2 Penis shaft Penis glans Coronal sulcus

Was the provided undergarment worn? Yes No

Was any moisturiser used prior to sexual intercourse? Yes No if yes, which one?

Has any washing taken place since intercourse? Yes No

Which stage in the menstrual cycle are you at during the time of intercourse
(Female participants only):

Any additional comments?

Figure 3.2.41: Section of Participant Completion Sheet.

The prepared persistence dotation kits contained the following items. Figure 3.2.42 displays an example donation kit.

- Participant Information Sheet
- Participant Completion Sheet with labelled envelope
- 6 x Durex Pleasure Me condoms

- 6 x Pairs of undergarments, each with a labelled plastic bag.
- 6 x Sets of 3 swabs labelled with different time intervals and plastic bag. 1 x control swab and 2 x blind vaginal swabs in each set.

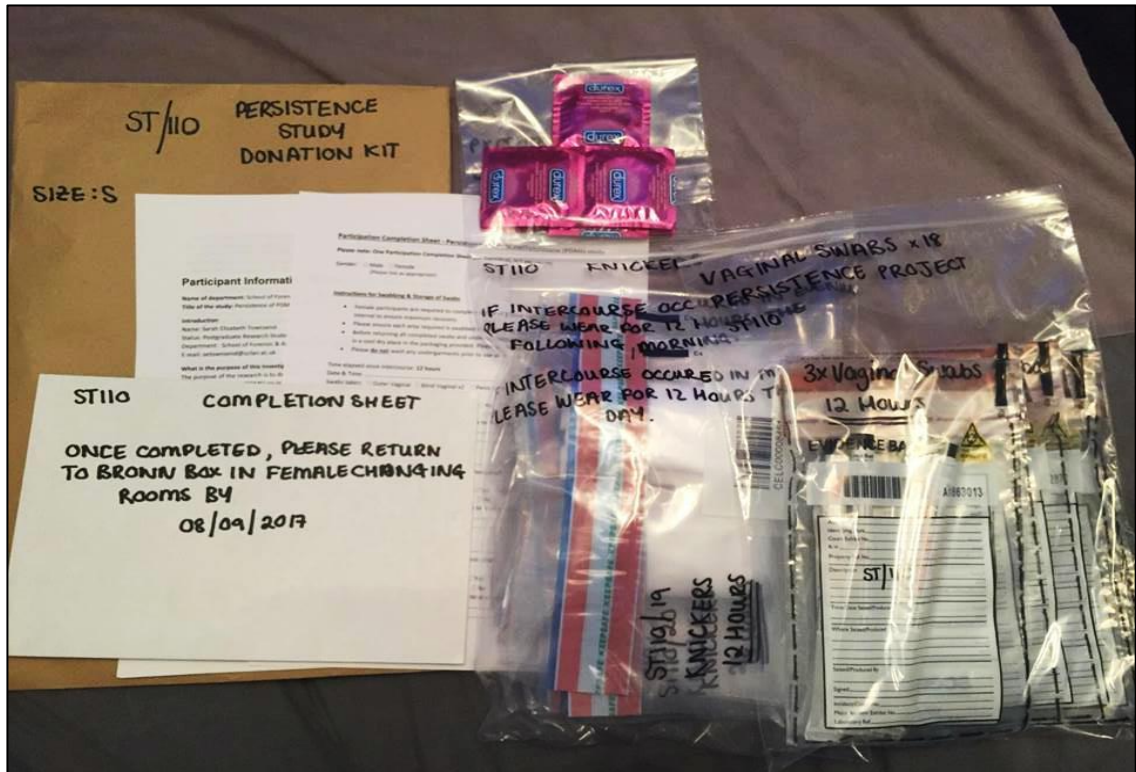


Figure 3.2.42: Persistence Participant Donation Kit.

Research into the persistence of PDMS in the vagina and on the penis gained ethical approval from University of Central Lancashire's Ethics Committee (STEMH). Consent forms were placed in the male and female changing rooms at Cellmark, Chorley. The donation kits were then prepared the following week after all consent forms had been completed and placed in the male and female changing rooms at Cellmark, Chorley.

Once swabs had been completed for each time interval, participants were asked to seal all samples in the labelled plastic packaging provided and place in the freezer in sample reception at Cellmark, Chorley. If swabs were taken at a weekend, participants were asked to store them in a cool dry place until returning to Cellmark. All intimate swabs were sperm eluted, followed by an extraction using isooctane.

3.2.5 Fabrics

All undergarments provided in the persistence study were purchased from Tesco. All knickers used in this study were 'F&F High Leg x4 Multipack' in white. They are all 95% cotton and 5% Lycra elastane. Control samples are to be taken prior to the start of the study to ensure no PDMS is present on the undergarments from manufacturing. This is crucial as PDMS is frequently applied to materials as a fabric conditioner.



Figure 3.2.5: F&F High Leg Knickers. The brand of knickers used throughout the project.

During the persistence study, participants were advised to wear the provided undergarments following sexual intercourse or solo sex to allow the recovery of PDMS from the penis, vaginal drainage or contact. This will greatly benefit the research; therefore the option for participants wearing the provided undergarment is strongly advised. Participants would wear the undergarment for approximately 12 hours following intercourse or the following day if intercourse occurred in the evening, then seal appropriately in the packaging provided.

3.2.6 Body Lotion Survey

A survey was created in order to determine the most popular body lotion and shower gels on the UK market. PDMS is a substance incorporated into many moisturisers to act as a skin protecting agent. Numerous body lotions and shower gels have been reported to still contain traces of polydimethylsiloxane despite the substance not being included on the ingredients list. The majority of individuals shower daily or every other day, therefore it is crucial PDMS is detected if present within shower gels. Furthermore, body lotions are often applied after showering and can remain on the surface of the skin once applied until washed away. The lotions can then rub off on to fabrics and undergarments which could lead to false results in sexual assault cases with regards to lubricant analysis if PDMS is present in the lotion.

The survey was compiled of the following questions:

- Are you male or female?
- Do you use body lotions?
- How often do you apply body lotions?
- What time of day do you apply your body lotion?
- Which body lotion(s) do you use most often?
- Do you use shower gel/cream?
- Which shower gel/cream do you use?

The survey was conducted by asking the questions to members of staff at Cellmark and students at the University of Central Lancashire. Also, the survey was shared on Facebook using Survey Monkey to allow a broader range of individuals to participate. A tally chart was produced to display the results for the body lotions and shower gels used by participants.

3.2.7 Body Lotion Samples

Using the results from the body lotions survey, a broad range of shower gels and body lotion samples were obtained from various households and tester samples in department stores. Samples were either squirted directly into a 2ml Eppendorf tube

or placed in the tube using a tongue depressor depending upon the packaging of the product. A total of 30 different body wash and shower gel samples and 26 different body lotions and butters of various brands were obtained. A maximum of three products from each brand was attained. The table below displays the samples acquired and whether PDMS is stated as a component within the ingredient list.

Table 3.2.7: A table to show body lotions and shower gels obtained and whether they contain PDMS.

Label	Body Lotion/Shower Gel	Brand	PDMS in Ingredients
ST25	Shower Gel	Hugo Boss Orange	No
ST26	Shower Gel	Paco Rabanne Invictus	No
ST27	Shower Gel	David Beckham Sport	No
ST28	Shower Gel	Lynx Black Night	No
ST29	Shower Gel	Lynx Gold Temptation	No
ST30	Shower Gel	Diesel Only the Brave	No
ST31	Shower Gel	Jack Wills for Men	No
ST32	Body Lotion	Ella & Rose Smoothie	Yes
ST33	Shower Gel	Ella & Rose Love to Bathe	No
ST34	Body Wash	Next Just Pink	No
ST35	Shower Gel	Pampered Coconut Smoothie	No
ST36	Body Oil	Garnier Ultimate Beauty Oil	No
ST37	Shower Gel	Radox Shower Smoothie Island Indulgent	No
ST38	Shower Gel	Lynx Attract for Her	No
ST39	Body Wash	Aussie Fresh Mate	No
ST40	Body Wash	The Body Shop Dreams Unlimited	No
ST41	Body Wash	Boots Extracts Coconut	No
ST42	Shower Gel	Impulse Hot Pink	No
ST43	Body Wash	Jack Wills Hope Cove	No
ST44	Shower Gel	Cussons Imperial Leather Fruit Salad	No
ST45	Body Lotion	Soap & Glory Butter Yourself	Yes
ST46	Body Lotion	Soap & Glory Body Butter	Yes
ST47	Body Lotion	Vaseline Intensive Care Essential Healing Lotion	Yes
ST48	Body Lotion	Tesco Loves Baby	No
ST49	Body Oil	Optima Raw Virgin Coconut Oil	No
ST50	Body Lotion	Aveeno Skin Relief Shea Butter	Yes
ST51	Body Butter	The Body Shop Deluxe Edition Chocomania	Yes
ST52	Body Lotion	Bodycare Deli Superfood Carrot & Mint	No
ST53	Body Lotion	Korres Sensitive	No
ST54	Body Lotion	Salcura Sensitive Hydrator	No

ST55	Body Lotion	Neals Yard Bee Lovely	No
ST56	Body Lotion	Seacret Pomegranate	Yes
ST57	Body Lotion	Bath House Velvet Orchid & Cardomon	Yes
ST58	Shower Gel	Sanex Zero	No
ST59	Body Oil	Sanctuary 2 day Moisture Oil	No
ST60	Shower Gel	Nivea Rich Moisture Smooth Shower Crème	No
ST61	Body Wash	Ted Baker Body Maintenance Kit	No
ST62	Body Scrub	Nuage Skin	Yes
ST63	Body Lotion	Cetaphil Moisturising Lotion	No
ST64	Body Lotion	Vaseline Intensive Rescue Moisture Locking Lotion	Yes
ST65	Shower Gel	Faith in Nature Watermelon	No
ST66	Body Lotion	Sanctuary Spa Silky Smooth	Yes
ST67	Shower Gel	Original Source Moisturising Milk	No
ST68	Body Wash	Sanctuary Spa White Lily & Damask Rose	No
ST69	Body Wash	Nivea in Shower Moisturiser Cocoa	No
ST70	Shower Gel	Original Source Peach & Apricot Shower Oil	No
ST71	Body Wash	Dove Nourishing Care & Oil	No
ST72	Body Wash	Dove Go Fresh	No
ST73	Body Lotion	Hawaiian Tropic After Sun Hydrating Lotion	Yes
ST74	Body Lotion	Hawaiian Tropic Silk Hydration	No
ST75	Bath Crème	Cussons Imperial Leather Bath Crème	No
ST76	Body Lotion	Olay Essentials Beauty Fluid	Yes
ST77	Body Lotion	Nivea Soft Moisturising Cream	Yes
ST78	Body Lotion	Nivea Crème	No
ST79	Body Butter	The Body Shop Shea Butter	No
ST80	Body Butter	The Body Shop Vitamin E Butter	No

None of the body wash or shower gels collected for analysis list PDMS in the ingredients. However, 53% of the body lotions do state that they contain PDMS. These body lotions are highlighted in bold in the table.

3.2.8 Condom Samples

As previous research states that PDMS is the lubricant applied to over 90% of condoms in the UK market, a range of different brands and designs of condoms were obtained from the high street. It is important to confirm that PDMS is still widely used as a condom lubricant, as if the volume is declining, this could have an impact on lubricant analysis in sexual assault cases. The table below lists the different condoms acquired for this study. No indication of the ingredients present within the lubricant was stated on any of the condom packaging. Multiple condoms from the

same brand have been collected to determine whether each manufacturer applies the same lubricants to a variety of their condoms.

Table 3.2.8: A table to show the brand, type and expiry of condom samples.

Condom	Expiry
Amor Extra Strong	02-2019
Amor Hot	03-2019
Boots Think Touch	08-2020
Boots Ultra-Fine	08-2014
Durex Extra Safe	03-2020
Durex Latex Free	07-2018
Durex Real Feel	06-2017
EXS Regular	02-2021
EXS Ribbed, Dotted & Flared	12-2020
EXS Smiley Face Condom	02-2021
Mates Natural	05-2021
Mates Original	03-2021
Mates Skyn Non-Latex	06-2013
Pasante Infinity	07-2019
Pasante Regular	08-2021
Skins Black Choc	04-2018
Skins Ultra-Thin	11-2021

3.3 Preparation of Swabs and Samples

All of the swabs used throughout this project are SceneSafe blank cotton sterile swabs with a black cap and plastic shaft. All of the swabs were obtained from SceneSafe Vaginal Swab Kits; however the lot and batch numbers vary between different sections of the project due to lack of stock to last the entire project. The swabs in this project are all assumed to contain no traces of PDMS unintentionally introduced during the manufacturing process.

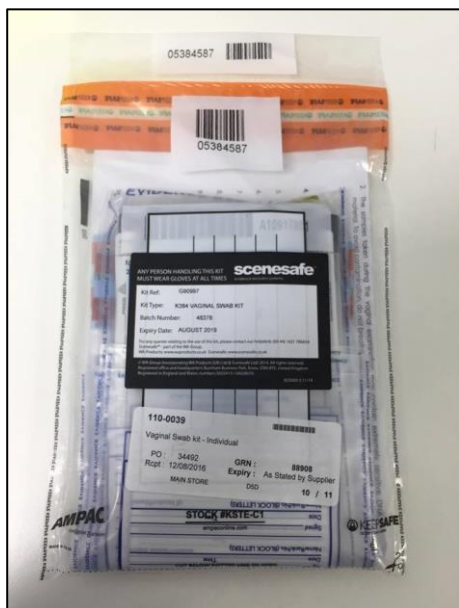


Figure 3.3: SceneSafe Vaginal Swab Kits

3.3.1 Blank Swabs

The analysis of blank cotton swabs was a requirement to ensure no PDMS is present which could compromise any findings made throughout the project. However, no preparation was required for these swabs prior to extraction and analysis. Previous research indicated that PDMS had been detected on the swab head, not in the adhesive or on the shaft. Therefore, only the swab head was required for analysis [53].

3.3.2 Limit of Detection

A total of eight different measurements of PDMS were seeded onto the SceneSafe swabs. These dilutions are 20 μ l, 10 μ l, 5 μ l, 1 μ l, 0.75 μ l, 0.5 μ l, 0.25 μ l and 0.125 μ l. Each run of swabs was repeated three times to increase the validity. A blank swab was also extracted to deduce whether the swabs were free from PDMS initially. The volumes of PDMS ranging between 20 μ l and 5 μ l were seeded using a micropipette. A new pipette tip was used in between each swab to prevent contamination and ensure an accurate volume of PDMS is applied. Volumes of PDMS ranging between 1 μ l and 0.125 μ l were seeded using a Gas Chromatography (GC) syringe. Before use and in between each swab, the GC syringe was flushed through with isooctane and wiped with a paper tissue.

3.3.3 Limit of Detection for Sperm Elution

The same process as described above was used to seed the PDMS onto SceneSafe blank cotton swabs. The same volumes were used, in addition to a swab containing no PDMS to ensure no contamination throughout the process. The same volume of semen was applied to each swab to increase the validity of the findings. Firstly, a dilution of semen was prepared at a 1:10 ratio with N-F water. N-F water has undergone Nano-Filtration which is a membrane water purification process which softens water. The dilution was then vortexed to ensure the semen and water was completely combined. This is because most males produce between 20-100 million sperm cells per 1ml of semen. Therefore, a dilution was required to ensure sperm cells can accurately be recognized during identification. A total of 50µl of the semen dilution was seeded onto each swab, including the control. The swabs were left to dry for approximately 1 hour before being placed in the freezer until further use. The PDMS was seeded on to the swab prior to semen to prevent contamination.

3.3.4 Body Fluid Matrix for Sperm Elution

A total of 5 different body fluids are often expected to be present on intimate swabs following a sexual assault. These body fluids are semen, saliva, blood, vaginal material and faecal material. Dependent upon case circumstances, these body fluids can be recovered in a variety of different combinations. 8 different possible combinations, as listed in Table 3.3.4, were seeded onto the blank cotton swabs. Three runs of each swab were seeded to increase the validity of the findings and allow any anomalous results to be ruled out.

Table 3.3.4: A table to show the combination of body fluids used for the body fluid matrix.

Swab Label			Body Fluid Combination
Run 1	Run 2	Run 3	
ST01	ST09	ST17	Semen
ST02	ST10	ST18	Semen, PDMS, Blood
ST03	ST11	ST19	Semen, PDMS, Saliva
ST04	ST12	ST20	Semen, PDMS, Vaginal Material
ST05	ST13	ST21	Semen, PDMS, Faecal Material
ST06	ST14	ST22	Semen, PDMS, Blood, Faecal Material

ST07	ST15	ST23	Semen, PDMS, Blood, Vaginal Material
ST08	ST16	ST24	Semen, PDMS, Saliva, Vaginal Material

Both the semen and the blood were diluted to a 1:10 ratio. The blood was required to be diluted to prevent clotting. Saliva was applied to the swabs neat. A 50µl volume of blood, semen and saliva was directly applied to each required swab to ensure all factors remained consistent except the variety of body fluids. 2µl of PDMS was applied to each swab as this volume is central within the range of dilutions analysed previously. PDMS was not applied to the control swabs containing just semen as this combination had been sperm eluted whilst determining the limit of detection. For combinations including vaginal or faecal material, the swabs in which the body fluid was obtained were the swabs used for the Sperm Elution, meaning vaginal/faecal material was the first fluid to be applied to the swab. PDMS was then applied to each swab, followed by semen. Each swab was left to dry for approximately one hour between the application of each additional body fluid. All swabs were then packaged in a sealed plastic bag and placed into the freezer until required.

3.3.5 Swabs for Direct Semen Extraction and Fast Differential

In forensic casework following the Sperm Elution process, the sample remains (SR), seminal pellet (SP) and epithelial pellet (XP) can be sent to Cellmark's DNA laboratory in Abingdon for further analysis if no sperm heads were identified. However, it is unknown whether lubricants are able to be extracted from the swab head following these processes. 9 blank scene safe cotton swabs were seeded with 2µl of PDMS followed by 50µl of a 1:10 dilution of semen. 3 swabs were to be sperm eluted only, 3 to be sperm eluted followed by Direct Semen Extraction and 3 to be sperm eluted followed by Fast Differential.

ST81 – ST83: Sperm Elution Only

ST84 – ST86: Sperm Elution and Direct Semen Extraction

ST87 – ST89: Sperm Elution and Fast Differential

The complete method for Direct Semen Extraction and Fast Differential is confidential and Copyrighted to Cellmark. This technique was performed by DNA analysts in Abingdon. Upon completion, the XP, SP and SR were all returned allowing the swab head to be extracted for PDMS.

3.3.6 Body Lotion Swabs

Applying each body lotion or shower gel directly onto the u-ATR was not possible due to the high water content present in each substance. The excess water displayed a very broad O-H peak between approximately $2400-3800\text{cm}^{-1}$ which masked the presence of other peaks. Therefore each sample was required to be applied to a swab, extracted and dried down prior to analysis allowing accompanying ingredients to be identified in addition to water. Approximately $20\mu\text{l}$ of each sample was applied to a blank cotton swab using a micropipette. However, a large proportion of the substances possessed a high viscosity; therefore it was not possible to use a micropipette to transfer all samples. As a result, the swabs were dipped into the Eppendorf containing the highly viscous substances to allow the transfer. As the aim of this section of the project is to determine whether PDMS is present in body lotions, the accuracy of the measurements of body lotions does not affect the outcome of the results. The swab heads were then extracted and left to dry down preceding analysis.

3.3.7 Condom Sample Swabs

The condom packaging was opened in the corner and the blank cotton SceneSafe swabs were inserted to collect a substantial volume of the lubricant from the condom. As the aim of this section of the project is to determine whether PDMS is present in the lubricants applied to condoms, the accuracy of the measurements of the lubricant from the condoms will not affect the outcome of the results. The swab heads were then extracted with isooctane and left to dry down prior to analysis using the u-ATR.

3.3.8 Persistence Knickers

Prior to knickers being issued for the persistence project, a 1cm² control section was cut away from the front right side waist area. This was then extracted with isooctane, left to dry down and analysed using the u-ATR. The results revealed that small traces of PDMS were present. This implied that if PDMS was to be detected in the gusset of the knickers following sexual intercourse with a lubricated condom, the peak intensity on the spectrum must be significantly higher for the results to be considered acceptable.

Once the knickers had been returned by participants, they were removed from the freezer and left to adapt to room temperature for an hour in the packaging. The knickers were then removed from the packaging and placed on a clean workbench. The knickers were cut at each side of the waist as shown in Figure 3.3.8 below. The gusset was then cut in half down the centre, with the left side being extracted with isooctane only, and the right side being sperm eluted, then extracted with isooctane. Each half of the gusset was cut into 1cm² segments to maximise the recovery of PDMS through extraction.

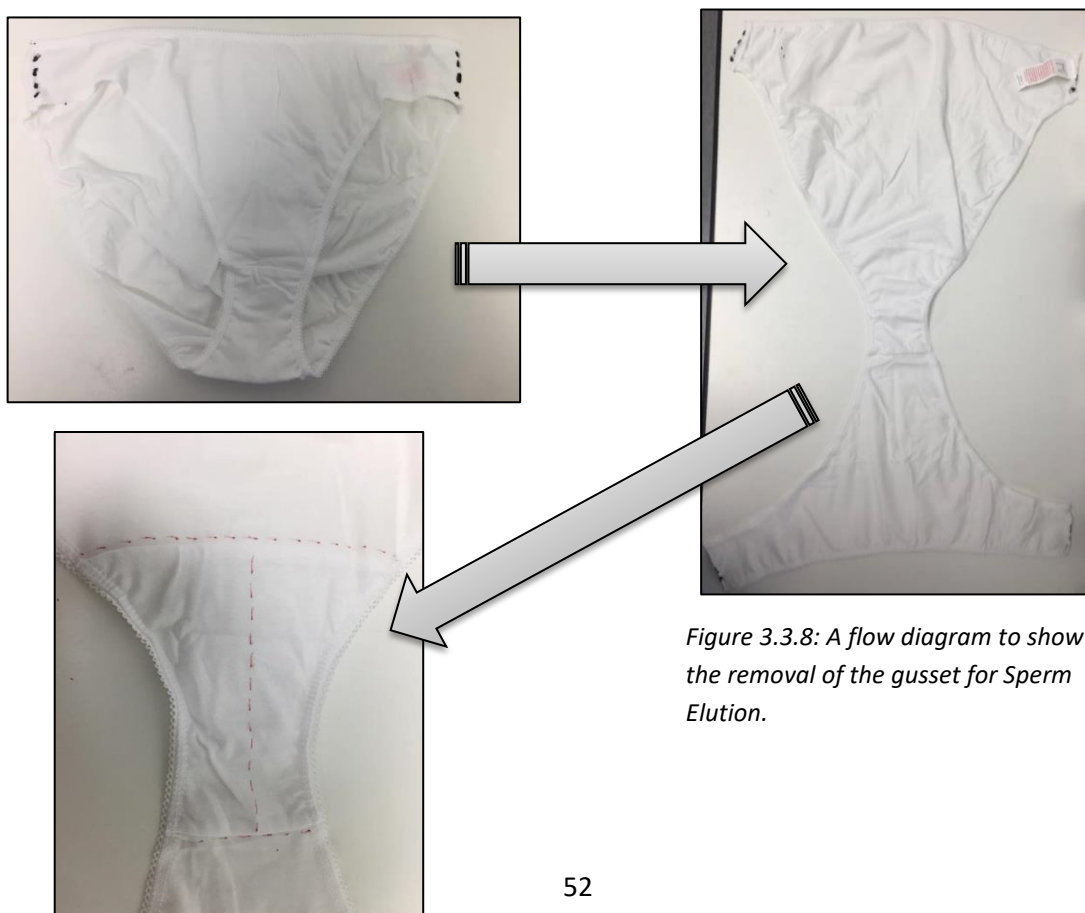


Figure 3.3.8: A flow diagram to show the removal of the gusset for Sperm Elution.

3.3.9 Washing of Knickers

Upon analysing the control areas taken from the knickers used in the persistence section of the project, PDMS was detected in minimal volumes. Unfortunately however, this could result in a false positive result in sexual assault cases. Although if volumes detected in the gusset are much higher compared to the control areas, Reporting Scientists would be more confident when presenting evidence in court that the traces are likely to have arisen from a condom.

By washing knickers, this could eliminate the PDMS from the fabric. Six control pairs of knickers, two of each size used in the persistence section of the project were obtained. This sample was then halved, whereby three pairs of knickers were to be washed using water only, and the second half to be washed using water and detergent. The washing machine used for this experiment was a 'Samsung EcoBubble WF431ABP'. The detergent used was 'P&G Professional – Fairy Non-Bio Pods'. PDMS was not indicated on the ingredients list as being present in the detergent.

Following each wash, the knickers were left to fully dry on a drying rack which had been wiped down with precept. A 1cm² section was cut away from the top inner layer of the gusset using a disposable scalpel. This was then extracted with isooctane, left to dry down and analysed using the u-ATR.

3.4 Sperm Elution Procedure

Before beginning the Sperm Elution procedure, all lab benches and seats were wiped down using a precept solution and paper towels. All equipment was thoroughly cleaned, including each well in the centrifuge. The work bench was covered with brown paper and a piece of sugar paper to complete the procedure.

The number of swabs processed per Sperm Elution varied depending on how many swabs were in each run. However, a maximum of 9 swabs were completed in each Sperm Elution. The Sperm Elution procedure is copyrighted by Cellmark Forensic

Services, therefore the exact procedure and composition of reagents used within is unable to be thoroughly documented.

- Mo Lite buffer – Dilution of phosphate buffered saline and Milli Q water.
- Mo Classic buffer – Sodium Dodecyl Sulphate stock and 0.01M Tris buffer.
- Proteinase K
- Mo Wash

Firstly, using the cap of the Eppendorf tube, the swab head was snapped from the shaft into the tube. 750µl of Mo Lite buffer was added to each Eppendorf tube using a micropipette and vortexed. The tubes were then left to incubate at room temperature for 30 minutes [59]. Following incubation, the tubes were vortexed for a second time and the swab head was transferred to a sterile spin basket using a disposable plastic needle, with the spin basket being returned to the Eppendorf tube. A different plastic needle was used per swab head to prevent contamination. Succeeding this, the tubes were centrifuged for 5 minutes at a speed of 13,000rpm. 600µl of the supernatant was then removed and transferred to an Eppendorf tube labelled 'XSN' (epithelial supernatant). The remaining pellet was then suspended in the remaining 150µl of supernatant and transferred to a separate Eppendorf tube [59].

The remaining swab heads were removed from the spin basket and placed into an Eppendorf tube containing 750µl of Mo Classic buffer and 12.5µl of Proteinase K. Tubes were initially vortexed and then left to incubate in a heat block at 56°C for 40 minutes, with samples being vortexed and at the 20 minute interval. Subsequent to this, tubes were sonicated for 5 minutes, vortexed and transported to a sterile spin basket using new plastic needles. Tubes were then centrifuged for 5 minutes at 13,000rpm [59]. The swab heads were then removed from the spin baskets and placed in the 'SR' (sample remains) tube. The supernatant was transferred to the 'SSN' (seminal supernatant) tube until approximately 70µl remained. 750µl of Mo Wash was added to the remaining seminal pellet and was re-suspended. Sample tubes were then vortexed and centrifuged for 5 minutes at 13,000rpm. The

remaining supernatant was transported to the 'SSN' tube, until the seminal pellet persisted in approximately 50µl [59].

The same procedure was performed for fabrics in addition to swabs. However, disposable forceps were used to transfer the sample between the spin baskets and tubes. Also, a maximum of four 1cm² pieces of fabric were placed into each tube. Overall, the sample preparation procedure for a Sperm Elution is complex. Each Sperm Elution procedure took approximately one working day to complete.

3.5 Direct Semen Extraction & Fast Differential

Direct Semen Extraction and Fast Differential are DNA extraction techniques performed when no sperm heads have been detected following the Sperm Elution procedure. Both methods are performed by DNA analysts at Cellmark Forensic Services, Abingdon. Upon completion of these processes, the sample remains are transferred back to Chorley via a courier to allow the swab heads to be further extracted with isooctane to determine whether PDMS had persisted on the swab head or had been removed through the second extraction.

3.5.1 Direct Semen Extraction

Direct Semen Extraction is the process undertaken when samples containing semen do not require any separation of epithelial cells and sperm cells. Therefore this is frequently a continuation method, often following the Sperm Elution procedure when no sperm heads have been identified on the slides using a microscope, or a process completed on a sample of neat semen. The instructions for completing this task contain similar steps as identified in the Sperm Elution; however a different buffer is created by combining Mo Classic A, Proteinase K and Dithiothreitol (DTT) and the method requires less time. A 'Robotic Workstation' is the system used to complete the extraction process once sample preparation is complete.

3.5.2 Fast Differential

Fast Differential is a further DNA process which can be completed after the Sperm Elution procedure if no DNA had been obtained. This technique allows nucleated

epithelial cells to be present, in addition to suspected sperm heads. Fast Differential is a much lengthier procedure in comparison to Direct Semen Extraction as the nucleated epithelial cells require to be lysed using the same buffer solution of Mo Classic A and Proteinase K, as in the Sperm Elution process. Following this, the Mo Classic A, Proteinase K and DTT buffer solution is created in order to isolate the seminal pellet. Samples are then placed in the BioRobot for the extraction to take place.

3.6 Slide Staining

In order to determine whether the Sperm Elution process has been completed successfully, microscopic examination of the epithelial and seminal pellet is required. In the epithelial pellet, both nucleated epithelial cells and sperm cells should be visible. If effective, the seminal pellet should contain only sperm cells with no cellular material present.

Approximately 150µl of supernatant remained alongside the pellet in the 'XP' tube. The volume of supernatant remaining in the 'SP' tube was dependent upon the size of the pellet. For an epithelial pellet slide, 5% of the total volume was transferred to the microscope slide. For a seminal pellet slide, 10% of the total volume was transferred to the microscope slide.

Table 3.6: A table to show the volume of supernatant to remain in the seminal pellet.

Size of Pellet	Volume of Supernatant to Remain
Small	30µl
Medium	50µl
Large	70µl

Primarily, all microscope slides were thoroughly rinsed with ethanol and wiped down using precision wipes. Slides were then placed on a hotplate set at 60°C for approximately 5 minutes to ensure they were completely dry and were then labelled appropriately. The sample tubes were then disturbed by drawing the supernatant

though the micropipette to ensure the pellet was re-suspended and evenly distributed within the supernatant. The appropriate volume was removed from each tube using a micropipette and released onto the corresponding microscope slide. The slides were then left to dry for a further 5 minutes on the hotplate prior to staining. The slides were then transported to a drying rack over the sink.

The XP slides were stained using Nuclear Fast Red and Picro-Indigo Carmine because the XP tubes displayed a larger pellet. The slide was covered with Nuclear Fast Red for 10 minutes and then rinsed with BH water. The slide was then covered with Picro-Indigo Carmine for 15 seconds and then rinsed with ethanol. The slides were then returned to the hotplate to dry.

The SP slides were stained using Haematoxylin and Eosin. This is because the SP tubes possessed a smaller pellet. The slide was covered with Haematoxylin for 90 seconds and then rinsed with BH water. The slide was then covered with Eosin for 30 seconds and then washed with BH water. The slides were then returned to the hotplate to dry.

Cover slips were then applied to the microscope slides to secure the stain in place. A wooden swab was snapped in half and used to place a small droplet of Entellan in the centre of a cover slip. The microscope slide was then firmly pressed onto the cover slip with the stain being central. The Entellan was left to dry before the slides were transferred to a microscope slide holder.

3.7 Slide Searching

Before being able to begin searching the microscope slides, it must be confirmed that the microscope is set up correctly to ensure all possible sperm heads can be identified. A Leica high powered microscope was used to complete the slide searching. Firstly, the microscope was switched on and the light was adjusted to the correct intensity. The smallest magnification, x10 was selected and the slide was placed on the stage. The right eye cup was removed and the condenser was opened until a heptagon with sharp edges could be seen through the eyepiece. The x400

magnification was then selected and this process was repeated. The specimen was then brought into focus by using the fine focusing knob. The eye tubes were then adjusted according to the interpupillary distance.

Slides were then searched using the X/Y stage control going from left to right and vice versa until the whole stain had been covered. The focus had to be adjusted depending on which stain had been used for the slide. Figures 3.71 and 3.72 show examples of an epithelial slide and a seminal slide. Both of these images are sections of the XP and SP sperm eluted from the same swab. It can be seen that larger volumes of sperm heads are recovered in the seminal pellet in comparison to the epithelial pellet. In Figure 3.71, the nucleated epithelial cells are stained green by the addition of Picro Indigo Carmine and the sperm heads are stained in red from Nuclear Fast Red. Figure 3.72 displays the seminal pellet whereby the sperm heads are stained purple by the Haematoxylin.



Figure 3.7: Leica High Powered Microscope

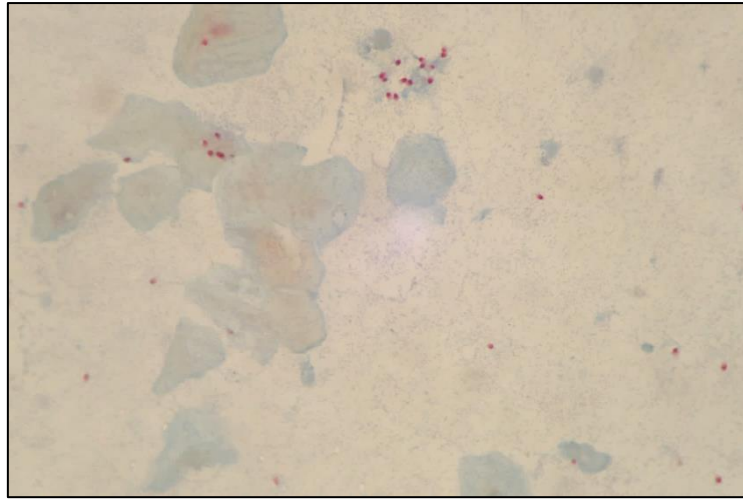


Figure 3.71: Epithelial Pellet containing nucleated epithelial cells and sperm heads after Sperm Elution.

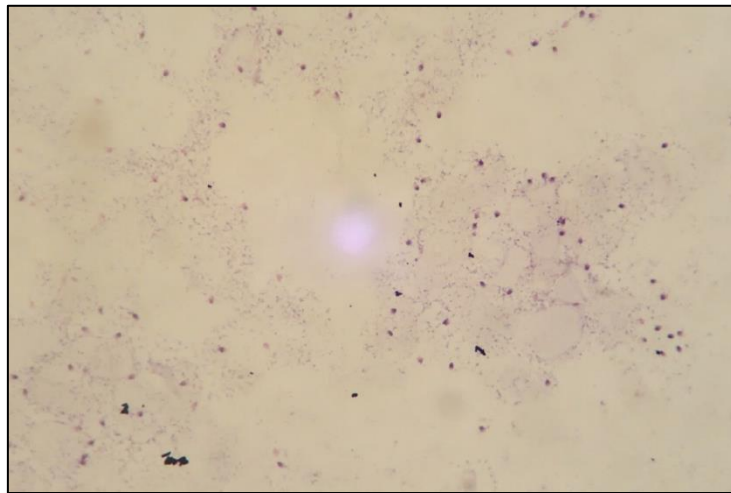


Figure 3.72: Seminal Pellet containing sperm heads after Sperm Elution.

Furthermore, when sperm eluted, the samples initially contain substantial volumes of PDMS. A portion of this is often detached from the swab during the process. PDMS can be visualised on microscope slides as small, clear and spherical globules. Although this is not a confirmatory method for identifying PDMS, the XP or SP pellet could then be analysed using FTIR to confirm this theory. Figure 3.73 displays a section of a seminal pellet from a sperm eluted swab initially containing 5 μ l of PDMS. Some of the globules of PDMS have been circled in black.

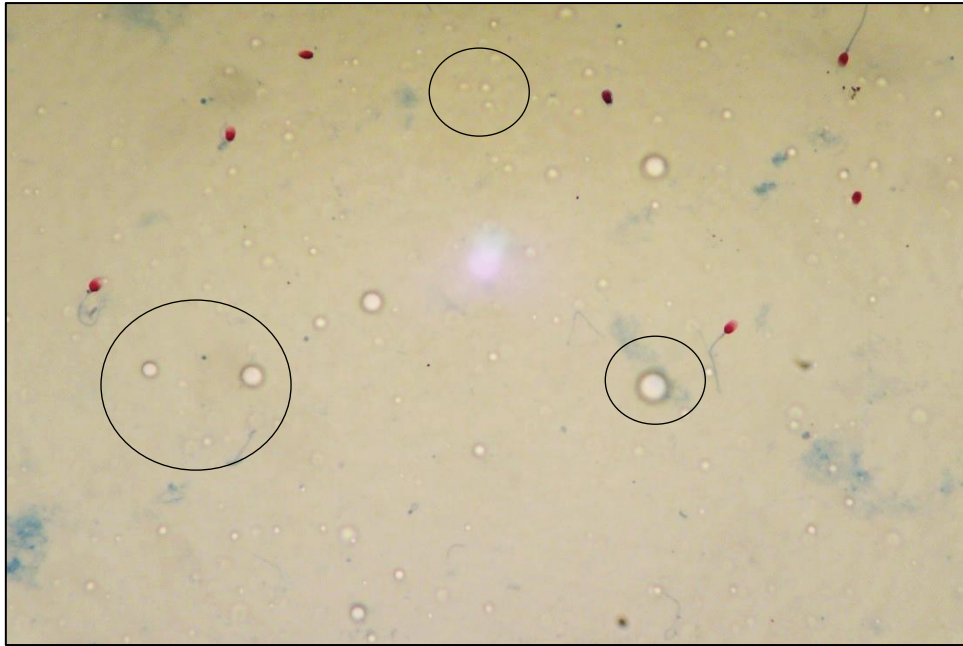


Figure 3.73: Seminal Pellet containing PDMS and sperm heads after Sperm Elution.

3.8 Isooctane Extraction

3.8.1 Extraction Preparation

Prior to analysis using either the u-ATR or FTIR microscope, all samples are required to be extracted. The extraction process took place in a fume cabinet at Cellmark Forensic Services to prevent any exposure to the chemicals used during the procedure. Before initiating the extraction, it was ensured the workspace is thoroughly cleaned down and all surfaces are wiped using a tissue moistened in isooctane solution. 10cm rimless test tubes, screw top glass chromatography (GC) vials and pipettes were all rinsed three times with isooctane. The third washing was retained and transferred to a GC vial labelled 'washings' to ensure any recovered substances are as a result of the sample not the glassware. All test tubes and GC vials were then labelled with the corresponding values to each sample using a permanent marker.

3.8.2 Extraction Process

Initially, the swab head was removed from the shaft of the swab by snapping the top of the shaft using the cap of the Eppendorf tube. This stage of the procedure was

skipped if the swab head had already been sperm eluted. The swab head was then placed into a clean, labelled test tube using a plastic needle. The needle was disposed of after every swab head to minimise the chances of any transfer of contamination between samples. 2ml of isooctane was then measured accurately using a micropipette and added to the test tube. The swab head was left to immerse in the solvent for approximately 2 minutes to ensure the solvent had fully combined with the swab head. The solvent was then drawn through the swab head three times using a Pasteur pipette. This was to ensure any possible substances within the swab head were released into the isooctane solution. The extracted swab head was then retained in the original Eppendorf tube and the solvent was transferred using a Pasteur pipette into a clean, labelled GC vial. The sample was then reduced down to dryness for roughly 24 hours in a fume cabinet and then screw capped to prevent contamination.

The same process was completed with fabric materials; however disposable forceps were used as an alternative to plastic needles to transfer the sample between the test tube and the Eppendorf tube.

3.9 Fourier Transform Infrared Spectroscopy

Before analysis using the FTIR, each sample required small additions of isooctane in order to be transferred to the glass window. The same brand of isooctane was used throughout the project to maintain consistency of any findings. Isooctane is the chosen solvent because it is less toxic compared to hexane which was used in previously conducted experiments. Blank samples are also completed in between each sample to prevent any false results arising. The initial sample completed on the FTIR at the start of each visit is an isooctane sample. This is to ensure no contaminants are present in the solvent which could provide incorrect results, hence reducing the reliability of the research. All glassware washings were analysed prior to any samples to ensure if any PDMS was to be detected, none of this arose from contamination within the equipment.

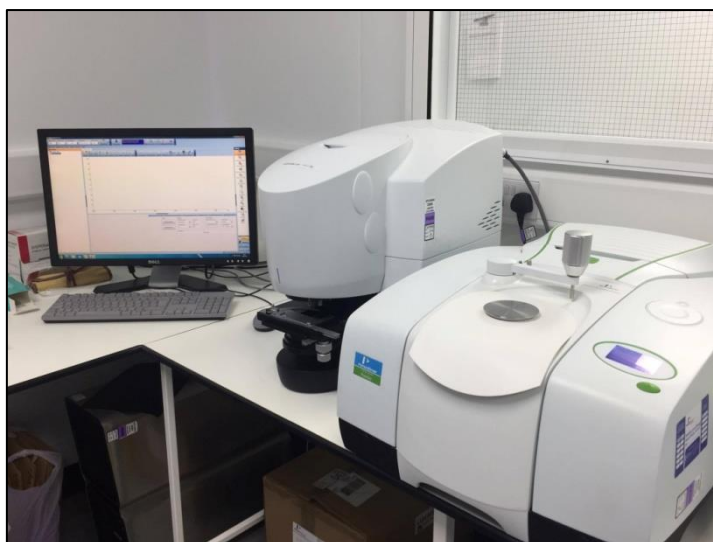


Figure 3.9: Perkin Elmer FTIR Equipment.

3.9.1 u-ATR FTIR

The u-ATR instrument used at Cellmark was a Perkin Elmer Frontier FTIR Spectrometer containing a Zn/Se crystal possessing a single reflection. Each spectrum represented a collection of 4 different scans and a wavelength within the range of 500cm^{-1} - 4000cm^{-1} . The instrument beam path was then set up and the instrument was aligned. The energy levels were then monitored as the required energy is above 600 for the technique to be successful. Only one spectrum was completed for each sample.

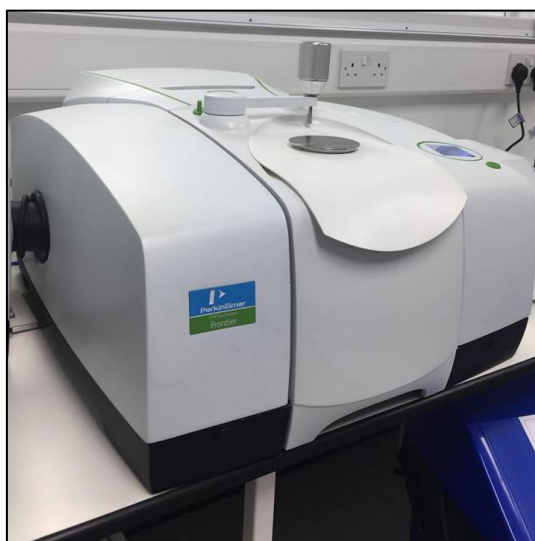


Figure 3.9.1: Perkin Elmer Frontier Universal Attenuated Total Reflectance.

