

Extreme Degradation of Human Hair by Keratinophilic and Keratinolytic Fungi

Walter F. Rowe

The George Washington University*

KEYWORDS

Forensic science, mycology, fungal degradation, keratinophilic, keratinolytic, fungus, light microscopy, scanning electron microscopy (SEM), transmitted light microscopy, DNA profiling

ABSTRACT

Head hairs from a 19-year-old female murder victim, who had been buried for 36 years, were examined by scanning electron microscopy (SEM) and transmitted light microscopy. The hairs exhibited a range of biodeterioration artifacts. In some areas on the hairs, the cuticular scales were intact; in others, scales had become loosened; and in yet other areas, scales had been completely lost, exposing the underlying cortical cells. Three different types of fungal tunnels were observed: thin, thread-like tunnels; conical tunnels that narrowed as they penetrated the shaft of the hair; and conical tunnels that increased in diameter as they penetrated the hairs. These fungal tunnels are consistent with the growth of both keratinophilic and keratinolytic fungi.

In some hairs, fungal hyphae had penetrated to the center of the hair and consumed the central medullary cells. In extreme cases, the hairs were reduced to hollow tubes resembling soda straws. A fungal penetrating organ was observed in situ, protruding from the shaft of the hair. The lack of cellular disruption surrounding this penetrating organ indicated that it had penetrated the hair by digesting the keratin of the cuticular and cortical cells. The extent of microbial attack on the hairs suggests that conditions (tempera-

ture, humidity and presence of a growth substrate) in the coffin were initially extremely favorable to fungal growth. The survival of substantial numbers of hair is most likely due to a fall in humidity due to loss of moisture from the coffin.

INTRODUCTION

From time to time, archaeologists and forensic scientists encounter hair samples (1-3). Hairs may be associated with human remains, may be loose in soil, may be loose on surfaces, and they may have been used to fabricate textiles and cordage. Examinations of suspected hairs will proceed in a step-by-step manner (4). Transmitted light microscopy will be used to determine (a) if the specimens are indeed hairs and (b) if so, what mammalian species the hairs came from. In the case of human hairs, microscopical examinations may determine population ancestry and somatic (bodily) origin of the hair.

In a forensic context, the hairs may be compared to known sources using a transmitted light comparison microscope. Before the advent of DNA profiling, microscopical hair comparisons were the end point of forensic hair examinations. Now, mitochondrial DNA (mtDNA) can be extracted from a specimen hair and its hypervariable regions sequenced (5). The resulting mtDNA sequence can be compared to corresponding regions in the mtDNA of a suspect or a crime victim. In the event that the potential source is not available to provide a reference DNA sample, an identical sample may be procured from a maternally related family member (6).

*Department of Forensic Sciences, The George Washington University, 2100 Foxhall Road NW, Washington, D.C. 20007



Figure 1. SEM micrograph showing intact cuticular scales, 1500x magnification at 25kV.

Environmental conditions can affect hairs. For example, exposure to sunlight can result in photochemical oxidation of hairs, leading to alterations in hair color (4). Hairs are also subject to biodegradation or bioerosion. Insects such as dermestid beetles can attack and consume hair (2). Microorganisms can also attack hair, destroying it or altering its microscopic appearance. Microbial growth on or within hair is favored in warm, moist environments, such as the insides of coffins. Fungal hyphae can grow on the surface of the hair, damaging or removing cuticular scales (7-14). They can also tunnel into the hair, disrupt its structure and even consume its structure from within (7-13, 15). Recognizing the microscopic artifacts of biodegradation can be useful in assessing the suitability of a particular hair for DNA profiling.

METHODS AND MATERIALS

The hairs used in this study were taken from the scalp of a 19-year-old Caucasian female murder victim who had been buried 36 years (16). The body had been embalmed and buried in a coffin. A random subsample of hairs was mounted in Permunt® (Fisher Scientific) and examined with transmitted light microscopy. An additional random subsample of hairs was mounted on aluminum SEM sample stubs (Ted Pella, Inc.) using colloidal graphite (Ted Pella, Inc.). The hairs were then coated with an approximately 367-Å layer of gold and palladium using a Polaron® SC7620 Mini Sputter Coater (Quorum Technologies, Ltd.). The gold-palladium-coated hairs were examined with a Hitachi S-2400 scanning electron microscope (Hitachi



Figure 2. SEM micrograph showing disrupted cuticular scales, 1000x magnification at 25kV.

