

In Search of The Boston Strangler: Genetic evidence from the exhumation of Mary Sullivan

DAVID R FORAN, PhD

Forensic Science Program, School of Criminal Justice and Department of Zoology, 560 Baker Hall, Michigan State University, East Lansing, MI 48824

JAMES E STARRS, LL.M.

Department of Forensic Sciences, Columbian College of Arts and Sciences, The George Washington University, 720 20th St. N.W., Washington, D.C. 20052

ABSTRACT

The Boston Strangler was one of the United States' most notorious serial killers, raping and strangling with decorative ligatures thirteen women in Boston during the early 1960s. Albert DeSalvo, never a suspect in the slayings, confessed in prison (where he was later murdered) to being the Boston Strangler, and the investigation largely ended. Mary Sullivan was the last victim of the Boston Strangler, found sexually assaulted and strangled in her Boston apartment in 1964. Recently, a team of forensic scientists undertook the exhumation and subsequent scientific analysis of Mary Sullivan's remains, in hopes of finding consistencies or inconsistencies between DeSalvo's confessed description of the murder and any evidence left behind. Included in these analyses was extensive DNA testing of all UV fluorescent material associated with the body. The large majority of results were negative, however, fluorescent material located on the underwear and entwined in her pubic hair generated two human mitochondrial DNA sequences. Neither of these matched the victim nor members of the forensic team who worked on the evidence. Most importantly, neither DNA sequence could have originated from Albert DeSalvo.

INTRODUCTION

On 13 October 2000, the remains of Mary Sullivan were exhumed from their burial place at the St Francis Xavier cemetery in Hyannis, Massachusetts. The exhumation was conducted by virtue of a permit issued by the Barnstable Town offices to Diane Sullivan Dodd, a younger sister of Ms Sullivan and the conservator of her estate. In view of Ms Dodd's relationship to Ms Sullivan and the absence of any otherwise controlling Massachusetts's statutes on the subject of exhumations

in such cases, no court order was required as a prerequisite to the exhumation.

Ms Dodd, through her son Casey Sherman, had previously contacted co-author James Starrs, asking him to investigate the death of her sister on 4 January 1964. Mary Sullivan was reputed to be the thirteenth and last victim of the notorious Boston Strangler – as the newspapers had dubbed him – a serial murderer who left victims with ligatures tied tightly around their necks. In the view of Ms Dodd and Mr Sherman, the person who had killed Mary Sullivan was still an open and a legally unresolved issue, even though Albert DeSalvo had given an audiotaped interview in which he confessed to killing Ms Sullivan, as well as the twelve other women. Oddly, Mr DeSalvo was never a police suspect in the murders, and was not convicted, nor even tried or indicted, for any of the thirteen murders attributed to the Boston Strangler; on the contrary he had been convicted of other, unrelated rapes of women. In 1973 Mr DeSalvo was murdered in the Walpole prison infirmary where he was confined.

MATERIALS AND METHODS

An investigation of the circumstances surrounding the death of Ms Sullivan, and related police and medical examinations of the murder, was initiated. The most comprehensive set of information came from a number of books devoted to the killings written over the years, including the latest, a 1995 text entitled



skull, however, 'acute traumatic injuries of both breasts' were observed at autopsy.

Apart from these contradictions and inconsistencies between the autopsy report and Mr DeSalvo's confession, the confession included one extremely incongruous feature, wherein he stated that during the assault upon Ms. Sullivan 'she had in her hand all the time' a small knife (not found at the scene), 'like you use for peeling vegetables in the kitchen,' and that 'she did not once lift that knife against me.' This odd declaration, the discrepancies between the autopsy report and confession, the lack of access to official information, and the inability to obtain possibly extant and inventoried evidence, convinced Ms Dodd and Mr Sherman that the only recourse to obtaining fresh evidence on the murder of Mary Sullivan was to authorize an exhumation of her remains. We agreed with the Sullivan family's assessment of the situation, while recognizing that finding such evidence in, or on, the remains of Ms Sullivan, some thirty-six years after her death, autopsy, and embalming, was bound to be a shot in the dark.

In view of the sexual assault claimed by Mr DeSalvo and confirmed, in part, by Dr Luongo, the possibility existed that a DNA-extractible seminal stain might be discovered on tissues obtained from the re-autopsy. Richard DeSalvo, the older brother of Albert DeSalvo, volunteered to provide the necessary reference samples, utilizing buccal swab and fingerstick kits (provided by Tri-Tech, Inc.). Richard DeSalvo agreed to cooperate because of his determined conviction that his brother was not the Boston Strangler and that our investigation might resolve the question once and for all, at least as to the death of Ms Sullivan.

