

**TECHNICAL NOTE**  
**CRIMINALISTICS**

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## Chemical Differences Are Observed in Children's Versus Adults' Latent Fingerprints as a Function of Time\*

**ABSTRACT:** The identification of aged latent fingerprints is often difficult, especially for those of children. To understand this phenomenon, the chemical composition of children's versus adults' latent fingerprints was examined over time using Fourier transform infrared microscopy. Hierarchical cluster analysis revealed that children's and adults' prints were distinguishable for up to 4 weeks after deposition, based on differences in sebum composition. Specifically, adults had a higher lipid content than children, but both decreased over time, attributable to the volatility of free fatty acids. The aliphatic CH<sub>3</sub>, aliphatic CH<sub>2</sub>, and carbonyl ester compositions changed differently in adults versus children over time, consistent with higher cholesterol and cholesteryl esters in children's prints and wax esters and glycerides in adults' prints. Thus, fingerprint composition changes with time differently in children versus adults, making it a sensitive metric to estimate the age of an individual, especially when the age of the print is known.

**KEYWORDS:** forensic science, latent fingerprints, chemical composition, children, Fourier transform infrared microscopy, hierarchical cluster analysis, wax esters, cholesteryl esters, squalene, cholesterol, free fatty acids

Traditional visualization methods of latent fingerprints, such as magnetic filings dusting, iodine, and cyanoacrylate fuming, are widely used in the forensic science field (1). These methods, though efficient, inexpensive, and relatively fast, can be limiting to an investigation when attempting to preserve valuable trace evidence found in a latent fingerprint. Thus, efforts are underway to find alternative latent fingerprint visualization methods, using instrumentation that is nondestructive to the fingerprint.

One particular situation where latent fingerprints are difficult to identify is the case of aged prints, especially those of children. Several studies have shown that children's prints often "disappear" faster than adults', making them inadequate for lifting after being dusted or fumed (2–5). For example, a seminal study using gas chromatography–mass spectrometry (GC–MS) showed that children's fingerprints disappeared faster than adults' because they contained more volatile fatty acids (3). In contrast, adults' fingerprint residue contained fatty acid esters, which are higher in molecular weight and have low volatility. This investigation spurred a slew of later experiments using various instrumentation techniques to investigate the chemical composition of latent fingerprints such as

GC–MS (6–8), ultraviolet fluorescence spectroscopy (9,10), and x-ray fluorescence microscopy (11).

Fourier transform infrared spectroscopy (FTIR) is a noninvasive and quantitative technique that has been used to study the composition of latent fingerprints based on their unique vibrational spectroscopic signatures. It has been used to determine the lipid composition in fingerprint residue (12,13) and as a biometric gauge of an individual's age (14). For the analysis of small and/or heterogeneous samples, IR light can be focused through an IR microscope for a spatial resolution of a few microns (15). Fingerprints are naturally heterogeneous materials, containing small particles of skin, droplets of sebum, and sweat residue. Thus, analysis of fingerprint composition with an FTIR microscope provides the added advantage of being able to examine individual fingerprint components separately. FTIR microscopy (FTIRM) has been used to examine fingerprint composition on various surfaces (16–18) and for contaminants (19,20).

In this study, the chemical composition of children's and adults' latent fingerprints was examined over the course of 4 weeks using FTIRM and these results were compared with conventional dusting methods. The goal of this work was to determine how specific components of fingerprint composition (i.e., skin, sebum, sweat) change over time in adults versus children, and how these changes influence the ability to predict the age of an individual based on his/her fingerprint.

### Materials and Methods

#### Sample Preparation

Six father (ages 35–45 years) and son (ages 7–10 years) pairs were asked to provide fingerprints for this 4-week study. For each

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participant, the hands were first washed with soap and water and dried thoroughly. Participants were then asked to touch their face with an index finger, and then place that finger onto an infrared-reflective (MirrIR) glass microscope slide (Kevley Technologies, Chesterland, OH). This procedure was then repeated 10 times, where subsequent prints were placed on conventional glass microscope slides. For each print, the same index finger was used and the face was touched between each deposition. A total of eleven fingerprints were collected per person (one on a MirrIR slide and 10 on conventional glass slides). All prints were stored at room temperature (22°C) and a relative humidity of 20% for the duration of the experiment. Fingerprints were analyzed by dusting and FTIRM twice a week for 4 weeks as described below. On week 4, the remaining prints were heated for 24 h at 43.3°C. This was done in order to test whether changes in temperature affect the latent fingerprint composition.