High-Technologies Corp.). In order to preserve adherent material such as fungal penetrating organe, none of the hairs were cleaned.

RESULTS

The SEM micrographs show a range of surficial alterations. In some areas, the cuticular scales were intact (Figure 1); in others some of the scales had become detached from the surface of the hair (Figures 2 and 3). Occasional masses of fungal hyphae could be seen (Figure 4). A fungal penetrating organ was found in situ penetrating perpendicularly into the shaft of a hair (Figure 5). There was also indirect evidence of the presence of fungi in the form of “craters” in the hair surface (Figure 6). Figure 6 is also notable for two additional features. First, the cuticular scales have been completely lost from the hair in this area. Second, the hair surface around the “crater” shows an interesting pattern of small, shallow pits.

Handling of the hairs in preparing them for microscopical examination showed them to be somewhat fragile with a tendency to break into short segments at specific weakened points. Microscopical examination confirmed that damage to the hairs tended to be focal (that is, confined to narrow specific regions of the hair shafts). Figure 7 shows a region of hair shaft in which all hair structures—cuticle, cortex and medulla—are intact. Figure 8 shows a region of damage extending almost completely through the hair shaft and having dimensions similar to the hair’s diameter. In the most extreme cases of damage, segments of hair were joined only by a thin bridge of cortical and cuticular cells



Figure 3. SEM micrograph showing peeling away of cuticular scales, 1500x magnification at 25kV.

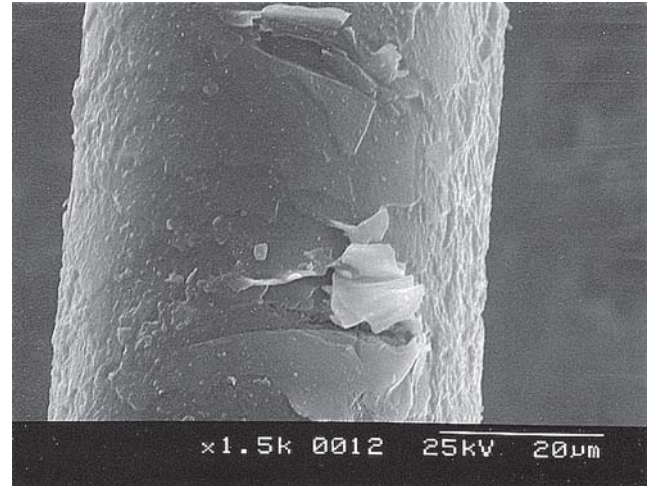


Figure 4. SEM micrograph showing an adherent mass of fungal penetrating organ, 2000x magnification at 25kV.

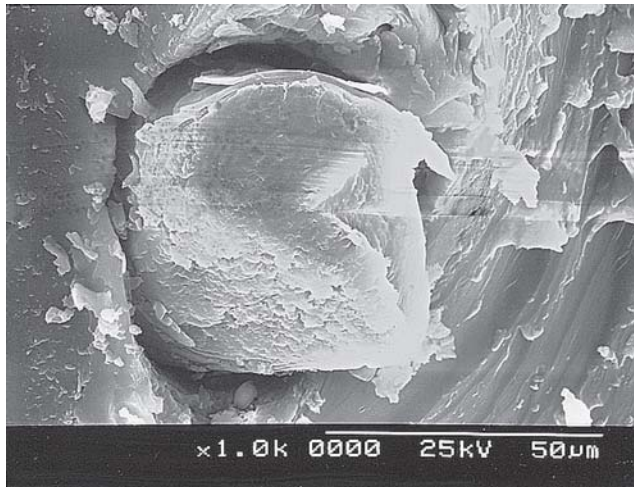


Figure 5. SEM micrograph showing a fungal penetrating organ penetrating the shaft of the hair, 1000x magnification at 25kV.