The exhumation

A team of forensic scientists was assembled that, beyond the authors, included a criminalist, pathologist, anthropologist, toxicologist, radiographer, geophysicist, and several other support staff members. When the well-drained sandy topsoil was removed from the monument-marked grave of Ms Sullivan, the tri-partite lid of a concrete grave liner, with eye hooks attached, was revealed.

The section of the concrete lid covering the head end was lifted to disclose a collapsed wooden coffin. The upper portion of the remains was seen to have been interred in a supine position. The skull, facing skyward, was plainly visible. The face seemed to be remarkably preserved for a thirty-six year old burial. Considering the fact that a body bag would be needed to remove the remains from the fragmented coffin, it was decided to replace the lid and return on the next day to complete the exhumation.

On 14 October 2000, upon the removal of the three parts of the concrete vault lid, it became immediately evident that the remains were almost fully skeletonized with little but fragments of adherent tissue attached, resembling a thin layer of firm plaster-like cardboard. The face, which the previous day had seemed to be well-preserved, was now disclosed to be covered with artifactual, cast-like material, probably a consequence of the mortuary's preparation of the remains for viewing. The remains were transported to the John Lawrence funeral home in Marston Mills, Massachusetts where a mortuary suite had been graciously provided for the re-autopsy.

The re-autopsy

All persons in contact with the remains at the grave site, as well as during the re-autopsy, were gloved and masked to protect against DNA cross-contamination. The plastic bag organs removed during the original autopsy was found to be lying in the abdominal region, as expected, without evidence of breakage. These organs and brain matter were in a remarkable state of preservation, even though the characteristic odour of formaldehyde preservative was not present. Samples of the organs were taken for toxicological study.

During the re-autopsy a particular interest was directed to the neck organs, since strangulation by three ligatures as well as a preceding manual strangulation had been claimed by Mr. DeSalvo and, at least as to the ligatures, confirmed by Dr Luongo's autopsy report. The hyoid bone was found to be unfractured and otherwise intact, as well as

completely fused, indicating that its epiphysis was complete (surprising for a person this young), and pointing away from a violent manual strangulation.

On the outside of the body, substances that dimly fluoresced under UV light were apparent in the head hair, while a slightly more intense fluorescence was seen about the pubic hair. Large masses of both types of hair, still in place as in life, were secured for further analysis. In addition, the deceased's underwear was retained for more exacting tests since it also fluoresced under UV light. Throughout the re-autopsy, samples taken for subsequent analysis were given access numbers and entered sequentially in a log book, with the sample containers marked with the same log book designations. All in all some sixty-four individual items were collected.

Laboratory procedures

Materials submitted to the laboratory for analysis included a ca. 12 × 12 cm segment of head hair, a ca. 8 × 8 cm segment of pubic hair, eyelashes, fingernails, viscera, and underwear, as well as soil, small coffin fragments, and other miscellaneous items from the exhumation. A central portion of the underwear had been cut away shortly after the exhumation and this section, as well as the major portion, were processed separately in the laboratory. The fingernails of Mary Sullivan, and their subsequent analysis, have been described (Cline et al., 2003).

Extreme care was taken when handling the evidence in order to minimize the chance of contamination. Gloves, gowns, and masks were worn, and all solutions and supplies were sterilized and subjected to short wavelength UV irradiation before use. Preliminary examination of the evidence included a complete inspection using a long wavelength UV light source. Areas that showed fluorescence underwent further analyses.

Tests for the presence of semen included standard assays for acid phosphatase, choline, prostate specific antigen (PSA or P30), and microscopic searches for sperm (Beachtel, 1988). Genetic testing focused on mtDNA, as its analysis is more likely to be successful in

aged or degraded samples (reviewed by Holland and Parsons, 1999). Reagent blanks and substrate controls were included throughout the process. DNA preparation was carried out in a separate room from subsequent PCR amplification and DNA analysis, and PCR reactions were set up in a HEPA filtered hood.

The two sections of underwear were analyzed independently by two individuals. UV fluorescent areas were cut out and soaked in a solution of 1% SDS, 50mM EDTA overnight. The liquid was then incubated with proteinase K (1μL of 20mg/mL per 100μL liquid) and DTT (3μL 1M per 100μL liquid) overnight at 55°C. Following incubation, the liquid was extracted with an equal volume of phenol/chloroform/isoamyl alcohol. Samples were precipitated using 1/10 volume of 3M NaAcetate, 2 volumes of 95% ethanol, dried, and resuspended in TE (10 mM Tris pH 7.5, 1 mM EDTA). This solution was further purified on a Microcon 100 column, resulting in a final volume of approximately 10μL.