Firstly, the crystal and surrounding area was thoroughly wiped down using isooctane and a lint free tissue. A preview scan was then viewed to ensure the crystal was contaminant free prior to running the background. If unexpected peaks were present, the crystal was carefully wiped for a second time. A background scan was then taken and completed in between each sample. Two Pasteur pipette drops of isooctane were transferred into the GC vial containing the sample and the liquid was rotated around the vial to allow any residue from the side of the vial to be reconstituted into the isooctane. Using a micropipette, the pipette tip was immersed into the sample resulting in a drop of solution gathering in the end of the pipette tip. The micropipette was used only to support the pipette tip. The sample was not drawn up the pipette tip as there would be too much liquid for the technique to be effective. The sample was then spotted directly onto the Zn/Se crystal and left to evaporate. A total of 5 spots were applied for each sample in increments. This is because if too much of the sample was applied to the crystal, the solvent would migrate away from the crystal towards the edge of the plate. Any sample which is not placed directly on the crystal would not be analysed.

3.9.2 FTIR Microscope

The FTIR instrument used at Cellmark was a Perkin Elmer FTIR with a Spotlight 150i microscope attachment. A microscope is required during the process because if any PDMS is present on the swabs, the sample would be too small to visualise with the naked eye, and therefore visual enhancement is necessary. The samples were viewed on a computer using a piece of software named Spectrum.



Figure 3.9.2: Perkin Elmer Spotlight 150 FTIR Microscope.

The method used to prepare the extracted sample for analysis using the FTIR is explained below:

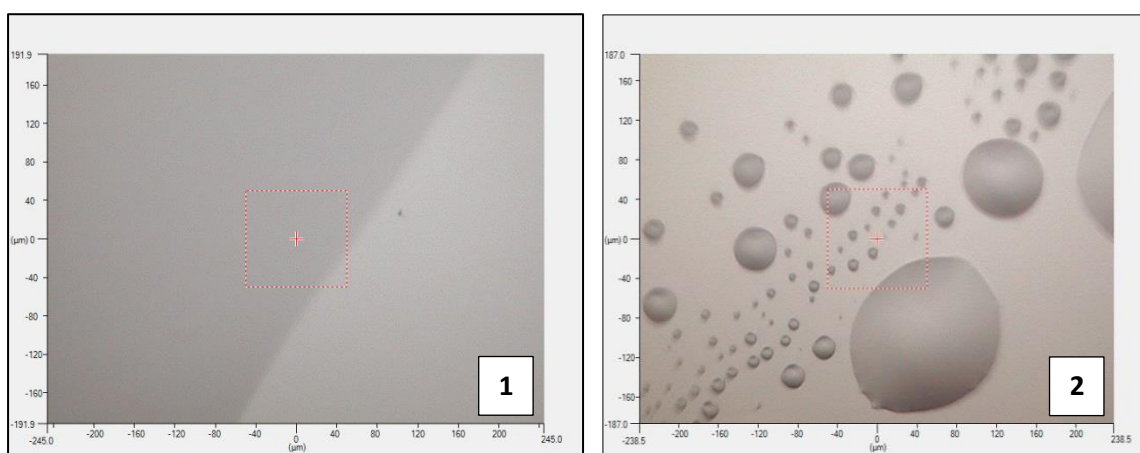
- The Spectrum software was opened and the microscope was aligned and calibrated. Secondly, the correction was set to $-500\mu\text{m}$ to increase the focus of the light beam from the microscope. This is because a 1mm BaF_2 glass window was used throughout the analysis which is approximately half the thickness of a standard glass window. Therefore, in order to accurately visualise the substances present, the correction is required to be reduced.
- A BaF_2 window was used as an alternative to the standard KBr window it contains water resistant properties against liquids, meaning no absorption of moisture from any of the samples.
- The FTIR was filled with approximately 4-5 funnel measurements of liquid nitrogen until an energy level between 3500-5000 is reached and remains fairly stable with no fluctuations. This takes approximately 10 minutes. The energy level was monitored throughout the samples to avoid any variations between results. It was important not to add any more liquid nitrogen than what is required. This would cool down the equipment too much and would be unable to maintain a stable energy level.

- Firstly, a background sample in air was run to confirm there are no contaminants present on the microscope lens.
- The BaF₂ window was thoroughly wiped down with isooctane solution and the microscope slide was placed on the stage to run the spectra. It was checked for unidentified peaks on the spectra. If present, the BaF₂ window was cleaned for a second time to guarantee all previous residues have been removed. The spectrum was then saved as a window blank.
- A background spectrum was then run on the blank window.
- Two Pasteur pipette drops of isooctane were added into the GC vial containing the sample and the liquid was rotated around the vial to allow any residue from the side of the vial to be re-suspended into the isooctane.
- Using a micropipette, the pipette tip was immersed into the sample resulting in a drop of solution gathering in the end of the pipette tip. The micropipette was used only to support the pipette tip. The sample was not drawn up the pipette tip as there would be too much liquid for the technique to be effective.
- The sample was then spotted onto the centre of the BaF₂ window from the pipette tip allowing the spot to evaporate in between spotting. Much care was taken to ensure the correct amount of solution was added to the window. If too much solution was added to the window, the solution would spread to the outer edges making it very difficult to identify substances within the solvent, especially in trace volumes.
- Once spotted, the slide was then placed on the microscope stage and viewed using the Spectrum software. The stage was adjusted until an area within the spot is located. A section close to the edge of the spot is preferred as if any PDMS is present; it migrates to the outer edge of the spot. The spectrum was then run on the chosen position.
- A different area within the sample was located and a total of three spectrums were run on each sample because if PDMS is present in a trace quantity, it may only be present in a specific position within the spot. Also, completing three spectrums allows any anomalous results to be identified.

- Samples containing contamination other than PDMS would then be compared to spectra on Cellmarks' internal database for comparison against known samples.

Each spectrum represented a collection of 16 different scans and a wavelength within the range of 650cm^{-1} - 4000cm^{-1} . Multiple scans are used to help eliminate background noise and interference caused by the equipment and the sample background environment. All spectra, including the background blanks, are saved in between each sample to prevent any loss of results if the software failed.

The images below show the view of samples using the FTIR microscope. The first image displays the view when $2\mu\text{l}$ of PDMS was spotted directly onto the u-ATR. A clear border is present whereby the sample can easily be distinguished. The second image illustrates the extract when a swab containing $5\mu\text{l}$ of PDMS had been extracted. Round globules with a darker circumference and paler centre can be observed. This is because PDMS often migrates to the outer edges of the spherical globules, resulting in a more opaque colour. The third image presents the view when a swab containing $0.125\mu\text{l}$ was extracted and analysed. The drops present are much fewer in quantity and smaller in volume, and are much lighter, thus containing lower amounts of PDMS. Finally, the fourth image contains the extract of a blank swab containing no PDMS. No spherical drops can be visualised, due to only phthalates, if any substances being detected in extremely small volumes.



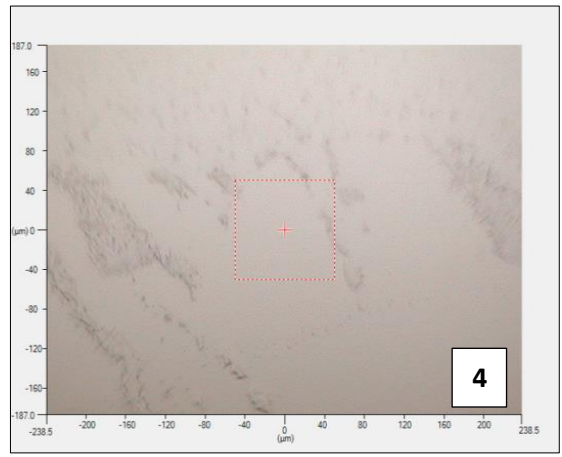
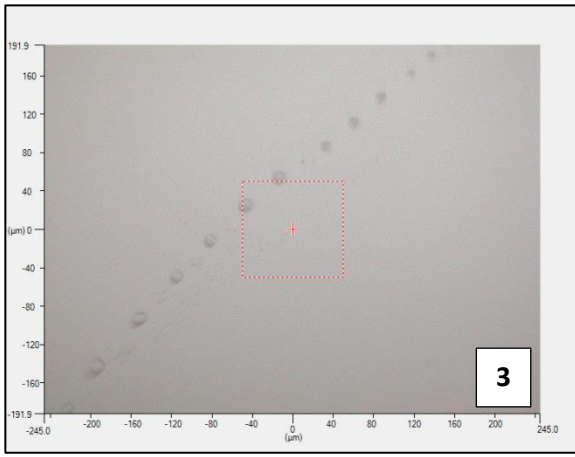


Figure 3.9.21: View of samples using an FTIR Microscope.

Chapter 4 – Results

4.1 Reference Spectrum of PDMS

The spectrum below, Figure 4.1, displays the reference sample of PDMS which was obtained using a sample of PDMS donated by 'Mates' which they apply to their condoms during the manufacturing process. Figure 4.11 shows the same spectrum which is focused on the fingerprint region. The peaks present have been labelled. It can be seen that the four peaks present at approximately 800cm^{-1} , 1020cm^{-1} , 1090cm^{-1} and 1260cm^{-1} are characteristic of PDMS.

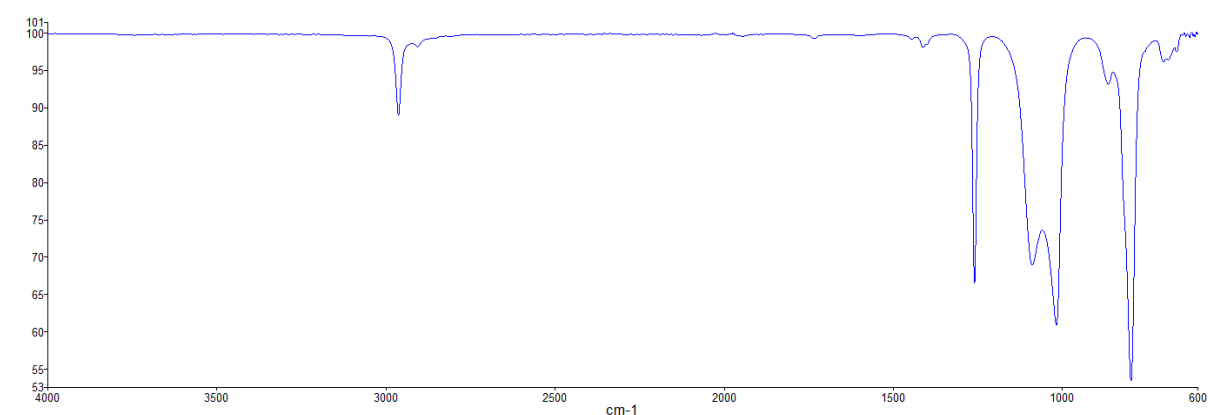


Figure 4.1: The reference spectrum of PDMS from the u-ATR.

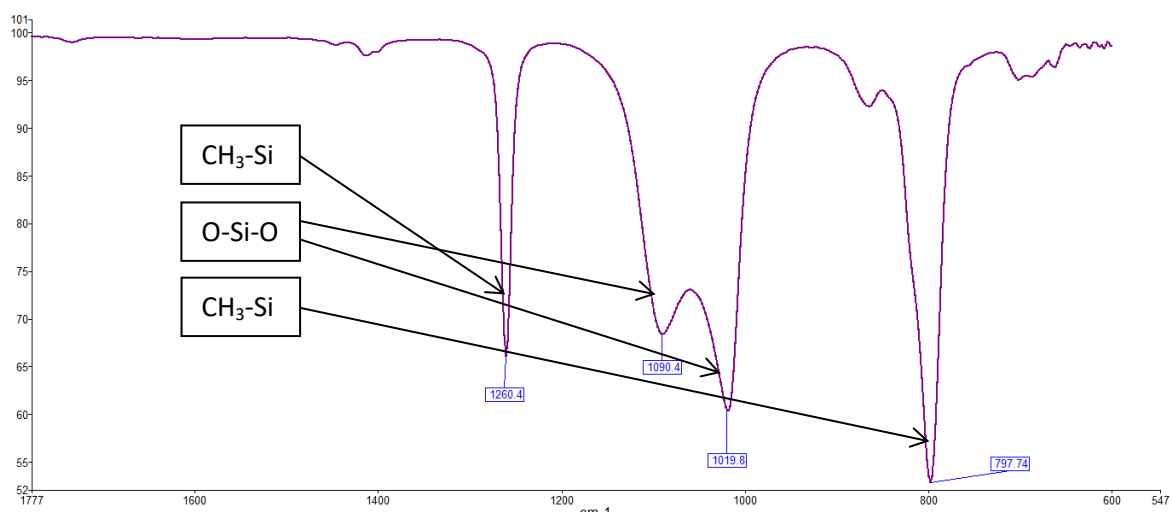


Figure 4.11: A zoomed in reference spectrum of PDMS from the u-ATR.

4.2 Glassware Washings & Window Blanks

4.2.1 Glassware Washings

The glassware washings from the equipment used throughout this project presented negative for PDMS. No unknown substances were identified through the glassware washings, only background noise could be seen from the spectrum. Figure 4.2.1 shows three examples of the spectra obtained from the glassware washings.

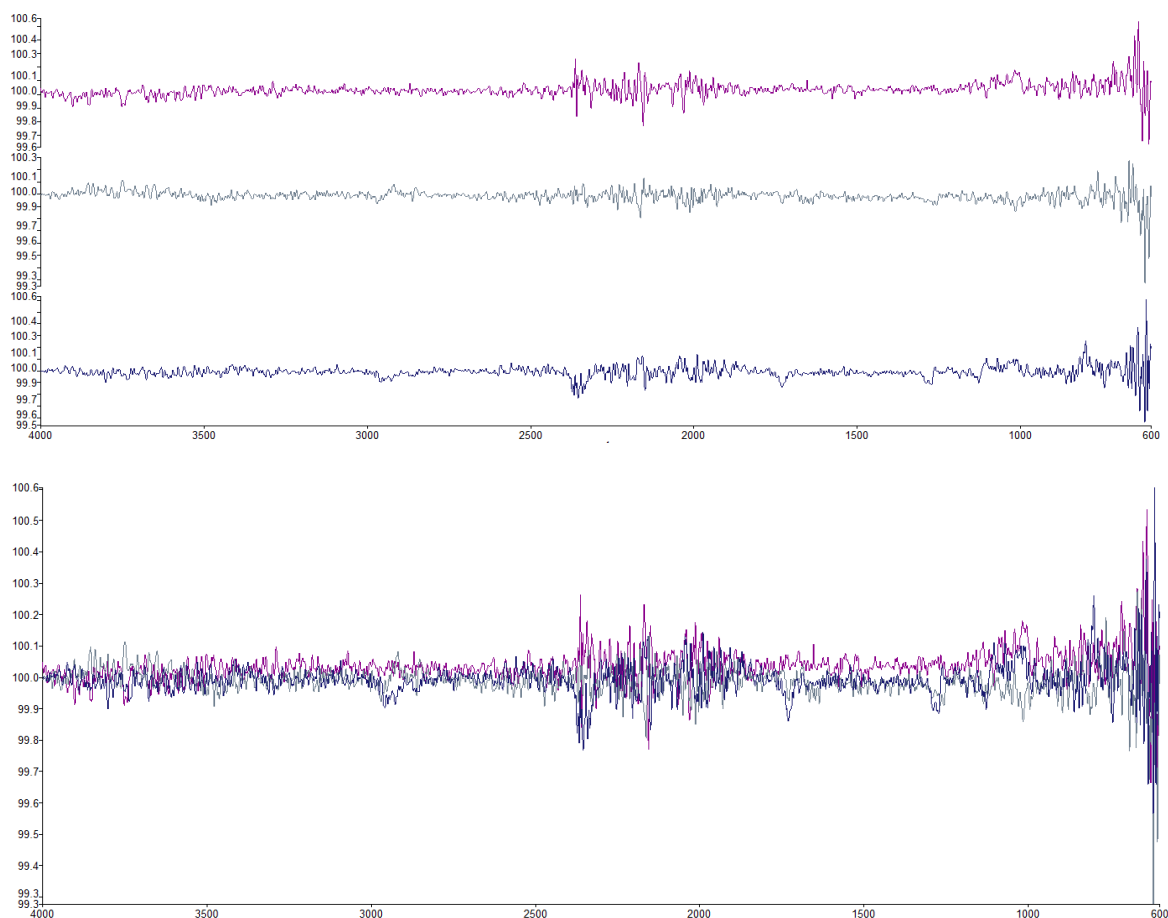


Figure 4.2.1: Spectra of 3 glassware washings samples.

4.2.2 BaF₂ Window Blanks

The spectra taken from both the u-ATR crystal and FTIR microscope prior to applying the sample were all blank. Figure 4.2.2 below shows an example of 6 different window blanks displaying very similar spectra with no peaks present in the fingerprint region.

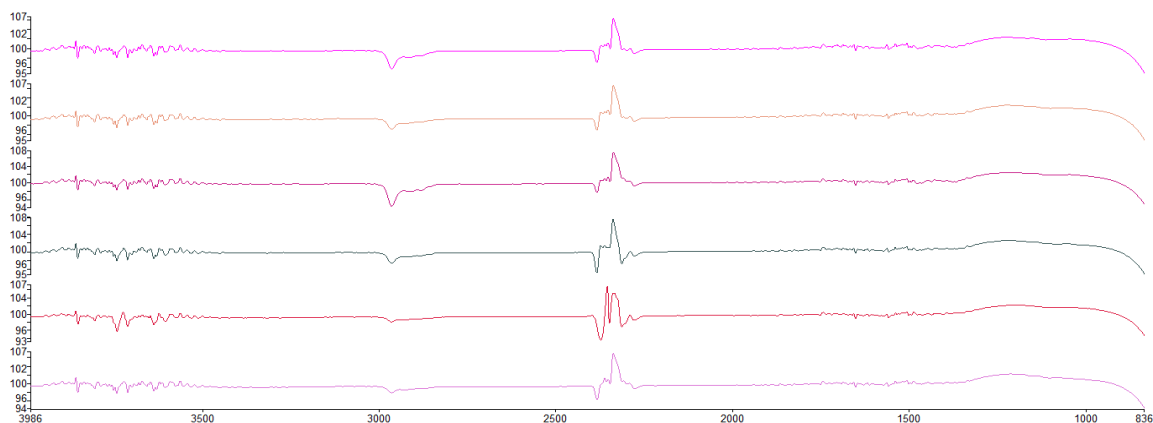


Figure 4.2.2: Spectra of 6 BaF₂ window blanks.

An impurity peak is present at approximately 2300-2400cm⁻¹. This peak represents CO₂ and arises as a result of the measuring conditions. For example, breathing whilst recording the sample would result in a CO₂ peak. The peak at approximately 2950cm⁻¹ is a C-H impurity.

4.3 Blank Swabs from Sexual Assault Kits

A total of 24 blank cotton Scene Safe swabs were obtained from 4 different Sexual Assault Kits donated to Cellmark from Merseyside Police and Merseyside Sexual Assault Referral Centre. All of the swabs present in the kits displayed negative for traces of PDMS. As previous research discovered PDMS had only been detected on the swab head, the shaft and adhesive were not analysed during this project.

4.3.1 Swabs Analysed using the FTIR Microscope

The spectrum below, Figure 4.3.1, shows three different areas of an extract taken from the same swab. Swab 11 from Sample 4 was selected at random from all of the swabs containing low levels of contamination. It can be seen from the zoomed in fingerprint region that the spectra between different areas of the extract are very similar, with little variation between the intensities.

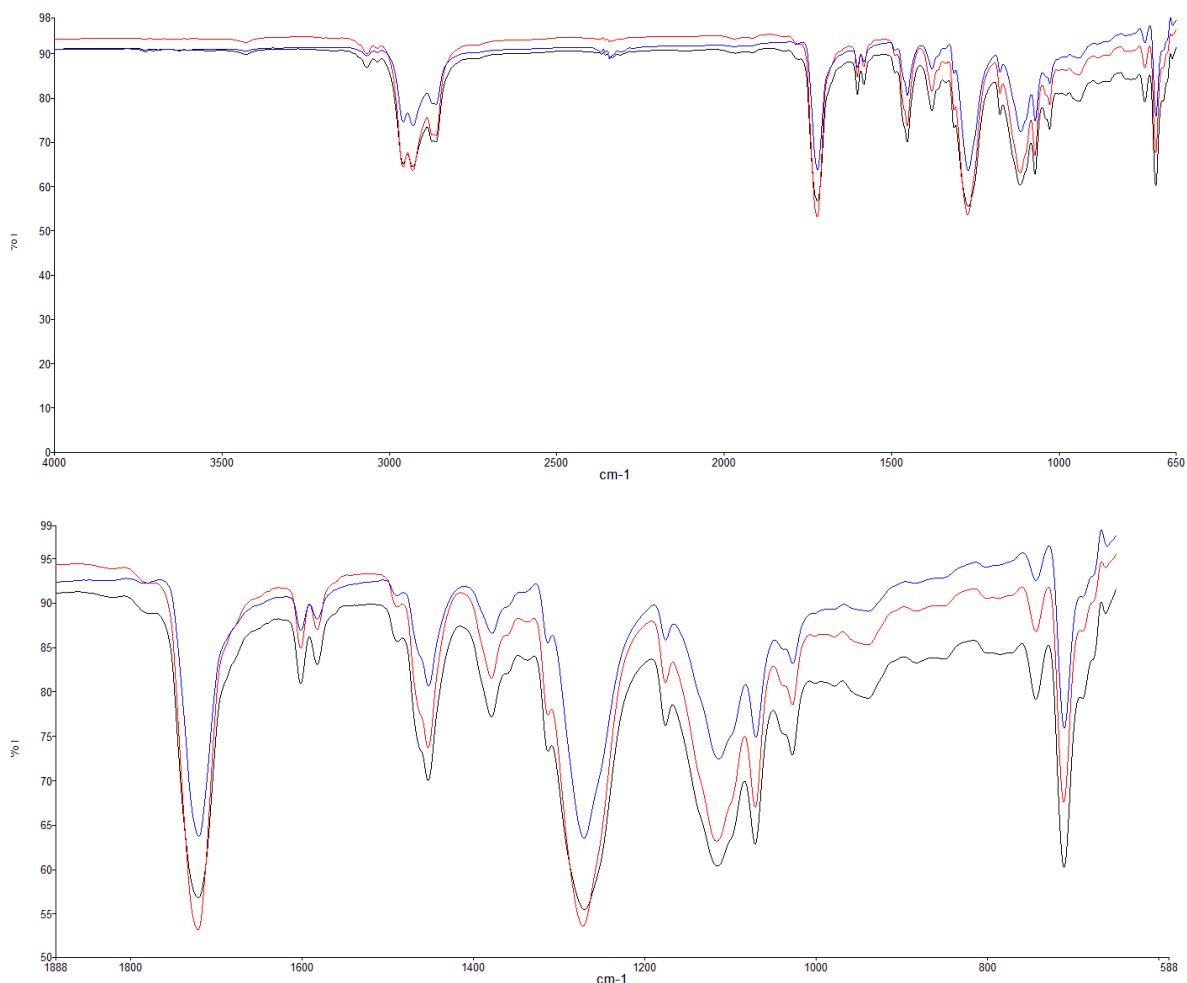


Figure 4.3.1: Spectra of a Blank Cotton Swab extract, Sample 4 Swab 11, and the focused fingerprint region.

An 86% match was made between this swab and Dibutyl phthalate. However, phthalates are present in many everyday items; therefore it is strongly likely that this is a result of the isooctane extracting the phthalates from the micropipette tips. Figure 4.3.12 shows the comparison between Sample 4 Swab 11 and Dibutyl phthalate. Although an 86% match is recorded, there are very few similarities between the peaks and the extract from the blank swab displays peaks with a much lower intensity.

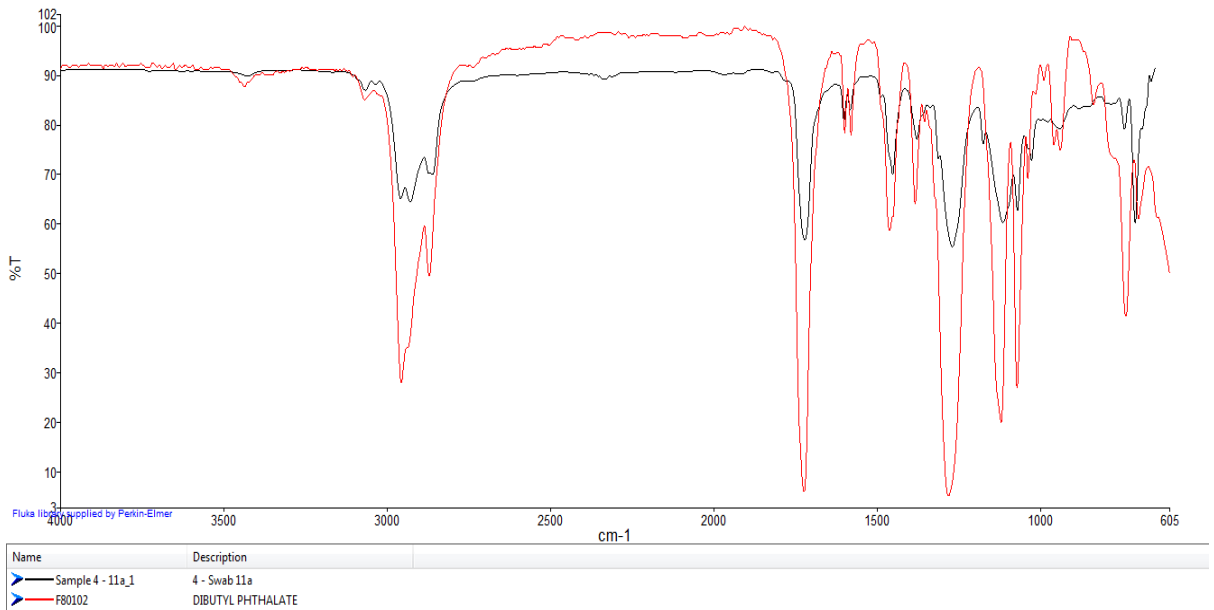


Figure 4.3.12: Spectra of Sample 4 Swab 11 compared to Dibutyl phthalate.

When Sample 4 Swab 11 was then compared to the reference spectrum of PDMS, it is clear there are no distinct similarities between the peaks meaning there is no PDMS contamination on the blank cotton swab.

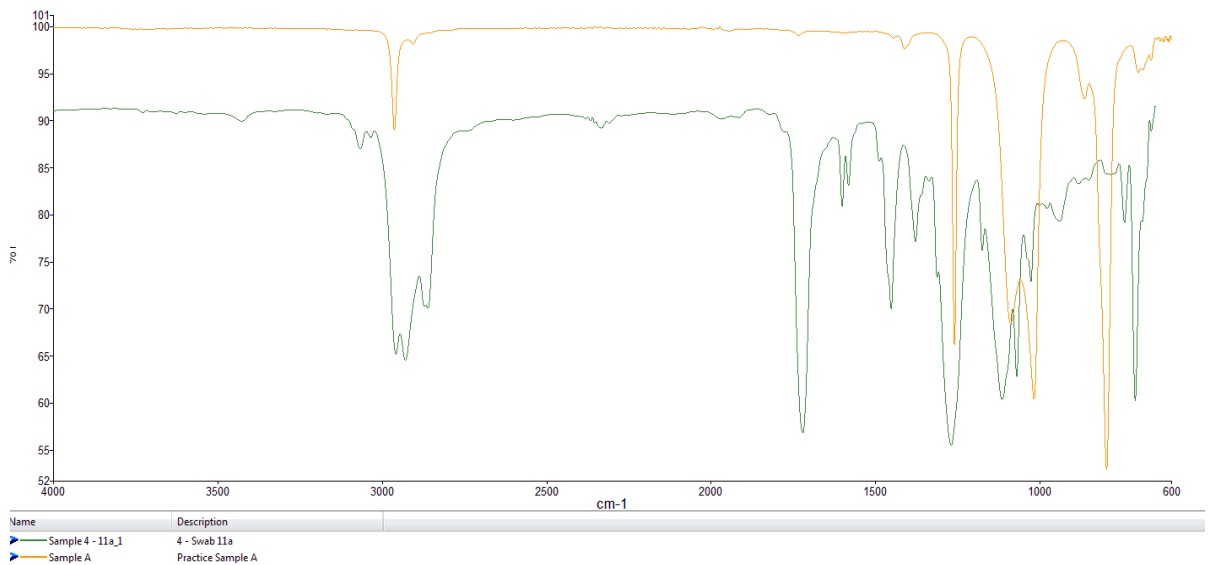


Figure 4.3.13: Spectra of Sample 4 Swab 11 compared to PDMS.

Below is a selection of 4 different blank swabs selected at random. All of the swabs demonstrate a very similar spectrum, but vary in intensity. All of the swabs display a very close match with Dibutyl phthalate.

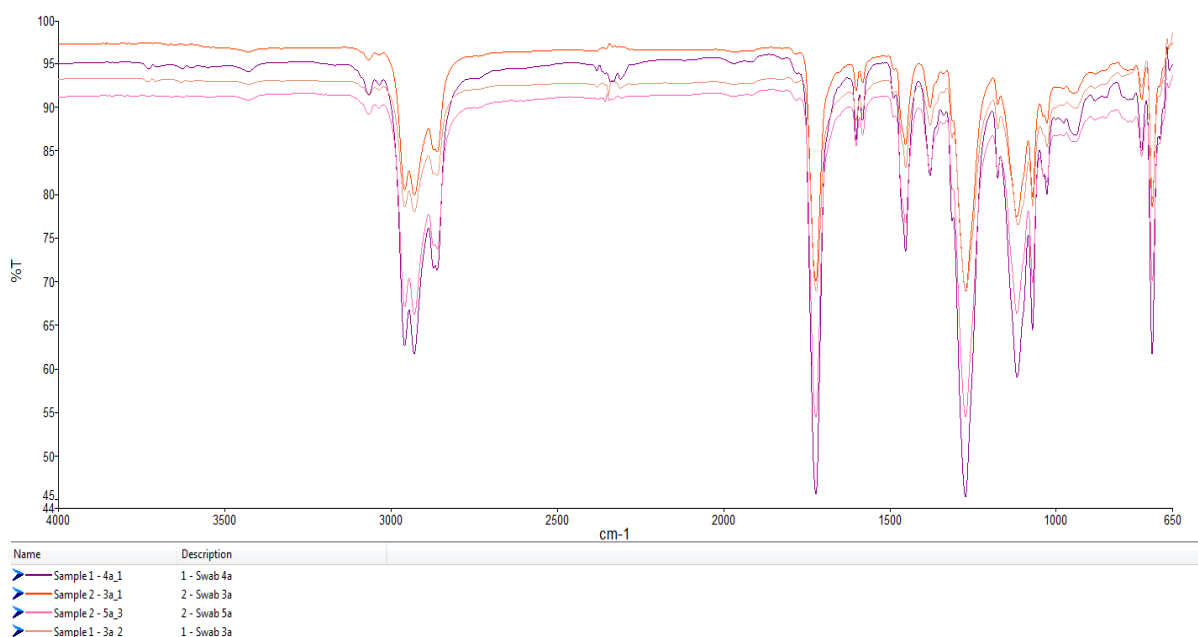


Figure 4.3.14: Spectra of 4 random blank cotton swabs using FTIR Microscope.

4.3.2 Swabs Analysed using the u-ATR Comparison

All of the swabs analysed using the u-ATR displayed the same peaks present on the spectra as when analysed using the FTIR microscope. Though, much variation occurred with the intensity of the peaks. Generally, the FTIR microscope produced spectra of a higher quality. Significant background noise and undefined peaks were observed when using the u-ATR when compared to the FTIR microscope.

Figure 4.3.2 illustrates a spectral comparison containing an example blank cotton swab taken from a sexual assault kit. The red spectrum was attained using the u-ATR and the blue using the FTIR microscope. Both spectrums observed were produced using extracts from the same swab. Although little background noise is observed using the u-ATR, very few peaks are able to be clearly identified and those which are visible possess a much lower intensity.

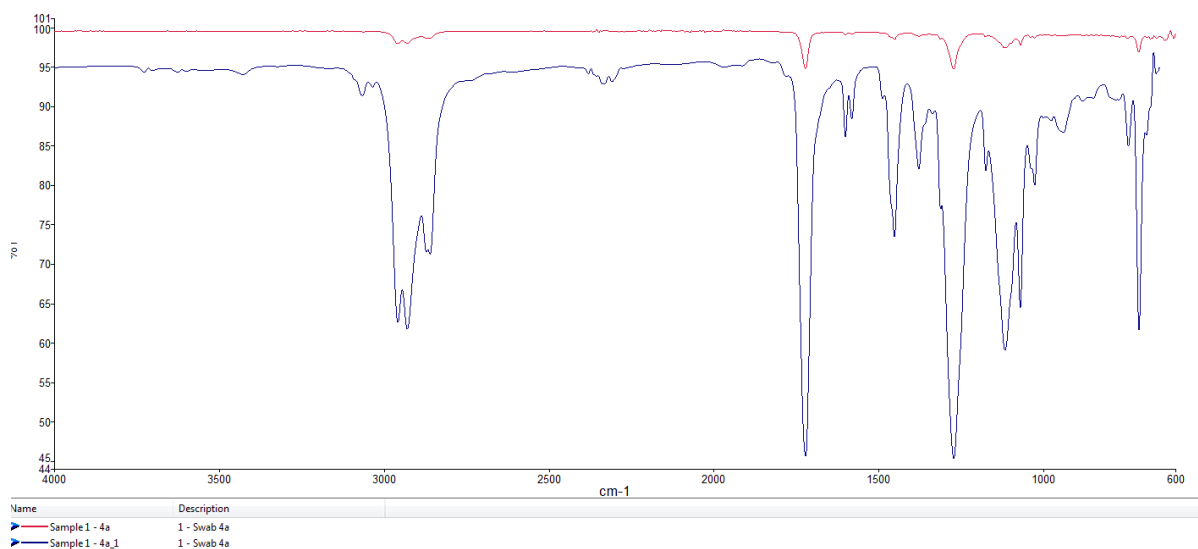


Figure 4.3.2: Spectra of Sample 1 Swab 4a – Comparison between FTIR microscope and u-ATR.

Figure 4.3.21 on the following page shows the spectrum of the same 4 random swabs as presented in Figure 4.3.14. However, the u-ATR was operated for the analysis as an alternative. Greater background noise is observed between the regions of 2000-2500 cm^{-1} , with the baseline as a whole not being as smooth when compared to the FTIR microscope. Furthermore, the intensity of the peaks is momentarily reduced with the lowest percentage transmission being approximately 85 compared to the FTIR microscope being roughly 45.

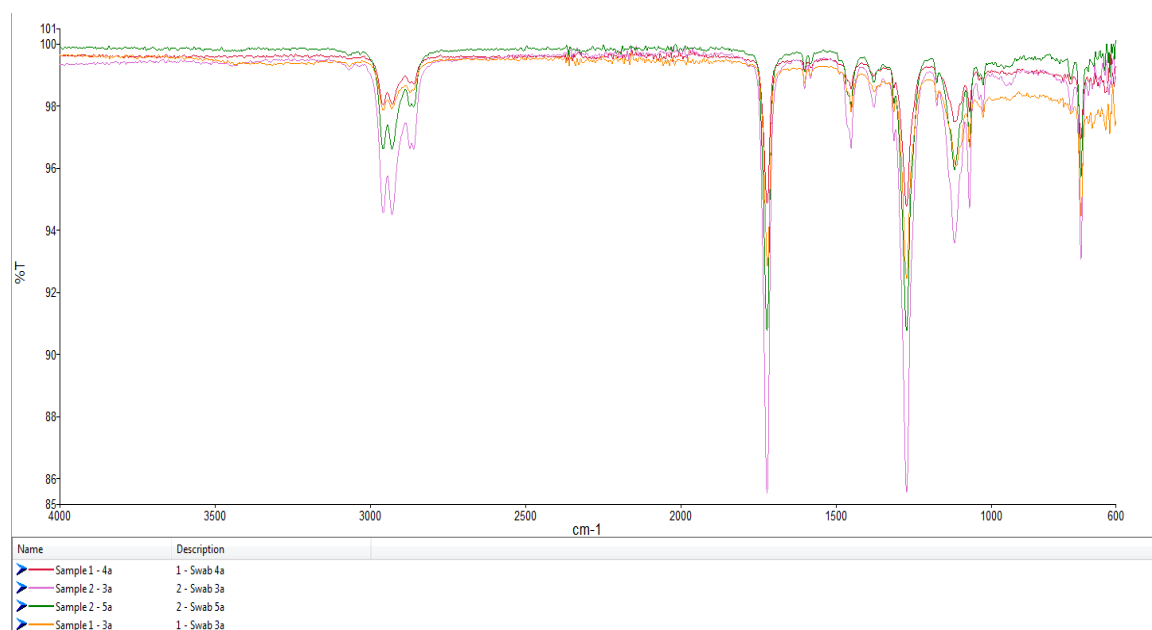


Figure 4.3.21: Spectra of 4 random blank cotton swabs using u-ATR.

Figure 4.3.22 below presents 2 different blank swabs from a sexual assault kit selected at random. The spectra shown in green represent the u-ATR and the purple signify the FTIR microscope. Again, the peaks are much more intense and sharper when using the FTIR microscope. However, the spectrum from the u-ATR still identified the blank swab as containing Dibutyl phthalate.

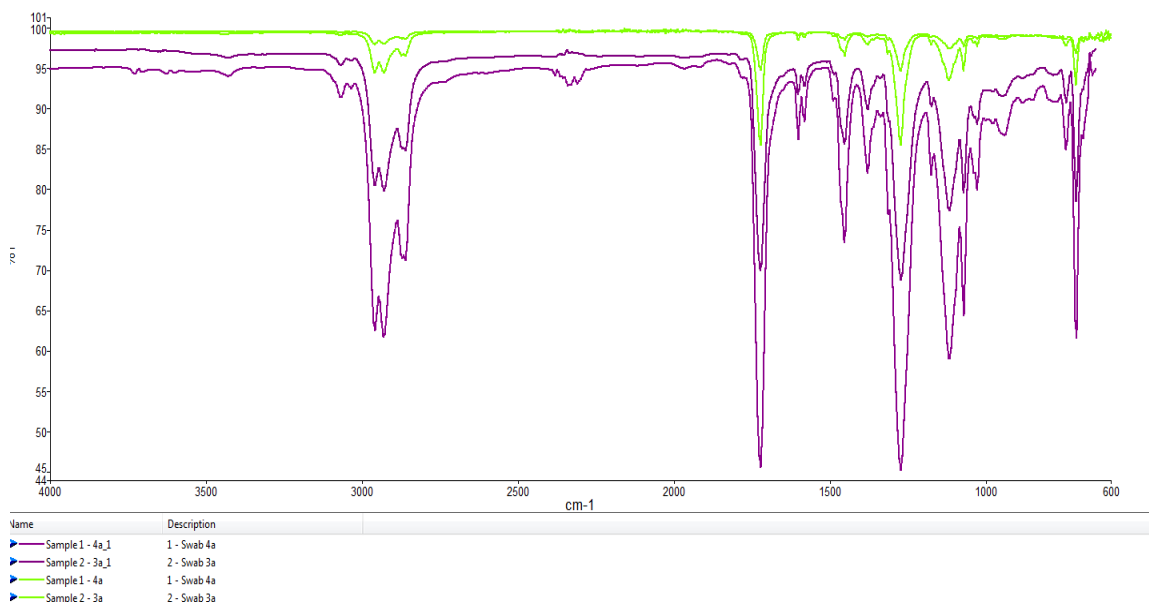


Figure 4.3.22: Spectra comparison of 2 different swabs from Sexual Assault Kits using u-ATR and FTIR Microscope.

4.4 Blank Swabs from Vaginal Swab Kits

A total of 3 Vaginal Swab Kits were obtained from Cellmark. Each kit contained 10 blank Scene Safe cotton swabs. All of the swabs present in the kits displayed negative for traces of PDMS. As previous research discovered PDMS had only been detected on the swab head, the shaft and adhesive were not analysed during this section of the project.

Figure 4.4 below shows 4 different blank swabs, with at least one taken from each Vaginal Swab Kit. The intensity of these peaks is very low with high levels of background noise. This indicates the contamination levels of these swabs are minimal. No PDMS was detected in any of the samples shown in Figure 4.4. Despite the same swabs being used in both the sexual assault kits and the vaginal swab kits,

the contamination levels appear to be less significant in the vaginal swab kits. This suggests that the volume of contamination may vary between different batches of swabs.

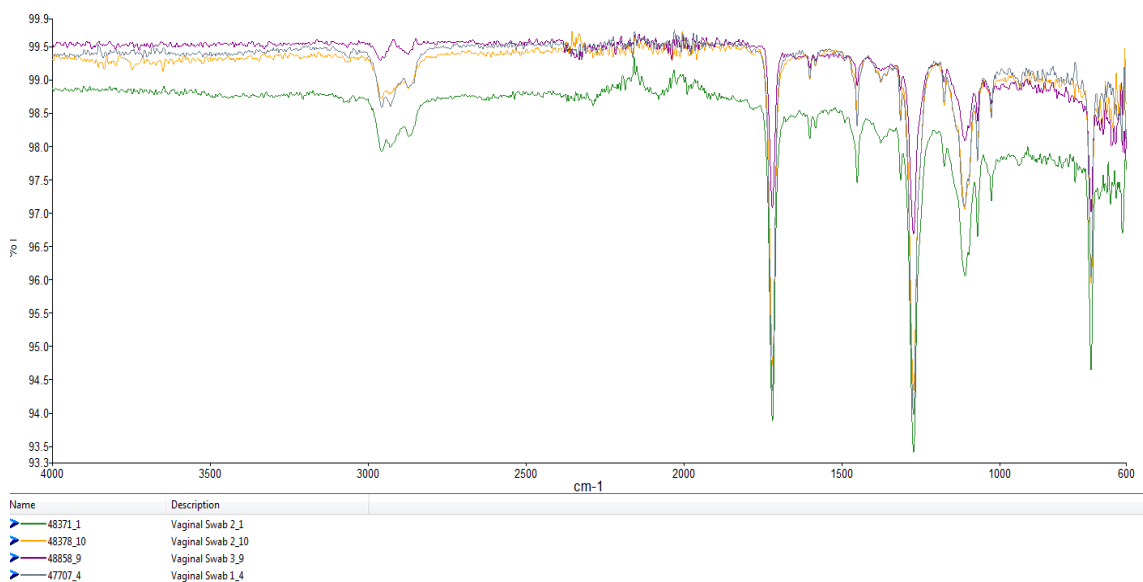


Figure 4.4: Spectra 4 blank cotton Scene Safe swabs from Vaginal Swab Kits.