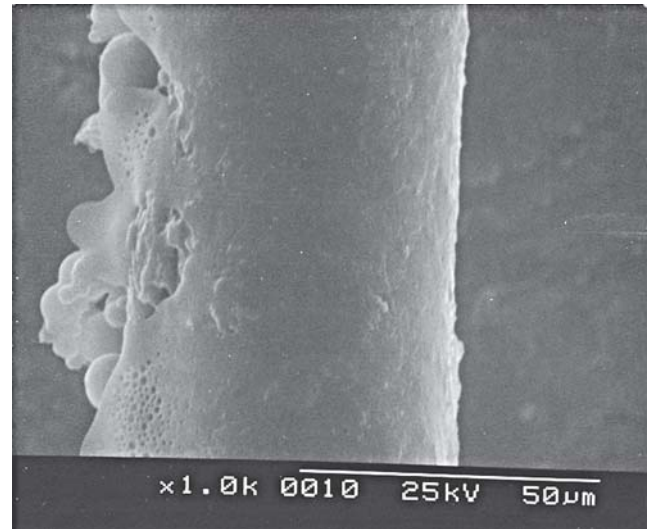


Figure 6. SEM micrograph showing "crater" produced by fungal attack, 1000x magnification at 25kV.

(Figure 9). Three types of fungal tunnels were observed: long, narrow tunnels having uniform diameters; broad conical tunnels having diameters that decreased with depth of penetration into the hairs; and tunnels having diameters that increased with depth of penetration (Figure 10). The broad conical tunnels appeared as lighter colored oval spots on the hair due to removal of pigment granules. Even hairs that retained a modicum of structural integrity often showed severe internal damage due to fungal tunneling. In these hairs fungal hyphae had penetrated to the centers of the hairs and then destroyed the center of the hair shaft, producing large cavities (Figure 11). Some hair shafts were

entirely cored out by hyphae, resulting in a "soda straw" effect (Figure 12).

DISCUSSION

The different types of biodeterioration observed in these hairs are consistent with those observed in laboratory studies and studies of archaeological hair samples. Hairs gradually lose their outer layers of scales. Fungi attach to small areas on the surfaces of the hairs; some of their hyphae grow along the hair surface and others penetrate perpendicularly into the shafts of the hairs. In laboratory studies, keratinophilic

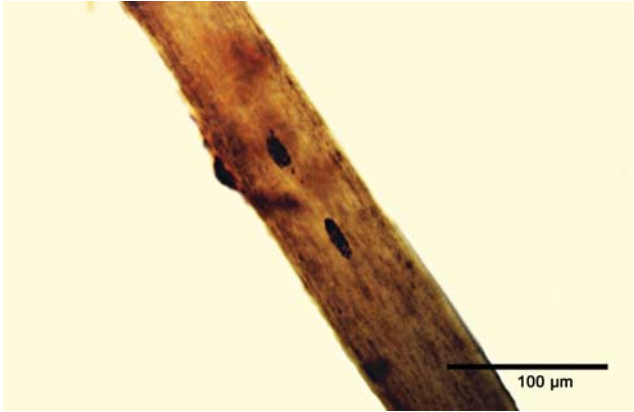


Figure 7. Human head hair showing an area in which no visible damage has occurred.

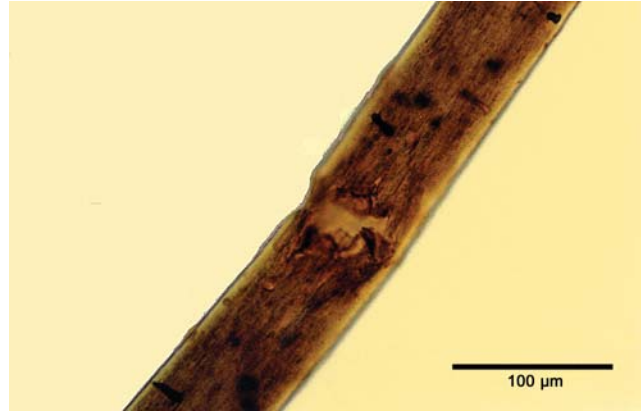


Figure 8. Human head hair showing focal destruction of the hair shaft.

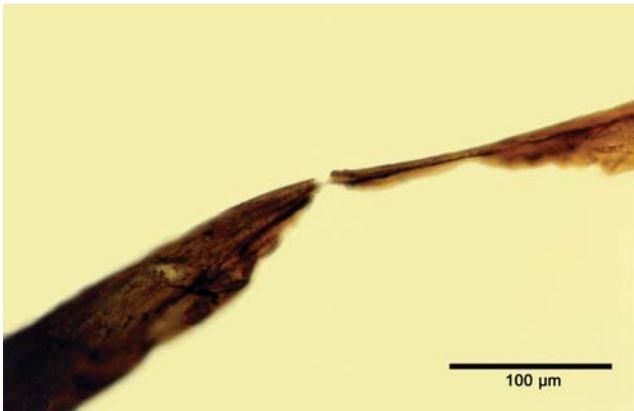


Figure 9. Human head hair held together by narrow strip of cells.