Fluorescent substances attached to hair were removed with sterile (flamed and cooled) forceps, or if they were too entangled with the hair, a section of the substance was cut away with a sterile scalpel, using a dissecting scope if needed. This allowed for avoidance of any hair, which could result in subsequent contamination. These substances were placed in digestion buffer (20mM Tris pH 7.5, 50mM EDTA, 0.1% SDS, and 1μL of 20mg/mL proteinase K and 3μL 1M DTT per 100μL liquid) and incubated at 55°C. If the substances were not fully digested after an overnight incubation, second aliquots of proteinase K and DTT were added, and digestion continued. Samples were then extracted, precipitated, and purified as above.

Multiple primers were used for mtDNA control region amplification and analysis (see Holland and Parsons, 1999) including F15989, F16190, F15, F82, R16410, R285, and R484 (sequences available through the Armed Forces DNA Identification Laboratory's web site; see references). PCR was conducted on Perkin-Elmer 2400 and 9600 thermalcyclers, with general cycling parameters of 94°C for 3 minutes, 33 – 38 cycles of 94° for 30 seconds,

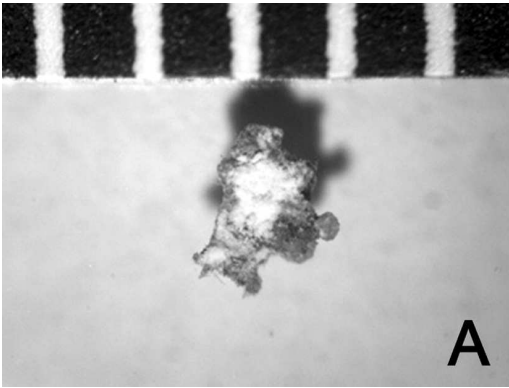


Figure 1A.

Representative particle found in head hair (scale is mm). A large number of these particles existed, although none generated a PCR product.

55° for 30 seconds, 72° for 45 seconds, followed by a final extension at 72° for 7 minutes. Amplification products were sequenced using a BigDye sequencing kit on an ABI 310 genetic analyzer according to the manufacturer's protocol.

In instances where no amplification product was generated, nested PCR (e.g., Strom and Rechitsky, 1998) was attempted. For this, outer primers (e.g., F15/R484) were used for an initial 20-30 cycles, and 1µL of this mix was added to a fresh PCR mix containing internal (nested) (e.g., F82/R285) or semi-nested (e.g., F15/R285) primers. The second tube was amplified for another 20 cycles. Owing to the high sensitivity (and therefore potential for contamination) of this technique, extreme care was taken in these procedures. Any indication of bands appearing in reagent blanks or negative controls negated the entire experiment; to be considered valid, results had to be replicable.

The same regions of mtDNA obtained from the evidence were sequenced for all individuals who handled either the body during the exhumation or the submitted evidence, to exclude them as a source of DNA. Reference samples were also obtained from Ms. Dodd and Richard DeSalvo, as they would have the same mtDNA haplotype as their siblings. MtDNA sequences were also obtained from a bone

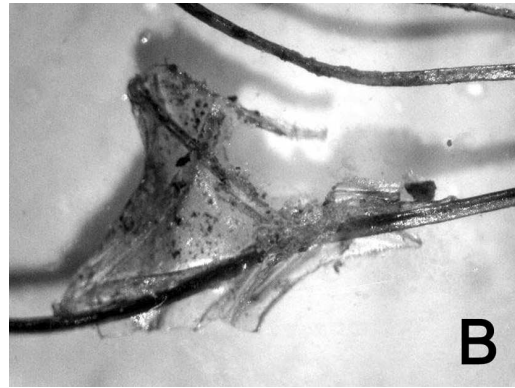


Figure 1B. Translucent material surrounding pubic hairs. The bottom portion of the material has been cut away with a scalpel for DNA analysis.

sample of Ms. Sullivan, and from a later exhumation of Albert DeSalvo. The DeSalvo reference samples were kept in a different building from that in which the evidence was processed until all tests of the evidence material had been completed. Only then were they brought to the lab for DNA typing.