### Dusting

Twice a week, one fingerprint from each participant was dusted using black magnetic fingerprint powder (Lightning Powder Company, Salem, OR). The print was lifted with transparent tape, placed into a notebook, and labeled. After each dusting, the darkness of the print was scaled by counting the number of distinguishable minutiae that were clearly visible.

### FTIR Microscopy

For each participant, the fingerprint that was deposited on the infrared-reflective glass slide was analyzed using a Perkin Elmer Spectrum Spotlight FTIR Imaging System. For each print, 3–5 skin particles and 3–5 sebum droplets were identified and their stage coordinates on the FTIR microscope were recorded so that the same regions could be analyzed for changes during the course of the experiment. Salt deposits from sweat were not probed in this study because there were insufficient areas in the children's fingerprints.

Spectra from skin and sebum areas were collected in transmittance mode from 4000 to 800  $\text{cm}^{-1}$  using a  $25 \times 25 \mu\text{m}$  square aperture. For each spectrum, 64 scans were co-added using a spectral resolution of 4  $\text{cm}^{-1}$ .

### Data Analysis

After data collection, spectra were exported to Thermo Nicolet Omnic Macros Basic for analysis. For each skin spectrum, the protein and lipid areas were integrated as shown in Table 1. For the sebum spectra, only the lipid areas (full,  $\text{CH}_3$ , and  $\text{CH}_2$ ) were calculated. After integration, a series of ratios were calculated for each spectrum: lipid/protein,  $\text{CH}_3$ /lipid,  $\text{CH}_2$ /lipid, and carbonyl ester/lipid. Ratios were generated in order to normalize to the total protein or lipid content. For the fathers and sons at each time point, ratios were averaged (mean  $\pm$  SE), and plotted as a function of time.

Hierarchical cluster analysis (HCA) was also performed on the averaged spectra at each time point using OPUS software (Bruker Optics, Billerica, MA). All spectra were first vector normalized to account for variations in sample thickness. Ward's algorithm was used to calculate the heterogeneity between clusters to generate a dendrogram. The heterogeneity between clusters indicates the degree of spectral similarity within the given spectral region. Clus-

TABLE 1—FTIR parameters used to analyze the skin and sebum spectra.

Region	Range ( $\text{cm}^{-1}$ )	Baseline ( $\text{cm}^{-1}$ )
Lipid	2750–3100	2750–3100
$\text{CH}_3$	2948–2953	2750–3100
$\text{CH}_2$	2918–2928	2750–3100
Carbonyl ester	1125–1210	1125–1210
Protein	1585–1480	1585–1480

### Results

Fingerprint residue is composed of three main components: skin, sebum, and sweat (5). All three components are visibly distinguishable with light microscopy. Moreover, since the chemical makeup of these components is different, they each have unique FTIR spectra, as can be seen in Fig. 1. Skin cells in a fingerprint are those sluffed from the outermost epidermis and consist mainly of protein. The repeating amide-bond backbone of proteins gives rise to their unique FTIR spectra (15). Specifically, the amide I band between 1700 and 1600  $\text{cm}^{-1}$ , arises from the C = O stretching vibration of the amide bond. The amide II band (1580–1480  $\text{cm}^{-1}$ ) is assigned to a combination of N-H bending and C-N stretching in the amide bond.

Sebum is an oily substance produced by the sebaceous glands and is composed largely of a variety of lipids (21). Sweat found in the residue of a fingerprint is secreted by eccrine sweat glands and is mainly composed of various organic and inorganic salts (12). The sebum spectrum contains characteristic lipid peaks, including the large  $\text{CH}_3$  and  $\text{CH}_2$  symmetric and antisymmetric peaks from 3100 to 2700  $\text{cm}^{-1}$  and peaks centered near 1740 and 1180  $\text{cm}^{-1}$  attributed to the C = O and C-O vibrations from carbonyl esters (e.g., wax esters and cholesteryl esters), respectively. Sweat spectra can also be identified by the characteristic carboxylic acid peak (COOH) from lactic acid between 1500 and 1600  $\text{cm}^{-1}$ . The broad peak centered around 3300  $\text{cm}^{-1}$  in both the skin and sweat spectra arises from hydrogen-bonded OH groups in both proteins and lactic acid.

Since individual peaks in an FTIR spectrum represent different chemical components, chemical images can be generated from an entire fingerprint. For example, by integrating the protein peak or lipid peak, chemical images of the entire fingerprint's skin (Fig. 2a) or sebum (Fig. 2b) were generated, respectively.