Figure 4.41 on the following page presents 2 different blank swabs from a vaginal swab kit selected at random. The spectrum shown in black represents the u-ATR and the red spectrum signifies the FTIR microscope. There is very little variation between the intensity of the peaks and the background noise from the two different techniques.

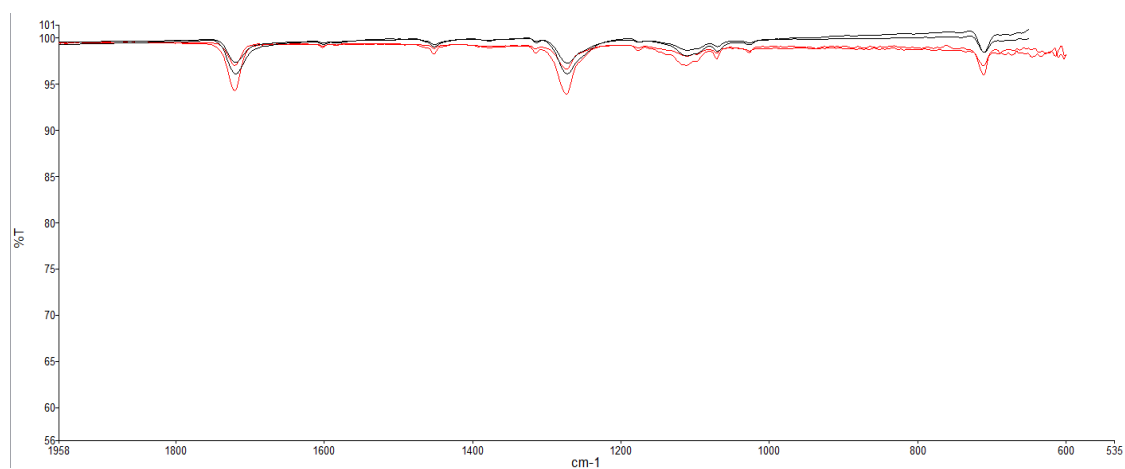


Figure 4.41: Spectra comparison of 2 different swabs from Vaginal Swab Kits using u-ATR and FTIR Microscope.

Vaginal Swab 1 was compared to Cellmarks internal database. An 86% match with Benzyl Benzoate was identified. Conversely, there are very few corresponding peaks in Figure 4.42 and the contamination levels are minimal implying this is a highly unlikely match.

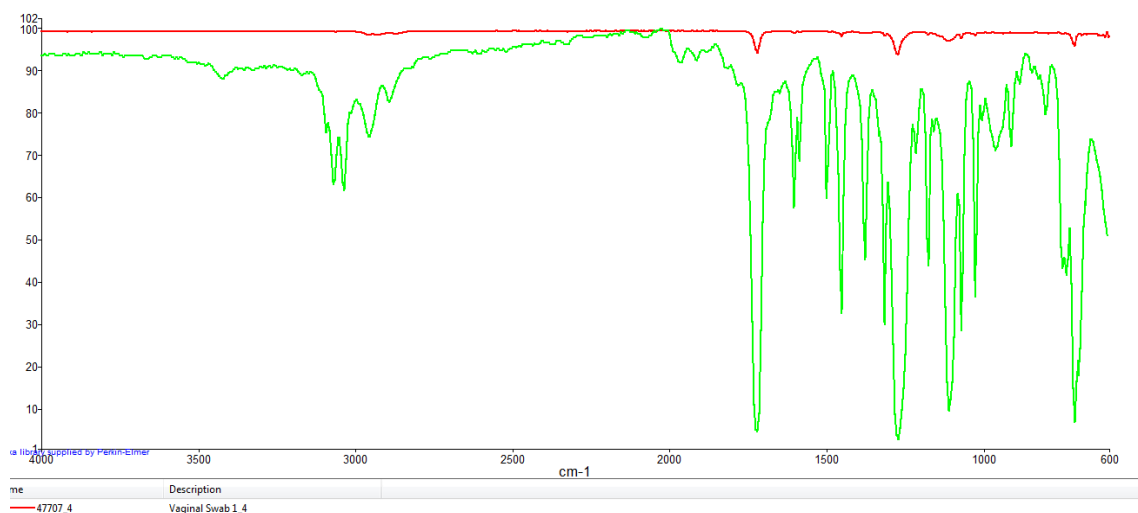


Figure 4.42: Spectra of Vaginal Swab 1 and Benzyl Benzoate.

Figure 4.43 displays 3 spectra from the same blank cotton swab using the FTIR microscope. The intensity varies in different areas of the swab extract. Using the FTIR microscope is a beneficial technique because of its high spatial resolution in comparison to alternative techniques. This allows substances to be recognized more simply. However, contamination can be easily missed as this technique doesn't analyse the whole extract.

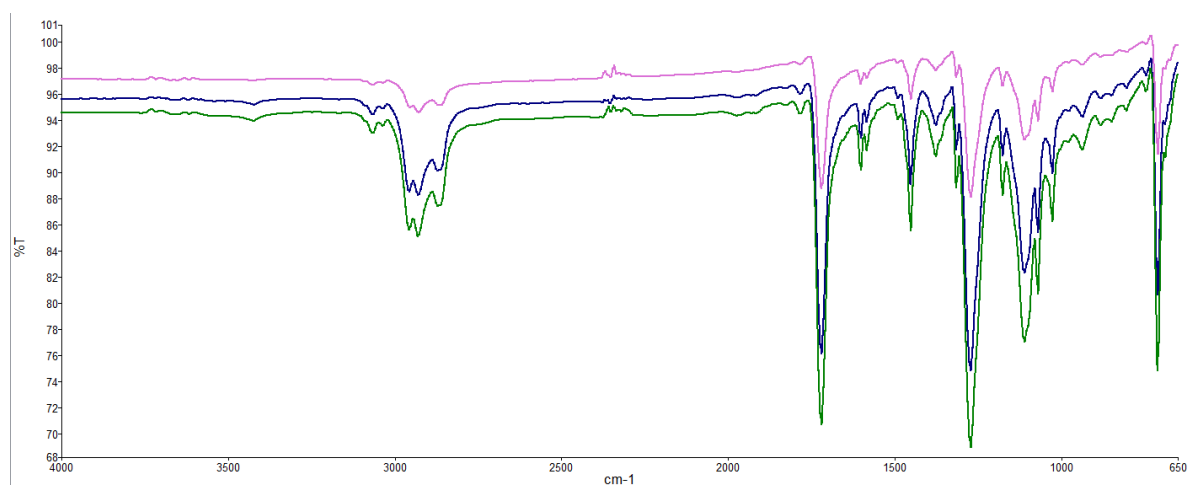


Figure 4.43: 3 Spectra from the same blank cotton swab using FTIR Microscope.

4.5 FTIR Microscope and u-ATR Comparison Summary

Overall, the FTIR microscope produces much clearer and distinguished peaks with a greater intensity in comparison to the u-ATR. Also, a reduced background noise is witnessed using the FTIR microscope. Nonetheless, from all samples analysed using the FTIR microscope, similar spectra with corresponding peaks were present when using the u-ATR also. Using the u-ATR as an alternative due to being a faster and simpler method didn't affect the detection of PDMS in any samples. All PDMS present on swabs up to volumes less than 0.125µl was detected on the u-ATR in addition to the FTIR microscope.

4.6 Lubricating Jelly from Sexual Assault Kits

Sexual Assault Kits include a lubricant to allow intimate swabs to be taken with the individual being provided minimal discomfort. Over recent years, the manufacturers have replaced the lubricant with a more cost-effective option. However, the ingredients within the lubricant are unknown to the forensic science community. The results indicated that both Aquagel and OptiLube are both water-based lubricants, with no traces of PDMS present. This is a crucial finding as the risk of external contamination of PDMS from Sexual Assault Kits is marginal.

4.6.1 Aquagel

Figure 4.6.1 compares PDMS with Aquagel. There are no corresponding peaks meaning PDMS is not present within the lubricant. Cellmarks internal database provided no close matches between the two lubricants.

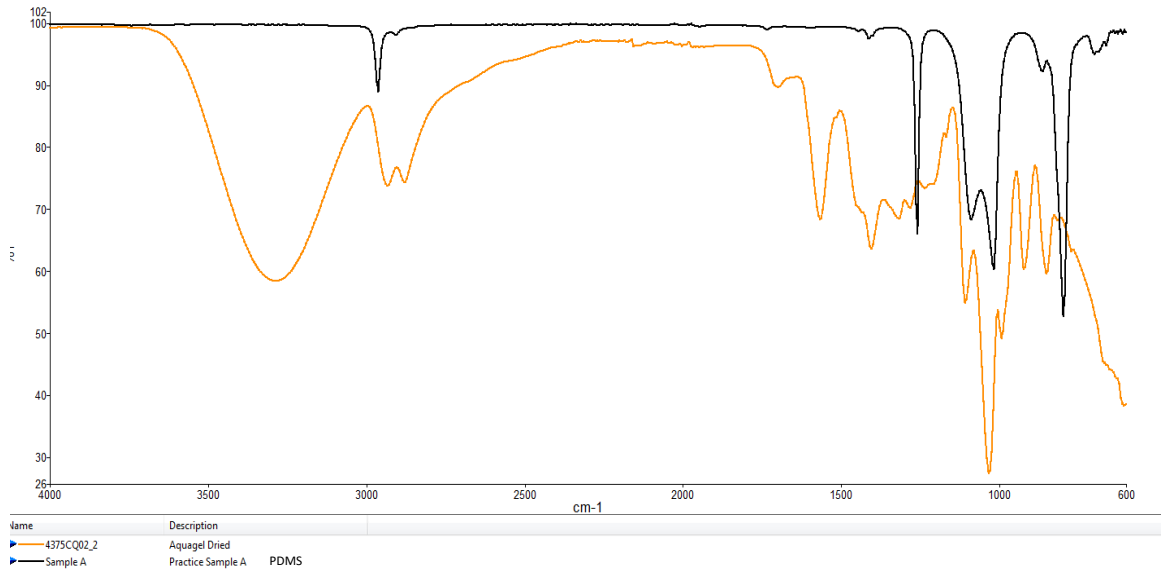


Figure 4.6.1: Comparison spectra of Aquagel and PDMS

However, when compared to Cellmark’s internal database, a 96% match was detected with glycerine. This indicates that Aquagel is a water-based lubricant containing no silicone. Figure 4.6.11 displays this spectrum with Aquagel being the pink spectrum, and glycerine being black.

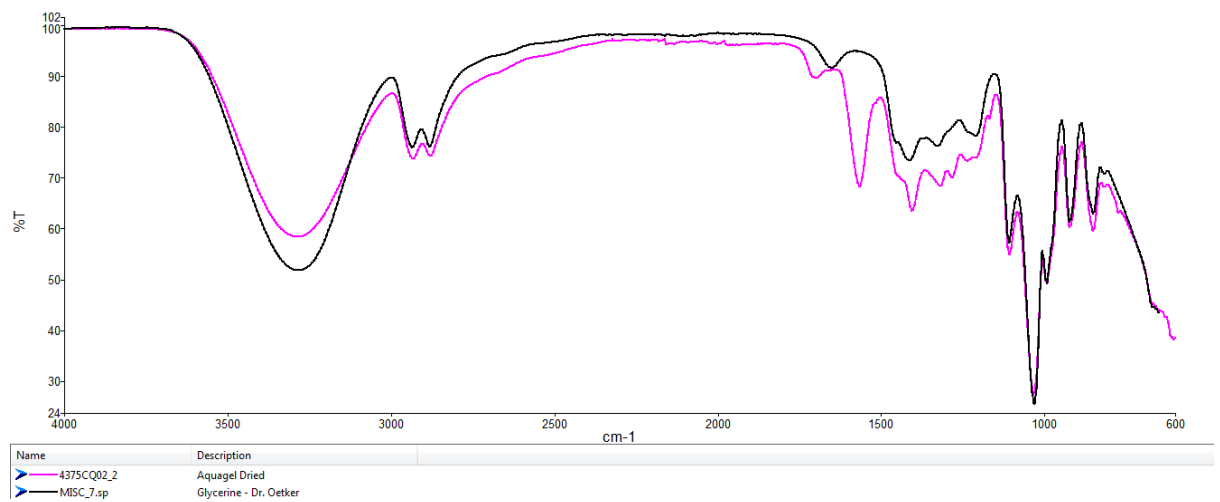


Figure 4.6.11: Comparison spectra of Aquagel and Glycerine

4.6.2 OptiLube

Figure 4.6.2 compares PDMS with OptiLube. There are no corresponding peaks meaning PDMS is not present within the lubricant.

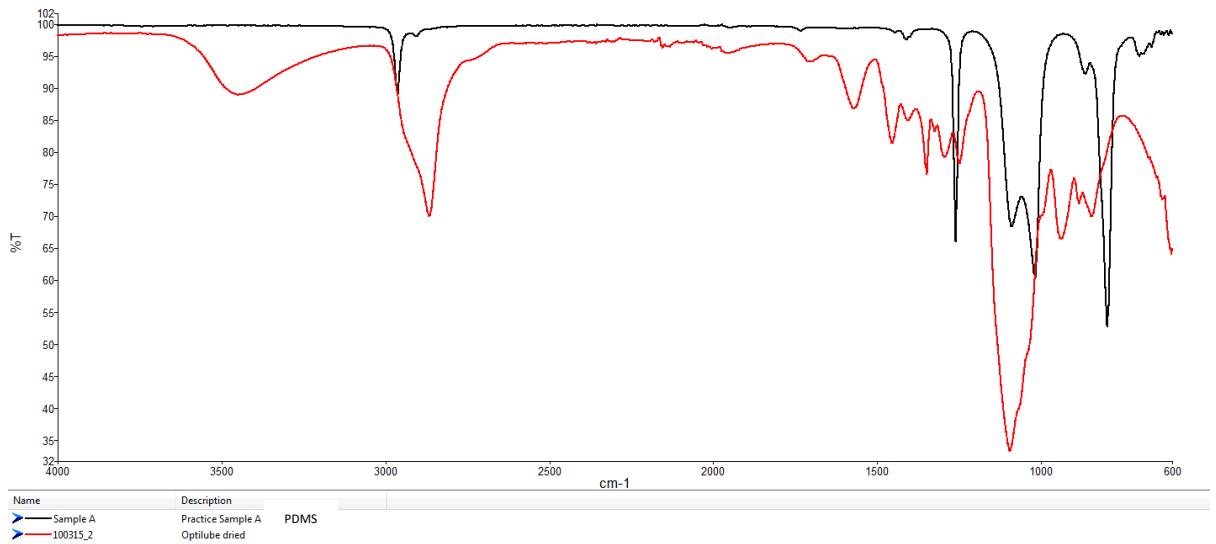


Figure 4.6.2: Comparison spectra of Optilube and PDMS

Figure 4.6.21 shows that Optilube (red) illustrates an 88% match with PEG 200. The spectrum isn't an exact match; however the positioning of the peaks is very similar. This indicates that Optilube is likely to contain some PEG containing compounds within. This suggests that Optilube is a water-based lubricant containing no silicone-based compounds.

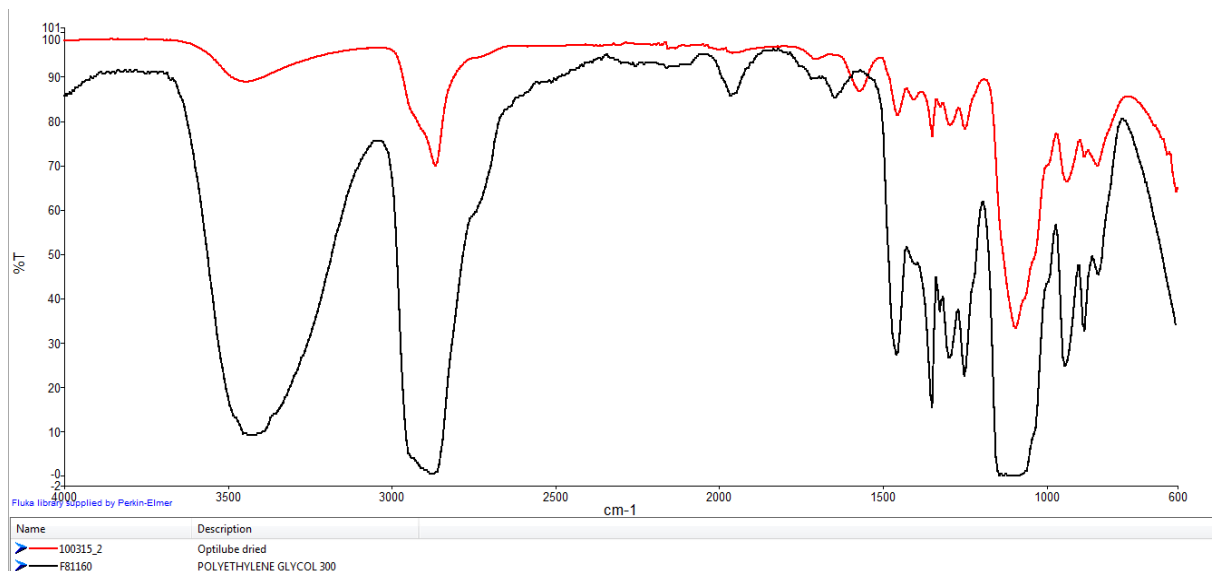


Figure 4.6.21: Comparison spectra of Optilube and PEG 200

The spectra of both OptiLube and Aquagel are very similar, with selected variations in the fingerprint region. This comparison is demonstrated in Figure 4.6.22 below.

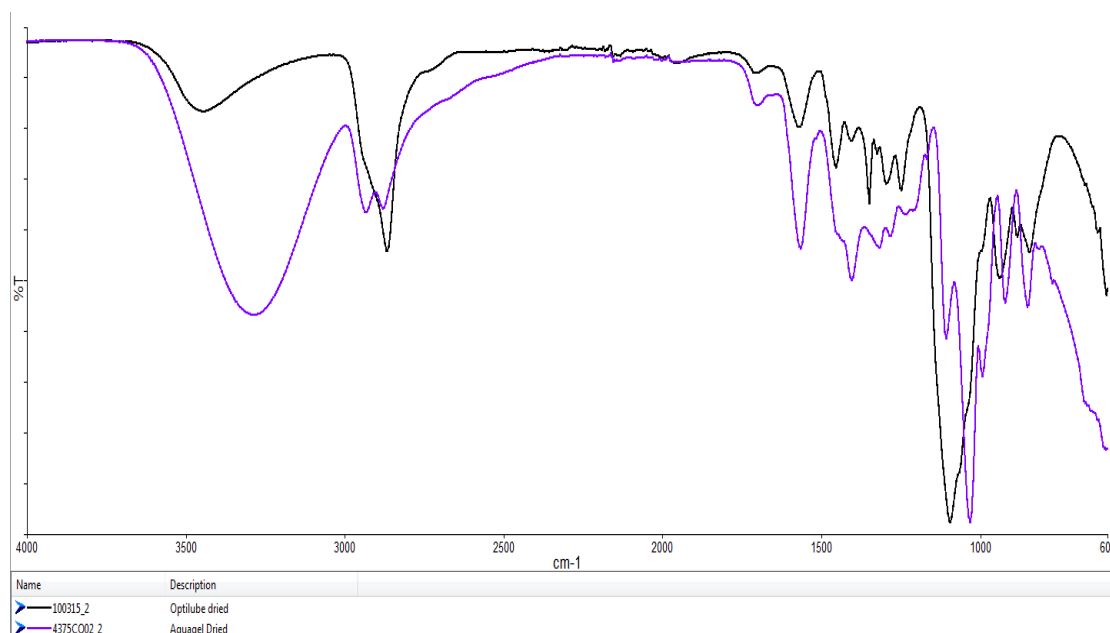


Figure 4.6.22: Comparison spectra of OptiLube and Aquagel

4.7 PDMS Dilutions

All analysis for the PDMS dilutions was completed using the u-ATR with the perception of this possibly being a quantitative method. PDMS was detected in the extracts of all dilutions ranging between 20 μ l to 0.125 μ l. As the smallest traceable sample of PDMS at 0.125 μ l was detected using the u-ATR, it was not deemed beneficial to repeat the analysis using the FTIR microscope.

Figure 4.7 on the following page shows the spectra for Run 1 of the PDMS dilutions. It can be clearly seen that there is great variation between the intensities of the peaks. The general trend of the peaks suggests that as the volume of PDMS seeded onto the swabs decreases, the intensity of the peaks on the FTIR spectrum also decreases. However, analysis using the u-ATR is only a semi-quantitative method.

Following recent suggestions as to fully quantify this method, the peak heights were recorded for the four characteristic peaks on the PDMS spectrum from all three separate runs. The measured peak height was recorded by measuring the bottom of

the peak and noting the percentage transmission at that given point. The average height of each peak was then calculated for each dilution. These results are displayed in Table 4.7.

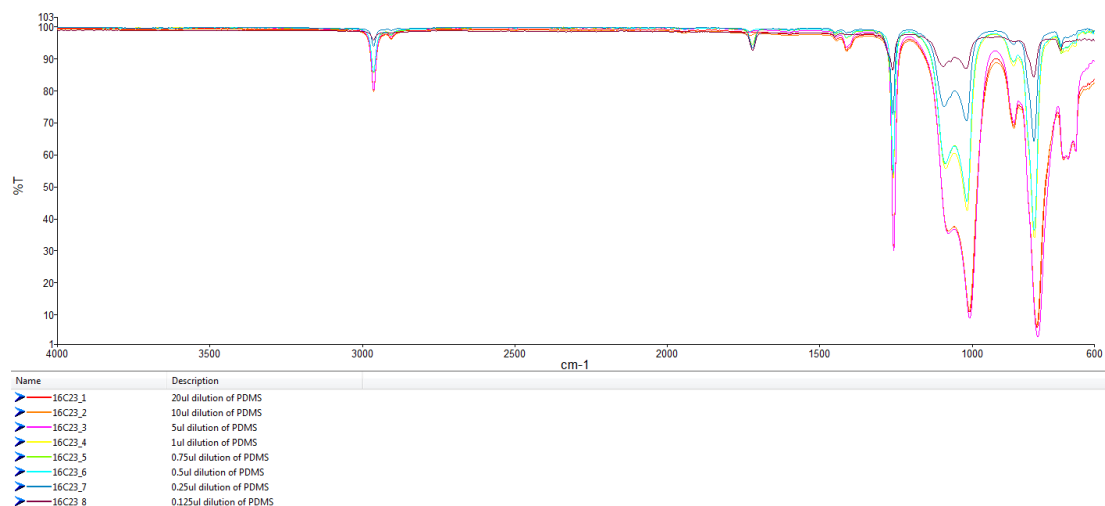


Figure 4.7: Spectra containing Run 1 of PDMS dilutions ranging between 20 μ l and 0.125 μ l.

Table 4.7: A collection of tables to show the Peak Heights for each run of PDMS volumes and the average peak heights.

Volumes	Run 1 - Peak Heights			
	1260	1090	1020	800
20 μ l	31.03	36.06	10.94	5.92
10 μ l	31.08	36.04	11.72	6.51
5 μ l	30.29	35.27	9.25	3.13
1 μ l	52.83	55.83	42.75	34.26
0.75 μ l	53.98	57.25	45.67	36.61
0.5 μ l	54.67	57.39	45.47	36.62
0.25 μ l	72.64	75.13	70.72	64.41
0.125 μ l	86.51	87.70	87.11	84.50
Blank	-	-	-	-

Volumes	Run 2 - Peak Heights			
	1260	1090	1020	800
20 μ l	29.82	35.26	8.57	3.38
10 μ l	31.26	36.11	10.08	4.98
5 μ l	48.01	51.31	36.92	27.40
1 μ l	54.91	57.92	46.71	37.68
0.75 μ l	40.33	44.10	25.46	17.26
0.5 μ l	77.31	79.21	75.08	69.38
0.25 μ l	88.11	89.28	87.99	85.03
0.125 μ l	93.57	94.29	93.39	91.80
Blank	-	-	-	-

Volumes	Run 3 - Peak Heights			
	1260	1090	1020	800
20 μ l	29.86	35.31	8.60	3.16
10 μ l	30.03	35.16	8.48	3.87
5 μ l	37.73	41.67	21.73	13.62
1 μ l	61.39	63.98	54.48	46.14
0.75 μ l	39.55	44.01	24.83	16.80
0.5 μ l	67.24	69.84	63.47	56.03
0.25 μ l	74.45	76.47	72.07	66.01
0.125 μ l	74.67	76.47	70.86	64.59
Blank	-	-	-	-

	Average			
	1260	1090	1020	800
20 μ l	29.85	35.29	8.59	3.23
10 μ l	30.44	35.48	9.01	4.24
5 μ l	41.16	44.88	26.79	18.21
1 μ l	59.23	61.96	51.89	43.32
0.75 μ l	39.81	44.04	25.04	16.95
0.5 μ l	70.60	72.96	67.34	60.48
0.25 μ l	79.00	80.74	77.38	72.35
0.125 μ l	80.97	82.41	78.37	73.66

Generally, the results presented in Table 4.7 demonstrate that as the volume of PDMS increases, the peak height decreases as expected. Though, there are some results in the tables which don't follow this trend, implying that the technique is unable to be completely quantified. An attempt at producing a quantitative curve for the average peak height for each peak is exhibited in Figures 4.71 to 4.74 underneath.

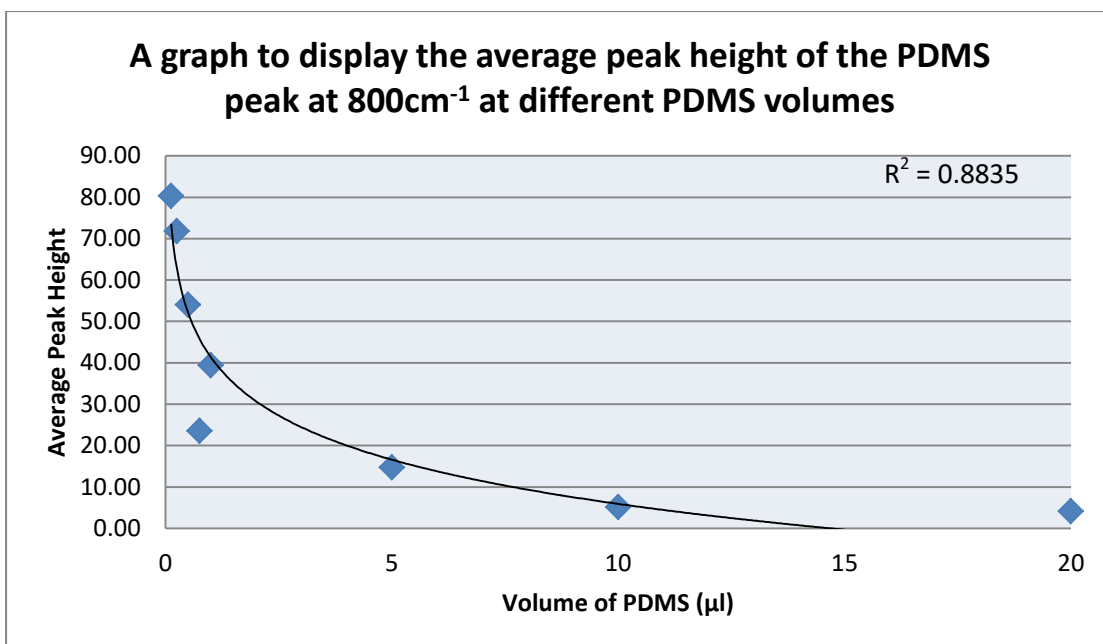


Figure 4.71: A graph to display the average peak height of the PDMS peak at 800cm⁻¹ at different PDMS volumes.

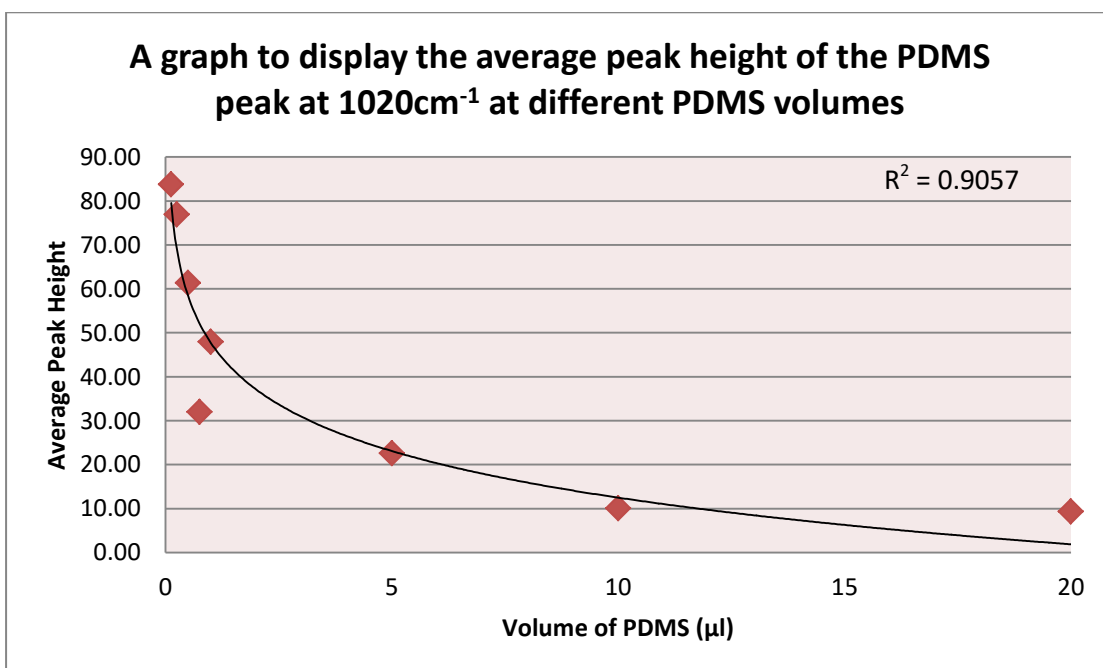


Figure 4.72: A graph to display the average peak height of the PDMS peak at 1020cm⁻¹ at different PDMS volumes.

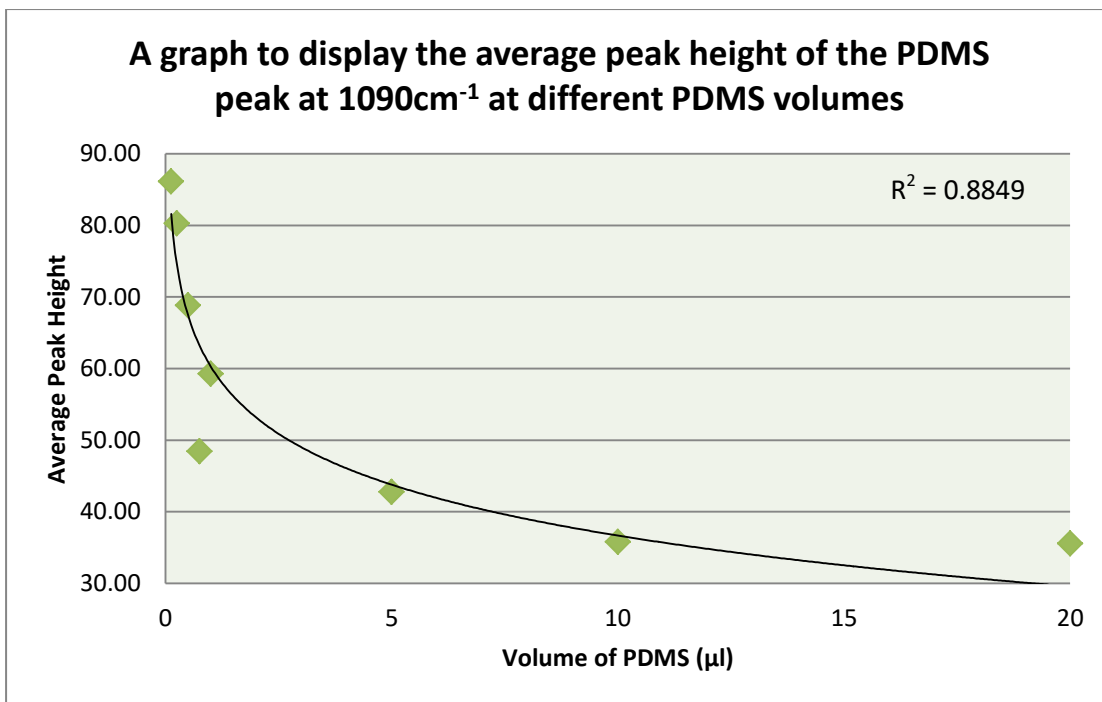


Figure 4.73: A graph to display the average peak height of the PDMS peak at 1090cm⁻¹ at different PDMS volumes.

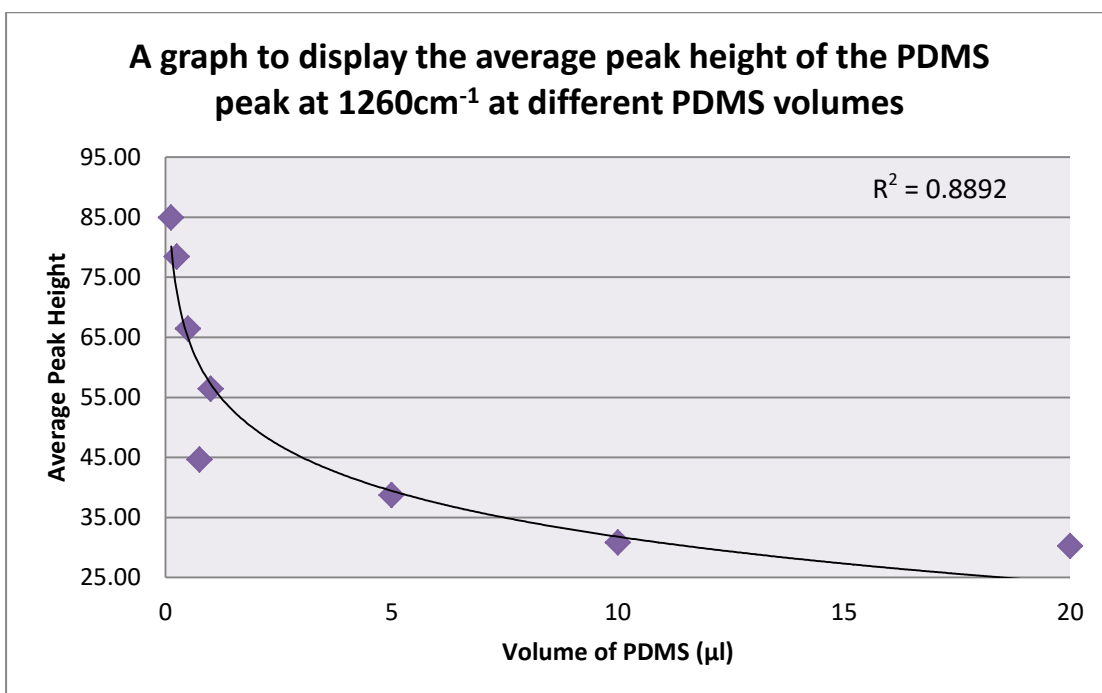


Figure 4.74: A graph to display the average peak height of the PDMS peak at 1260cm⁻¹ at different PDMS volumes.

The curves are very consistent for each of the four peaks which can be seen in the R^2 values. Conversely, the average peak height for the dilution of 0.75μl does not fit in with the overall trend of either peak. However, this could just be an anomalous

result or an accuracy issue; further repeats would be required to clarify this. The trendline on the graph is a logarithmic curve. This was chosen because the curve best fit the results obtained, and the values tend to decrease rapidly at the start of the curve, then level out towards the end. Overall, these results can only be categorized as semi-quantitative. Additional equipment and sampling techniques are required in order for a fully quantitative curve to be formed.

Upon completion of this analysis, it was suggested that a plot of volume against absorbance should have been produced, rather than percentage transmission. This is because the percentage transmission is not proportional to concentration. The Beer Lambert Law, $A = \epsilon cl$, states that there is a linear relationship between concentration and absorbance. If the percentage transmission had been converted into absorbance using a logarithm, a linear graph would have resulted, rather than a curve.

$$\%T = \frac{I}{I_o} \times 100$$

$$A = \log_{10} \left(\frac{I_o}{I} \right)$$

Figure 4.75 displays the spectra of the 20 μ l and 0.125 μ l PDMS dilutions. There are clear visible differences between the intensities of the peaks. On the other hand, the 0.125 μ l spectrum still clearly displays PDMS being present, indicating that smaller volumes of the lubricant are likely to be detected.

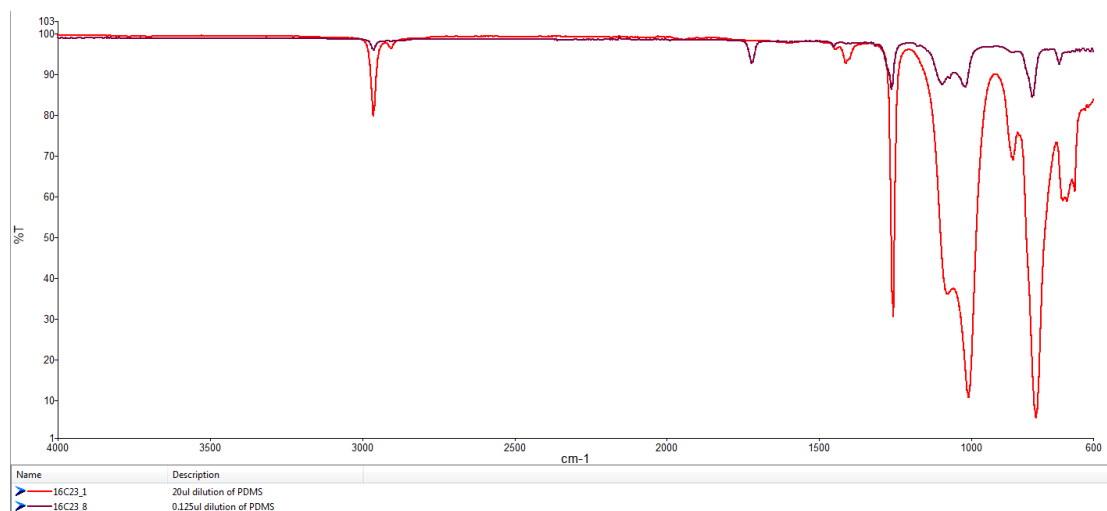


Figure 4.75: Spectra to display the variation between the largest and smallest volume of PDMS

4.8 PDMS Dilutions after Sperm Elution

The same volumes of PDMS used for the PDMS dilutions with extraction alone were used in the Sperm Elution batch of swabs to maintain consistency. Results indicated that PDMS remained present on all of the swab heads after Sperm Elution for all dilutions. But, PDMS was also detected in a collection of the epithelial pellets and seminal pellets too, implying that some of the lubricant is removed from the swab head and into the supernatant during the Sperm Elution process.

The average peak height of each run of dilutions is displayed in Table 4.8. The same procedure as displayed in Section 4.7 is fulfilled in order to create a curve.

Table 4.8: A collection of tables to show the Peak Heights for each run of PDMS volumes after Sperm Elution, and the average peak heights.

Volumes	Run 1 - Peak Heights			
	1260	1090	1020	800
20µl	43.19	46.82	29.53	20.35
10µl	45.31	48.60	31.95	23.31
5µl	88.75	89.62	87.36	84.15
1µl	88.97	90.22	89.25	85.66
0.75µl	90.34	91.29	89.48	86.90
0.5µl	92.23	93.64	93.31	92.34
0.25µl	97.87	98.44	97.93	98.21
0.125µl	97.11	97.81	98.06	97.84
Blank	-	-	-	-

Volumes	Run 2 - Peak Heights			
	1260	1090	1020	800
20µl	38.99	42.44	22.90	14.16
10µl	57.35	60.07	48.41	40.58
5µl	59.90	62.61	52.37	43.93
1µl	95.91	96.43	95.36	94.64
0.75µl	88.85	89.81	88.10	84.82
0.5µl	95.10	95.74	95.58	94.13
0.25µl	94.89	95.50	95.04	93.86
0.125µl	98.38	98.99	98.90	98.69
Blank	-	-	-	-

Volumes	Run 3 - Peak Heights			
	1260	1090	1020	800
20µl	56.56	59.34	48.24	39.43
10µl	80.53	81.80	77.17	72.47
5µl	69.80	72.05	65.54	58.25
1µl	89.34	90.53	88.93	86.24
0.75µl	43.53	48.42	33.13	23.74
0.5µl	88.37	89.73	88.57	85.36
0.25µl	88.16	89.13	86.93	83.70
0.125µl	95.01	95.78	95.30	94.15
Blank	-	-	-	-

	Average			
	1260	1090	1020	800
20µl	50.70	53.71	29.85	23.26
10µl	72.80	74.56	50.69	46.38
5µl	66.50	68.90	45.86	40.11
1µl	91.53	92.50	68.31	66.78
0.75µl	58.64	62.22	38.59	33.08
0.5µl	90.61	91.73	68.18	66.21
0.25µl	90.40	91.25	67.23	65.32
0.125µl	96.13	96.85	72.38	71.75

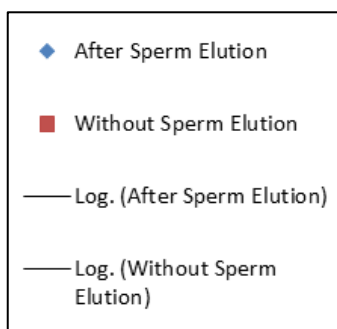


Figure 4.8: Key to the peak height graphs.

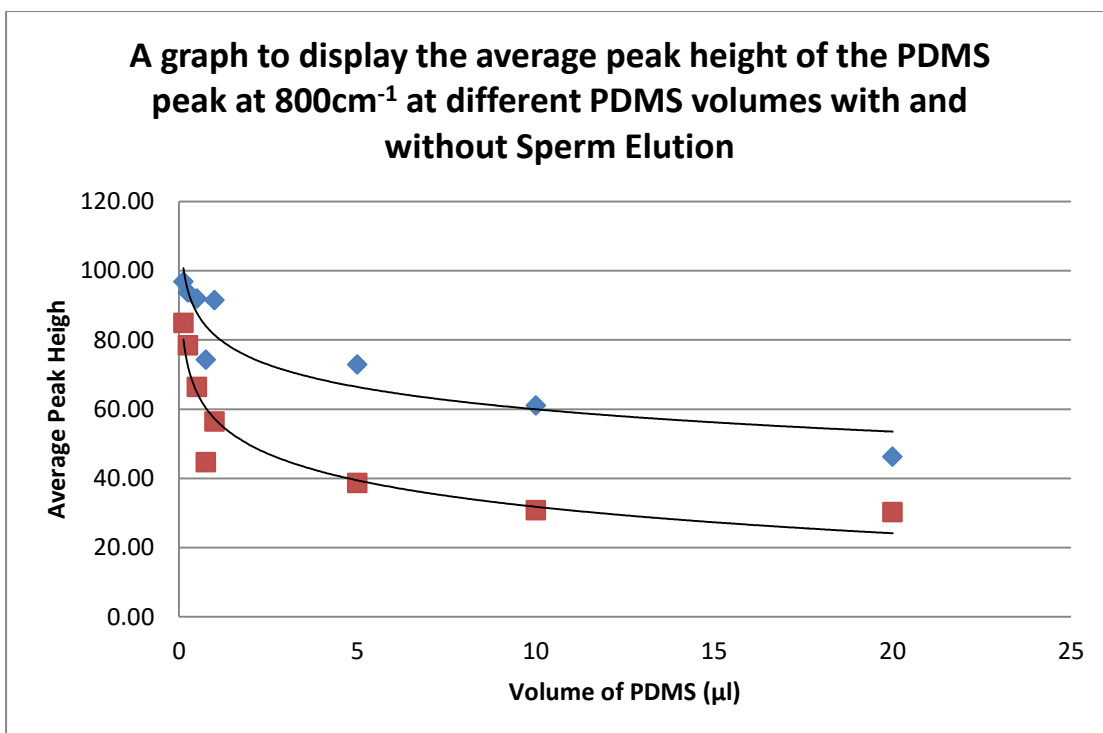


Figure 4.81: A graph to display the average peak height of the PDMS peak at 800cm^{-1} at different PDMS volumes both with and without Sperm Elution.