RESULTS AND DISCUSSION OF LABORATORY ANALYSES

The exhumation of Mary Sullivan revealed a complete skeleton, consistent with 36 years of

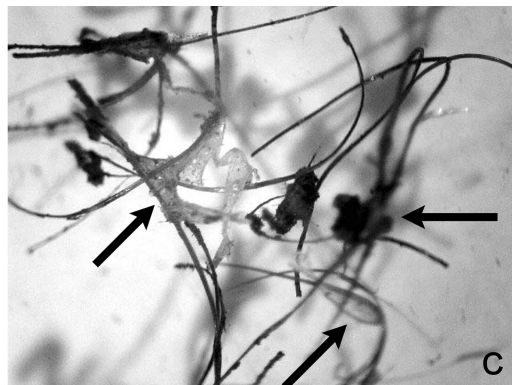


Figure 1C. Translucent material (arrows on left) and dirt particles (e.g., right arrow) surrounding pubic hairs.

interment. Grave goods, as well as a bone fragment match of mtDNA control region sequences to the deceased's sister, confirmed the body as that of Mary Sullivan. As mentioned, the abdominal organs and brain were situated inside a plastic bag in the abdomen, but unlike other non-skeletal parts of the body, these showed a remarkable degree of preservation, being readily identifiable, including distinct organ structures. However, in no instances could DNA be isolated/amplified from these tissues. It is possible the organs were treated with some preservative or fixative at the time of autopsy, protecting the tissues while chemically modifying the DNA or otherwise allowing its decay, although this is unclear.

Visual and serological examination of the evidence

UV fluorescent material was found in three places: on both sections of underwear (as irregular areas of weak fluorescence a cm or two in diameter), in the head hair (as 1-2mm, irregularly shaped, hard objects; Figure 1A), and in the pubic hair (as somewhat translucent patches of 1-3mm, generally engulfing a hair or hairs; Figure 1B and 1C). Dirt and debris were also associated with the hair. The head hair fluorescent substances, which were very hard, could be removed easily using forceps, while the pubic hair substance, given its entanglement, was generally removed using a scalpel, being careful to avoid any hair. The pubic hair substance had a less brittle texture; it did not chip when cut with a scalpel, but sliced cleanly.

Presumptive and confirmatory tests for semen were negative in all cases, although the exact relevance of this is not clear. These tests are based on the high concentrations of specific (non-DNA) bio-molecules or intact sperm in semen, but unlike DNA methods that allow for its utilization from aged and degraded material, no amplification procedure can be used to enhance them. We do not know if they could possibly withstand the conditions of burial, nor the time frame that this case entails. In contrast, DNA is routinely recovered from samples far older than this, indicat-

ing that, if present, it would be among the most probable sources of evidence to be recovered following exhumation.

DNA testing of the evidence

Repeated efforts to obtain and amplify DNA from the head hair material (Figure 1A) were negative. These particles were found throughout the head hair, and are presumed to be of fungal or some other non-human origin.

The two sections of underwear both had UV fluorescent areas, which were processed by individuals working separately. None of the DNA extracts produced PCR product using standard amplification procedures, but when nested PCR was employed, 203 bp amplicons (basepairs 82 to 285 of the human mtDNA sequence) from both sections were obtained. All controls were negative. These segments of underwear produced identical sequences, differing from the Anderson/Cambridge Reference Sequence (Anderson et al., 1981) at 151T, 152C, 263G, and a deletion at 249.

The UV fluorescent matter found in the pubic hair (Figure 1B and C) was very slow to dissolve in the digestion buffer containing proteinase K and DTT. After two to three days a portion of the substance generally remained. In two separate cases (i.e., the substance was obtained from different regions of pubic hair) segments of mtDNA were successfully amplified using nested or semi-nested PCR. Sequences from positions 22-309 were obtained, and both differed from the Cambridge Reference Sequence only at position 263 (G).

DNA sequences from this same mitochondrial region were generated for all individuals potentially in contact with the evidence, including those at the exhumation and in the laboratory. In all instances members of the project could be excluded as sources of the DNA isolated from the evidence. Further, the two sequences detailed above were not the same as Ms Sullivan/Ms Dodd.

The final step was to isolate DNA from the Richard DeSalvo reference sample, to see if the DNAs obtained from the evidence could have originated from Albert DeSalvo. HVII sequences from Richard DeSalvo differed by one or more basepairs from both the under-

wear and pubic hair substance sequences. Subsequent mtDNA sequencing of a bone fragment from a later Albert DeSalvo exhumation confirmed this finding.

CONCLUSIONS

The effort to exhume, autopsy, and analyse evidence from Mary Sullivan, the last victim of the Boston Strangler, involved a large number of scientists, as well as the cooperation of both the victim's and the accused's families. Our goal was to test any and all evidence as thoroughly as possible, to see if Albert DeSalvo might in any way be identified, or even suggested, as her murderer. Certainly the most straightforward results would have been to find DNA consistent with Mr DeSalvo on Mary Sullivan, thus implicating him as the assailant. The controversy could perhaps then be ended.