Conventional dusting of the adults' and children's fingerprints over the 4-week duration of the experiment is shown in Fig. 3. At all time points, the fathers' prints (Fig. 3a) dusted darker than the sons' prints (Fig. 3b). As time progressed, the fathers' prints remained visibly unchanged, while the fine minutiae of the sons' prints became more difficult to visualize.

HCA was performed on the time-point-averaged father and son spectra in the lipid, protein, and carbonyl ester regions (Fig. 4). The resulting dendrograms clearly show two distinct clusters separating the fathers' and sons' lipid spectra for both skin (Fig. 4a) and sebum (Fig. 4b) regardless of time (i.e., fingerprint age). A similar trend was observed for the carbonyl ester dendrogram from the sebum spectra (Fig. 4c). The fathers' and the sons' spectra did not cluster separately in the protein spectral region from the skin spectra (Fig. 4d).

Once it was clear that the fathers' and sons' prints were distinguishable through HCA, further analysis was performed to evaluate the change in fingerprint composition over time and how it influenced the ability to distinguish a child's from an adult's finger-

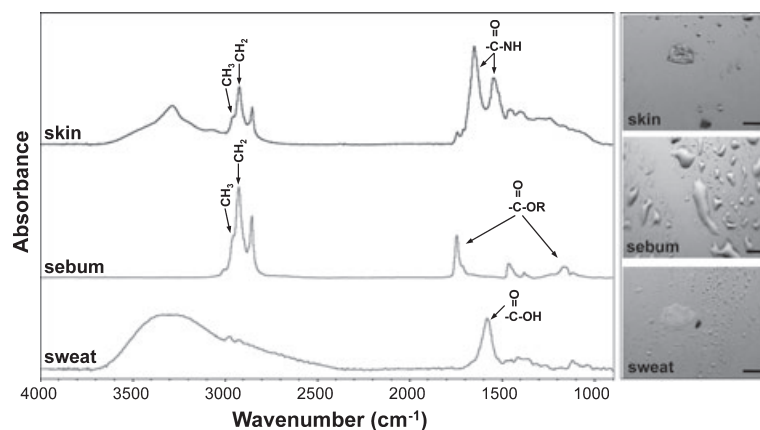


FIG. 1—FTIR spectra of skin, sebum, and sweat with their corresponding light micrographs. Scale bar: 20 μm.

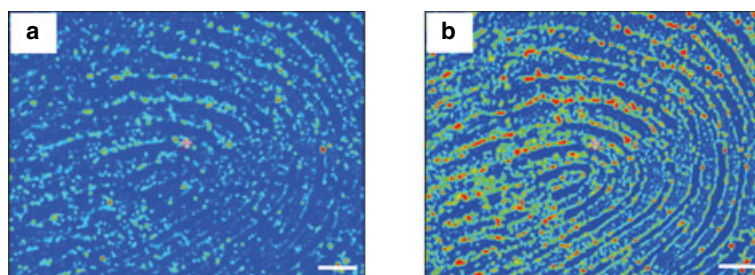


FIG. 2—FTIR images of a child's fingerprint generated by integrating the (a) protein and (b) lipid spectral regions. Scale bar: 500 μm.

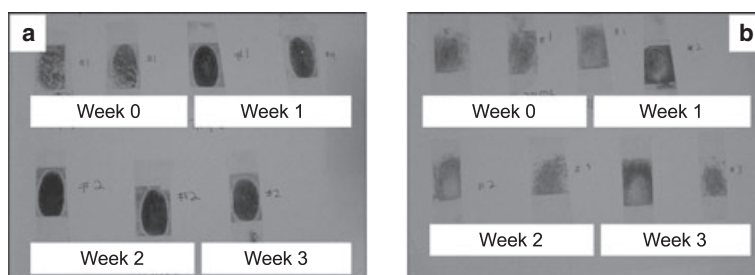


FIG. 3—Light micrographs of the dusting results over the course of the experiment for the (a) fathers and (b) sons.

fathers had a significantly higher lipid content in the skin than the sons at all time points. Over the 4-week duration of this study, both the fathers' and sons' lipid/protein ratio decreased at a similar rate.