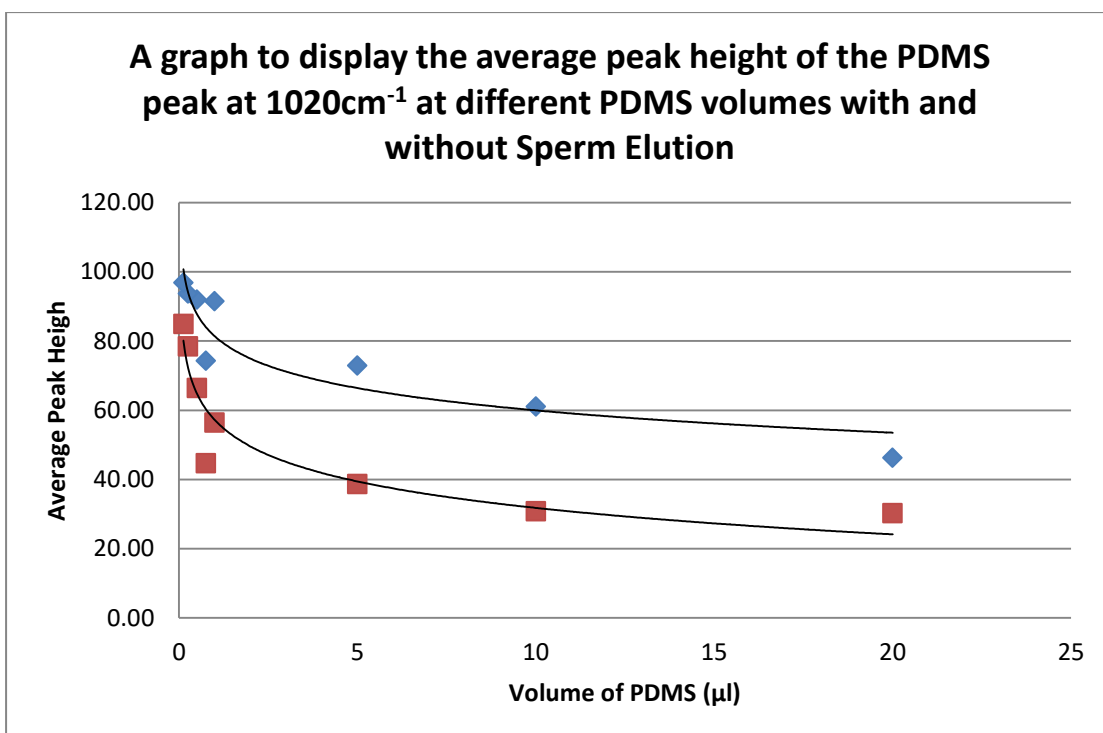


Figure 4.82: A graph to display the average peak height of the PDMS peak at 1020cm^{-1} at different PDMS volumes both with and without Sperm Elution.

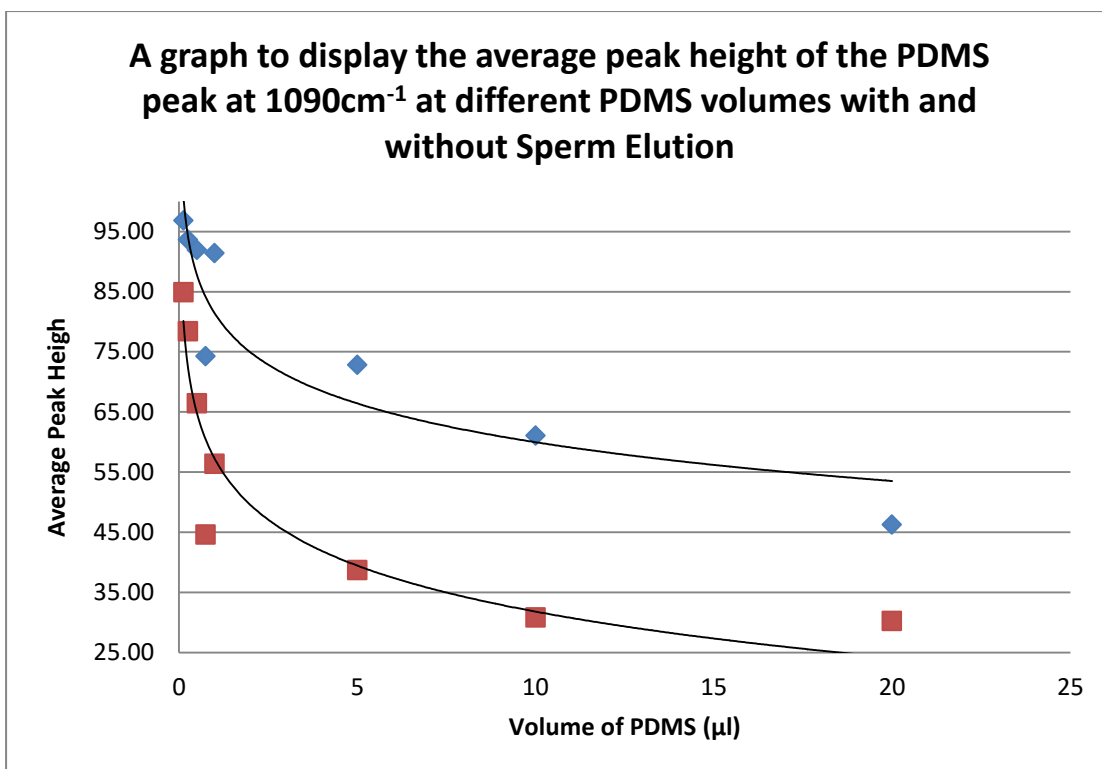


Figure 4.83: A graph to display the average peak height of the PDMS peak at 1090cm^{-1} at different PDMS volumes both with and without Sperm Elution.

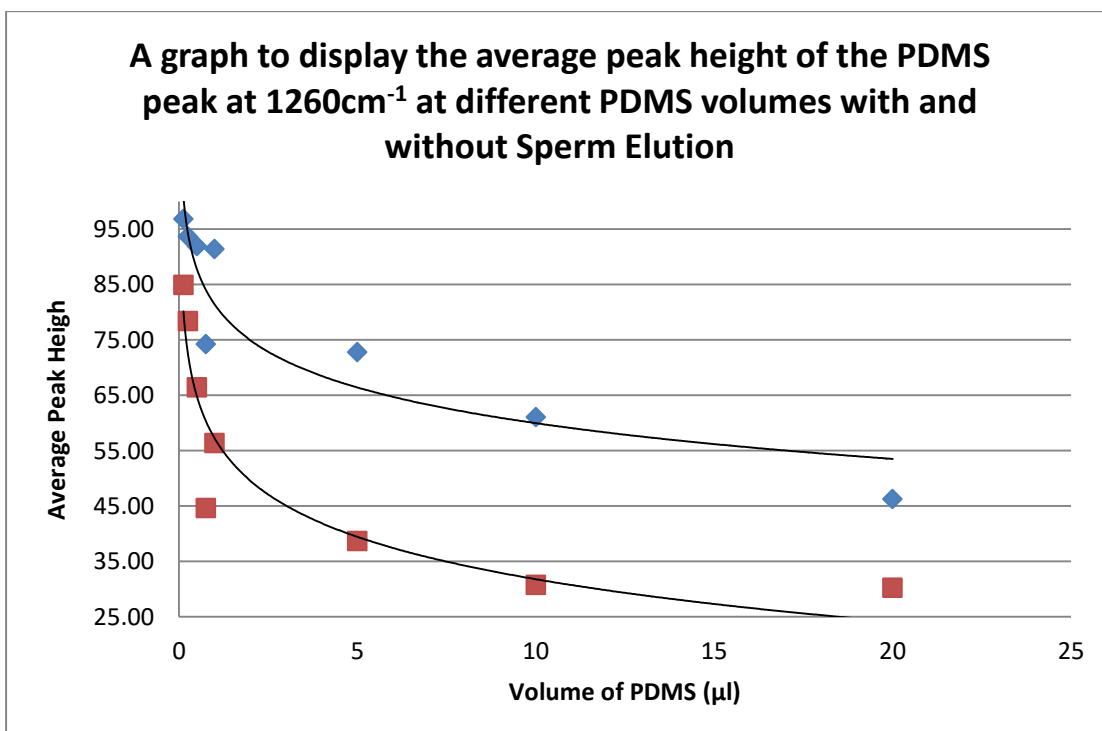


Figure 4.84: A graph to display the average peak height of the PDMS peak at 1260cm^{-1} at different PDMS volumes both with and without Sperm Elution.

The graphs confirm a great reduction in intensity for all four characteristic peaks after Sperm Elution. The approximate difference between each peak is between 30 and 40. Also, the shape of the curve remains consistent, indicating that the loss of PDMS affects each peak similarly in the spectrum.

As noted in the previous section, graphs of volume against absorbance should have been plotted instead of volume against percentage transmission.

4.8.1 Sample Remains

Figure 4.8.1 displays the spectra for the swabs containing both the largest and smallest volumes of PDMS prior to Sperm Elution. It is still evident that there is a distinct difference between the intensities of the peaks. However, when comparing to Figure 4.75 of PDMS extracted alone without Sperm Elution, the overall intensities of the peaks are significantly lower. This is especially the case for the 20 μ l volume of PDMS as the difference between the intensities is approximately 35. This result also correlates with the suggestion that some PDMS is lost into the supernatant during the Sperm Elution process.

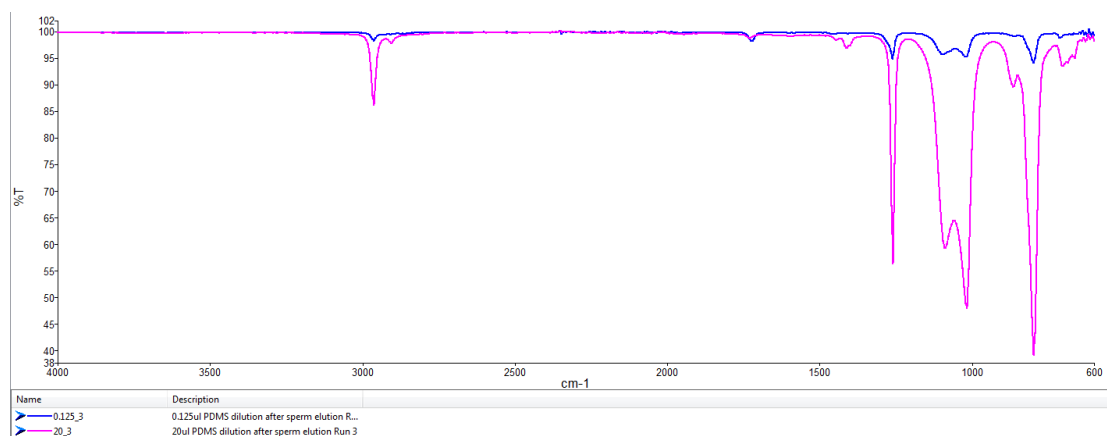


Figure 4.8.1: Spectra to show the difference between 20 μ l and 0.125 μ l of PDMS after Sperm Elution.

On the other hand, the peak intensities don't remain constant for each dilution. This indicates that different amounts of PDMS could be lost during the Sperm Elution process for each swab initially seeded with the same volume of the lubricant. For example, in Figure 4.8.11, the three swabs, one from each run, each seeded with 10 μ l of PDMS, all displayed spectra with large variation in peak intensities.

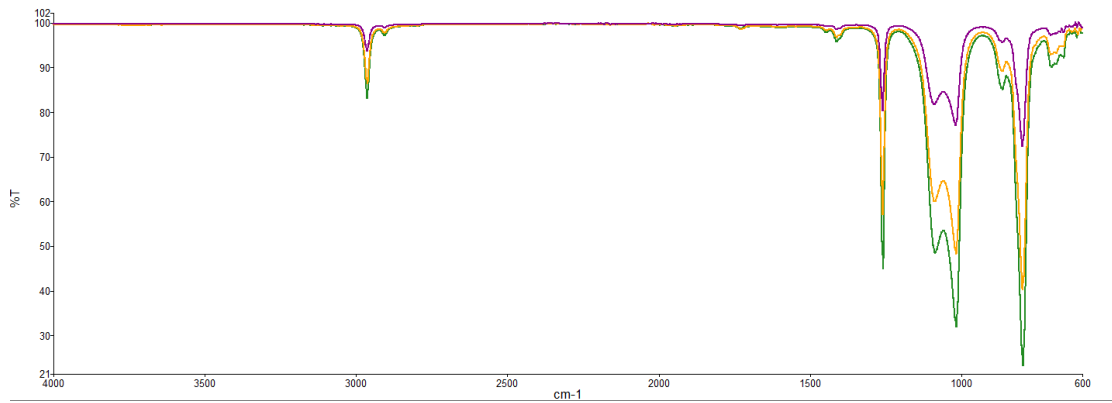


Figure 4.8.11: Spectra showing three different swabs initially containing 10µl of PDMS.

Figure 4.8.12 supports the suggestion that the method isn't a quantitative technique as there are many variations between the peak heights of one run of dilutions. This could be due to the dissimilar amounts of PDMS being transferred into the supernatant during Sperm Elution. This is because, for PDMS in larger volumes, less of the lubricant is able to remain attached to the swab head and extracts off. However, the exact amount is unknown and will fluctuate dependent upon the location of the PDMS on the swab head, and whether it is spread out.

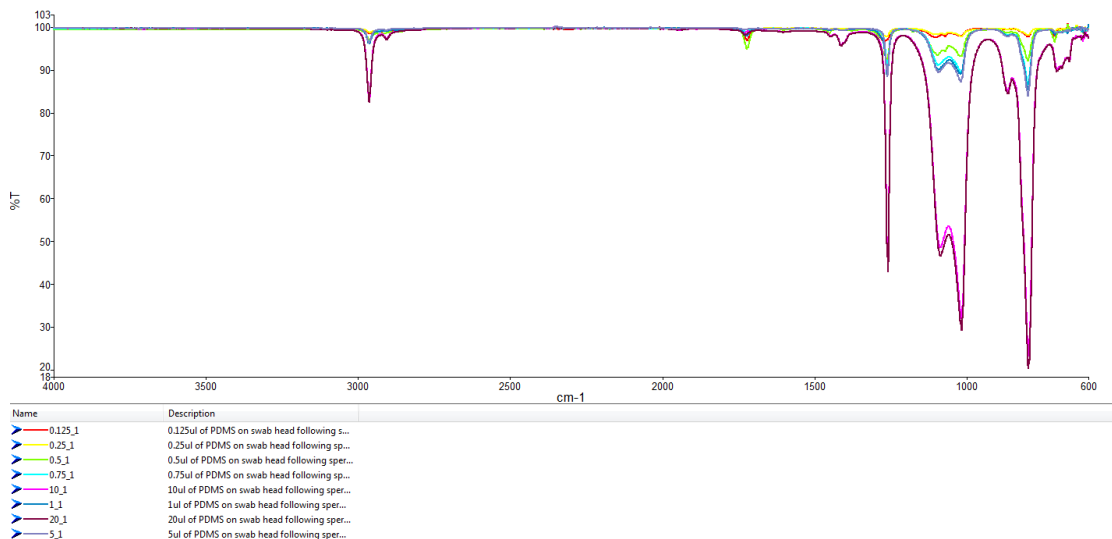


Figure 4.8.12: Spectra showing one run of PDMS dilutions after Sperm Elution.

4.8.2 Epithelial Pellet and Seminal Pellet

Results have indicated that a substantial portion of PDMS is transferred from the swab head into the supernatant during the Sperm Elution process. However, this has only been detected in certain samples which are shown in Table 4.8.2.

Table 4.8.2: Tables to show whether PDMS was detected in the Epithelial and Seminal Pellet during Sperm Elution.

XP	PDMS Detected?			SP	PDMS Detected?		
Vol. of PDMS	Run 1	Run 2	Run 3	Vol. of PDMS	Run 1	Run 2	Run 3
20µl	Yes	Yes	Yes	20µl	Yes	Yes	Yes
10µl	Yes	Yes	Yes	10µl	Yes	Yes	Limited
5µl	Yes	Yes	Yes	5µl	Yes	Yes	Yes
1µl	Yes	No	Yes	1µl	Limited	Limited	Yes
0.75µl	Yes	No	No	0.75µl	No	No	Limited
0.5µl	No	No	No	0.5µl	No	No	No
0.25µl	No	No	No	0.25µl	No	No	No
0.125µl	No	No	No	0.125µl	No	No	No

Results in these tables show that very little, if any, PDMS is detected in both the epithelial pellet and the seminal pellet in PDMS dilutions smaller than 1µl. Limited evidence of PDMS is where all peaks for PDMS are visible on the spectrum, however the peaks at 1020cm⁻¹ and 1090cm⁻¹ haven't entirely separated. As a whole, PDMS is lost from the epithelial pellet to the seminal pellet, meaning PDMS is released into the seminal supernatant. On the other hand, 1µl of PDMS in Run 2 was marginally detected in the seminal pellet, but not identified in the epithelial pellet. This suggests a fully quantitative method is required as PDMS must be present in the epithelial pellet in order to be transferred into the seminal pellet.

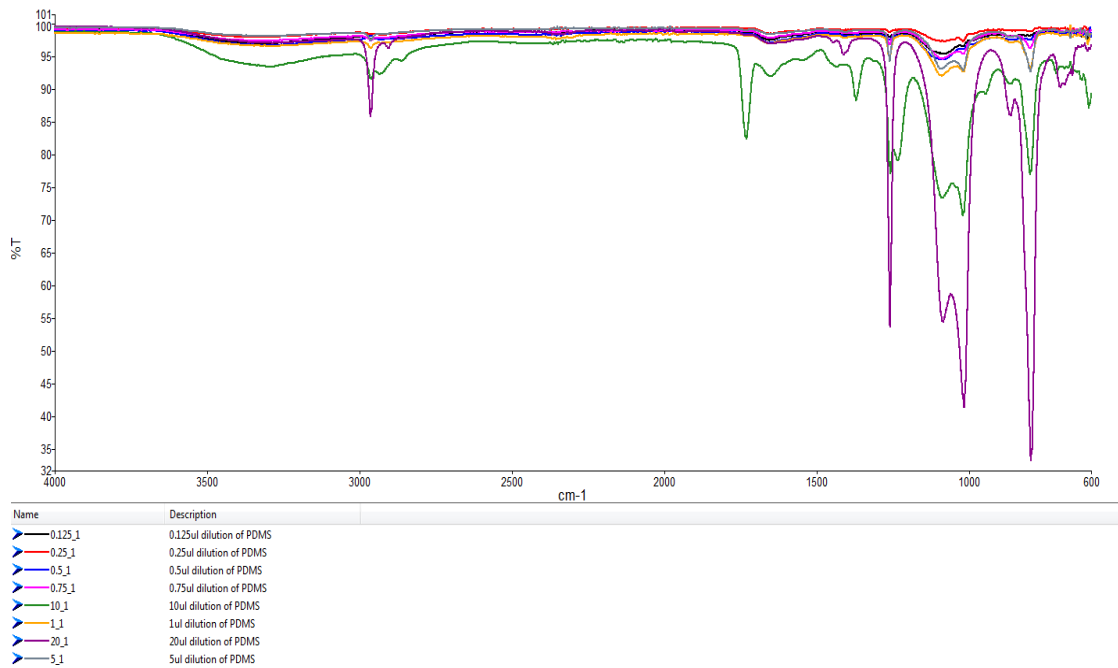


Figure 4.8.2: Spectra to show the epithelial pellets of Run 1 of PDMS dilutions after Sperm Elution.

The intensity of the peaks is much lower for the epithelial pellet in comparison to the sample remains. However, the peaks remain sharp and clear for the larger volumes of PDMS, indicating a lower affinity for the lubricant to remain on the swab head in greater volumes.

Figure 4.8.21 displays the seminal pellet for Run 3 of the PDMS dilutions. There are larger quantities of background noise present on the spectra; nevertheless the peaks are still able to be accurately distinguished. As a whole, the peaks in the epithelial pellet are stronger than the seminal pellet. In addition to this, the separation between the 1020cm^{-1} and 1090cm^{-1} is dramatically reduced; meaning in forensic casework, reporters would be unable to confidently state that PDMS is present in the smaller volume samples.



Figure 4.8.21: Spectra to show the seminal pellets of Run 3 of PDMS dilutions after Sperm Elution.

4.8.3 Assessment of Sperm Elution Reagents

In order to positively confirm the reliability of results, the verification that the reagents used for the Sperm Elution contain no traces of PDMS is a requirement. Mo Classic A, Mo Lite, Proteinase K and Mo Wash were all analysed using the u-ATR. The spectra revealed that no PDMS was identified in any of the reagents. When compared to Cellmark’s internal database, Proteinase K corresponded a 99% match with glycerine. This is because glycerine is a highly water-based substance added to the Proteinase K to minimise the strength of the digestive enzyme and to enable the material to work effectively during DNA extraction. However, if this sample was to be dried down further, prior to FTIR analysis, other components present in smaller volumes would be detected.

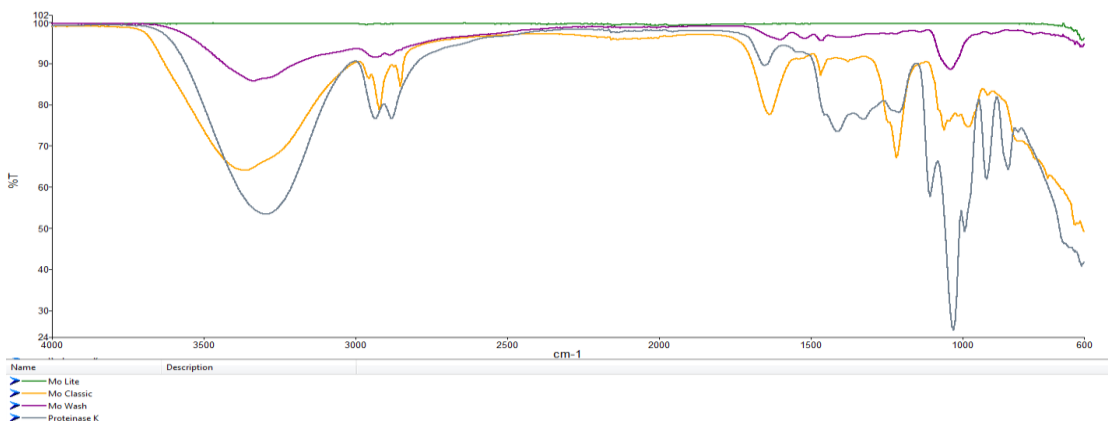


Figure 4.8.3: Spectra of Mo Classic A, Mo Lite, Proteinase K and Mo Wash.

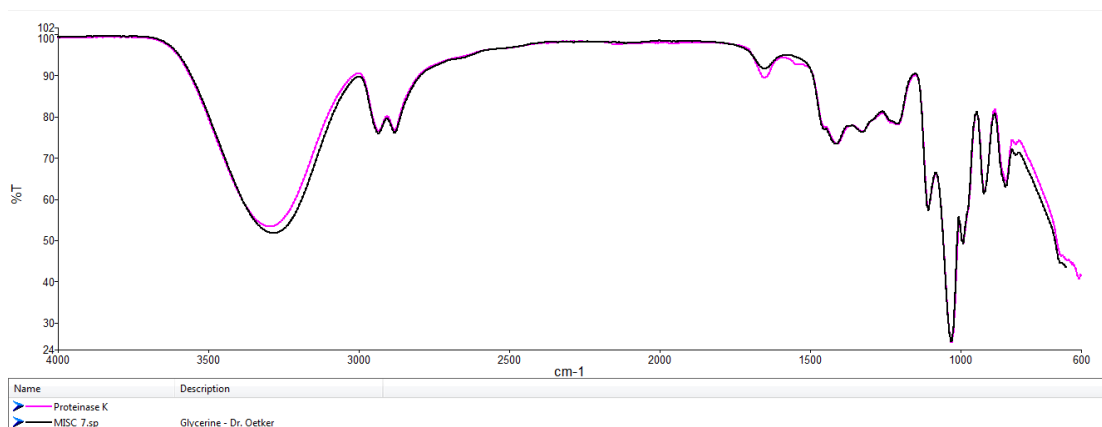


Figure 4.8.31: Spectral comparison of Proteinase K and Glycerine.

4.9 Sperm Elution using a Body Fluid Matrix

PDMS was detected on all the different body fluid matrix combinations. Although the intensities of the peaks of each spectrum varied slightly, all peaks were clear at confirming the presence of PDMS. However, some combinations displayed stronger peaks compared to others.

Table 4.9 displays the results for the detection of PDMS after Sperm Elution using a Body Fluid Matrix.

Table 4.9: A table to show the detection of PDMS using a Body Fluid Matrix.

Body Fluid Combination	PDMS Detected?		
	Run 1	Run 2	Run 3
Semen	Yes	Yes	Yes
Semen, PDMS & Blood	Yes	Yes	Yes
Semen, PDMS & Saliva	Yes	Yes	Yes
Semen, PDMS & Vaginal Material	Yes	Yes	Yes
Semen, PDMS & Faecal Material	Yes	Yes	Yes
Semen, PDMS, Blood & Faecal Material	Yes	Yes	Yes
Semen, PDMS, Blood & Vaginal Material	Yes	Yes	Yes
Semen, PDMS, Saliva & Vaginal Material	Yes	Yes	Yes

Run 1 of 3 is shown in Figure 4.9. It can be seen that the combination of body fluids situated on a swab does have an effect on the detection and recovery of PDMS. Even though PDMS is distinctively present on all samples, there is a variation in the peak height of approximately 5-30.

The combination of semen, PDMS and faecal material displays peaks with the lowest intensity of PDMS from all three runs. The volume of faecal material present on the swab is dependent on the individual taking the swab. For example, the time interval since the individual last visited the lavatory and the time the swab was taken. Also, the thoroughness of the swabbing affects the recovery of faecal material.

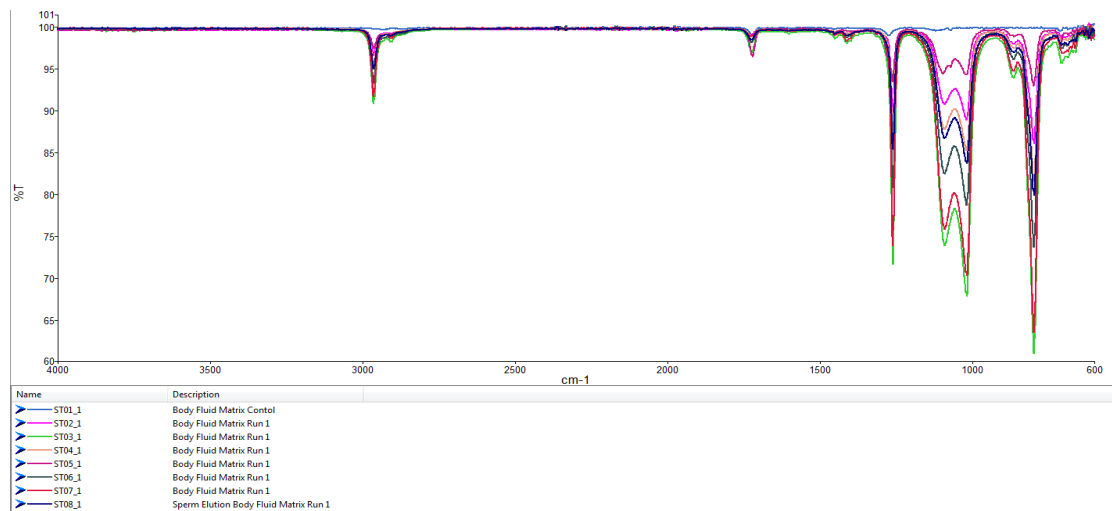


Figure 4.9: Spectra of Run 1 of Sperm Elution Body Fluid Matrix.

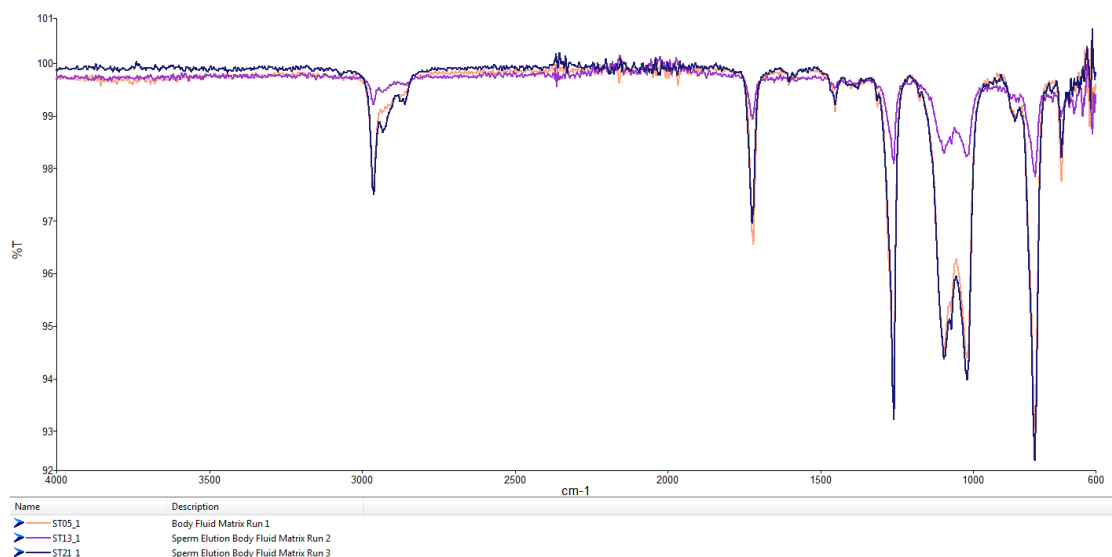


Figure 4.91: Spectra of each sample containing PDMS, semen and faecal material.

As a whole, the semen, PDMS and saliva combination displays the most intense peaks. This is demonstrated in Figure 4.92. Although there is still dissimilarity between the peak heights from all of the runs, the peaks are much clearer and defined and display an overall stronger intensity.

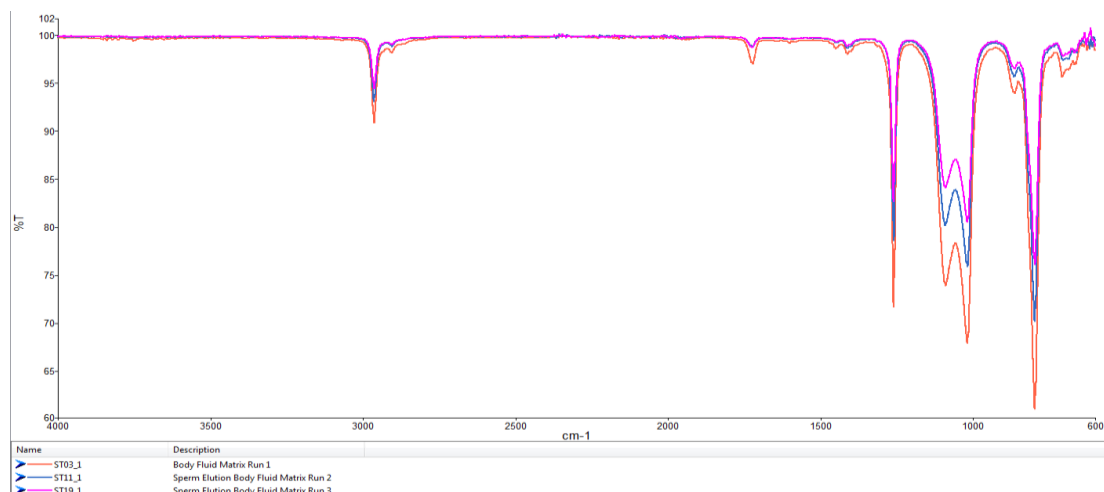


Figure 4.92: Spectra of each sample containing PDMS, semen and saliva.

The combination of PDMS, semen, saliva and vaginal material also shows well defined peaks despite there being three body fluids present on the swab. Furthermore, the vaginal material was present on the swab prior to the addition of PDMS. This suggests PDMS is able to remain on the swab head during the Sperm Elution process, even with the extraction of DNA material which is present underneath the lubricant.

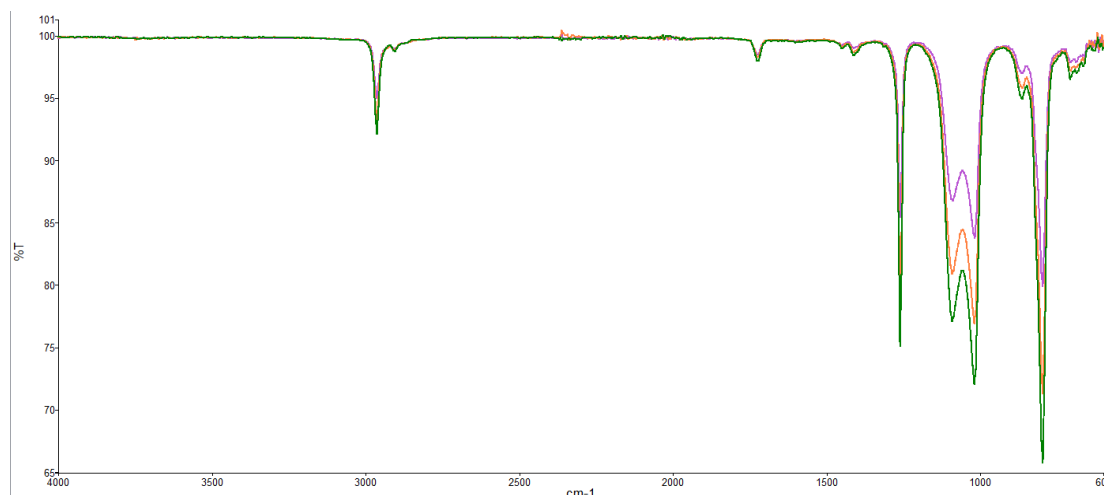


Figure 4.93: Spectra of each sample containing PDMS, semen, saliva and vaginal material.

In conclusion, the volume of body fluids on a swab has little effect on detection of PDMS. The variation of the volume of PDMS detected is more likely due to the extraction process and the position of PDMS in the vial during analysis using the u-ATR compared to the presence of additional body fluids.

4.10 Body Lotion and Shower Gel Analysis

Through completing a survey involving asking individuals which shower gels and body lotions they use on a regular basis, the results in Table 4.10 were attained. A total of 71 responses were received, with approximately half of individuals alternating the brand of body lotion and shower gels they used on a regular basis. The results revealed that The Body Shop, Soap and Glory, Nivea, Dove and Aveeno are the most common brands of body lotions used by individuals completing the survey. Also, Radox, The Body Shop and Dove are the most common shower gels used.

Table 4.10: A table to show the number of responses from the survey asking which body lotions and shower gels individuals use most regularly.

Brand of Body Lotion	No. of Individuals	Brand of Shower Gel	No. of Individuals
Astral	I	Original Source	III I
Aveeno	III II	Lynx	III I
Avon	I	Radox	III III II
Boots	I	Soap & Glory	III I
Clarins	II	Dove	III III I
Coco Chanel	I	Boots	II
Coconut Oil	I	The Body Shop	III III
Dermalogica	I	Imperial Leather	I
Dove	III II	Aveeno	I
E45	I	Nivea	III I
Garnier	I	Avon	I
Hawaiian Tropic	I	Dermalogica	I
Jack Wills	I	Neals Yard	III
Johnsons	II	NSpa	I
Lancôme	I	Molton Brown	II
Lush	II	Lush	I
Neals Yard	III	Sanex	III
Nivea	III III I	Mooster Bubble	I
NSpa	II	Faith in Nature	II
Palmers	IIII	Lancôme	I
Sanctuary	IIII	Palmolive	I
Soap & Glory	III III		
Superdrug	I		
Ted Baker	I		
The Bath House	II		
The Body Shop	III III III I		
The White Company	I		
Vaseline	IIII		

The results from Table 4.10 have been entered into pie charts and displayed in Figure 4.10 and Figure 4.101. Although the results clearly indicated there are more popular brands on the market, there is a wide disperse of results. It is crucial to determine whether these body lotions and shower gels contain PDMS. Upon the collection of this evidence, a wide collection of samples of body lotions and shower gels were obtained as explained in Section 3.2.7. Following analysis of these samples using the u-ATR, the results table was updated explaining whether PDMS was detected in the substances.

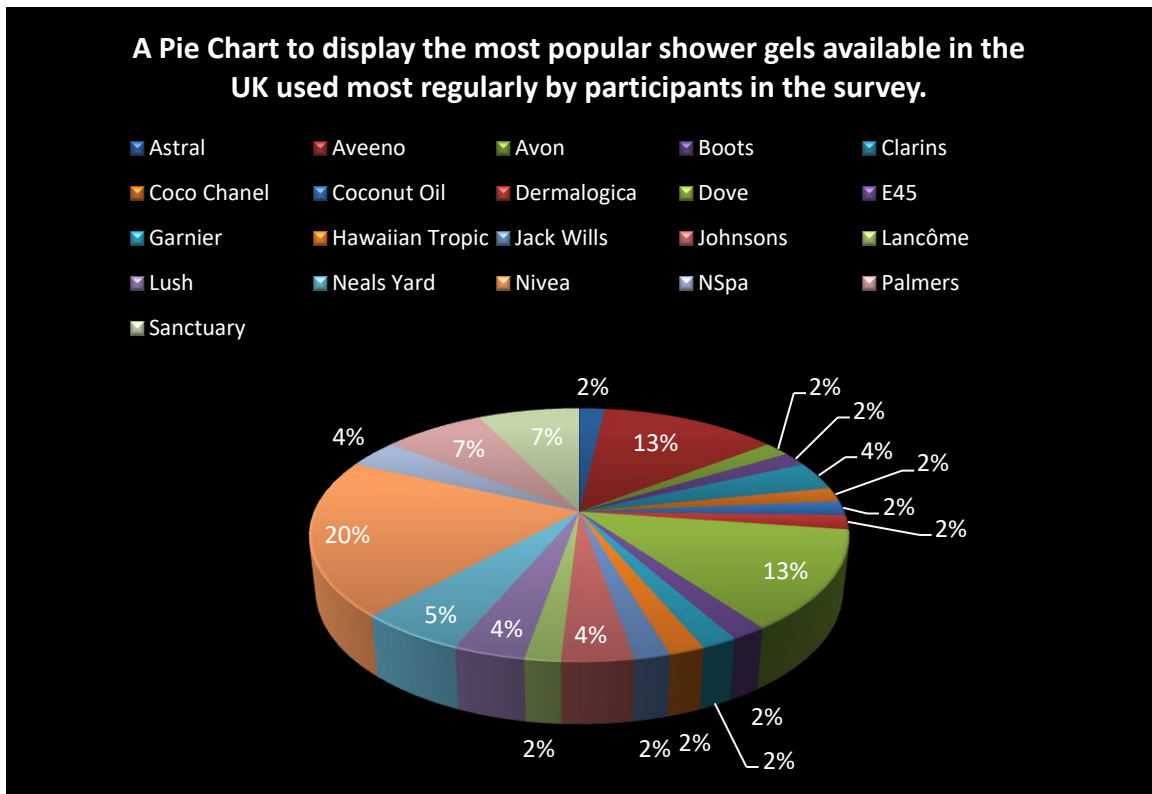


Figure 4.10: A pie chart to show the most popular shower gels used by participants in the survey.

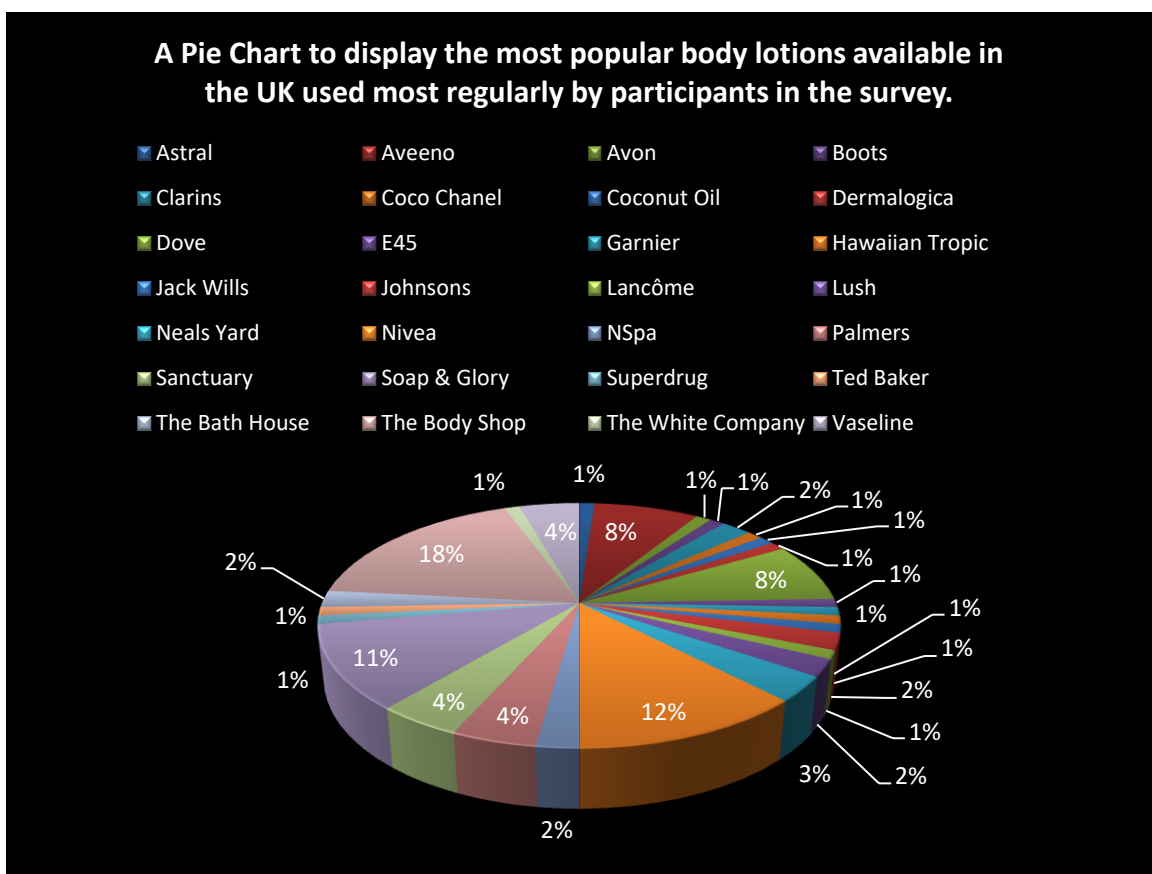


Figure 4.10.1: A pie chart to show the most popular body lotions used by participants in the survey.