However, after wide-ranging and exhaustive attempts, we found no indication of biological material from Albert DeSalvo on the exhumed body of Mary Sullivan. We did, on the other hand, find mtDNA from two other, unidentified individuals – results that were repeatable. This does not mean that both or either of these DNAs came from her murderer of course; we cannot conclude that. The DNA sequence obtained from the underwear is uncommon, occurring in five individuals of the 4839 (0.1%) in the FBI's database (Monson et al., 2002). The sequence is not seen in the larger Caucasian, African American or Hispanic databases, and only appears in Asians (Japan, China, Korea) at a frequency of 5.9%. The mtDNA obtained from the UV fluorescent substance throughout the pubic hair is more common, found in 2.6% of all individuals, and is very common in Caucasians, occurring in more than 15% of individuals. Regardless of their general frequencies however, it is clear that neither source of DNA belonged to Albert DeSalvo.

From whom then did these substances originate? Certainly one or both could have come from Mary Sullivan's murderer(s). Unfortunately there were reference samples we could not obtain for this study, including those from persons who handled the body immedi-

ately following her death. In the early 1960s the police, medical examiner, etc., could not be aware of future, ultra-sensitive DNA typing techniques, and bodies were dealt with differently than they are today. We do know that the victim's body was discovered unclothed, and the underwear described here, while hers, was placed on her after her death. It was unquestionably handled by others, and potential sources of contamination are numerous. Yet given that she was sexually assaulted, one very feasible source of the fluorescent material remains her attacker.

The substance engulfed in the pubic hair was even more intriguing because it was unusual, and seemed visually consistent with dried and very old semen. The forensic community does not, of course, have the luxury of conducting decades-long experiments designed to learn how evidence behaves over that amount of time. We do not know how dried semen should appear after thirty-six years of burial, nor if any standard presumptive or confirmatory tests for the presence of semen could possibly work after that interval. We were, however, able to retrieve a single mtDNA sequence from the pubic hair material on two occasions. From whom that DNA originated remains unknown.

ACKNOWLEDGEMENTS

The authors would like to thank Danielle Bernier, Laura Cannon, Rachel Cline, James DiFrancesco, Jennifer Dreier, Priscilla Foran, Leah Ford, Sylvia Gill, Nicole Laurent, Elizabeth Nies, Gina Sola, and Dr. Walter Rowe for their participation in this endeavour.

REFERENCES

- Anderson S., Bankier A.T., Barrell B.G., de Bruijn M.H., Coulson A.R., Drouin J., Eperon I.C., Nierlich D.P., Roe B.A., Sanger F., Schreier P.H., Smith A.J., Staden R. and Young I.G. (1981) Sequence and organization of the human mitochondrial genome. *Nature* **290**, 45765.
- Armed Forces DNA Identification Laboratory <http://www.afip.org/Departments/oafme/dna/>
- Beachtel F.S. (1988) The identification and individualization of semen stains. In: Saferstein R., (ed) *Forensic Science Handbook Vol. II*. Upper Saddle River, NJ, Prentice Hall, 34792.
- Cline R.E., Laurent N.M. and Foran D.R. (2003) The fingernails of Mary Sullivan: developing reliable methods for selectively isolating endogenous and

- exogenous DNA from evidence. *J. For. Sci.* **48**, 32833.
- Frank G. (1966) *The Boston Strangler*. New York, NY, New American Library.
- Holland M.M. and Parsons T.J. (1999) Mitochondrial DNA sequence analysis – validation and use for forensic casework.. *For. Sci. Rev.* **11**, 2250.
- Kelly S. (1995) *The Boston Stranglers: The Public Conviction of Albert DeSalvo and the True Story of Eleven Shocking Murders*. Secaucus, NJ, Carol Pub. Group.
- Kelly S. (2002) *The Boston Stranglers*. New York, NY, Kensington Pub. Corp.
- Monson K.L., Miller K., Wilson M.R., DiZinno J.A. and Budowle B. (2002) The mtDNA Population Database: An integrated software and database resource for forensic comparison. *For. Sci. Comm.* **4**.
- Rae G.W. and DeSalvo A. (1967) *Confessions of the Boston Strangler*. New York, NY, Pyramid Books.
- Strom C.M. and Rechitsky S. (1998) Use of nested PCR to identify charred human remains and minute amounts of blood. *J. For. Sci.* **43**, 696-700.