To characterize the type of lipids present in the sebum, the CH<sub>3</sub> and CH<sub>2</sub> peak areas were calculated and normalized to total lipid content. Specifically, the relative abundances of short- and long-chain lipids, and branched versus straight-chain lipids, were evaluated by calculating the CH<sub>3</sub>/lipid and CH<sub>2</sub>/lipid peak areas as a function of time (Fig. 5b–d). At early time points, the sons had a lower CH<sub>3</sub>/lipid ratio than the fathers (Fig. 5b). However, this ratio increased steadily over time for the sons, such that they were indistinguishable from the fathers after 4 weeks. For the fathers, this ratio remained unchanged over time. For the CH<sub>2</sub>/lipid ratio, the fathers' values were consistently lower than the sons', and both increased slightly over time (Fig. 5c). Analysis of

the amount of carbonyl esters increased in the sons while they decreased in the fathers. Thus, after 4 weeks, the sons and fathers had significantly different carbonyl ester content in their fingerprint residue.

## Discussion

Sebum is the main oily component of latent fingerprints and it is composed mainly of 30% free fatty acids, 33% glycerides, 22% wax esters, 10% squalene, and 5% cholesterol and hydrocarbons (22). The fingerprint dusting technique relies on the mechanical adherence of fingerprint powder to the moisture and sebum components of the skin ridge deposits such that the dusting intensity is determined by the moisture and oil concentration of the print donor (23). A number of studies have shown that children produce less sebum than adults until they reach puberty (24–29) when the seba-

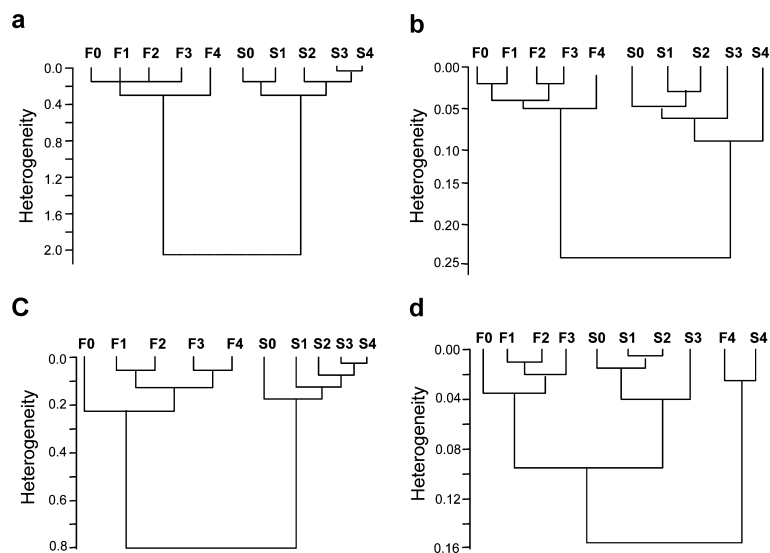


FIG. 4—HCA for the (a) lipid composition in the skin, (b) lipid composition in the sebum, (c) carbonyl ester composition in the sebum, and (d) protein composition in the skin.

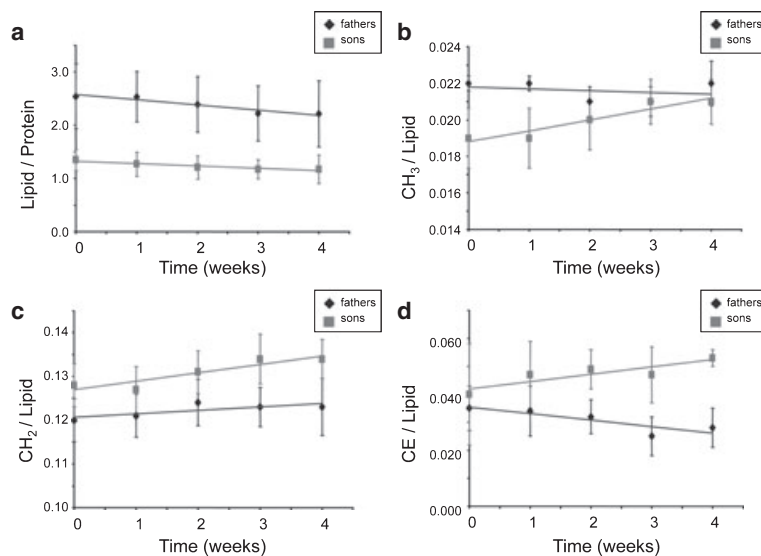


FIG. 5—FTIR parameters (mean  $\pm$  SE) for the fathers ( $\blacklozenge$ ) versus sons ( $\blacksquare$ ) as a function of time for the (a) lipid/protein ratio, (b)  $\text{CH}_3$ /lipid ratio, (c)  $\text{CH}_2$ /lipid ratio, and (d) carbonyl ester (CE)/lipid ratio.

adult prints dusted darker across all time points and the lipid/protein ratio was consistently higher, respectively.