Table 4.10.1: A table to show body lotions and shower gels obtained and whether PDMS was detected using the u-ATR.

Label	Body Lotion/Shower Gel	Brand	PDMS in Ingredients	PDMS Detected?
ST25	Shower Gel	Hugo Boss Orange	No	No
ST26	Shower Gel	Paco Rabanne Invictus	No	No
ST27	Shower Gel	David Beckham Sport	No	No
ST28	Shower Gel	Lynx Black Night	No	No
ST29	Shower Gel	Lynx Gold Temptation	No	No
ST30	Shower Gel	Diesel Only the Brave	No	No
ST31	Shower Gel	Jack Wills for Men	No	No
ST32	Body Lotion	Ella & Rose Smoothie	Yes	No
ST33	Shower Gel	Ella & Rose Love to Bathe	No	No
ST34	Body Wash	Next Just Pink	No	No
ST35	Shower Gel	Pampered Coconut Smoothie	No	No
ST36	Body Oil	Garnier Ultimate Beauty Oil	No	No
ST37	Shower Gel	Radox Shower Smoothie Island Indulgent	No	No
ST38	Shower Gel	Lynx Attract for Her	No	No
ST39	Body Wash	Aussie Fresh Mate	No	No
ST40	Body Wash	The Body Shop Dreams Unlimited	No	No
ST41	Body Wash	Boots Extracts Coconut	No	No
ST42	Shower Gel	Impulse Hot Pink	No	No
ST43	Body Wash	Jack Wills Hope Cove	No	No
ST44	Shower Gel	Cussons Imperial Leather Fruit Salad	No	No
ST45	Body Lotion	Soap & Glory Butter Yourself	Yes	No
ST46	Body Lotion	Soap & Glory Body Butter	Yes	No
ST47	Body Lotion	Vaseline Intensive Care Essential Healing Lotion	Yes	Yes
ST48	Body Lotion	Tesco Loves Baby	No	No
ST49	Body Oil	Optima Raw Virgin Coconut Oil	No	No
ST50	Body Lotion	Aveeno Skin Relief Shea Butter	Yes	Yes
ST51	Body Butter	The Body Shop Deluxe Edition Chocomania	Yes	No
ST52	Body Lotion	Bodycare Deli Superfood Carrot & Mint	No	No
ST53	Body Lotion	Korres Sensitive	No	No
ST54	Body Lotion	Salcura Sensitive Hydrator	No	No
ST55	Body Lotion	Neals Yard Bee Lovely	No	No
ST56	Body Lotion	Seacret Pomegranate	Yes	No
ST57	Body Lotion	Bath House Velvet Orchid & Cardomon	Yes	No
ST58	Shower Gel	Sanex Zero	No	No

ST59	Body Oil	Sanctuary 2 day Moisture Oil	No	No
ST60	Shower Gel	Nivea Rich Moisture Smooth Shower Crème	No	No
ST61	Body Wash	Ted Baker Body Maintenance Kit	No	No
ST62	Body Scrub	Nuage Skin	Yes	Yes
ST63	Body Lotion	Cetaphil Moisturising Lotion	No	No
ST64	Body Lotion	Vaseline Intensive Rescue Moisture Locking Lotion	Yes	Yes
ST65	Shower Gel	Faith in Nature Watermelon	No	No
ST66	Body Lotion	Sanctuary Spa Silky Smooth	Yes	No
ST67	Shower Gel	Original Source Moisturising Milk	No	No
ST68	Body Wash	Sanctuary Spa White Lily & Damask Rose	No	No
ST69	Body Wash	Nivea in Shower Moisturiser Cocoa	No	No
ST70	Shower Gel	Original Source Peach & Apricot Shower Oil	No	No
ST71	Body Wash	Dove Nourishing Care & Oil	No	No
ST72	Body Wash	Dove Go Fresh	No	No
ST73	Body Lotion	Hawaiian Tropic After Sun Hydrating Lotion	Yes	Yes
ST74	Body Lotion	Hawaiian Tropic Silk Hydration	No	No
ST75	Bath Crème	Cussons Imperial Leather Bath Crème	No	No
ST76	Body Lotion	Olay Essentials Beauty Fluid	Yes	Yes
ST77	Body Lotion	Nivea Soft Moisturising Cream	Yes	Yes
ST78	Body Lotion	Nivea Crème	No	No
ST79	Body Butter	The Body Shop Shea Butter	No	No
ST80	Body Butter	The Body Shop Vitamin E Butter	No	No

The results in the table show that although PDMS was stated in the ingredients of 46% of the body lotions samples, when using the u-ATR, PDMS was only detected on 50% of these samples. This indicates that although PDMS may be incorporated into body lotions during manufacturing, it is present in volumes which are too small for detection, or other substances are present in much larger volumes preventing the exposure of PDMS.

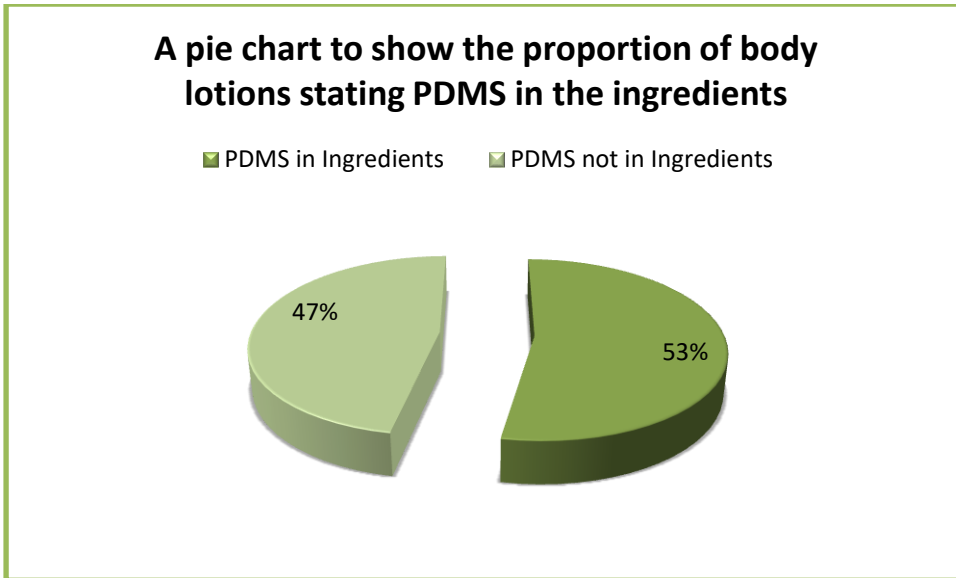


Figure 4.10.2: A pie chart to show the proportion of body lotions stating PDMS in the ingredients

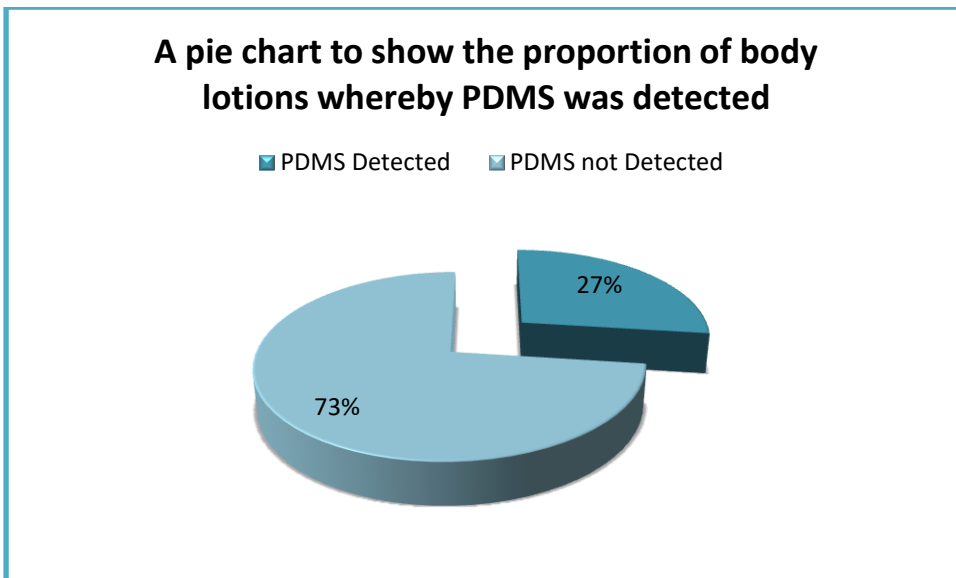


Figure 4.10.3: A pie chart to show the proportion of body lotions whereby PDMS was detected

Positively, PDMS was not detected on any of the body lotions stating that PDMS was not in the ingredients, or any shower gel samples. This is a positive result because if victims of alleged sexual assault have showered since the attack, it is highly unlikely contamination would have occurred from shower gels and body washes.

The Ella & Rose Smoothie Body lotion states dimethicone (PDMS) as being present in the ingredients list, however, when compared to a known sample of PDMS, it is clear the substance is not detected clearly with fully separated peaks, therefore indicating

a very small volume must be included. This is the same case as with the Soap and Glory body butter crème. As the peaks are unable to be evidently defined, these substances would be regarded as negative if recovered in a forensic case.

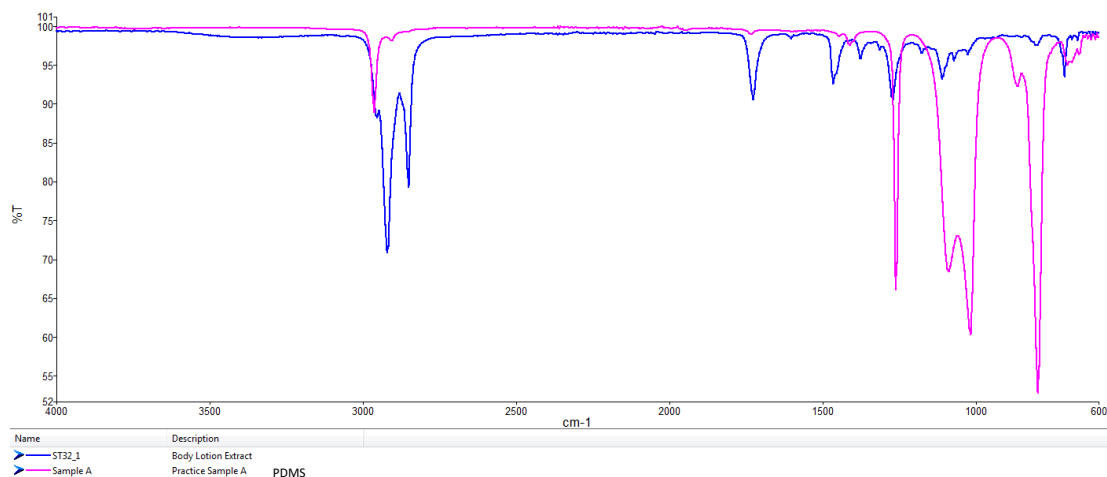


Figure 4.10.4: Spectrum of Ella & Rose body lotion compared to PDMS.

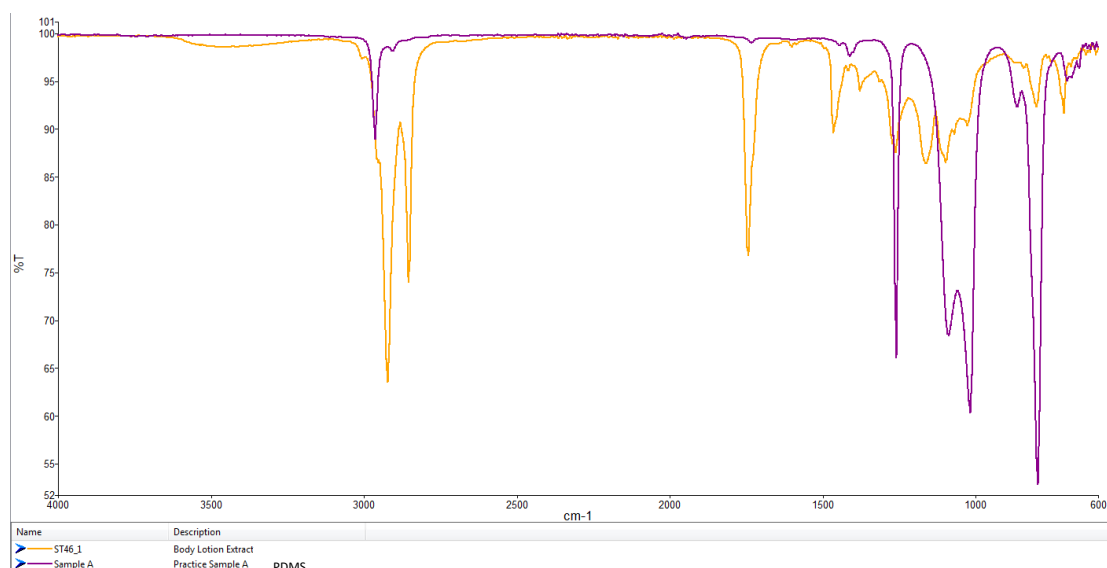


Figure 4.10.5: Spectrum of Soap and Glory body butter crème compared to PDMS.

When the Soap and Glory body butter crème was compared to Cellmark’s internal database, the closest match observed was against Butyl Stearate with an 89% match. Butyl Stearate is a known spreading agent often applied to hand creams and personal care products. This indicates that Butyl Stearate is likely to be present in a much higher quantity compared to PDMS which isn’t strongly detected in the body butter.

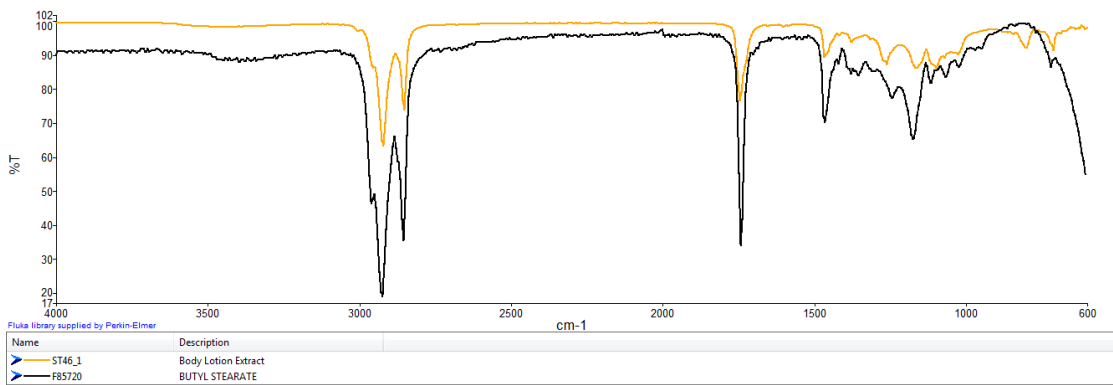


Figure 4.10.6: Spectrum of Soap and Glory body butter crème compared to Butyl Stearate.

Figure 4.107 below shows a random selection of body lotions with the presence of PDMS. The peaks are easily distinguishable but contain contamination peaks due to the other substances present within the lotions. Furthermore, there is a reasonable level of background noise indicating only a low volume of PDMS is able to be sensed.

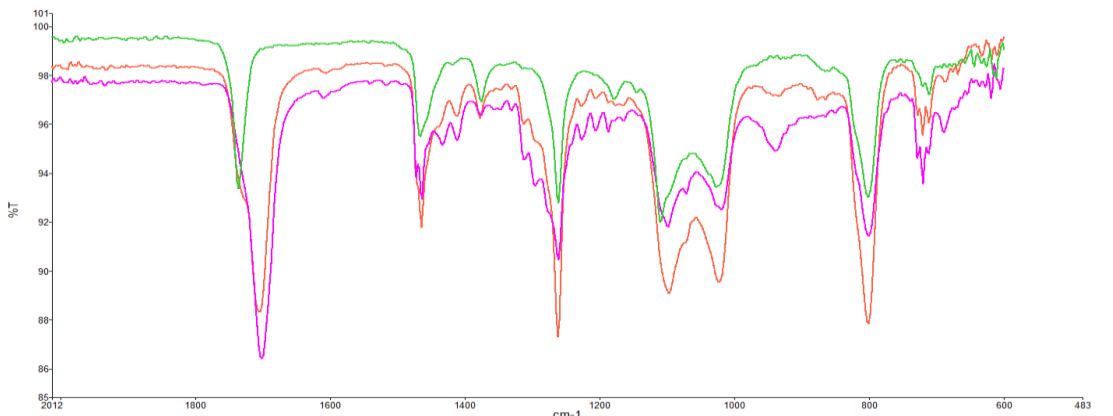


Figure 4.10.7: Fingerprint region of three body lotions containing PDMS.

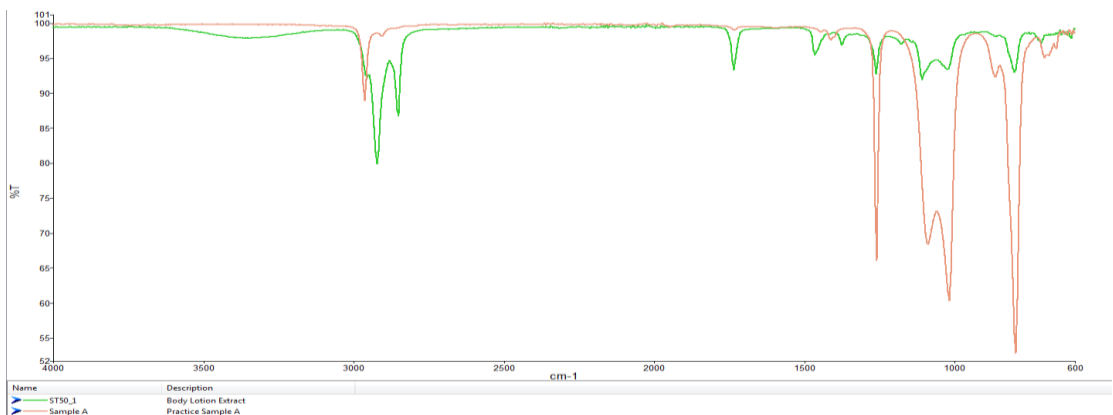


Figure 4.10.8: Sample of body lotion containing PDMS compared to a known PDMS sample.

A random selection of 4 shower gels were chosen from the sample. The spectra were compared and discovered to all possess very similar peaks. In addition to this, none of the peaks correspond to PDMS.

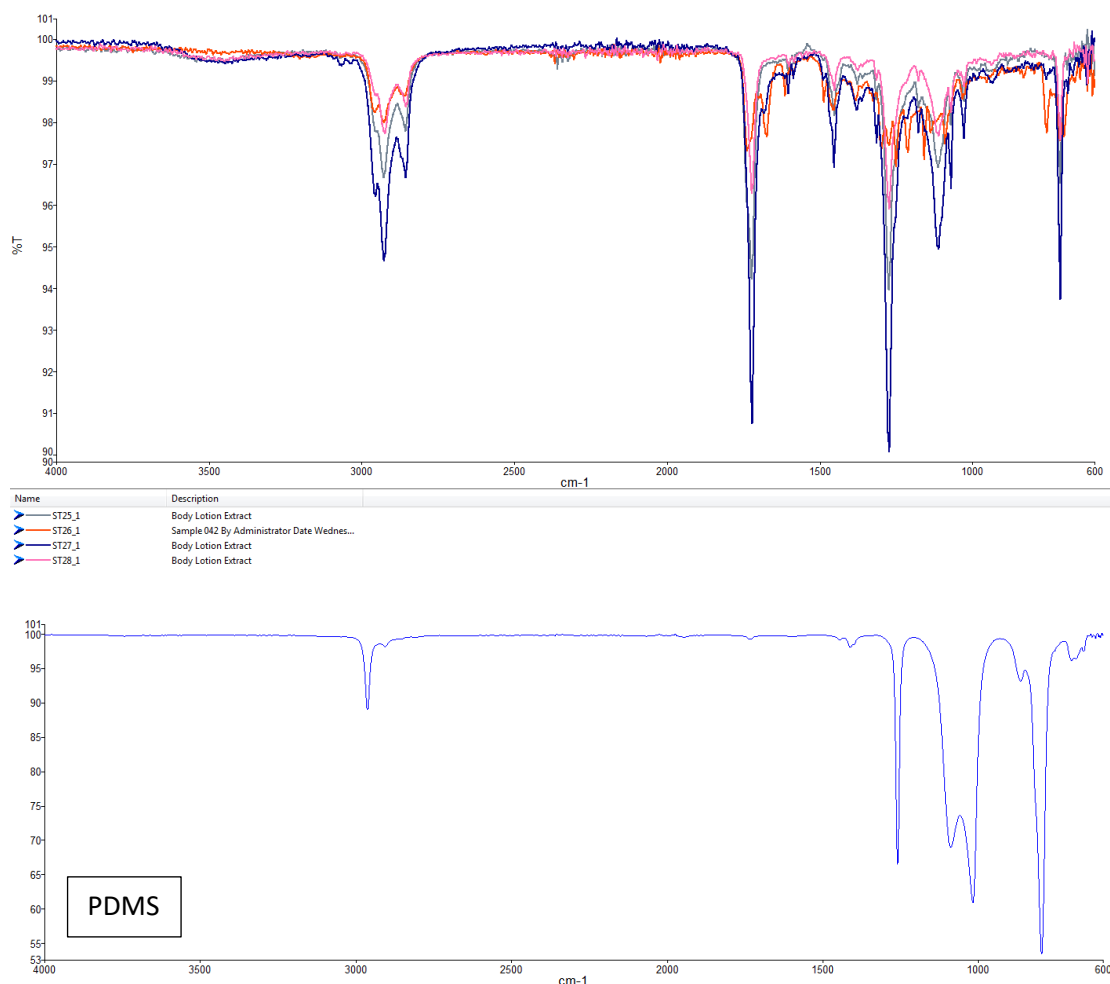


Figure 4.10.9: Selection of shower gel spectra and a comparison with PDMS.

4.11 Direct Semen Extraction and Fast Differential Swabs

All 9 swabs in this area of the project contained 2µl of PDMS and 50µl of diluted semen. PDMS was detected on all swabs after Sperm Elution and either Direct Semen Extraction or Fast Differential.

Table 4.11: A table to show the processes undertaken by each swab and whether PDMS was detected on the swab head following the procedures.

Process Undertaken	Label	PDMS Detected?
Sperm Elution Only	ST81	Yes
	ST82	Yes
	ST83	Yes
Sperm Elution & Direct Semen Extraction	ST84	Yes
	ST85	Yes
	ST86	Yes
Sperm Elution & Fast Differential	ST87	Yes
	ST88	Yes
	ST89	Yes

There is a variation in the peak intensities between all methods, including Sperm Elution alone. On average, Sperm Elution alone displays the most intense peaks, however all methods are easily able to identify PDMS. The overall peak heights are much lower after the Direct Semen Extraction. Figure 4.11 shows the spectra after samples were sperm eluted only. Figure 4.111 displays the spectra after a Direct Semen Extraction, and Figure 4.112 portrays the results after Fast Differential.

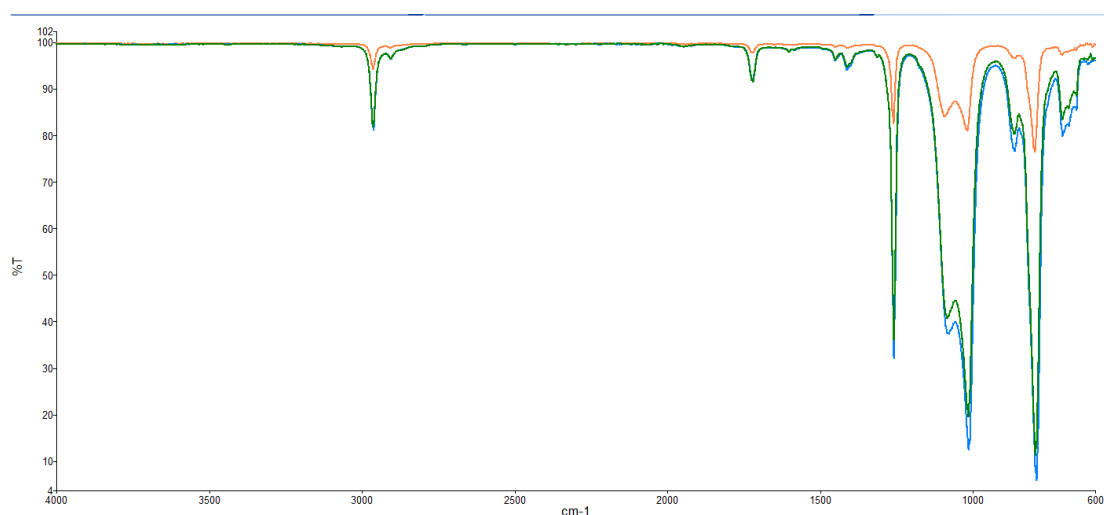


Figure 4.11: Spectra to display the presence of PDMS after Sperm Elution only.

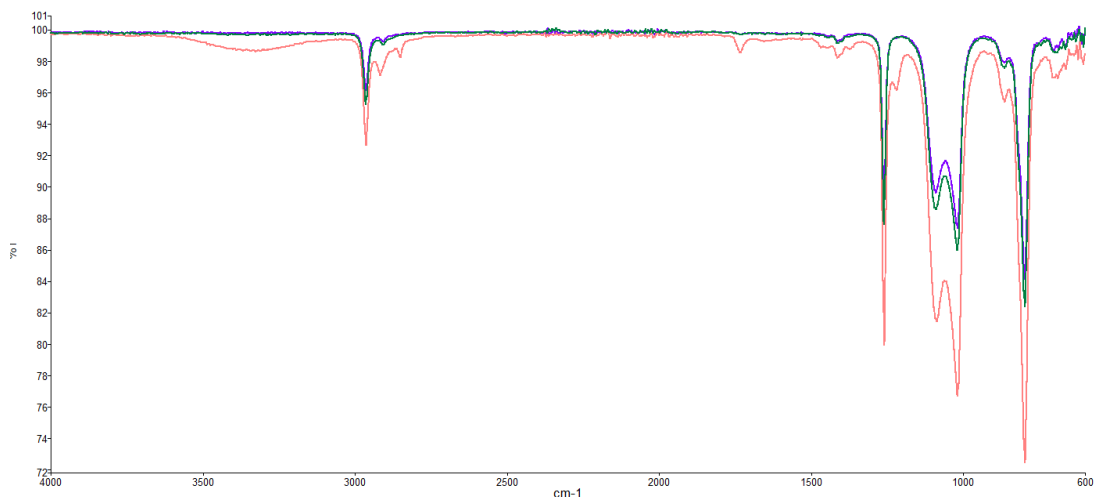


Figure 4.11.1: Spectra to display the presence of PDMS after Sperm Elution and Direct Semen Extraction.

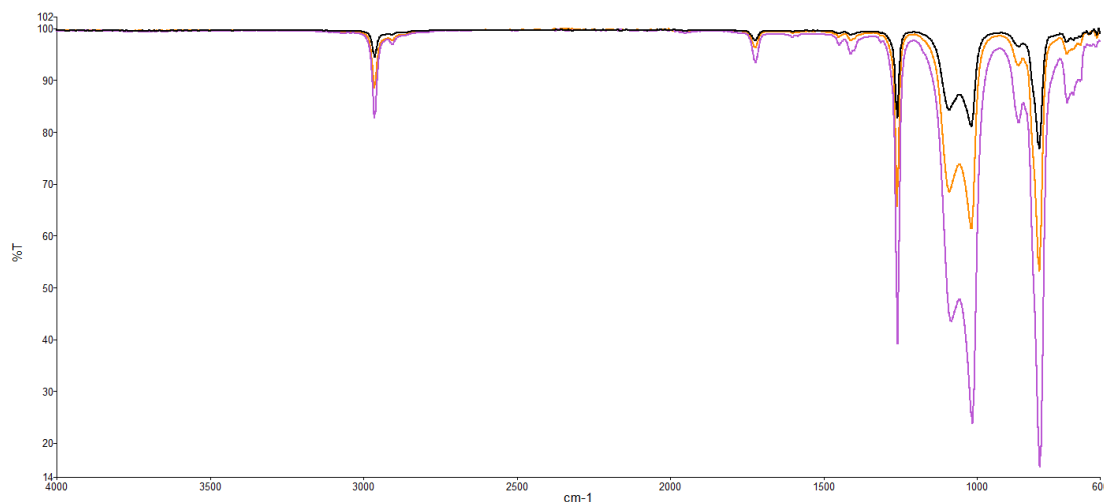


Figure 4.11.2: Spectra to display the presence of PDMS after Sperm Elution and Fast Differential.

4.12 Condom Survey Analysis

All condoms from the sample analysed, except Amor Hot, displayed positive for PDMS.

Table 4.12: A table to show the detection of PDMS on a sample of condoms.

Condom	Expiry	PDMS Detected?
Amor Extra Strong	02-2019	Yes
Amor Hot	03-2019	No (Water-based Lubricant)
Boots Thin Touch	08-2020	Yes
Boots Ultra-Fine	08-2014	Yes
Durex Extra Safe	03-2020	Yes

Durex Latex Free	07-2018	Yes
Durex Real Feel	06-2017	Yes
EXS Regular	02-2021	Yes
EXS Ribbed, Dotted & Flared	12-2020	Yes
EXS Smiley Face Condom	02-2021	Yes
Mates Natural	05-2021	Yes
Mates Original	03-2021	Yes
Mates Skyn Non-Latex	06-2013	Yes
Pasante Infinity	07-2019	Yes
Pasante Regular	08-2021	Yes
Skins Black Choc	04-2018	Yes
Skins Ultra-Thin	11-2021	Yes

Although Amor Hot didn't contain PDMS, Amor Extra Strong did contain traces of the silicone-based lubricant. This indicates that the same brand uses more than one type of lubricant for their range of condoms. However, all other brands from this sample remained consistent at incorporating PDMS. Amor Hot is said to give the user and their partner a warming sensation during intercourse which could be a reason as to why a different lubricant was incorporated. Figure 4.12 shows the spectral differences in two Amor condoms.

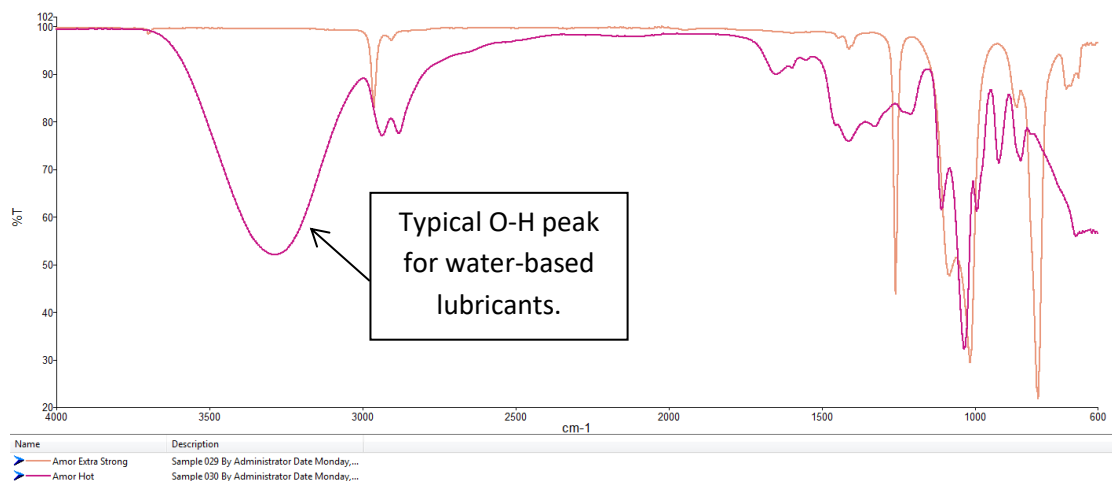


Figure 4.12: Comparison Spectra between Amor Hot and Amor Extra Strong.

When Amor Hot was compared to Cellmark's internal database, a 95% match was displayed with glycerine. Glycerine is a substance often applied in water-based lubricants.

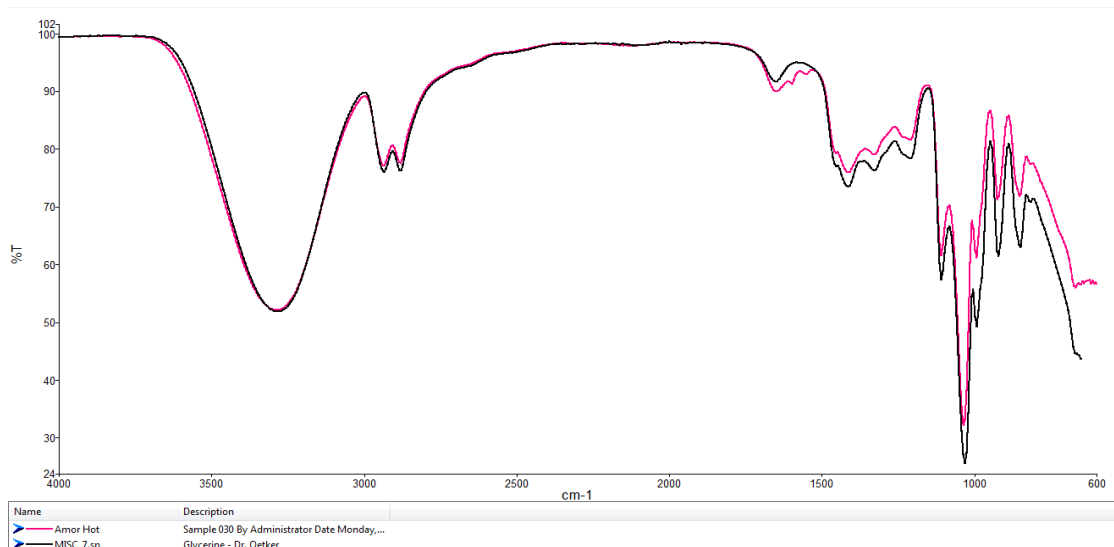


Figure 4.12.1: Comparison Spectra between Amor Hot and Glycerine.

Whilst observing the packaging of Boots Thin Touch and Boots Ultra-Fine, Pasante was stated as the manufacturer. This indicates that specific companies may produce condoms for a wide range of department stores and supermarket own brands. When the spectra for Pasante condoms and Boots condoms were compared, all corresponding peaks for PDMS were clearly present, with no visible differences between each peak. This suggests that the exact same lubricant containing identical additives are combined.

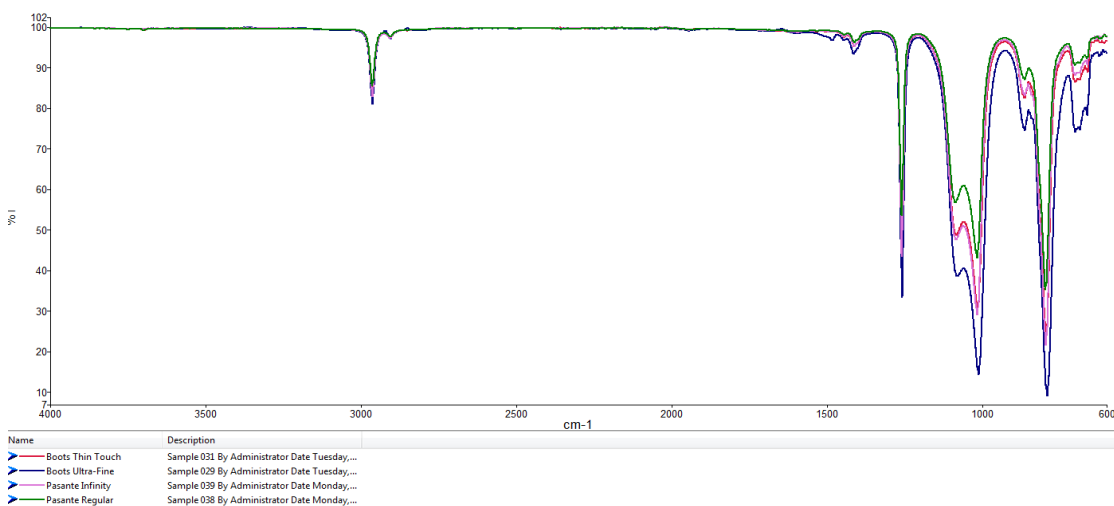


Figure 4.12.2: Comparison Spectra between Pasante and Boots condoms.

Durex is the most popular condom manufacturer on the UK market. This research shows that PDMS is the chosen lubricant applied to all their condoms analysed in this

sample. Figure 4.123 displays the vast similarity of the lubricant applied between a combination of Durex condoms.

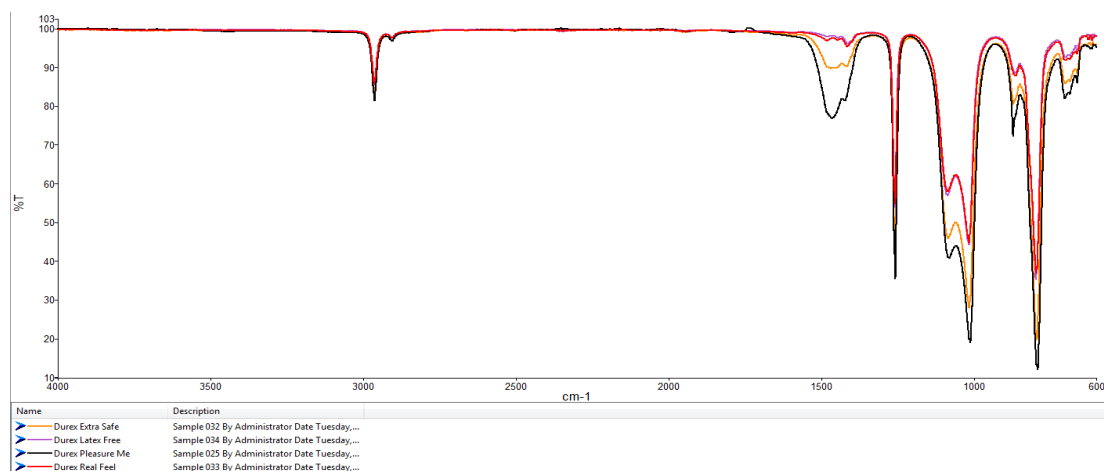


Figure 4.12.3: Comparison Spectra between Durex condoms.

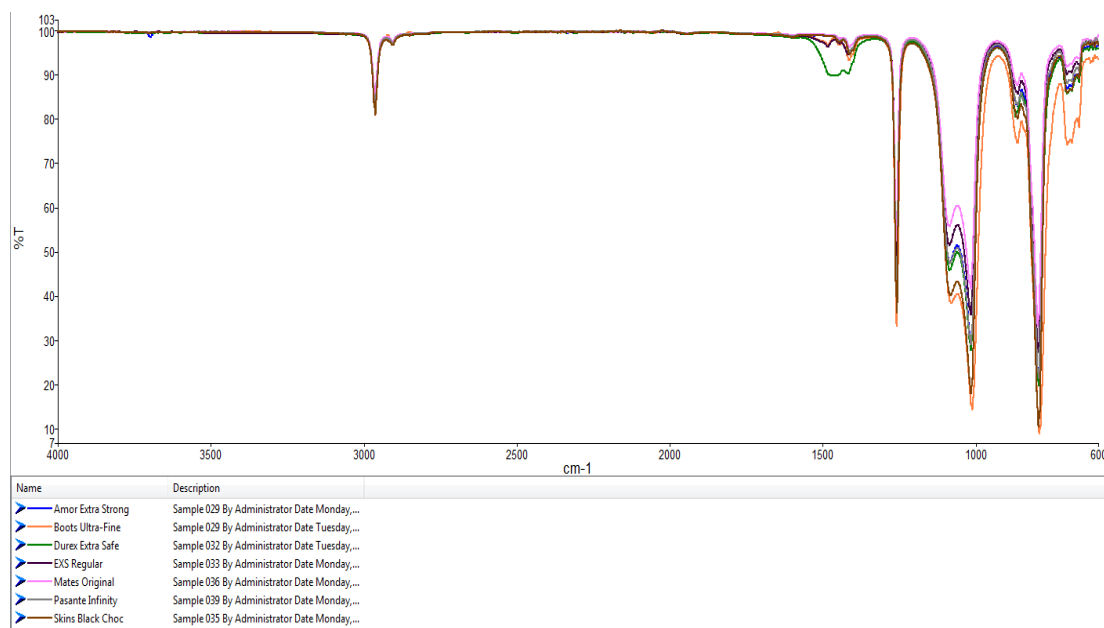


Figure 4.12.4: Comparison Spectra between one condom from each brand.

Figure 4.124 shows the combination of one spectrum from each condom brand within the sample analysed. It can be seen that PDMS is consistently used as a lubricant by condom producers. Furthermore, the spectrum suggests that most manufacturers obtain their lubricants from the same suppliers as there is very little variation between the additional peaks which aren't the four main characteristic peaks of PDMS alone.

4.13 Fabrics

4.13.1 Control Knickers

Prior to determining whether PDMS deposits onto the knicker gusset through vaginal drainage following sexual intercourse with a silicone based lubricated condom, controls from the undergarments must be taken to minimise external contamination. Three pairs of knickers were used as controls, one pair of each size used in the persistence study. Two control areas were recovered from each pair of knickers. All 6 control areas presented positive for PDMS

Table 4.13.1: A table to show the detection of PDMS on control areas of knickers.

Item	Controls	
	Area 1	Area 2
Size 10 Knickers	Positive	Positive
Size 14 Knickers	Positive	Positive
Size 16 Knickers	Positive	Positive

PDMS was detected in all three pairs of knickers in small intensities, however clearly identifiable as the lubricant. Contamination levels differ very slightly between each control area. If a similar level of PDMS is detected in the gusset of the knickers, any findings of the substance would be deemed insignificant. Furthermore, there is a slight correlation between size of knickers and volume of PDMS. However, the volumes detected are very low initially and more control samples would be required to confirm this suggestion. The presence of PDMS on the knickers is because the substance is used as a fabric finish during manufacturing to ensure the product is smooth and durable for use, whilst remaining soft after multiple washes.