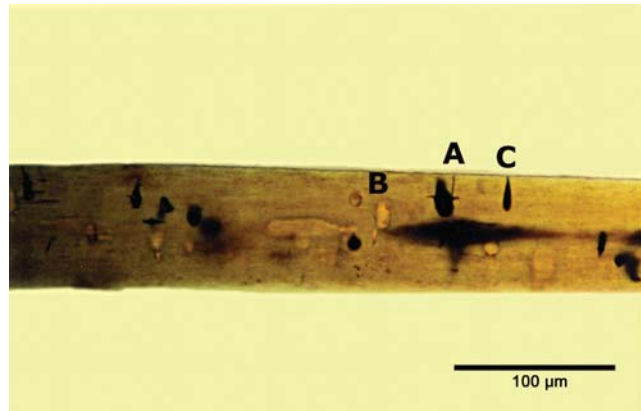


Figure 10. Human head hair showing three types of tunnels: Narrow tunnel with uniform diameter (A); broad, conical tunnels (B); and tunnel with diameter increasing with depth of penetration (C).

fungi have been observed to produce narrow, thread-like tunnels (13, 17). These fungi grow on hair but do not consume the keratin comprising hair. In contrast, keratinolytic fungi produce wide, conical tunnels. These fungi actually digest keratin. Once the hyphae of keratinolytic fungi reach the center of the hair they grow laterally, first attacking the medullary cells and then the inner surface of the cortex.

The SEM micrograph of a fungal penetrating organ (Figure 5) provides insight into the mechanism by which keratinolytic hyphae penetrate hair shafts. The disruption of both the cuticular scales and the cortical cells at the point of entry is minimal. The enzymes produced by the penetrating organ have dissolved the cuticular and cortical cell keratin at the point of contact. Such localized dissolution of keratin may also account for the pattern of small, shallow pits seen in Figure 6.

The extent of fungal attack on the specimen hairs provides some insight into post-burial environmen-

tal conditions in the deceased person's coffin. Fungal growth requires moisture, favorable temperatures and a growth substrate which provides both energy and carbon for structural proteins and enzymes. The degree of fungal attack on the specimen hairs shows that conditions initially favorable for fungal growth were present. Then, at some point, growth ceased. Because the hairs were not completely consumed they clearly were not the limiting resource for fungal growth in this case. The deceased in this study died in January and was probably buried shortly thereafter. Clearly, the temperature inside the coffin did not completely inhibit fungal growth; it started but eventually came to a stop.

The most plausible factor limiting fungal growth in this case is moisture. Although modern embalming techniques remove most of the blood and the fluids from internal organs, embalmed human remains still retain substantial amounts of water in soft tissue.



Figure 11. Human head hair showing large cavities.

Water from the remains would initially raise the relative humidity of the air in the coffin and promote fungal growth. Eventually, much of the moisture would escape from the coffin into the surrounding soil, and fungal growth would cease.

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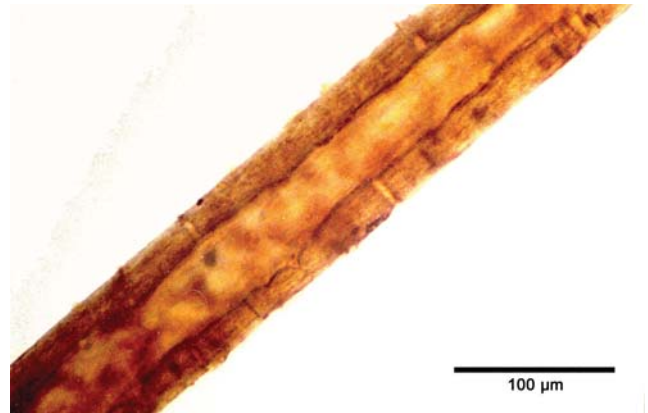


Figure 12. Human head hair reduced to hollow tube or soda straw.

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