Not only does sebum concentration change with age in the fingerprints, the lipid composition has also been shown to differ between adults and children (24,26,27). In this study, we used HCA to examine whether these differences could be observed with FTIRM. The HCA performed on the lipid and carbonyl ester regions demonstrated that there was a significant difference between adults' and children's sebum and skin, especially in the lipid and carbonyl ester spectral regions. Similar findings were observed using principal components analysis (PCA) recently (14).

To further understand the specific compositional differences, the individual contributions from the  $\text{CH}_3$  and  $\text{CH}_2$  groups were examined in the FTIRM spectra. For short-chain and/or highly branched

long- and/or straight-chain lipids should have a higher fraction of  $\text{CH}_2$  groups in the lipid spectral region.

In the sebum of postpubertal individuals, more highly branched lipids have been observed, consisting of squalene, wax esters, and branched fatty acids (14,27). In the FTIRM data, these highly branched lipids resulted in a higher fraction of  $\text{CH}_3$  groups and lower fraction of  $\text{CH}_2$  groups in the adult sebum. Conversely, children's sebum has been shown to have high concentrations of long-chain fatty acids, cholesterol, and cholesteryl esters (30,31). Here, the children's prints were found to have a higher fraction of  $\text{CH}_2$  groups and a lower fraction of  $\text{CH}_3$  groups than the adults, consistent with a greater number of straight-chained lipids and cholesterol.

Over time, the lipid concentration and composition changed in

spectrometry studies and attributed to the volatility of low molecular weight components (2,3,32,33). These components are most likely free fatty acids, which represent ~30% of the sebum composition (22) and have been shown to be the most volatile components by thin layer chromatography (33). More specifically, the free fatty acids are primarily straight-chain aliphatic molecules that vary in length, where the shorter chain molecules have the highest volatility. The FTIRM data show an increased CH<sub>2</sub> fraction over time especially in children's prints, consistent with a disappearance of the shorter chain fatty acids and retention of longer fatty acids, the latter of which have a higher fraction of CH<sub>2</sub> groups per lipid molecule.

There were noticeable compositional differences between the fathers' and sons' prints over the course of the experiment. For example, we found that the fraction of CH<sub>3</sub> groups in the children's sebum increased but remained constant in adults. Children's prints have a high concentration of cholesterol, unlike adults (30,31). Since cholesterol is not observed in aged prints (33), and its molecular structure has a low CH<sub>3</sub>/lipid ratio, we suggest that the increased CH<sub>3</sub> fraction in children's prints over time may arise from the disappearance of cholesterol over time. Conversely, adults' prints have very little cholesterol, which is consistent with the unchanged CH<sub>3</sub> fraction over time.

We find that the carbonyl ester fraction was similar between adults and children at the start of the experiment, but became increasingly different over the course of 4 weeks. This was likely due to different sources of carbonyl esters in adults' versus children's prints. Specifically, the carbonyl ester contribution from the children's prints is attributable to a high concentration of cholesteryl esters, which are highly stable over time (2,24,28). On the other hand, the carbonyl ester contribution in the adults' prints arises primarily from wax esters. Wax esters are composed of a wide range of esterified fatty acids, making their volatility more variable than cholesteryl esters. Since the adults' carbonyl ester contribution decreases over time, these findings suggest the disappearance of more volatile wax esters.

In summary, this study showed that fingerprints change composition significantly over time, and these changes are different in children versus adult prints. Specifically, the results indicate that all sebum contains a high content of volatile fatty acids that cause fingerprints to "disappear" over time. However, children's sebum also contains a higher proportion of cholesterol, cholesteryl esters, and straight-chain fatty acids that are different in stability from the squalene, wax esters, and branched fatty acids contained in adults' sebum. Based on these differences, children's prints can still be distinguished from adults' prints even 4 weeks after deposition. These findings support recent work demonstrating the ability to classify the age of a person based on the FTIR spectrum of his/her fingerprint (14), and confirm that this can even be done for aged prints. However, care should be taken when gauging the age of an individual through FTIR analysis, where the age of the fingerprint itself should first be determined if possible. Thus, FTIRM is a complementary tool to conventional dusting methods because its non-invasive nature makes it useful for preserving trace evidence, while the spectral features provide unique information on sebum composition.

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