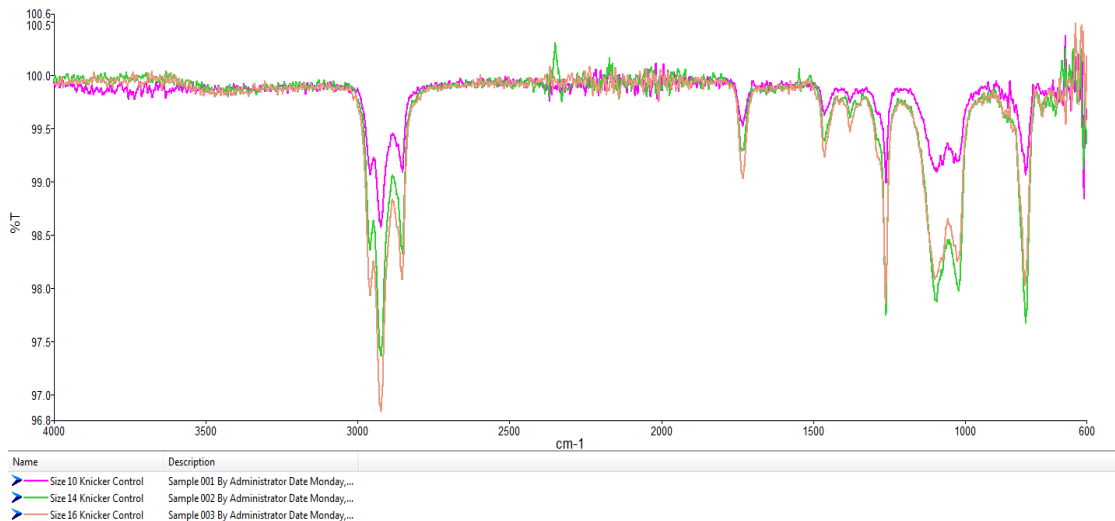


Figure 4.13.1: Spectra of knickers controls displaying PDMS.

Following on from this, the knicker controls were compared to a 0.125 μ l sample of PDMS which had been sperm eluted. The knicker controls are shown in red and the sperm eluted sample of PDMS in black. The 0.125 μ l sample of PDMS after Sperm Elution fluctuates with intensity much more. However, displays a similar average intensity of PDMS compared to the knicker controls.

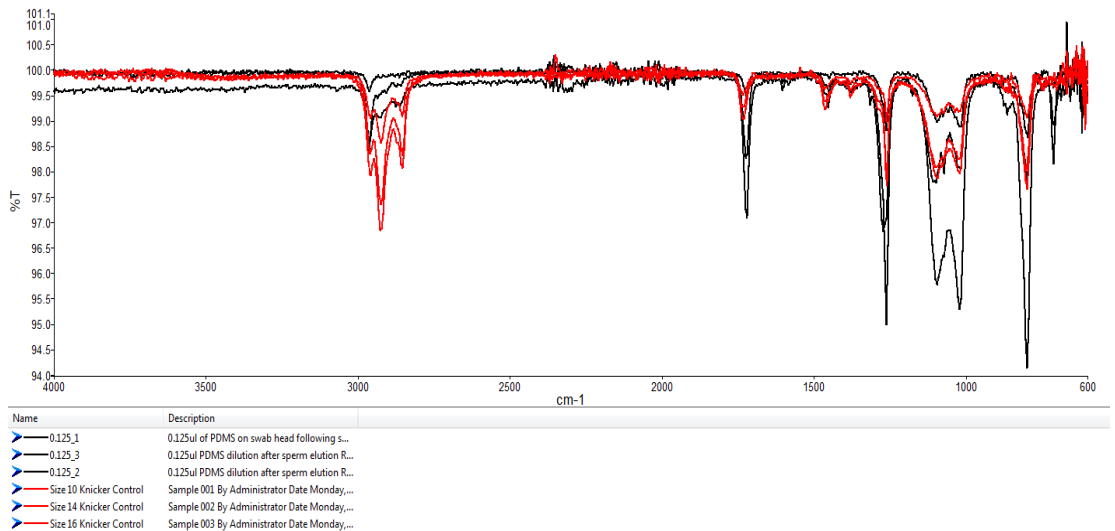


Figure 4.13.11: Spectra of knickers controls compared to a sperm eluted sample of 0.125 μ l sample of PDMS.

4.13.2 Washing Knickers

As PDMS was detected on the knickers to be used in this study, determining the persistence of PDMS on the fabric of knickers would be useful in forensic casework. Two further pairs of knickers, one of each from sizes 10, 14 and 16 were acquired and split equally into two groups. Run 1 was washed with water only and run 2 were washed with water and detergent. A total of 6 washes were completed on each run of knickers.

Table 4.13.2: A table to show the effect of washing knickers on the detection of PDMS.

Run 1 - Wash with Water Only			Run 2 - Wash with Water and Detergent		
Before Washing			Before Washing		
Size	PDMS Detected?	Average Peak Intensity	Size	PDMS Detected?	Average Peak Intensity
10	Positive	99	10	Positive	98.54
14	Positive	97.68	14	Positive	98.49
16	Positive	97.87	16	Positive	98.24

Wash 1			Wash 1		
Size	PDMS Detected?	Average Peak Intensity	Size	PDMS Detected?	Average Peak Intensity
10	Positive	96.92	10	Positive	97.92
14	Positive	96.18	14	Positive	98.52
16	Positive	99.03	16	Positive	98.9

Wash 2			Wash 2		
Size	PDMS Detected?	Average Peak Intensity	Size	PDMS Detected?	Average Peak Intensity
10	Positive	93.46	10	Positive	97.92
14	Positive	97.44	14	Positive	97.16
16	Positive	98.55	16	Limited	-

Wash 3			Wash 3		
Size	PDMS Detected?	Average Peak Intensity	Size	PDMS Detected?	Average Peak Intensity
10	Positive	99.27	10	Limited	Limited
14	Positive	93.39	14	Limited	-
16	Positive	97.72	16	Limited	-

Wash 4			Wash 4		
Size	PDMS Detected?	Average Peak Intensity	Size	PDMS Detected?	Average Peak Intensity
10	Positive	97.47	10	Limited	-
14	Positive	94.31	14	Negative	-

16	Positive	99.41
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16	Negative	-
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Wash 5		
Size	PDMS Detected?	Average Peak Intensity
10	Positive	97.44
14	Positive	98.39
16	Positive	96.45

Wash 5		
Size	PDMS Detected?	Average Peak Intensity
10	Negative	-
14	Negative	-
16	Negative	-

Wash 6		
Size	PDMS Detected?	Average Peak Intensity
10	Positive	97.19
14	Positive	97.64
16	Positive	97.63

Wash 6		
Size	PDMS Detected?	Average Peak Intensity
10	Negative	-
14	Negative	-
16	Negative	-

The results in the tables reveal that washing knickers with water alone displays a very little, if any effect on the removal of PDMS from fabrics. Washing knickers with the use of a detergent removes PDMS from fabrics after approximately 3 washes. Limited recovery of PDMS is where peaks are present in the characteristic positions of PDMS; however the peaks have not fully separated and are unable to be confidently identified as the lubricant. This provides valuable evidence towards forensic casework because if a pair of knickers have been submitted for analysis following an alleged sexual assault with the use of a condom, PDMS is unlikely to arise as contamination from a fabric conditioner if the knickers appear to have been well worn prior to the incident. If these details are obtained for the victim, this could aid in the confidence of any findings if PDMS is detected.

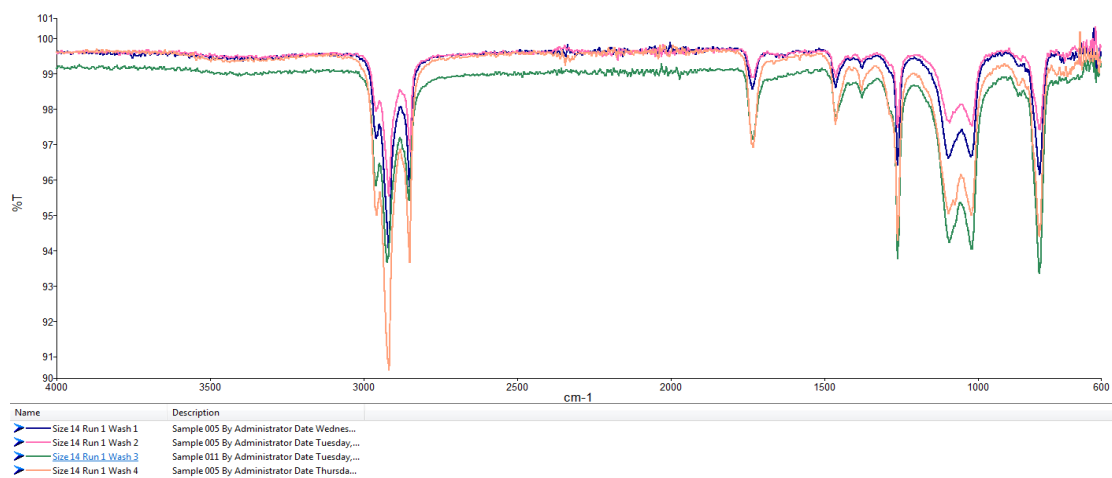


Figure 4.13.2: Spectra of size 14 knickers after 1-4 washes with water only.

Figure 4.13.2 displays an example of Size 14 Knickers having undertaken 1-4 washes with the use of water only. PDMS is still clearly present after 4 washes. There is also no correlation between the volume of PDMS present and the number of washes. Besides, this proposes that the volume of PDMS applied to different areas of knickers may not be constant.

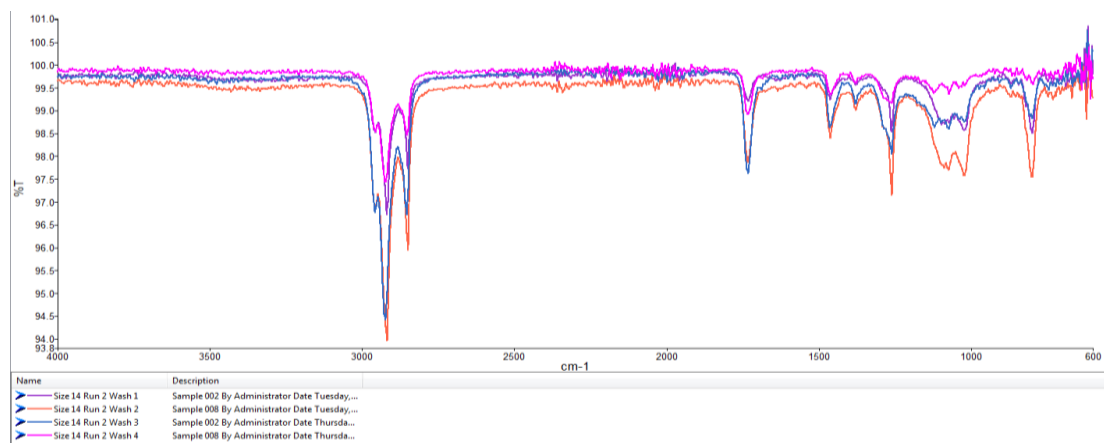


Figure 4.13.21: Spectra of size 14 knickers after 1-4 washes with water and detergent.

Figure 4.13.21 above illustrates the same four washes on the second run of size 14 knickers; however detergent was used this time. It can be seen that there isn't complete correlation between the number of washes and the depletion of PDMS. However, after wash 4, PDMS is no longer detectable on the knickers, meaning PDMS can be removed from fabrics with the use of detergents. Additionally, the peak intensity of the spectra from the knickers washed with detergents is much weaker with higher levels of background noise compared to the knickers washed with water alone.

4.14 Persistence

4.14.1 Persistence Swabs

The results for the persistence of PDMS in the vagina following sexual intercourse are illustrated in Table 4.14.1. Unfortunately, no males volunteered to participate in this study, therefore there are no results present for penile swabs. Fortunately, all of the outer vaginal control swabs displayed negative, meaning no external factors contributed to the detection of PDMS.

PDMS has been detected on all blind vaginal swabs taken at the 6 hour time interval. However, swabs from one participant contained no PDMS at 0 hours, which is a time interval where PDMS is highly likely to be present. This indicates that external factors, e.g. thoroughness of swabbing, have affected the quality of the findings and may have resulted in anomalous results occurring. PDMS was marginally detected on one swab at 12 hours. However, this was only detected on the first blind vaginal swab, meaning that the lubricant was only present in very small volumes, hence not being recovered on the second blind vaginal swab. Following on from this, it is highly improbable that PDMS will be detected from sexual assault victims from time intervals greater than 12 hours after the offence.

Resulting from time restraints, participant ST117 did not complete the final 3 time intervals. However, this was deemed unnecessary with PDMS not being detected from that participant at 12 hours.

All sample remains were initially analysed using the u-ATR, however, for any swab heads where PDMS was not detected, the sample was additionally analysed using the FTIR microscope due to being a more sensitive technique. However, this technique was able to confirm the result as no extra PDMS was detected using the FTIR microscope.

Table 4.14.1: A table to show the detection and persistence of PDMS at different time intervals following sexual intercourse of a Cellmark employee with their partner with a PDMS lubricated condom.

Persistence Swabs						
Time Interval	Item Number	Item Description	Item Returned	Item Analysed	PDMS Detected u-ATR?	PDMS Detected Microscope?
0 Hours	ST110/13	Outer Vaginal Control Swab	Yes	Yes	Negative	Negative
	ST110/14	Blind Vaginal Swab 1	Yes	Yes	Positive	-
	ST110/15	Blind Vaginal Swab 2	Yes	Yes	Positive	-
	ST116/13	Outer Vaginal Control Swab	Yes	Yes	Negative	Negative
	ST116/14	Blind Vaginal Swab 1	Yes	Yes	Positive	-
	ST116/15	Blind Vaginal Swab 2	Yes	Yes	Positive	-
	ST117/13	Outer Vaginal Control Swab	Yes	Yes	Negative	Negative
	ST117/14	Blind Vaginal Swab 1	Yes	Yes	Negative	Negative
	ST117/15	Blind Vaginal Swab 2	Yes	Yes	Negative	Negative
6 Hours	ST110/16	Outer Vaginal Control Swab	Yes	Yes	Negative	Negative
	ST110/17	Blind Vaginal Swab 1	Yes	Yes	Positive	-
	ST110/18	Blind Vaginal Swab 2	Yes	Yes	Positive	-
	ST116/16	Outer Vaginal Control Swab	Yes	Yes	Negative	Negative
	ST116/17	Blind Vaginal Swab 1	Yes	Yes	Positive	-
	ST116/18	Blind Vaginal Swab 2	Yes	Yes	Positive	-
	ST117/16	Outer Vaginal Control Swab	Yes	Yes	Negative	Negative
	ST117/17	Blind Vaginal Swab 1	Yes	Yes	Positive	-
	ST117/18	Blind Vaginal Swab 2	Yes	Yes	Positive	-

12 Hours	ST110/1	Outer Vaginal Control Swab	Yes	Yes	Negative	Negative
	ST110/2	Blind Vaginal Swab 1	Yes	Yes	Positive	-
	ST110/3	Blind Vaginal Swab 2	Yes	Yes	Negative	Negative
	ST116/1	Outer Vaginal Control Swab	Yes	Yes	Negative	Negative
	ST116/2	Blind Vaginal Swab 1	Yes	Yes	Negative	Negative
	ST116/3	Blind Vaginal Swab 2	Yes	Yes	Negative	Negative
	ST117/1	Outer Vaginal Control Swab	Yes	Yes	Negative	Negative
	ST117/2	Blind Vaginal Swab 1	Yes	Yes	Negative	Negative
	ST117/3	Blind Vaginal Swab 2	Yes	Yes	Negative	Negative
18 Hours	ST110/4	Outer Vaginal Control Swab	Yes	Yes	Negative	Negative
	ST110/5	Blind Vaginal Swab 1	Yes	Yes	Negative	Negative
	ST110/6	Blind Vaginal Swab 2	Yes	Yes	Negative	Negative
	ST116/7	Outer Vaginal Control Swab	Yes	Yes	Negative	Negative
	ST116/8	Blind Vaginal Swab 1	Yes	Yes	Negative	Negative
	ST116/9	Blind Vaginal Swab 2	Yes	Yes	Negative	Negative
	ST117/4	Outer Vaginal Control Swab	No	N/A	N/A	N/A
	ST117/5	Blind Vaginal Swab 1	No	N/A	N/A	N/A
	ST117/6	Blind Vaginal Swab 2	No	N/A	N/A	N/A
24 Hours	ST110/7	Outer Vaginal Control Swab	Yes	Yes	Negative	Negative
	ST110/8	Blind Vaginal Swab 1	Yes	Yes	Negative	Negative
	ST110/9	Blind Vaginal Swab 2	Yes	Yes	Negative	Negative
	ST116/4	Outer Vaginal Control Swab	Yes	Yes	Negative	Negative
	ST116/5	Blind Vaginal Swab 1	Yes	Yes	Negative	Negative
	ST116/6	Blind Vaginal Swab 2	Yes	Yes	Negative	Negative
	ST117/7	Outer Vaginal Control Swab	No	N/A	N/A	N/A

	ST117/8	Blind Vaginal Swab 1	No	N/A	N/A	N/A
	ST117/9	Blind Vaginal Swab 2	No	N/A	N/A	N/A
36 Hours	ST110/10	Outer Vaginal Control Swab	Yes	Yes	Negative	Negative
	ST110/11	Blind Vaginal Swab 1	Yes	Yes	Negative	Negative
	ST110/12	Blind Vaginal Swab 2	Yes	Yes	Negative	Negative
	ST116/10	Outer Vaginal Control Swab	Yes	Yes	Negative	Negative
	ST116/11	Blind Vaginal Swab 1	Yes	Yes	Negative	Negative
	ST116/12	Blind Vaginal Swab 2	Yes	Yes	Negative	Negative
	ST117/10	Outer Vaginal Control Swab	No	N/A	N/A	N/A
	ST117/11	Blind Vaginal Swab 1	No	N/A	N/A	N/A
	ST117/12	Blind Vaginal Swab 2	No	N/A	N/A	N/A

As previously covered, all outer vaginal control swabs tested negative for PDMS. A handful of examples are shown below in Figure 4.14.1. There are large amounts of background noise present. This is because only low levels of contamination are present with undefined peaks. When compared to Cellmarks internal database, the closest match was 81% with Dibutyl phthalate; however, the intensity comparison was extremely weak displaying no correlating peaks in the controls.

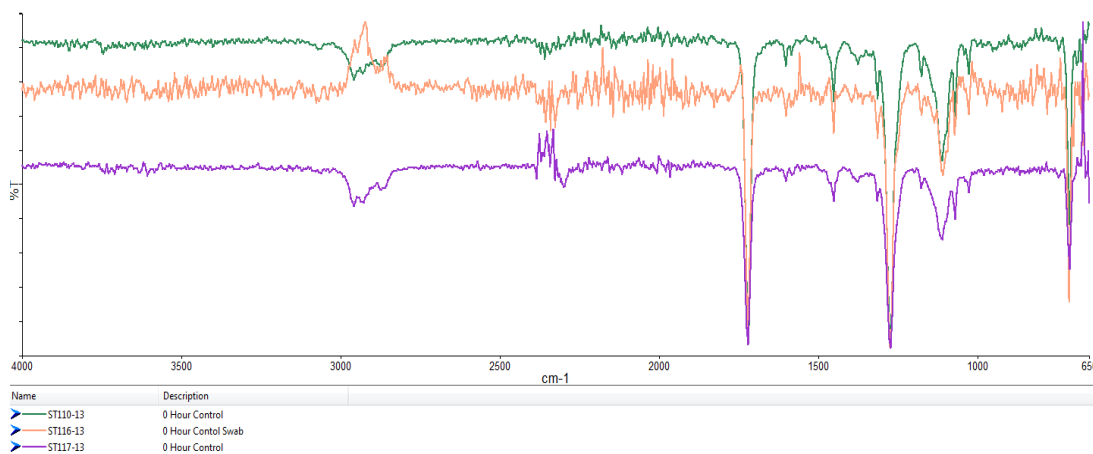


Figure 4.14.1: Spectra of 0 hour outer vaginal control swabs.

All swabs taken from 18 hours onwards were negative for PDMS. Examples of each time interval are shown in Figure 4.14.11. All three spectra exhibited in Figure 4.14.11 possess great similarities between their peaks. Medium peak intensities are present, indicating significant levels of external substances are present. However, this does not affect the detection of PDMS. The differences between blind vaginal swab 1 and blind vaginal swab 2 are minimal, with only one variance at 12 hours.

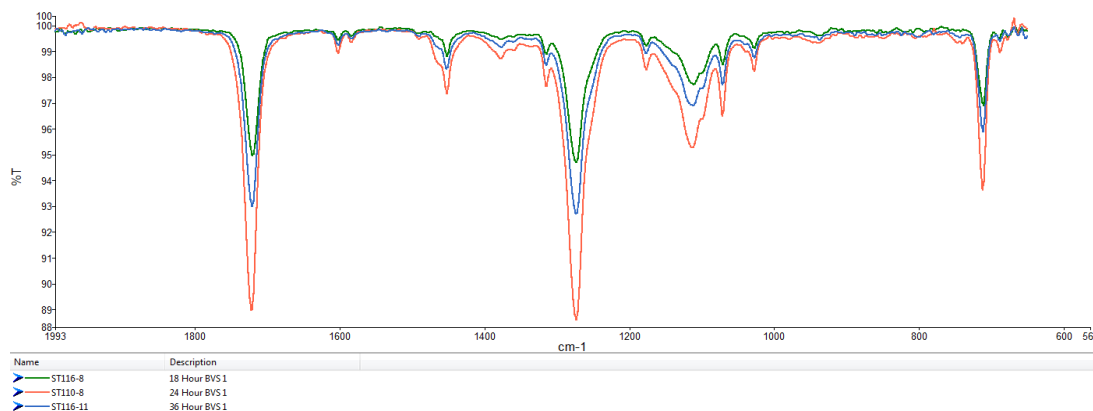


Figure 4.14.11: Spectra of 18, 24 and 36 hour blind vaginal 1 swabs.

Blind vaginal swabs taken at 0 hours are shown in Figure 4.14.12. There is little consistency between the intensity of the peaks. As stated prior, PDMS is not detected on 2 of the 6 swabs. This is unexpected at 0 hours as you would expect to find PDMS on all swabs taken immediately after intercourse using a PDMS lubricated condom. This raises a second question of whether condom manufacturers apply a specific known volume of lubricant to each condom. Also, it is unknown how much lubricant is lost to the packaging when removing for use.

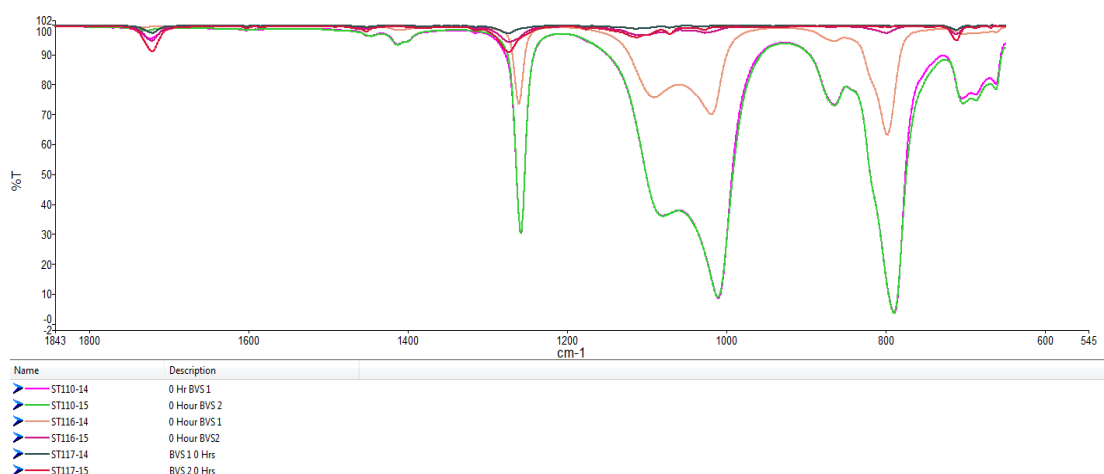


Figure 4.14.12: Spectra of 0 hour blind vaginal swabs.

Figure 4.14.13 displays that PDMS is clearly detected on all the 6 hour swabs. However, only a limited amount of PDMS was identified at 12 hours. This suggests that smaller time intervals may provide a more accurate limit for the recovery of PDMS.

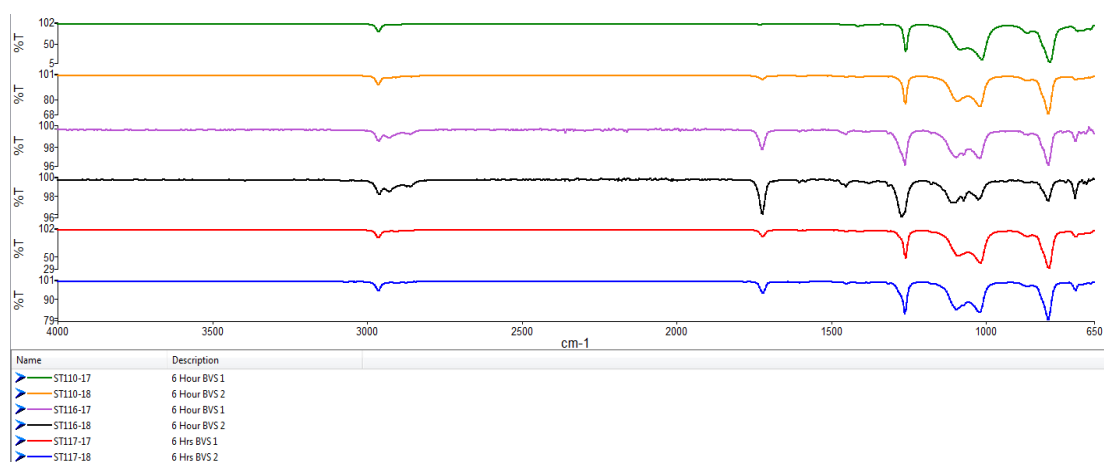


Figure 4.14.13: Spectra of 6 hour blind vaginal swabs.

Finally, it would be expected that, the first vaginal swab would tend to contain a slightly larger volume of PDMS compared to the second. This is likely to be because the PDMS is removed onto the first swab, leaving very little remaining for the second swab. However, as seen in Figure 4.14.14 this is not the case as both spectra for volunteer ST/110 are very similar. On the other hand, this could be due to the PDMS being present in high volumes at 0 hours.

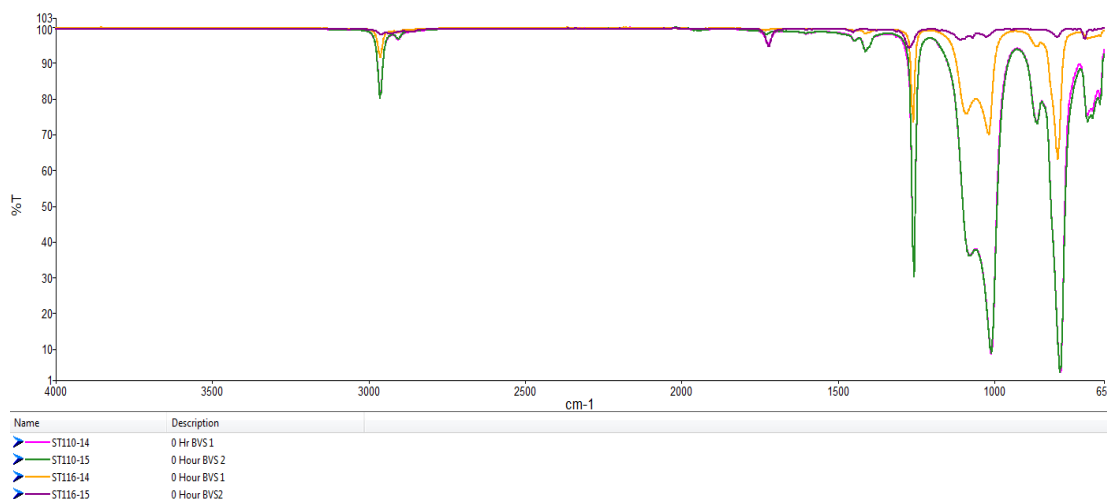


Figure 4.14.14: Variation spectra of 0 hour blind vaginal swabs.

4.14.2 Persistence Knickers

PDMS was able to be detected on the gusset of all knickers which were extracted with isooctane only, and all but 1 pair which was sperm eluted and extracted. However, there is no visible indication as to the location of PDMS in the gusset of the knickers. For example, with sample ST116/23, all the lubricant could have been present on the left hand side of the gusset, hence no detection after Sperm Elution. Although PDMS was identified as being present within the fabric of the knickers, the lubricant detected in this section of the project was present in a significantly higher volume. As a whole, the success rate of detecting PDMS in the gusset of knickers following intercourse with a lubricated condom could be very useful in sexual assault cases because if too much time has passed since the alleged assault, the knickers could provide an alternate form of evidence if they hadn't been washed since the time of the offence.

Table 4.14.2: A table to show the detection of PDMS in knicker gussets at different time intervals following sexual intercourse.

Persistence Knickers						
Time Interval	Item Number	Item Description	Item Returned	Item Analysed	PDMS Detected? Extraction Only	PDMS Detected? After Sperm Elution
0 Hours	ST110/23	Size 10 White Cotton Knicker Gusset	Yes	Yes	Positive	Positive
	ST116/23	Size 16 White Cotton Knicker Gusset	Yes	Yes	Positive	Negative
	ST117/24	Size 14 White Cotton Knicker Gusset	Yes	Yes	Positive	Positive
6 Hours	ST110/24	Size 10 White Cotton Knicker Gusset	Yes	Yes	Positive	Positive
	ST116/24	Size 16 White Cotton Knicker Gusset	Yes	Yes	Positive	Positive
	ST117/23	Size 14 White Cotton Knicker Gusset	Yes	Yes	Positive	Positive
12 Hours	ST110/19	Size 10 White Cotton Knicker Gusset	Yes	Yes	Positive	Positive
	ST116/19	Size 16 White Cotton Knicker Gusset	Yes	Yes	Positive	Positive
	ST117/19	Size 14 White Cotton Knicker Gusset	Yes	Yes	Positive	Positive
18 Hours	ST110/20	Size 10 White Cotton Knicker Gusset	Yes	Yes	Positive	Positive
	ST116/20	Size 16 White Cotton Knicker Gusset	Yes	Yes	Positive	Positive
	ST117/20	Size 14 White Cotton Knicker Gusset	No	N/A	N/A	N/A
24 Hours	ST110/21	Size 10 White Cotton Knicker Gusset	Yes	Yes	Positive	Positive
	ST116/21	Size 16 White Cotton Knicker Gusset	Yes	Yes	Positive	Positive
	ST117/21	Size 14 White Cotton Knicker Gusset	No	N/A	N/A	N/A
36 Hours	ST110/22	Size 10 White Cotton Knicker Gusset	Yes	Yes	Positive	Positive
	ST116/22	Size 16 White Cotton Knicker Gusset	Yes	Yes	Positive	Positive
	ST117/22	Size 14 White Cotton Knicker Gusset	No	N/A	N/A	N/A

All but one pair of knickers displayed positive for PDMS after Sperm Elution. The spectra below in Figure 4.14.2 displays the knickers provided for the 0 Hour time interval. The peak at approximately 800cm^{-1} is missing. The closest peak present to this is at 742cm^{-1} . A possible reason for no detection could be because all the PDMS had been swabbed at 0 hours, meaning no PDMS could have been transferred to the knickers via drainage or contact.

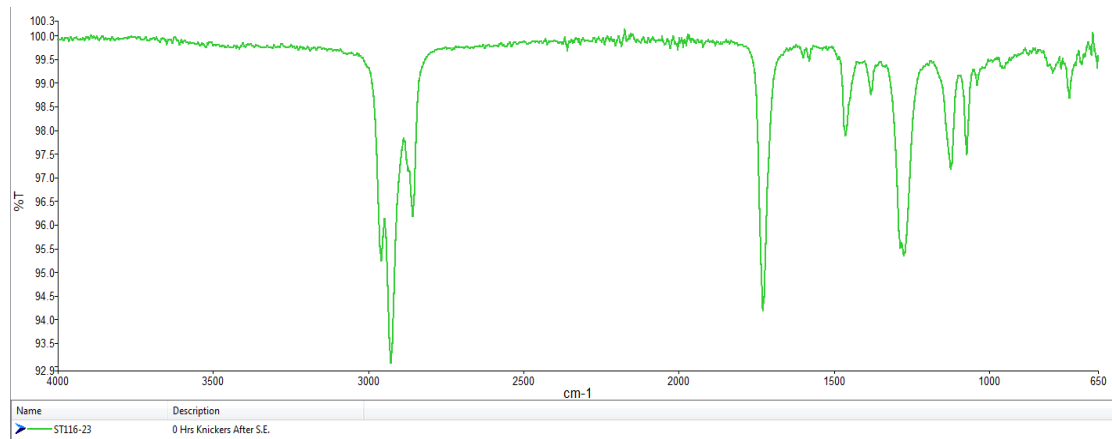


Figure 4.14.2: Spectra of 0 hour knicker gusset after Sperm Elution.

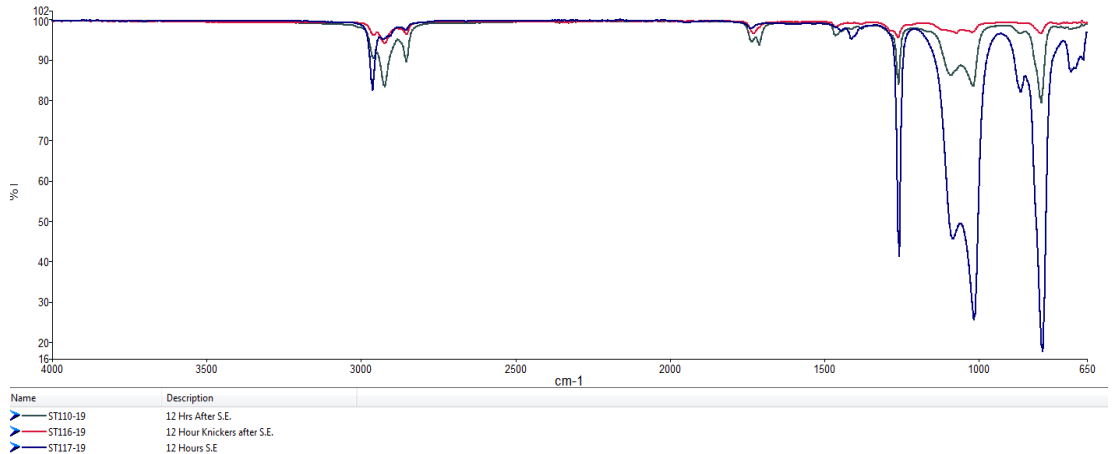


Figure 4.14.21: Spectra of 12 hour knicker gusset after Sperm Elution.

Figure 4.14.21 displays the knickers worn for 12 hours by each participant. Varying levels of PDMS have been detected in each pair of knickers; however the lubricant is still visibly present in each pair. This could be due to either vaginal drainage and/or direct contact. Also, external factors including the different stages in menstrual cycle could affect the amount of vaginal drainage. Alternatively, dissimilarity could result from the accuracy of the spotting technique for analysis.

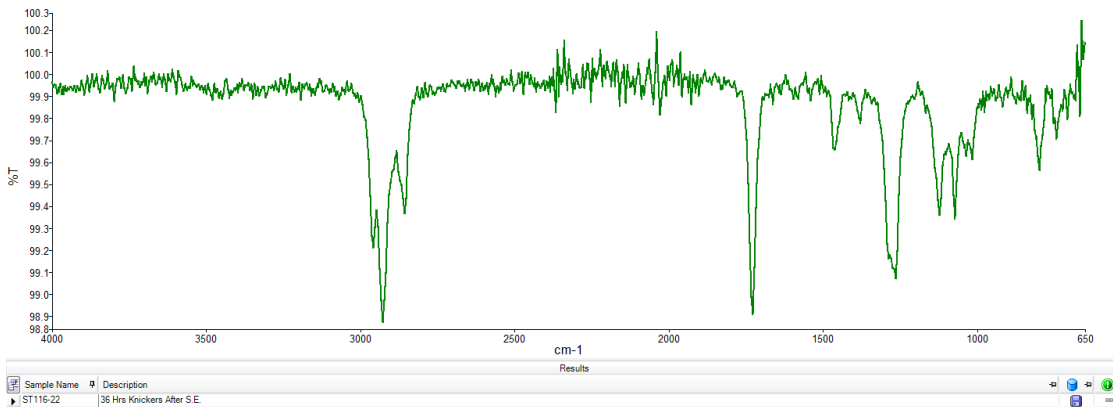


Figure 4.14.22: Spectra of 36 hour knicker gusset after Sperm Elution.

Very limited evidence is recovered for the 36 hour time interval. The peaks present aren't very clear and concise and this spectrum is unlikely to be confidently accepted as PDMS in forensic casework. Therefore, not all PDMS is drained into the gusset. As previously mentioned, the PDMS could be present in larger volumes on the other half of the gusset used for the extraction only.

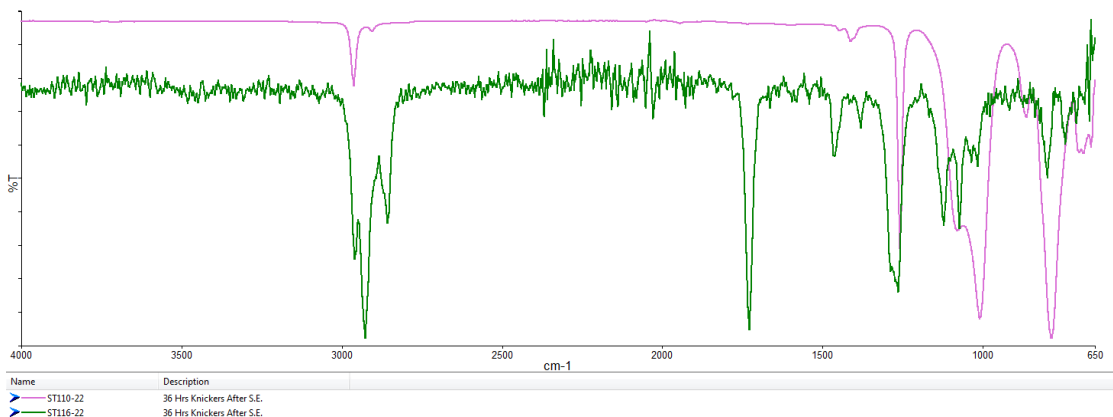


Figure 4.14.23: Comparison spectra of 36 hour knicker gusset after Sperm Elution.

There is a wide range of individual differences seen between the 36 hour pair of knickers. More volunteers are needed to produce accurate results. However, this variation could be contributed to by the activity of the individual. If volunteer ST116 was more active than volunteer ST110, the traces of PDMS could have been wiped away from the gusset. It may otherwise be suggested that more movement could increase the likelihood of PDMS transferring into the gusset via vaginal drainage.

Figure 4.14.24 demonstrates the spectra for one volunteer's 0-36 hour time intervals following the Sperm Elution procedure. There are immense differences between the

peak intensities for each pair of knickers. Nonetheless, no correlation is observed between the swabbing time intervals and the volume of PDMS detected.

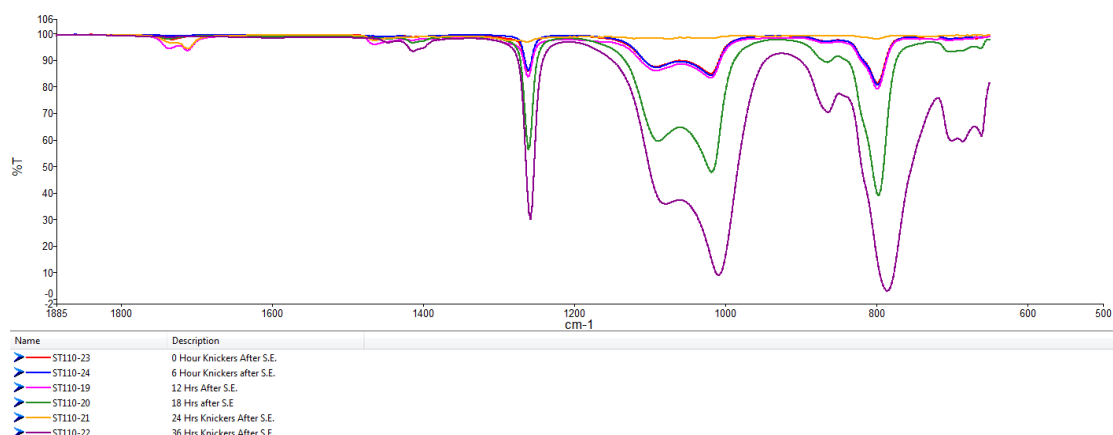


Figure 4.14.24: Spectra of participant ST110 0-36 hour swab intervals.

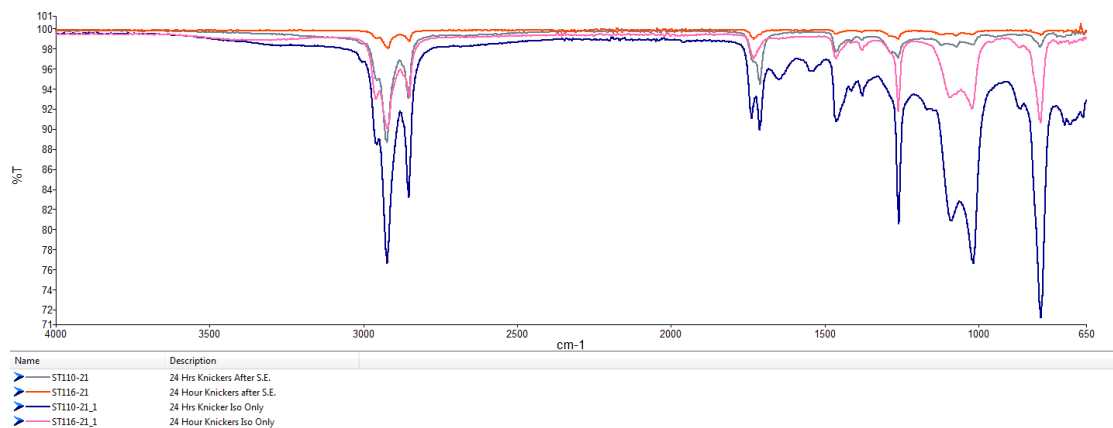


Figure 4.14.25: Spectra of participant ST110 24 hour swab intervals.

Figure 4.14.25 presents the spectra of knickers worn for 24 hours. 2 pairs were sperm eluted and two pairs were extracted with isooctane only. It can be seen that some PDMS is likely to have been lost during Sperm Elution as the peak intensity is much greater for the isooctane extraction only. On the other hand, the PDMS may have been present on the opposing area of the gusset prior to analysis.

Chapter 5 – Discussion

5.1 PDMS Identification

Figure 5.1 illustrates an example of an Infra-red spectrum of PDMS retrieved during this project. There are four characteristic peaks of PDMS in the fingerprint region that make up the spectrum. A symmetric CH_3 deformation formed as a singlet peak at 1260cm^{-1} . Anti-symmetric Si-O-Si stretching displays a doublet peak at 1090cm^{-1} and 1020cm^{-1} . Si- CH_3 rocking is present as a singlet at 800cm^{-1} . These values concur with the peak assignments identified for PDMS from multiple alternate sources [75]. Nevertheless, deviation within this outline may occur. For example, peak intensities may alter dependent upon the volume of the lubricant, and often a neighbouring peak attached to the baseline at 800 cm^{-1} arises. In addition to this, the wavenumber can fluctuate slightly depending upon the accuracy of the equipment, and whether any contaminants are present within the substance. These minor dissimilarities are standard and the spectrum would be regarded as PDMS providing all four of the characteristic peaks are existent and distinguishable.

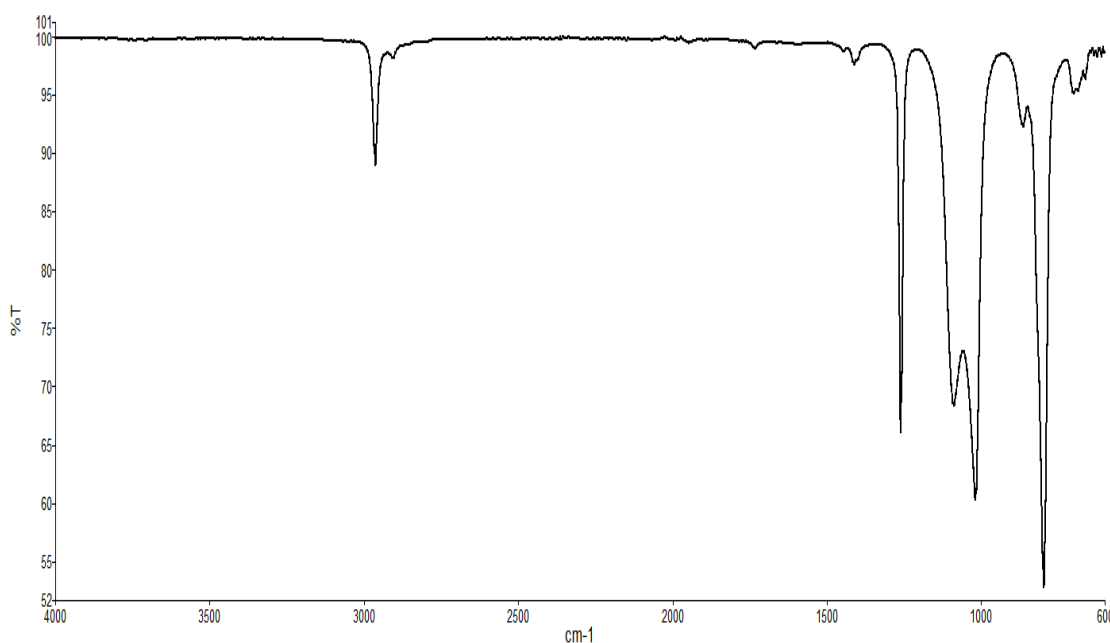


Figure 5.1: An Infra-red Spectrum of polydimethylsiloxane.

This arrangement of peaks is specific to PDMS and if observed by a scientist, this configuration would directly specify PDMS is present. Each of the four peaks frequently displays comparable peak intensities, with the peak height slightly

decreasing as the wavenumber increases. The peaks are proportionally shaped with sharp, narrowing rounded peak points.

To allow for an affirmative response for the presence of PDMS, all peaks must be present. Insignificant concealing of peaks may arise due to the attendance of additional silicones with comparable absorbances. However, if the peak is still able to be determined, the lubricant may still be confirmed. Figure 5.11 shows PDMS present in a low volume. Here, the peak at 1090 cm^{-1} differs in appearance compared to a standard PDMS spectrum. This is because the peak has further split and isn't as sharp and clear due to little substance available for analysis. Nonetheless, the doublet is evidently noticeable meaning PDMS is able to be identified.

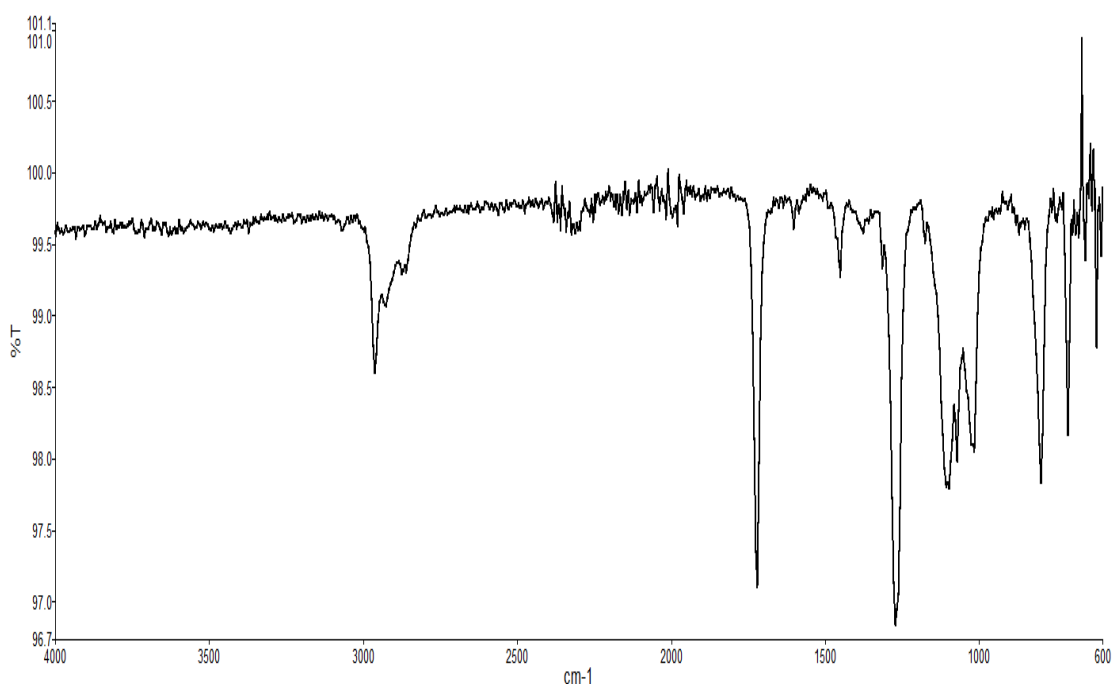


Figure 5.11: An Infra-red Spectrum of a small volume of polydimethylsiloxane.

Alternate siloxane materials possess similar characteristics and are often incorporated for similar roles as PDMS. These substances display alike spectra to PDMS, however variations in the fingerprint region allow the different substances to be differentiated. Cyclotetrasiloxane is a substance integrated into a number of items in the pharmaceutical and cosmetic industry as it possesses features suited as a conditioning agent [76]. It is simple to distinguish between PDMS and

cyclotetrasiloxane because the peak at approximately 1050cm^{-1} is a singlet rather than a doublet, and the peak is broader and more rounded.

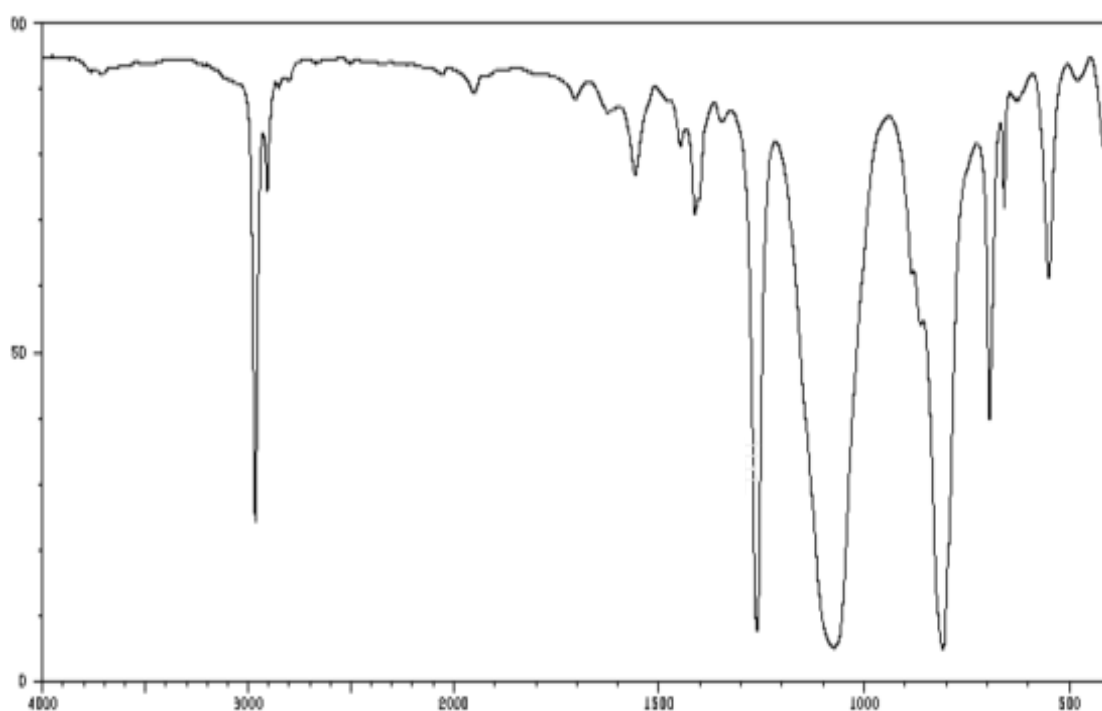


Figure 5.12: An Infra-red Spectrum of Cyclotetrasiloxane [77].

Although a sample of PDMS was used to complete the dilution samples, the original source of this PDMS is from Mates; though its viscosity was unknown. Although the viscosity of lubricants can vary between manufacturers, the average remains at approximately 200 centistokes. However, by contacting Mates, an exact viscosity may be determined. A sample of PDMS obtained from additional condom suppliers including Durex, Pasante and Skyn may have increased the reliability of these results by comparing a broad range of PDMS samples. This would allow for any distinct differences between several brands of condom lubricants to be identified. On the other hand, when the Mates sample of PDMS was compared to Cellmark's internal database, a 99.4% positive match was recovered with a Durex condom. This indicates that the viscosity or origin of the PDMS, displays a very little effect on the outcome of the FTIR spectrum. However, if the viscosity of the PDMS was able to be determined via another technique alongside FTIR, such as Desorption Chemical Ionisation, the chain length would assist in identifying the source when compared to wider known databases.

To ascertain whether a condom was used during an alleged sexual assault, analytical methods, such as GCMS and MALDI, could have been completed to test for the presence of spermicides. Spermicides, especially nonoxynol-9 are common materials incorporated within condom lubricants. These substances operate by destroying sperm cells. The presence of this compound could act as substantial evidence in an alleged sexual assault case if there is no detection of semen or PDMS. For example, spermicides are also applied to condoms lubricated with polyethylene glycol. As the persistence of water-based lubricants is much shorter in comparison to silicone-based lubricants, the identification of a spermicide proposes the perpetrator wore a condom.

5.2 Anti-Contamination

Strict anti-contamination procedures were put into place to minimise the likelihood of any findings being compromised. Prior to beginning any examination, all equipment was thoroughly wiped down with isooctane for extraction procedures or precept for Sperm Elution. Furthermore, all glassware was rinsed with isooctane in triplicate to ensure any contaminants are removed. The glassware washings were collected and analysed once dried down to ensure all glassware was free from PDMS. All workbenches were covered with brown paper, followed by a sheet of sugar paper which was disposed of after each use.

Separate laboratories were used to complete different sections of the project. All swab seeding and Sperm Elution processes were completed in a biology laboratory. The extraction and analysis of samples was completed in an analytical laboratory. The donated sample of PDMS was packaged in multiple layers of protective materials and stored in a separate location away from the laboratories where the project was carried out. The PDMS was stored in a rounded bottom glass vial with a black crew capped lid. This was then wrapped in two layers of bubble wrap which was placed inside a clear resealable bag and into an envelope.

Although all of the equipment was thoroughly cleaned prior to use, glassware was rinsed, and all the Sperm Elution reagents were examined for the presence of PDMS, additional equipment including screw cap vials and Eppendorf tubes were not tested. Swabbing and extracting a selection of each apparatus would minimise the risk of contamination from the equipment if PDMS was detected.

The introduction of a verbal scale could have allowed for an in depth description of the observations to ensure consistency between all samples. For example, the following scale could have been employed to describe the levels of PDMS or contamination detected: extremely low, low, moderately low, moderate, moderately high, high, and extremely high. Furthermore, the presence of any residue in the glass vials should have been noted. The results suggested that the numerous vials containing residue often presented greater volumes of contamination.

5.3 Blank Swab Analysis

The findings obtained from the blank swab analysis showed that there was no PDMS present on any of the blank swabs. This is a beneficial outcome because it is now improbable that apparent sexual assault cases are compromised by the accidental addition of siloxane contaminants on cotton swabs during manufacture. Further to this, no polyethylene glycol was detected on any of the swabs. Though a possible origin of the contamination is now able to be eliminated, additional investigation is needed to allow alternate sources of contamination if alleged false positive results continue to occur.

When gathering the blank cotton SceneSafe swabs, vaginal swab kits with the same batch numbers were attempted to be obtained to keep the sample consistent in all areas possible. This reduces the contamination by additional factors. All swab kits were collected towards the start of the project to allow all kits to be from same batches. However, variation of the batch numbers would have provided a more diverse set of results covering a larger selection of swabs.

Only three sexual assault kits were extracted and analysed for the presence of PDMS. This is not a representative sample size to be able to confidently state that PDMS is not able to be detected on blank cotton swabs in sexual assault kits. Although an encouraging result, a much larger sample size is required to allow a representative impression as to whether PDMS is unintentionally applied to blank cotton swabs. If swab manufacturers were able to provide a guarantee confirming their swabs are free from silicone, the risk of contamination would be drastically reduced allowing scientists to confidently apply the use of swabs for the recovery and analysis of lubricants.

A suggestion for the basis of the PDMS appearing on contaminated swab heads proposes that the lubricating oils applied to grease the machinery during the manufacturing of swabs contain traces of PDMS. The likelihood is that this substance has transferred onto the swabs through either leaking or coming into contact with the swab head during production. However, this is unlikely to be a common occurrence, therefore only a small quantity of swabs will contain this impurity. Contacting swab manufacturers and establishing which lubricants they apply to their machinery could determine whether or not this is a possible explanation for the contamination.

Are SceneSafe the only company producing swabs used for the recovery of evidence from sexual assault victims? Contacting numerous police forces across the United Kingdom would determine if any other kits are used. This would be beneficial as false positives could be arising from other brands, not just SceneSafe.

A portion of blank swabs contained higher levels of contamination compared to others. Testing a range of brands of swabs with a plastic shaft could determine whether there is a direct link between the brand and levels of contamination. Individual differences during manufacture could potentially be the cause and may address the source of contamination. In addition to this, during the extraction of blank swabs, experimental variation including cutting closer to the shaft when removing the swab head could increase the amount of contamination. This is because more adhesive would be present in the sample. The results revealed that

vials containing a greasy residue once dried down provided higher levels of unanticipated substances. This is likely to be traces from the glue which was carried into the extracted sample. It is therefore important that each vial is rotated before analysis to ensure all residues are transferred into the isooctane.

To improve the strength of the outcomes, a broader range of swabs in addition to different brands should have been explored, as well as the type of swab. Tapered tip, ultrafine and micro ultrafine flexible tips are frequently used for urethral and vaginal examinations after alleged sexual assaults. These types of swabs weren't studied for PDMS traces. As a result of the delicate nature of materials in the ultrafine tips, different adhesives may have been applied throughout manufacturing; and so additional analysis into the diverse ranges of swabs could have strengthened the outcomes.

5.4 Lubricating Jelly from Sexual Assault Kits

Findings indicated that both Aquagel and OptiLube are both water-based lubricants, with no traces of PDMS present. This is a crucial finding as the risk of external contamination of PDMS from Sexual Assault Kits is marginal. However, it is currently unknown whether SceneSafe are the only Sexual Assault Kit provider throughout the United Kingdom. If alternate suppliers source these kits, different lubricants may be included which could contain silicone-based compounds. Furthermore, suggests should be made for the manufacturers to state the ingredients on the packaging of the lubricants. If PDMS should be present in these lubricants, this could immensely compromise alleged sexual assault cases.

5.5 PDMS Dilutions

PDMS was detected on all eight of the PDMS dilutions with both extraction only and extraction after Sperm Elution. This shows that volumes less than 0.125µl of PDMS is able to be detected using u-ATR after Sperm Elution even though some PDMS is lost

into the supernatant during the Sperm Elution procedure. Smaller volumes of PDMS should have been applied to the swabs. However, the smallest volume able to be seeded onto a swab using the Gas Chromatography syringe at Cellmark is 0.1 μ l. Should this process be repeated in the future, more precise equipment allowing smaller volumes to be seeded onto the swabs should be obtained. This doesn't affect the validity of previous work because different Gas Chromatography syringes were used in previous research. Alternatively, the polydimethylsiloxane could be diluted and then transferred directly to a swab for the application of volumes less than 0.1 μ l.

Only one sample of semen was used throughout the project. The neat semen was stored in the freezer and defrosted for use when required. A 1:10 dilution was prepared and stored in the refrigerator for up to 15 days and discarded when required. It may be questioned that one sample of semen is not representative. However, semen is only applied to the PDMS dilutions swabs to ensure the Sperm Elution had been completed correctly. The presence of sperm heads in both the epithelial and seminal pellet confirm that the Sperm Elution has been effective and if PDMS is able to be detected following this process it is as a result of the substance persisting on the swab head.

In a real forensic case, the volume of lubricant on a vaginal swab will be unknown to forensic scientists. In the majority of forensic cases, all vaginal swabs from alleged sexual assaults will be sperm eluted, unless it becomes known to officials the identity of the perpetrator and that DNA evidence is not required. This project revealed that although a large portion of PDMS remains on the swab head following Sperm Elution, a percentage of the lubricant is still removed into the supernatant. Therefore, if a vaginal swab is sperm eluted in casework, both the initial and residual volumes of PDMS will remain unknown. Contacting condom manufacturers and establishing the volume of PDMS applied to condoms during production could aid in determining the most appropriate dilutions to consider.

Using the equipment currently available for lubricant analysis at Cellmark, FTIR analysis must remain a semi-quantitative technique. If a 'well' attachment was

incorporated to allow the complete sample to be analysed, full quantification would occur. A 'well' is an attachment whereby the sample is loaded into a groove and the IR light passes through allowing the whole sample to be analysed. Due to cost and time restraints, this was not possible during this project. Attempts were made to produce a quantitative curve, however the results obtained were not consistent enough to allow an accurate curve to be created. A possible explanation for this is that not all PDMS is transferred from the vial to the u-ATR. For example, a large volume of PDMS could be present in the vial. If the isooctane isn't rotated around the vial thoroughly to gather all of the PDMS, only a portion of the sample will be collected for analysis.

Spectra have shown that when 20 μ l and 10 μ l of PDMS is extracted, the peak intensities are very high with a percentage transmission of approximately 5. This peak intensity is almost at a maximum. This shows that 20 μ l is a high volume for lubricants and any PDMS analysed between 20 μ l and 10 μ l is likely to display peaks with similar intensities. If this experiment was to be repeated, smaller dilution intervals are required between 5 μ l and 10 μ l.

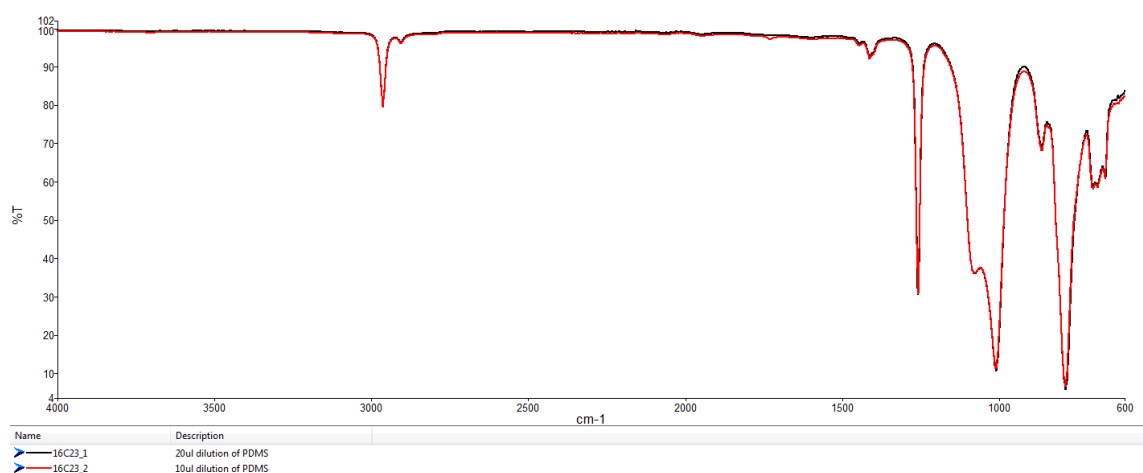


Figure 5.5: A comparison spectrum of 20 μ l and 10 μ l of PDMS.

The results for 0.75 μ l don't follow the trend as with all the other dilutions. This is the case for the dilutions both with and without Sperm Elution. This questions the accuracy of the Gas Chromatography syringe. Perhaps some isooctane was left in the syringe whilst cleaning before use, or human error whilst spotting the extract onto

the u-ATR crystal. On the other hand, this method just may not be able to quantify results. Further repeats are required to confirm these suggestions.

As the intensity of the peaks for the PDMS dilutions after Sperm Elution was slightly less, the epithelial and seminal pellets were analysed to determine whether PDMS was extracted into the supernatant from the swab head. The results revealed that PDMS was detected in both the epithelial and seminal pellet up to approximately the 1µl dilution. However, the clarity of the spectra isn't very sharp with broad peaks being present. Drying down the epithelial and seminal pellets and extracting with isooctane would provide sharper, more established peaks. This is because water-based reagents are incorporated during Sperm Elution, hence the broad O-H peak at approximately 2700-3600cm⁻¹. The spectrum in Figure 5.51 displays the seminal pellet of a 0.5µl dilution of PDMS after Sperm Elution. Although it is known the substance present is PDMS, this would not be accepted as a positive identification in forensic casework as the peaks at 1090cm⁻¹ and 1020cm⁻¹ have not separated. If this sample had been fully dried, the singlet peak at approximately 1110cm⁻¹ would have split into a doublet.

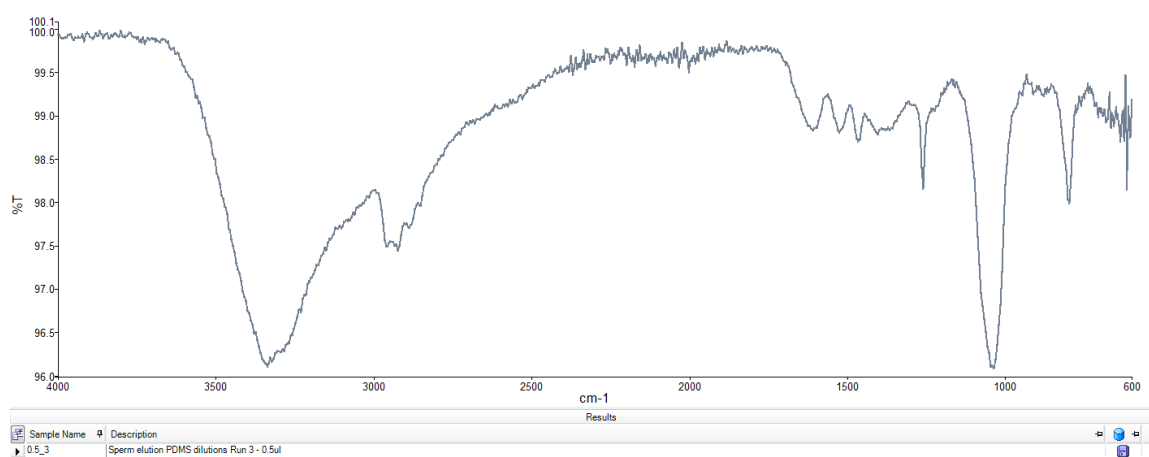


Figure 5.51: A spectrum of the seminal pellet of a 0.5µl dilution after Sperm Elution.

5.6 Body Fluid Matrix

Fortunately, all of the 8 different body fluid combinations tested positive for PDMS following Sperm Elution. 50µl of each saliva, semen and blood was seeded onto the

required swabs. However, the volumes of faecal and vaginal material on each swab remain unknown. This volume is dependent upon the thoroughness of the swabbing by the donor. Furthermore, the volume of faecal material varies greatly dependent on the time since the donor passed faeces and how carefully they wiped clean. Biological factors including the stage of the menstrual cycle affect the volume of vaginal material recovered. Varying quantities of vaginal material is released during different intervals of the cycle. For example, if the donor has swabbed within 5 days of menstruation, a reduced volume of vaginal material will be obtained in comparison to donors who swabbed after this time period.

The spectrum displaying the average lowest peak intensities is the body fluid combinations including faecal material. This could be due to a large quantity of faecal material being present on the swab. PDMS is seeded on to the swab over the faecal material. Perhaps PDMS should have been applied to the swab head prior to swabbing to maintain consistency through the order of the application of body fluids. However, this suggestion was decided against because of hygiene purposes and to prevent any lubricant being removed from the swab head during swabbing.

To enhance the validity of the findings, this project could be repeated by applying the lubricant to the swab in varying orders. For example, applying PDMS before the addition of any body fluids, placing the lubricant after the addition of semen and transferring PDMS once all body fluids had been applied. It could then be determined whether PDMS is still able to persist on the swab head after body fluids have soaked into the cotton tip or whether it is extracted into the supernatant. Alternatively, PDMS could be combined with body fluids. For example, by combining the PDMS with semen and then applying the mixture to the swabs. This is more likely to be expected in forensic casework.

2 μ l of PDMS was the chosen volume to be applied to all swabs in this section of the project excluding controls. This volume was selected as a median which would expect to be detected on the swab head following Sperm Elution. However, it is unknown whether this volume is representative and whether less PDMS would be

expected to be recovered during sexual assault cases. Therefore, a range of smaller volumes of PDMS would be required to increase the accuracy of the findings.

The epithelial and seminal pellets should have been extracted, dried down and analysed to discover if PDMS was extracted into the supernatants or whether the additional body fluids assisted in the persistence on the swab head. The volume of supernatant could have prevented PDMS being detected; hence drying the extract down would be required. As the mixture of PDMS, semen and faecal material displayed the lowest intensity of peaks for PDMS remaining on the swab head, this could indicate a larger proportion of the lubricant was extracted into the supernatant. Completing the analysis of the seminal and epithelial pellet would allow the outcome of this theory.

For volunteers to be able to donate blood for this section of the project, individuals were advised to pin prick their finger and recover blood into a bijoux tube. Volunteers should have initially been directed to refrigerate the sample immediately to prevent clotting. Although a pin prick to the finger may be seen as unethical, the method was deemed the most appropriate source as an alternative to blood being drawn from a vein, as only a small volume was required.

5.7 Body Lotion & Shower Gel Survey & Analysis

A larger sample size of volunteers to complete the body lotion and shower gel survey would have given more significant and representative data. Although a total of 71 responses were received, this is a combination of both males and females, and all within a similar location radius. The survey was advertised on Facebook, however many of the individuals likely to have participated are situated in Lancashire. A question to determine the location of the participants should have been asked. If this survey was to be advertised externally, a wider sample range is more likely to have been obtained. With this in mind, the volume and range of volunteers participating in this survey is improbable to be representative of the United Kingdom.

Following on from this, demographics and specific features are characteristic of the typical survey respondent. Research suggests that females are more likely to participate in surveys than males [78]. This correlates with the study as 91% of volunteers who completed the body lotion and shower gel survey were female. Approximately 1 in 20 women are raped in their lifetime compared to 1 in 71 men. Therefore, there are more examinations of vaginal swabs than penile swabs. It is therefore more important to determine the most common body lotions used by women in the UK, as these are likely to provide false positive results for condom lubricants. On the other hand, more males are regularly applying body lotions due to media engagement. However, reports suggest that the only varying features between body lotions marketed for males is the fragrance. As a result, many men resort to using female body lotions due to easier access.

Numerous body lotions were found to contain PDMS. This raises the question as to whether body lotions containing PDMS have an effect on the identification of silicone-based lubricants and whether the two different sources of PDMS are able to be distinguished between. Is it feasible to propose that a body lotion comprising of PDMS is able to contaminate the gusset of a pair of knickers which are subsequently extracted and examined during an examination for lubricants? If so, this would enable the PDMS in the body lotion to deliver a false positive identification for the presence of condom lubricants. To prevent this issue arising, analysis of materials using additional analytical techniques could allow the viscosity of each sample of PDMS to be determined and allow different samples of PDMS to be differentiated between. As previously mentioned, PDMS incorporated within the cosmetics industry possess smaller chain lengths in comparison to PDMS used as a condom lubricant [44]. As a result, the two different forms of PDMS would be able to be differentiated and lower viscosities could be discounted in interpretation.

From the selection obtained, a proportion of body lotions listed PDMS in their ingredients. However, during the analysis of these products, the substance was not able to be positively detected as the peaks weren't clear enough on the Infra-red spectrum to confidently state PDMS was present. This indicates that the lubricating

material could be present in very small quantities, or many other similar substances are present in much higher volumes which have masked the exposure of PDMS.

Following on from the extraction of swabs containing shower gels and body lotions, the glass vials containing more residues once the extracts had dried down displayed higher levels of PDMS during the examination. This is a likely outcome because PDMS is an oil often applied as a lubricant, hence the reason a greasy residue is left behind and doesn't evaporate. However, this was not always visualised in samples containing low levels of the contaminant. This presents an explanation for the absence of PDMS in shower gels. The role of shower gels is to clean the skin by removing grease without leaving a layer of mineral residue on the skin. If PDMS was incorporated into these articles, the skin would be nourished but an oleaginous film is likely to remain on the surface of the skin.

In addition to examining a range of body lotions and shower gels for the presence of PDMS, sanitary wear should also be considered as a contaminant. As PDMS is often applied as a fabric conditioner, the substance could be applied to sanitary wear to provide comfort to the user. If present, this could transfer to the gusset of the knickers causing contamination in sexual assault cases. Furthermore, primary transfer between sanitary wear and the user via contact could occur. It is therefore crucial to examine items of sanitary wear to identify or rule out this concept as a source of contamination.

Although a large sample of body lotions and shower gels were obtained for analysis, the diversity of the samples wasn't fully representative. A broad range of materials located on the high street and in supermarkets were collected from the low to middle price range. However, very few upmarket examples were acquired. Due to cost and availability of tester samples, this was not a possible action. However, the most common lotions and gels used by participants in the survey were high street items available in most department and superstores. Also, recovering samples from tester pots in department stores increases the risk of introducing contaminants. However, this is only a very slight risk and the volume of both primary and secondary

transfer from securing samples via this method is unknown, especially for items with a screw cap lid as an alternative to a dispensing pump.

As PDMS was unable to be detected in any of the shower gels obtained, further research into analysing a greater volume of body lotions is required. Not only acquiring a broader range of brands, but also varying the types of lotions produced by the same manufacturer. For example, a 'deeply nourishing' or 'silky smooth' lotion is more likely to contain silicone-based materials compared to standard body lotions. Following on from this, could shower creams, used as a substitute to shower gels contain PDMS? Shower creams often contain oil based thickening agents used to hydrate, smooth and enrich the skin in addition to cleansing. Therefore the likelihood of manufacturers integrating PDMS into their shower creams is probable. This recommends the analysis of shower creams to account for whether this assumption is justified.

To allow these propositions to be answered, additional research is necessary. Yet, from the data achieved in this study, it is evident that if PDMS is detected in cases of sexual assault, it cannot automatically be assumed that the source of the substance is as a result of the use of a condom. Attentive examinations and case scenarios must be taken into account to accurately identify the initial source.

5.8 Fast Differential & Direct Semen Extraction

The forensic laboratories at Cellmark, Chorley, do not possess equipment for DNA analysis. Therefore, if any further investigation is required following a standard DNA extraction for identification, all exhibits are required to be transferred to Cellmark, Abingdon DNA unit. As a result, the environment in which the procedure for Fast Differential and Direct Semen Extraction is completed is unknown. Also, the examination is completed by a DNA analyst in Abingdon, not by myself. This means the exact details regarding the environmental factors and procedure is confidential and remains unreported. However, Cellmark is a highly reputable and accredited company recognised for producing valid results of high quality; therefore the results

from any samples sent for Fast Differential and Direct Semen Extraction are trusted to be reliable.

As mentioned in the methods section, the procedures for Fast Differential and Direct Semen Extraction display similar features to Sperm Elution. However, an extra step involving the addition of Dithiothreitol (DTT) is present. A sample of this substance should have been requested and analysed to allow it to be excluded as a contaminant. Although, DTT does not contain any silicone within its structure and, therefore, is unlikely to contaminate any of the findings.

Observations made during the isooctane extraction revealed that the sample remains which had undertaken Direct Semen Extraction produced a frothy substance whilst the isooctane was being drawn through the pipette. This could be because not all of the material had been extracted from the swab head during Direct Semen Extraction, or the isooctane is reacting with the DTT. Strangely, frothing only occurred on swab heads which were sent for Direct Semen Extraction, not Fast Differential. This is an unusual result due to the similarities between the stages and reagents applied in each of the two processes.

5.9 Condom Survey

Although a varied range of branded condoms were obtained for this study, a larger variety of condoms produced by the same manufacturer are required. For example, Durex are currently the largest manufacturer of condoms in the UK, however only 3 of their broad selection were analysed. There are currently 13 different condom types on the official Durex website, however, the variety varies greatly over time depending upon popularity and marketing in the media. 3 condoms is not representative to be able to conclude that PDMS is the only lubricant applied to condoms manufactured by Durex.

It could be suggested that although PDMS is the lubricant readily applied to the majority of condoms, condoms which possess specific characteristics i.e. warming, cooling or enhanced pleasure, could require the use of alternate lubricants to allow

these characteristics to be sensed. Amor hot gives the user and their partner a warming sensation which could be a reason as to why a water-based lubricant was incorporated. Testing a range of warming condoms could determine whether this suggestion is accurate.

This section of research would benefit if the volume of lubricant applied to condoms was known. Condoms manufacturers should have been approached to determine a volume. This would have allowed a more representative quantity of lubricant to be used throughout the study. Furthermore, requesting a sample of lubricants from multiple suppliers would allow variation between lubricants if additional analytical techniques were incorporated. This would enhance the findings because if PDMS was to be detected on intimate swabs following an alleged sexual assault case, being able to identify the source of the lubricant directly to a specific condom would massively advance the confidence of lubricant analysis in forensic casework. However, most silicone-based condom lubricants are expected to display very similar features, with very little variation in the infra-red spectrum. Therefore, the likelihood of determining a specific condom is minimal.

Most supermarkets and department stores hire external manufacturers to produce their supplies as an alternative to minimise their costs. However, on the packaging of own branded condoms, it rarely informs users of the source of manufacture. If this was known, comparisons between the lubricants of different branded condoms produced by the same manufacturer could be made to determine whether the same substances are incorporated.

5.10 Fabrics

All of the undergarments purchased for use in this study have been detected to contain PDMS within their fabric. The most plausible reason for this is that the silicone-based lubricant is incorporated into the conditioners employed for fabric finishes on newly produced garments.

A larger volume of control areas should have been taken from different sections of the knickers. This is because it is suspected that different positions contain varying levels of PDMS contamination. Furthermore, research revealed that there is no correlation between the size of the knickers and the volume of PDMS detected. However, accuracy was not maintained through cutting the control areas. Approximately 1cm² portions were utilised as controls, but this may vary by a couple of millimetres as no ink markings were drawn on the garments as this could contaminate the material. Following on from this, the control areas were all taken from just below the waistband. If controls were taken from the gusset also, it could then be concluded whether the volume of PDMS present is consistent throughout the whole garment.

All of the knickers used in this study were of the same brand, style and colour. The only variation was the size of the undergarment. If extra time was available, obtaining undergarments of a diverse range of style, material, brand and colour would have benefitted this research. This would have allowed comparisons to determine whether different manufacturers apply different fabric finishes to their garments and whether this is dependent upon the style and colour. Furthermore, varying fabric finishes containing PDMS could be present in different volumes and display varying persistence times after laundering.

Polydimethylsiloxane is a substance which is tremendously insoluble in water [79]. This insinuates that the material is not easily detached throughout washing. Consequently, the outcomes of laundering items contaminated with PDMS must be considered. It would be predicted if PDMS is present on apparel, the whole stain would not be thoroughly eliminated after cleansing but, alternatively, gradually reduce in volume in correlation to the amount of launders implemented.

In forensic cases, the condition and age of an undergarment needs to be taken into account. If a pair of knickers appears to be well worn, the likelihood of detecting PDMS as a contaminant is very low. However, if the underwear seems to be in 'new' condition, caution must be taken in addition to control areas. Although the substance is highly unlikely to transfer from fabric to user and lead to a false positive

result on outer vaginal swabs, a false positive could arise in the gusset and mislead examiners into perceiving the PDMS is present as a result of vaginal drainage.

Although many fabric finishes are known to contain silicone-based ingredients, it is currently unknown whether these are present in a large proportion of fabric conditioners and detergents. To minimise the likelihood of this affecting the outcome of any findings, no fabric conditioners were used during this study. The ingredients on the outer packaging of the detergent implied that no silicone based products were combined within the detergent. However, to confirm this statement, a sample of the detergent should have been examined using FTIR. In order to enhance the outcomes of this research, a range of brands of fabric conditioners and detergents should have been gathered and tested for the presence of PDMS. Furthermore, determining whether all detergents are capable of removing the PDMS from the fabric finishes on garments. Results revealed that washing with water alone was unable to extract the substance from the fabric; therefore this could suggest that mild detergents may not completely remove the PDMS. It is also currently unknown whether the temperature of the wash affects the removal of PDMS. It can be assumed that the greater the temperature, the more effective the removal of the fabric finish. However, the temperature could affect the effectiveness of the detergent. As a result, completing further analysis into the research involving the use of alternative detergents and varying temperatures will provide a more precise outcome.

Following on from this, it is unknown whether all the detergent is removed from the washing machine after each wash. During the washing study, the knickers which were washed with just water still released a mild fragrance of the detergent. This indicates that a proportion of the detergent remains in the system and is transferred to the proceeding wash. This also supports the suggestion that weak detergents may be unable to remove PDMS from fabrics as all of the knickers laundered with water only displayed positive for PDMS.

Reversing the project could discover whether PDMS is able to be transferred onto garments during laundering, as opposed to being removed from the fabric. If

underwear known to be contaminant free from PDMS was washed with varying combinations of detergents and conditioners known to contain silicone based compounds, it could be determined whether the PDMS deposits onto the fabric during washing. If so, this could produce false positive results in alleged sexual assault cases.

5.11 Persistence

The results from the persistence area of the project indicate that PDMS is unable to be detected on vaginal swabs for times greater than 12 hours following Sperm Elution. However, for all but one pair of knickers worn during the study, PDMS was able to be detected after Sperm Elution. Unfortunately, there were only a small number of volunteers participating in this study, therefore these results aren't representative. A reason for the reduced sample size is that only members of staff at Cellmark were able to volunteer as their DNA is currently stored on an internal database, therefore wouldn't interfere with forensic casework. If external candidates were able to volunteer, it would be a lengthy process to approve their DNA to the database and send all the donation kits and they would be unable to return their swabs and undergarments directly to the freezer in sample reception at Cellmark, Chorley. Unfortunately, no males volunteered to participate in this research. As a result, the persistence of PDMS on penile swabs is unable to be determined. Should there have been more time available within the project timeline, recruiting external participants may have been a possibility. If incentives were provided by Cellmark to encourage staff to volunteer, the likelihood of an increased sample size would have been more probable but may encourage coercion of partners to take part. The solo sex option should, however, eliminate this possibility.

As a result of time restraints and personal commitments, not all volunteers were able to complete all time intervals for the persistence study. However, the only intervals missed were 18, 24 and 36 hours. This is unlikely to affect the outcome of the findings as no PDMS was detected after 12 hours from any other participants. On

the other hand, as only 2 participants fully completed the study, the results aren't valid and no statistical analysis is able to be completed.

To allow individuals without a partner to participate in this study, the option of solo sex was available. However, there were no volunteers who opted for this alternative. Perhaps if all equipment was provided to female volunteers to allow solo sex to be performed, this could have attracted a greater number of volunteers.

The thoroughness of the swabbing from each participant is a factor which is unable to be controlled. Despite an approximate depth indicated to participants, some volunteers will be more thorough at swabbing than others; therefore not all PDMS may be recovered onto the swabs. No PDMS was detected on a 0 hour swab from one volunteer, however was detected on the knickers worn following the swabbing. This is likely to be an anomalous result and the PDMS could have been transferred from the swab to the supernatant during the Sperm Elution procedure. However this indicates the substance is only present in very minimal amounts prior to Sperm Elution and extraction. In addition to this, the assumption of participants swabbing at the precise time interval may not always be accurate. If an individual is driving or working for example, there may be variation between the times the swabs were taken.

On the completion sheet, Appendix 8, participants were asked whether they had showered in between intercourse and taking swabs. This is unlikely to affect the persistence of PDMS in female participants; however silicone-based materials can be present in shower gels and body lotions which could cause false positive results for PDMS on the intimate swabs. Participants are also asked if any moisturisers are used, and if so, a sample can be retrieved and tested for PDMS. No participants used any form of moisturiser for the duration of this study.

The type of clothing participants wear during this study needs to be considered. Fortunately at Cellmark, most members of staff wear scrubs which is a loose form of clothing and isn't likely to cause friction and rub away any PDMS. Furthermore, all participants opted to wear the undergarments which also aren't a tight fit. Volunteers are asked their stage in the menstrual cycle. This is because the volume

of vaginal discharge varies at different stages in the cycle. If higher volumes of vaginal discharge are occurring, there will be a greater loss of PDMS from the vagina.

It needs to be considered whether participants completed each time interval on a weekday or at weekend. As all participants were employed by Cellmark, this isn't an active job; therefore the likelihood of PDMS being lost due to activity is very low. On the other hand, if intercourse and swabbing occurred at the weekend, activity levels vary and there is a greater chance of PDMS being removed as a result of increased activity. On the completion sheets returned by participants, no strenuous or out of the ordinary activities were recorded. Participants should have also been asked whether they went swimming during the study. The presence of chlorine in the pools could aid in the removal of PDMS. Unless this research is completed in a controlled environment, it is not possible to regulate all external factors. However, this isn't a realistic scenario and wouldn't be expected from victims in an alleged sexual assault.

The time of intercourse is a factor which may affect the persistence of PDMS, especially in the smaller time intervals. If intercourse occurred late in the evening, PDMS is unlikely to pass through in vaginal drainage if the volunteer is going to sleep shortly afterwards. If intercourse took place in the morning, the participant is likely to be more active, meaning PDMS is more probable to pass through to the knickers via vaginal drainage.

It is highly likely that PDMS is transferred onto the knicker gusset by a combination of both contact and drainage. This is because PDMS was detected on all knickers at the 0 hour time interval, which indicates all PDMS present is likely to have passed through contact because PDMS from the vagina will have been recovered via swabbing initially. On the other hand, it is highly probable that PDMS present on knickers worn for 12 hours is a result of vaginal drainage. If participants were asked to wear the undergarments for a longer period of time, this could increase the recovery of PDMS. Conversely, there is a longer period of time whereby the lubricant could be rubbed away. Extending the length of time participants wore the undergarment is not possible because this is unhygienic and unethical.

The whole gusset from the knickers should now be sperm eluted in further research, not just half. This is because we now know that PDMS is likely to be present on undergarments following intercourse with a silicone-based lubricated condom and this would simulate a real case as we now know that PDMS is still able to be detected following the Sperm Elution procedure in volumes as low as 0.125µl, if not lower. Sperm eluting the whole gusset is essential because PDMS and/or semen may only deposit on a certain area of gusset depending on how the knickers are worn. DNA is stronger evidence in many cases, especially when the offender is unknown; therefore Sperm Elution is often conducted if case circumstances reveal it is unknown whether a condom was worn.

If this project was to be repeated, the knickers should be washed until there is no detection of PDMS prior to distributing to volunteers as traces of PDMS could pass onto the participant. It could be suggested that PDMS may have been present in the gusset initially, hence will definitely be detected during the study. However, when compared to controls, the volume of PDMS detected was present in much larger quantities. One pair of knickers was found to be negative for PDMS 0 hours after swabbing following Sperm Elution. This could be because the PDMS present was swabbed away, therefore not present for vaginal drainage. This means that not all PDMS is passed through contact with the knickers. Also, this indicates that PDMS may not be present in the gusset of the knickers from the manufacturing process, or the PDMS could have come into contact with or drained onto the other half of the gusset which was extracted with isooctane only.

5.12 External Sources of PDMS

It is critical that the context for each potential sexual assault case is taken into account prior to any conclusions being created from the results of the examined evidence. For example, on what occasion did the victim last undertake any sexually related activity involving the use of a condom or lubricant previous to the supposed assault? If this was in the region of 3 days earlier, any detected PDMS could have

remained in the vagina since the occasion with approval. Consequently, the recovered PDMS would act as strong evidence towards the defence.

Examiners and investigators exploring a crime scene frequently wear gloves comprising of latex. This is to protect both themselves and the scene from contamination. It is vital that it is determined whether the latex gloves worn in cases involving alleged sexual assaults are non-lubricated. If the gloves are lubricated, this could contaminate the recovered vaginal swabs and bed sheets if these were also recovered for further examination. However, if the viscosity of the PDMS is determined, the original source of the PDMS could be identified. In addition to this, some examiners are more in-depth than others when collecting evidence using swabs. This denotes that not all traces of PDMS may be recovered from a victim if the examiner isn't thorough.

Numerous environmental features have the potential to hold traces of PDMS which may provide an account as to why particular incorrect results have emerged. For example, if a sexual assault happened in a hotel room, several elements could contribute to the false positive recovery of PDMS. As previously mentioned, PDMS is often used as a fabric finish on many materials in addition to garments. An example of this is on bedsheets supplied to hotels. These conditioners aid the material to remain soft despite frequent use. Consequently, if a bedsheet is recovered from a hotel for analysis following an alleged sexual assault, it is significant that various regions of the exhibit are examined as control samples. This is to confirm any detected PDMS is from a condom and isn't from the fabric finish applied during production. Despite the use of a condom, semen may still be identified on a bedsheet following intercourse. Both semen stains and PDMS aren't always extracted from fabrics when washed with detergents. As a result, if PDMS and/or semen are recovered, they may have been deposited onto the fabric on a previous occasion. Specifically on hotel bedding, numerous individuals could have performed sexual activity, meaning DNA evidence would be required to confirm any evidence recovered was from a particular alleged offence.

To eradicate these diverse factors as the supplier of PDMS in alleged sexual assault cases, the chain length of the PDMS could be determined. For example, PDMS applied as a condom lubricant often displays a viscosity of roughly 200 centistokes. However, items in the cosmetic industry feature viscosities approximately in the region of 5 centistokes [44]. To conclude, a PDMS viscosity not within the range of 100 and 300 centistokes suggests that PDMS is extremely likely to have originated from a different source, not a condom lubricant.

5.13 Analytical Techniques & Equipment

The internal FTIR library at Cellmark does not feature a broad range of substances. Comparison with a superior, well recognised database containing a larger volume FTIR spectra with a variety of known materials readily found in forensic casework would have increased the accuracy of any findings. A match of or greater than 99.5% is required to confidently confirm an accurate match between a known and an unknown. However, despite comparing a known sample of PDMS to Cellmarks internal database, a match of only 93% was established with a durex condom.

When analysing samples using the FTIR microscope, three spectra were produced from three different areas of each sample. This allowed for any anomalous results to be identified. Furthermore, thick oil based substances, principally PDMS, often migrate towards the outer edge of the droplet; therefore it is essential to analyse various areas to increase the likelihood of detecting any lubricant which may be present. It is possible to complete more than one run of each sample on the u-ATR also, however this is not readily completed in casework and would increase the time and cost dedicated towards a case. In addition to this, a larger volume of the sample would be consumed which may later be required for further investigation. When spotting a sample onto the u-ATR, the substance is less likely to spread around the crystal as far in comparison to a sample being spotted onto a glass window. However, if the sample is not directly placed onto the crystal and spreads, the substance won't be thoroughly analysed and could prevent accurate identification. The surface of the u-ATR crystal was roughly 1.5mm in diameter, this caused

difficulty when applying extracts and keeping samples retained within a limited surface area.

If too much of a liquid-based sample was spotted onto the glass, the sample would spread leaving no distinct outline visible of the spot. As stated above, if PDMS is present within a material, the compound usually gathers to the outer edge of the spot, therefore it is crucial the boundary can be distinguished using the FTIR microscope to prevent any PDMS from being undetected. However, if there are minute traces of PDMS or any contaminants within the sample, the spot evaporates rapidly on the glass window leaving a trace sample which is extremely difficult to visualise using the microscope. As a result, more spotting is required; however this increases the likelihood of the sample spreading across the microscope window. In comparison to the FTIR microscope there is potentially less chance of human error when using the u-ATR, as samples are directly applied to the surface of the crystal and are then scanned.

To prevent any accidental contamination from the external factors, the same bottle of isooctane was used throughout the entire research. However, the results may have been more accurate if the quantity of the isooctane was measured more accurately, especially for the transfer of the sample for analysis under the FTIR. Approximately two drops of isooctane using a teat pipette was added to the glass vial in order to wet the sample before being spotted on to the BaF₂ window. To ensure each sample was diluted equally, this should have been accurately measured using a micropipette. In addition to this, the number of spots added to the BaF₂ window should have been monitored with the same amount being added for each sample. Each sample was aimed to be spotted onto the BaF₂ window five times, however if a small volume of material was present within the sample, these spots evaporated quickly and required further spotting. Too much isooctane was spotted onto the slide with some samples, which resulted in the solution spreading to the outer edge of the window. This made the identification of the substances difficult as contamination usually gathers towards the circumference of the droplets.

To increase the accuracy of any findings, the use of a 'well' attachment would be a more appropriate method to allow quantitative results. There are many factors, including the skill of the examiner, which prevent the FTIR microscope and the u-ATR from being quantitative techniques. Examples include, the accuracy of the spotting onto the BaF₂ window, the accuracy of spotting onto the u-ATR crystal, the volume of isooctane added to the vial to wet the extract, the thoroughness of drawing through the solvent from the swab head/fabric, whether too much/not enough solvent was used for the extracting from material, and PDMS could have been present in the glass vial, however was not collected by the pipette tip. A 'well' would allow the whole sample to be placed within the attachment and analysed. Furthermore, the Infra-red light source would travel through the whole sample allowing the overall volume to be accounted for.

Due to the nature of the techniques used for this project there will always be some variation in the spectra produced. Regarding peak intensities, the spectra from both the u-ATR and the microscope are not able to be reproduced.

Results from the PDMS dilutions section questions the accuracy of the gas chromatography syringe. The 0.75µl dilution displays spectra with peak intensities much greater than expected and doesn't fit with the projected trend. Although the method used is not a quantitative technique, the GC syringe may be a contributory factor for the fluctuating results. By using a micropipette to transfer a sample and diluting could ensure the correct volume is being applied to the swab.

Glass Refractive Index Measurement (GRIM) is a process frequently carried out at forensic laboratories by examiners for the analysis of glass fragments. To allow this technique to function correctly, silicone oil is required to be heated to different temperatures to allow investigators to determine the refractive index value of a sample of glass [80]. Though laboratories are thoroughly cleaned weekly to minimise the possibility of contamination, there is always a chance that the silicone oil could pass through to other laboratories as extensive personal protective equipment is not required in all glass workrooms.

A limitation of using FTIR to produce accurate spectra is the presence of multiple substances in one sample. Mixtures of materials often produce spectra with excessive peaks which can be challenging to interpret. Assigning individual peaks to characteristic materials becomes testing as it is probable for minor PDMS peaks to be disguised by more intense peaks from substances with similar features. Moreover, if a sample isn't dried down, the presence of water in a sample produces a broad peak at approximately 3200cm^{-1} [68]. This was an issue with shower gel samples as water peaks masked the presence other materials in the sample.

Overall, FTIR was a successful technique for detecting PDMS after completing a range of different processes. However, if additional techniques such as GCMS and NMR had been used alongside FTIR, more accurate results would have been obtained. Furthermore, the chain length could have been identified via these techniques allowing the source of PDMS to be considered, and the viscosity could be compared to those from condom lubricants.

Chapter 6: Conclusions

Firstly, the analysis of blank swabs revealed that no PDMS was able to be detected on swab heads following an isooctane extraction. Previously, where false positives had been encountered, this was only in a small proportion of the swabs. This suggests that it is highly unlikely that PDMS is unintentionally transferred onto blank cotton swabs during manufacture.

When the lubricating jellies obtained from sexual assault kits were analysed, PDMS was found not to be present within either of the two lubricants. Both OptiLube and Aquagel are water-based lubricants containing no silicone-based materials. This means no PDMS contamination occurs during the recovery of intimate swabs as a result of the lubricating jellies.

PDMS was able to be detected on swab heads at trace samples as low as 0.125µl after an isooctane extraction and both Sperm Elution and an isooctane extraction. This suggests that if both lubricants and semen could be present on a swab head, Sperm Elution could be initially completed to recover DNA evidence, followed by lubricant analysis secondly. A correlation was observed between the volume of PDMS seeded onto swabs and the peak intensity of the spectra recorded. However, the method of analysis using an FTIR microscope and u-ATR is not a fully quantitative technique due to many factors, including that the whole extract is unable to be analysed. As a result, the volume of PDMS recovered from an intimate swab will remain unknown. An estimate may be produced, but the accuracy would be questionable.

From the eight different body fluid combinations applied to the swabs alongside 2µl of PDMS, it was discovered that PDMS was able to be detected on all of the swabs following Sperm Elution. This is an advantageous result as in many sexual assault cases; multiple body fluids are frequently present. However, only one volume of PDMS was used during the body fluid matrix, and the volume of lubricant expected on intimate swabs following intercourse with a condom is unknown. To increase the validity of these findings, the same method should be repeated using varying volumes of PDMS until no longer detected.

A broad selection of shower gels and body lotions were collected and examined to determine whether PDMS is incorporated into these products. The results showed that no PDMS was present in any of the shower gel samples. This is a positive finding because if a sexual assault victim had showered since the alleged attack, they are unlikely to introduce PDMS via washing. However, approximately half of the body lotion samples tested positive for PDMS. It is therefore crucial that victims of sexual assault are thoroughly questioned regarding which cosmetic products they have applied in the hours prior to and after the incident.

Nine blank cotton swabs were seeded with PDMS and semen. All swabs were sperm eluted, and six swabs were sent for further analysis. Three swabs underwent Direct Semen Extraction and three undertook Fast Differential. Findings displayed that following all three extraction methods, PDMS was still able to be detected on all nine swabs. This is a positive finding because if no sperm heads are detected in the epithelial and seminal pellet after Sperm Elution, the sample remains can be sent for further extraction. If no DNA evidence is recovered, lubricant residues are likely to remain on the swab head, if present initially.

PDMS is frequently applied to undergarments as a fabric finish. If underwear is provided to examiners from an alleged sexual assault victim, control areas must be examined to confidently state that if PDMS is to be detected, it has not originated from a fabric conditioner. This project revealed that when new, shop bought knickers were laundered with water alone, PDMS persisted on the fabric. However, when washed with the addition of detergent, PDMS was gradually extracted from the material and was unable to be detected on all undergarments after approximately four washes.

Previous research regarding the persistence of lubricants is limited, yet it is an important piece of information when conducting an examination in a sexual assault case. This study involved volunteers undertaking sexual intercourse with their partner using a lubricated condom. Results showed that PDMS was detected on blind vaginal swabs up to 6 hours, which is a reduced time frame compared to the 24 hours which forensic experts generally advise. All participants opted to wear the

provided undergarments. PDMS was recovered in the gusset of all but one pair of knickers worn during this project. This suggests that PDMS may be transferred onto the knickers via a combination of contact and drainage.

In conclusion, this research has uncovered multiple findings to aid forensic examiners during the analysis of lubricants in sexual assault cases.

Acknowledgements

I would firstly like to thank Cellmark Forensic Services for providing me with the opportunity to conduct this research. Their support by supplying all the necessary equipment and materials for this research has made the progression of this project achievable.

In particular I would like to thank Caroline Eames and Paul Ryder for proposing the study of polydimethylsiloxane and for their continuous guidance during the project. I would like to express appreciation to Nic Alexander and Charlotte McCarthy for training me to complete Sperm Elution and operate the FTIR.

I wish to thank my academic supervisor Isobel Colclough for all the help, time and assistance she has offered me over the past year.

Finally, I would like to thank all the staff at Cellmark Forensic Services who kindly volunteered to take part in my project and donated body fluids. Without this participation, this project would not have been possible.

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Appendices

Appendix 1: Research Programme Approval Confirmation

Date: 4th May 2017

Sarah Townsend
(G20587891)
Email: SETownsend@uclan.ac.uk



Research Student Registry
University of Central Lancashire
Preston PR1 2HE
United Kingdom
Telephone 01772 895085
Fax 01772 892930
www.uclan.ac.uk

Dear Sarah,

RESEARCH PROGRAMME APPROVAL FOR THE AWARD OF RESEARCH DEGREE OF THE UNIVERSITY OF CENTRAL LANCASHIRE

I am pleased to inform you that the School of Forensic and Investigative Sciences has approved your application for Research Programme Approval on a FULL time basis for the degree of MSc (by Research)

Title of Programme of Research

A study to investigate the persistence of Polydimethylsiloxane and determine the validity of sperm elution on intimate swabs using a body fluid matrix.

Collaborator(s)

Cellmark Forensic Services

Supervisors

Director of Studies: Isobel Colclough
School of FAS

Second Supervisor 1: Dr Jennifer Readman
School of PSC

Programme Start Date and Duration

The expected programme length is 12 months (full-time) with effect from 1st January 2017, subject to conditions specified in the University Regulations.

The expected date for submission of your final thesis is 31st December 2017.

Ethical Approval of your Project

Your application for RPA has been approved. However, please note that until you have gained ethical clearance (where you answer "No" to all questions on the Ethics checklist and clearance is confirmed by the ethics committee) or ethical approval (where you answer "Yes" to any question on the Ethics checklist and submit an application for full ethical approval which is subsequently approved by the ethics committee) you are not permitted to do any data collection or fieldwork, or participant surveys. To do so will mean you are uninsured, in breach of the Code of Conduct for Research, and liable for disciplinary action.

Examination Arrangements

- a) The arrangements for examining you on your programme of work.
- b) The external and internal examiners to be appointed.



INVESTOR IN PEOPLE

P.T.O

These arrangements should be submitted no later than 4 months before you propose to submit your thesis for examination. Please note that you will not be able to submit your thesis until examination arrangements have been approved.

Please feel free to contact me about any aspect of your research programme or with any other queries you may have.

Yours sincerely

pp. 

Clare Altham
Senior Administrative Officer (Research)
Research Student Registry
Harris Building room HB104

Copies: Isobel Colclough
Jennifer Readman
Jai Singh

Appendix 2: Body Fluid Matrix Ethical Approval



10 May 2017

Isobel Colclough / Sarah Elizabeth Townsend
School of Forensic & Applied Sciences
University of Central Lancashire

Dear Isobel / Sarah

Re: STEMH Ethics Committee Application
Unique Reference Number: STEMH 636

The STEMH ethics committee has granted approval of your proposal application 'Investigation into the validation of sperm elution on intimate swabs used in sexual assault cases using a body fluid matrix'. Approval is granted up to the end of project date*.

It is your responsibility to ensure that

- the project is carried out in line with the information provided in the forms you have submitted
- you regularly re-consider the ethical issues that may be raised in generating and analysing your data
- any proposed amendments/changes to the project are raised with, and approved, by Committee
- you notify roffice@uclan.ac.uk if the end date changes or the project does not start
- serious adverse events that occur from the project are reported to Committee
- a closure report is submitted to complete the ethics governance procedures (Existing paperwork can be used for this purposes e.g. funder's end of grant report; abstract for student award or NRES final report. If none of these are available use [e-Ethics Closure Report Proforma](#)).

Yours sincerely

A handwritten signature in blue ink that reads 'William Goodwin'.

William H Goodwin
Chair
STEMH Ethics Committee

* for research degree students this will be the final lapse date

NB - Ethical approval is contingent on any health and safety checklists having been completed, and necessary approvals as a result of gained.

Appendix 3: Body Fluid Matrix Participation Information Sheet



Participant Information Sheet

Name of department: School of Forensic & Applied Sciences, University of Central Lancashire

Title of the study: Validation of Sperm Elution on intimate swabs using a body fluid matrix.

Introduction

Name: Sarah Elizabeth Townsend

Status: Postgraduate Research Student

Department: School of Forensic & Applied Sciences, University of Central Lancashire

E-mail: setownsend@uclan.ac.uk

What is the purpose of this investigation?

Polydimethylsiloxane is a silicone based compound which is applied to the majority of condoms in the UK to act as a lubricant. If a condom is used during a sexual assault, the likelihood of obtaining semen from an intimate swab is very low; therefore a forensic scientist may consider undertaking lubricant analysis. The purpose of the research is to determine whether the presence of other bodily fluids, in addition to semen, including blood, saliva and vaginal cells affect the detection of polydimethylsiloxane (PDMS) on intimate swabs following the sperm elution process. Furthermore, to identify whether the presence of extra fluids affects whether the PDMS remains on the swab head throughout the process or whether it is extracted into the supernatant. This would be an extremely beneficial technique to have validated for use in forensic casework as scientists are often faced with scenarios where multiple body fluids are present on each intimate swab. For example in sexual offence investigations, especially rape, bleeding may have occurred during the assault.

What will you do in the project?

This project aims to demonstrate the effect of multiple body fluids on the detection of PDMS on intimate swabs following the sperm elution procedure. Participation in the project would require a donation of a range of biological samples: semen, saliva, blood, faecal material and vaginal cells (semen free). The project will take place at Cellmark Forensic Services Laboratories in Chorley and will last up to 10 months, ending in October 2017.

Do you have to take part?

No, the decision to take part in this research is voluntary. You can refuse to take part without providing a reason.

Why have you been invited to take part?

Any adult, male or female, over the age of 18 who is a member of staff at Cellmark are able to donate samples.

How to donate

If you are willing to donate any of the required samples, please complete the consent form which is supplied in the sample boxes. Semen and saliva donations will be obtained by producing the sample into a bijoux tube. Faecal material and vaginal material will be obtained by swabbing either the anus or vagina. Blood will require a prick to the finger with the sample being placed into a bijoux tube. An example of the consent form is attached to this email. Upon completion of your donation please seal the form in the envelope provided with the sample boxes which states the unique reference number. Please ensure all donated samples provided are labelled with the unique reference number and placed in the bottom drawer of the transfer freezer in the receipting area of sample reception. Once samples have been processed you are unable to withdraw from the research. All donated samples will remain anonymous. All samples will be either processed or destroyed at completion of the project.

If you are donating vaginal material, please ensure that these are semen free with no internal vaginal ejaculation for at least 7 days prior to the donation date.

Where and when to donate

Sample donation kits will be located in the male and female changing rooms at Cellmark Forensic Services, Chorley from **11th May 2017 until 17th May 2017**.

What happens to the information in the project?

All consent forms will be kept confidential and will be stored securely at Cellmark. All consent forms will be discarded at the end of the research. All data included in the project will be fully anonymised.

Cellmark Forensic Services is registered with the Information Commissioner's Office who implements the Data Protection Act 1998 and the Freedom of Information Act 2000. All personal data on participants will be processed in accordance with the provisions of the Data Protection Act 1998.

Thank you for reading this information – please contact me with any questions if you are unsure about any information regarding this study.

Researcher contact details:

Sarah Townsend
MSc(Res) Forensic Science Research Student
University of Central Lancashire
setownsend@uclan.ac.uk

First Supervisor details:

Isobel Louise Colclough
School of Forensic & Applied Sciences
University of Central Lancashire
JB Firth Building
Preston PR12HE
ilcolclough@uclan.ac.uk

If you have any questions, concerns or complaints during or after the project or wish to contact an independent person to whom any questions may be directed or further information may be sought from, please contact Isobel Colclough on:
ilcolclough@uclan.ac.uk or the [University's Office for Ethics, email: \[roffice@uclan.ac.uk\]\(mailto:roffice@uclan.ac.uk\)](#)

Appendix 4: Body Fluid Matrix Consent Form



Consent Form

Name of department: School of Forensic & Applied Science, University of Central Lancashire

Title of the study: Validation of Sperm Elution on intimate swabs using a body fluid matrix.

- I confirm that I have read and understood the information sheet for the above project and the researcher has answered any queries to my satisfaction.
- I understand that my participation is voluntary and that I can refuse to take part without detriment.
- I understand that anonymised data (i.e. data which does not identify me personally) cannot be withdrawn once they have been included in the study. Data can be withdrawn at any time up to that point.
- I understand that any information recorded in the investigation will remain confidential and no information that identifies me will be made publicly available.
- I consent to providing a biological sample(s) and understand that they will remain in the custody of Cellmark Forensic Services. Samples donated specifically for this project will be destroyed upon completion of the project.
- The sample(s) I have donated towards this research are (please tick as appropriate):
 - Semen
 - Saliva
 - Blood
 - Vaginal Cells
 - Faecal material

Sample Reference Number (as stated on donation kit):

Please either sign below or add your Cellmark Donor Number.

.....

Appendix 5: Persistence Ethical Approval



16 June 2016

Isobel Colclough/Sarah Townsend
School of
University of Central Lancashire

Dear Isobel and Sarah

Re: STEMH Ethics Committee Application
Unique Reference Number: STEMH 636

The STEMH ethics committee has granted approval of your proposal application 'To determine the persistence of polydimethylsiloxane (PDMS) in the vagina and on the penis'. Approval is granted up to the end of project date*.

It is your responsibility to ensure that

- the project is carried out in line with the information provided in the forms you have submitted
- you regularly re-consider the ethical issues that may be raised in generating and analysing your data
- any proposed amendments/changes to the project are raised with, and approved, by Committee
- you notify roffice@uclan.ac.uk if the end date changes or the project does not start
- serious adverse events that occur from the project are reported to Committee
- a closure report is submitted to complete the ethics governance procedures (Existing paperwork can be used for this purposes e.g. funder's end of grant report; abstract for student award or NRES final report. If none of these are available use [e-Ethics Closure Report Proforma](#)).

Yours sincerely

A handwritten signature in black ink, appearing to read "Emma".

Emma Sandon-Hesketh
University Officer Ethics
STEMH Ethics Committee

* for research degree students this will be the final lapse date

NB - Ethical approval is contingent on any health and safety checklists having been completed, and necessary approvals as a result of gained.

Appendix 6: Persistence Participation Information Sheet



Participant Information Sheet

Name of department: School of Forensic & Applied Sciences, University of Central Lancashire

Title of the study: Persistence of PDMS in the vagina and on the penis.

Introduction

Name: Sarah Elizabeth Townsend

Status: Postgraduate Research Student

Department: School of Forensic & Applied Sciences, University of Central Lancashire

E-mail: setownsend@uclan.ac.uk

What is the purpose of this investigation?

The purpose of the research is to determine the persistence time of the lubricant polydimethylsiloxane (PDMS) on intimate swabs following sexual intercourse with the use of a lubricated condom. Previous research regarding the persistence of PDMS in the vagina and penis is very outdated and limited. Conducting a new study involving volunteers undertaking consented sexual intercourse with their partner with the use of a silicone lubricated condom would provide a more accurate time frame of the persistence of PDMS. This information could greatly influence the forensic science market as currently the majority of forensic providers only recommend the analysis of intimate swabs for the presence of lubricants to be 24 hours. However, if this time frame could be extended, it may be of use in more forensic cases. This is valuable as many victims of alleged sexual assault don't come forward immediately after the assault.

Additionally, the detection of PDMS in fabrics will be analysed. Following sexual intercourse, vaginal drainage occurs in which PDMS may be excreted from the vagina into the undergarment. If PDMS is unable to be detected on the vaginal swabs, this could be due to the substance passing through to the underwear. Furthermore, PDMS would be present on the penis following the use of the lubricated condom. If PDMS is unable to be detected on intimate swabs, this is likely to be because the PDMS has transferred onto the underpants following intercourse.

What will you do in the project?

This project aims to determine persistence time of PDMS in the vagina and on the penis

following consensual sexual intercourse with the use of a silicone lubricated condom. Participation in the project would require the volunteer to have sexual intercourse with their partner using the provided condoms. Male participants will then be required to swab the penis in the following three areas: shaft, glans and coronal sulcus at the stated time intervals on the Participant Completion Sheet using the swabs provided. Female participants are required to complete two blind vaginal swabs at the same stated time intervals. Female participants are also required to take a swab outside the vagina up to one hour prior to sexual intercourse to allow any external contaminants to be identified. Sexual Intercourse will be required 6 times for this study, one act of intercourse per time interval.

Furthermore, for individuals who would like to participate in the study who aren't in a relationship or whose partners aren't wishing to participate in this study, solo sex is an option whereby female or male participants masturbate whilst wearing the provided condom. The same conditions apply as the intercourse whereby intimate swabs are taken at the stated time intervals and a 7 day break is required to prevent contamination.

Male and female participants will be advised to wear the provided undergarment following the sexual intercourse to allow the recovery of PDMS from vaginal drainage and the penis. This will greatly benefit the research; therefore wearing the provided undergarment is strongly advised. Participants would wear the undergarment for approximately 12 hours following intercourse or the following day if intercourse occurred in the evening then package appropriately in the packaging provided.

The project will take place at Cellmark Forensic Services Laboratories in Chorley and will last for approximately 2 months, ending in October 2017.

Do you have to take part?

No, the decision to take part in this research is voluntary. You can refuse to take part without providing a reason.

Why have you been invited to take part?

Any adult, male or female, over the age of 18 who is a member of staff at Cellmark are able volunteer to take part in this research. Partners of staff members at Cellmark who are not employed by the company are required to consent to undertaking sexual intercourse, however are unable to submit any swabs or DNA material towards the study.

How to participate

If you are willing to participate in this study, the volunteer and their partner are required to complete a consent form which is supplied in the sample boxes in the male and female changing rooms at Cellmark, Chorley. The partner is not required to submit any swabs or DNA material towards the study; however are required to consent to undertaking sexual intercourse. Upon completion, please seal the consent form in the envelope provided with the sample boxes which states the unique reference number. Please ensure all swabs provided are labelled with the unique reference number and time since intercourse and placed in the bottom drawer of the transfer freezer in the receipting area of sample reception, Cellmark, Chorley.

If you **are** a Cellmark employee and do not know your unique donor number, please also complete the QAP0065 Donor consent form in order to obtain your donor number from HR.

Male and female participants opting to wear the provided undergarments should seal the item in the packaging provided and place in the bottom drawer of the transfer freezer in the receipting area of sample reception upon completion of the study. If the participant opted to wear the undergarment, the size required is requested on the consent form. Please **do not** wash the undergarment prior to use or after use as this will negatively impact the validity of the results. Once samples have been processed you are unable to withdraw from the research. All samples will be either processed or destroyed at completion of the project. Participation in this project will take approximately 2 months. All Participation Completion Sheets are to be sealed in an envelope in the sample boxes upon completion of the study.

It is advised that individuals with a latex allergy are not permitted to participate in this study.

If you are participating in this research, please ensure that no sexual intercourse with the use of a lubricated condom has taken place for at least 7 days prior to participating in this study. Also, a 7 day interval is required between each course of sexual intercourse to ensure no PDMS is being transferred to the next time interval. Therefore, this study is expected to last approximately 10 weeks. No additional sexual intercourse is permitted in between time intervals with or without the use of a condom to prevent the transfer of PDMS. Both heterosexual and homosexual couples are able to participate in this study.

Where and when to donate

Consent forms will be available in the male and female changing rooms at Cellmark Forensic Services, Chorley from **Friday 7th July 2017 until Friday 21st July 2017**. Participation kits will be located in the male and female changing rooms at Cellmark Forensic Services, Chorley and available for collection from **Wednesday 26th July 2017**.

Please ensure all participation in this study is completed with all swabs and undergarments returned to the transfer freezer in Sample Reception, Cellmark, Chorley and all Completion Forms sealed in an envelope in the male/female changing rooms, Cellmark, Chorley by **Friday 20th October 2017**.

What happens to the information in the project?

All consent forms will be kept confidential and will be stored securely at Cellmark. All consent forms will be discarded at the end of the research. All data included in the project will be fully anonymised.

Cellmark Forensic Services is registered with the Information Commissioner's Office who implements the Data Protection Act 1998 and the Freedom of Information Act 2000. All personal data on participants will be processed in accordance with the provisions of the Data Protection Act 1998.

Thank you for reading this information – please contact me with any questions if you are unsure about any information regarding this study.

Researcher contact details:

Sarah Townsend
MSc(Res) Forensic Science Research Student
University of Central Lancashire
setownsend@uclan.ac.uk

First Supervisor details:

Isobel Louise Colclough
School of Forensic & Applied Sciences
University of Central Lancashire
JB Firth Building
Preston PR12HE
ilcolclough@uclan.ac.uk

If you have any questions, concerns or complaints during or after the project or wish to contact an independent person to whom any questions may be directed or further information may be sought from, please contact Isobel Colclough on: ilcolclough@uclan.ac.uk or the University's Office for Ethics, email: roffice@uclan.ac.uk

Appendix 7: Persistence Consent Forms



Consent Form

Name of department: School of Forensic & Applied Science, University of Central Lancashire

Title of the study: Persistence of PDMS in the vagina and on the penis.

- I confirm that I have read and understood the information sheet for the above project and the researcher has answered any queries to my satisfaction.
- I understand that my participation is voluntary and that I can refuse to take part without detriment.
- I understand that anonymised data (i.e. data which does not identify me personally) cannot be withdrawn once they have been included in the study. Data can be withdrawn at any time up to that point.
- I understand that any information recorded in the investigation will remain confidential and no information that identifies me will be made publicly available.
- I consent to participating in this research and understand that samples provided will remain in the custody of Cellmark Forensic Services. Samples donated specifically for this project will be destroyed upon completion of the project.
- I consent to not participate in the study if I have a latex allergy.
- I consent to undertake minimal activity, not wear tight clothing/underwear and to continue the usual amount of washing in intimate areas for the duration of the study.
- I consent to not use any intimate body moisturisers for the duration of the study (due to polydimethylsiloxane (PDMS) contamination).

Donation Reference Number (please remember this number and this will indicate the correct donation kit to collect from Wednesday 26th July 2017):

I am willing to wear the provided undergarment: Yes No

In this study I am consenting to: Sexual Intercourse Solo Sex

Gender: Male Female

Please state the size of undergarment required: Small Medium Large

Cellmark Donor Number/Signature:



Partner Consent Form – Non-Cellmark Employee

Name of department: School of Forensic & Applied Science, University of Central Lancashire

Title of the study: Persistence of PDMS in the vagina and on the penis.

- I consent to not use any intimate body moisturisers for the duration of the study (due to polydimethylsiloxane (PDMS) contamination).
- I understand that any information recorded in the investigation will remain confidential and no information that identifies me will be made publicly available.
- I understand that my participation is voluntary and that I can refuse to take part without detriment.
- I consent to not participate in the study if I have a latex allergy.
- I consent to undertaking sexual intercourse with my partner with the use of a provided condom for a total of 6 times within a 10 week period.
- I consent to not providing swabs or DNA evidence towards this study.

Signature:

Appendix 8: Persistence Participant Completion Sheet

Participation Completion Sheet - Persistence of polydimethylsiloxane (PDMS) study

Please note: One Participation Completion Sheet per individual, not per couple.

Gender: Male Female
(Please tick as appropriate)

I am undertaking: Sexual intercourse Solo sex

Instructions for Swabbing & Storage of Swabs

- Female participants are required to complete a total of **2** blind vaginal swabs for each time interval to ensure maximum recovery.
- Please ensure each area required is swabbed thoroughly for approximately 5 seconds.
- Before returning all completed swabs and undergarments to Cellmark, Chorley, please store in a cool dry place in the packaging provided. Please do not freeze.
- Please **do not** wash any undergarments prior to use or after use.

Time elapsed since intercourse/solo sex: **12 hours**

Date & Time:

Swabs taken: Outer Vaginal Blind Vaginal x2 Penis shaft Penis glans Coronal sulcus

Was the provided undergarment worn? Yes No

Was any moisturiser used prior to sexual intercourse? Yes No if yes, which one?.....

Has any washing taken place since intercourse? Yes No

Which stage in the menstrual cycle are you at during the time of intercourse (Female participants only):.....

Any additional comments?.....

Time elapsed since intercourse/solo sex: **18 hours**

Date & Time:

Swabs taken: Outer Vaginal Blind Vaginal x2 Penis shaft Penis glans Coronal sulcus

Was the provided undergarment worn? Yes No

Was any moisturiser used prior to sexual intercourse? Yes No if yes, which one?.....

Has any washing taken place since intercourse? Yes No

Which stage in the menstrual cycle are you at during the time of intercourse (Female participants only):.....

Any additional comments?.....

Time elapsed since intercourse/solo sex: **24 hours**

Date & Time:

Swabs taken: Outer Vaginal Blind Vaginal x2 Penis shaft Penis glans Coronal sulcus

Was the provided undergarment worn? Yes No

Was any moisturiser used prior to sexual intercourse? Yes No if yes, which one?.....

Has any washing taken place since intercourse? Yes No

Which stage in the menstrual cycle are you at during the time of intercourse (Female participants only):.....

Any additional comments?.....

Time elapsed since intercourse/solo sex: **36 hours**

Date & Time:

Swabs taken: Outer Vaginal Blind Vaginal x2 Penis shaft Penis glans Coronal sulcus

Was the provided undergarment worn? Yes No

Was any moisturiser used prior to sexual intercourse? Yes No if yes, which one?.....

Has any washing taken place since intercourse? Yes No

Which stage in the menstrual cycle are you at during the time of intercourse (Female participants only):.....

Any additional comments?.....

Time elapsed since intercourse/solo sex: **48 hours**

Date & Time:

Swabs taken: Outer Vaginal Blind Vaginal x2 Penis shaft Penis glans Coronal sulcus

Was the provided undergarment worn? Yes No

Was any moisturiser used prior to sexual intercourse? Yes No if yes, which one?.....

Has any washing taken place since intercourse? Yes No

Which stage in the menstrual cycle are you at during the time of intercourse (Female participants only):.....

Any additional comments?.....

Time elapsed since intercourse/solo sex: **60 hours**

Date & Time:

Swabs taken: Outer Vaginal Blind Vaginal x2 Penis shaft Penis glans Coronal sulcus

Was the provided undergarment worn? Yes No

Was any moisturiser used prior to sexual intercourse? Yes No if yes, which one?.....

Has any washing taken place since intercourse? Yes No

Which stage in the menstrual cycle are you at during the time of intercourse (Female participants only):.....

Any additional comments?.....

Please ensure all swabs provided are labelled with the unique reference number and time since intercourse and placed in the bottom drawer of the transfer freezer in the receipting area of sample reception, Cellmark, Chorley. Participants opting to wear the provided undergarments should seal the item in the packaging provided and return to the bottom drawer of the transfer freezer in the receipting area of sample reception upon completion of the study. Please can all swabs and undergarments be returned by **Friday 20th October 2017**.

Thank you for your participation. Please sign and date this sheet below, seal in an envelope and place in the sample boxes in the male and female changing rooms, Cellmark, Chorley by **Friday 20th October 2017**.


Cellmark Donor Number or Signature:

Date:

Appendix 9: CoSHH Form

CoSHH RISK ASSESSMENT FORM. (Page 1 of 2)



<u>Faculty/Department</u>	<u>Assessors Name(s)</u>	<u>Job Title/Position</u>	
School of Forensic & Applied Science	Sarah Townsend	MSc (Research) Student	

Briefly describe the task/process. (description, use, users)

To successfully complete the sperm elution process using a body fluid matrix. Analyse samples to determine the presence of PDMS after the process.

Substances (used or produced as by-products or wastes)	Quantity	Hazard Class	WEL	Exposure Route(s)	Frequency and Duration of Exposure	Known Health Effects:
Liquid Nitrogen (LN ₂)	Up to 50 litres in dewars	Asphyxiant	N/A	Inhalation Skin contact	Variable. Generally very short duration.	Asphyxiation resulting in death. Cold burns, frostbite – skin damage.
Isooctane	Upto 5 litres	Danger, Flammable Irritant	N/A	Skin contact,	Upto 6 hours	H225 - Flammable liquids, H315 - Skin irritation Specific target organ toxicity H336 - Central nervous H304 - Aspiration hazard H400 - Acute aquatic toxicity

						H410 - Chronic aquatic toxicity
Ethanol	Upto 2 litres	Danger, Flammable, Irritant	1,000 ppm 1,920 mg/m ³	Skin contact, Eye contact	30 mins	H225 Highly flammable liquid and vapour. H319 Causes serious eye irritation
Results of Relevant Health Surveillance				Results of Exposure Monitoring		
N/A				N/A		

Control Measures				
<input type="checkbox"/> Elimination	<input type="checkbox"/> Substitution	Reduction	<input type="checkbox"/> Isolation	Eng. Control
<i>Details</i>	<i>Details</i>	<i>Details</i> Use smallest volumes possible for each activity.	<i>Details (glovebox)</i>	<i>Details (LEV, fumehood)</i> Fume cupboard where appropriate and available.
<p>Further Details (if required)</p> <p>Where possible, insulate all exposed cold surfaces. NEVER travel in a passenger lift with a Dewar of liquid nitrogen. Report all leaks immediately and remove the Dewar from the building to an outdoor location (e.g. ventilated gas store bay in Stewart Building). Follow all local written procedures for the handling of LN2. Users instructed never allow skin to be in direct contact with LN2 Low oxygen detection and alarm</p> <p>Training: Appropriate information and/or training on risk of asphyxiation, fire hazards, cold burns, frostbite and hypothermia for staff who decant and transport LN2.</p>				
Personal Protective Equipment				
X Gloves	X Eye protection	X Coverall/lab coat	X Foot protection	<input type="checkbox"/> Respiratory protection
<i>Details</i> Nitrile gloves supplied and	<i>Details</i> All students and staff required to	<i>Details</i> All students and staff required to	<i>Details</i> All students and staff required to	<i>Details:</i> Dust mask to be use when

should be worn	wear eye protection	wear lab coats	wear sturdy shoes	weighing and transporting Lead-A
<input type="checkbox"/> Health Surveillance required		<input type="checkbox"/> Exposure monitoring required		

Emergency Arrangements

First Aid:			
Eyes	Seek medical attention immediately.		
Skin	Cold burns: loosen any clothing that may restrict blood circulation and seek medical attention for all but superficial injuries. Do not try to remove clothing that is frozen to the skin. Do not apply direct heat to affected parts, but if possible, place in tepid water. Clean plastic kitchen film or sterile dry dressings should be used to protect damaged tissue from infection or further injury, but they should not be allowed to restrict blood circulation. Alcohol and cigarettes should not be given. Where exposed skin is stuck to cold surfaces such as un-insulated cryogenic pipework, isolate the source of the cold liquid and thaw with copious amounts of tepid water until the skin is released.		
Ingestion	If cryogenic fluid is ingested. Seek emergency medical attention immediately.		
Inhalation	Asphyxiation: Seek emergency medical attention immediately. Summon local first aider. Transient exposure to very cold gases produces discomfort and can also trigger an asthma attack in susceptible people. Remove to fresh air and seek medical attention. If you suspect a person has been asphyxiated (i.e. they are in an area where a low oxygen alarm is sounding) and they cannot exit the room without assistance: * DO NOT ENTER THE ROOM. Dial 999 and request emergency services, stating building, room no and casualty trapped in a low oxygen/ asphyxiant environment. * Nitrogen gas is colourless and odourless. Do not rely solely on sense of smell to detect leaks!		
Fire: Extinguisher Type			
<input type="checkbox"/> Water	X Foam	X Powder Use dry powder for fire	X CO ₂
Spillage/release:			
Evacuate all personal from area and ventilate naturally and allow to dissipate. Check oxygen level prior to re-entry.			

Waste Disposal procedure

Excess or waste LN2 must not be poured down the drain. It must be allowed to evaporate naturally either outdoors or in a fume cupboard.

Persons likely to be exposed

<input checked="" type="checkbox"/> Staff	<input checked="" type="checkbox"/> Student	<input type="checkbox"/> Visitor	<input type="checkbox"/> Contractor
<input type="checkbox"/> Public	<input type="checkbox"/> Other (specify)		

Additional risks: for example circumstances where work will involve exposure to more than one substance hazardous to health, consider the risk presented by exposure to such substances in combination. Also, non-routine maintenance may present additional risk of exposure.

Authorised by (sign):		Review date due:	
Date:			

Notes:

Hierarchy of control

<i>Change the task or process so that the hazardous substance is not required or generated.</i>
<i>Replace the substances with a safer alternative.</i>
<i>Totally isolate or enclose the process.</i>
<i>Partially enclose the process and use local exhaust ventilation.</i>
<i>Ensure good general ventilation.</i>
<i>Use a system of work that minimises the chance and degree of exposure.</i>
<i>Provide personal protective equipment (PPE).</i>
<i>Train and inform staff in the safe system of work and risks.</i>
<i>Additional supervision.</i>
<i>Examination, testing and maintenance of engineering controls and/or PPE.</i>
<i>Monitoring of exposure.</i>
<i>Health Surveillance.</i>

