Nghiên cứu phân tích và đánh giá mức độ phơi nhiễm, đặc trưng tích lũy hydrocacbon thơm đa vòng (PAHs) trong mẫu tóc và móng tay người

TÓM TẮT

Hydrocarbon thơm đa vòng (PAHs) có khả năng tích lũy trong một số mẫu sinh phẩm người không xâm lấn và các loại mẫu này có thể được sử dụng làm chỉ thị sinh học để đánh giá mức độ phơi nhiễm các độc chất này ở người. Trong nghiên cứu này, mẫu tóc và móng tay của một nhóm sinh viên đại học tại Việt Nam được thu thập và phân tích nhằm cung cấp các thông tin quan trắc sinh học về mức độ tích lũy và đặc trưng ô nhiễm PAHs. Đây là nghiên cứu đầu tiên tại Việt Nam đánh giá đồng thời PAHs trong tóc và móng tay trên cùng một đối tượng nghiên cứu. Hàm lượng tổng của 16 PAHs trong mẫu tóc dao động từ 258 đến 811 (trung bình 493) ng/g, trong khi hàm lượng trong móng tay dao động từ 29 đến 667 (trung bình 335) ng/g. Mặc dù hàm lượng tổng PAHs trong tóc có xu hướng cao hơn nhưng móng tay lại giàu các PAHs khối lượng phân tử lớn có độc tính cao, dẫn đến giá trị độ độc tương đương quy về benzo[a]pyrene (BaP-EQ) cao hơn so với mẫu tóc. Ngược lại, một số mẫu tóc chủ yếu tích lũy các PAHs khối lượng thấp như naphthalene và phenanthrene, phản ánh sự phơi nhiễm từ môi trường không khí hoặc do sự tích lũy nội sinh trong quá trình hình thành sợi tóc. Một số mẫu móng tay, đặc biệt ở nữ giới, ghi nhận hàm lượng BaP-EQ cao hơn nhiều lần so với mẫu tóc tương ứng, cho thấy vai trò của cơ chế hấp phụ ngoại sinh từ bụi và sự tiếp xúc tay với các bề mặt có nguy cơ ô nhiễm PAHs.

Từ khóa: *PAHs, tóc, móng tay, Việt Nam, GC-MS.*

Study on Analysis and Assessment of Exposure Levels and Accumulation Profiles of Polycyclic Aromatic Hydrocarbons (PAHs) in Human Hair and Fingernail Samples

ABSTRACT

Polycyclic aromatic hydrocarbons (PAHs) can accumulate in some non-invasive human samples, which can serve as bioindicators for assessing human exposure to these contaminants. In this study, hair and fingernail samples collected from a group of university students in Vietnam were collected and analyzed to provide biomonitoring information on the accumulation and contamination characteristics of PAHs. This is the first study in Vietnam to simultaneously analyze PAHs in hair and fingernails obtained from the same study subjects. Total concentrations of 16 PAHs in the hair samples ranged from 258 to 811 (mean 493) ng/g, while those in fingernails ranged from 29 to 667 (mean 335) ng/g. Although total PAH levels in the hair samples were slightly higher than in the fingernails, the latter matrix preferably accumulated high-molecular-weight PAHs with higher toxicity, resulting in higher benzo[a]pyrene equivalents (BaP-EQ) in the fingernails than in hair samples. In contrast, many hair samples accumulated low-molecular-weight PAHs such as naphthalene and phenanthrene, implying the exposure from the air environment and/or endogenous accumulation during hair growing process. Some fingernail samples, especially in women, showed BaP-EQ levels several times higher than the corresponding hair samples, suggesting the role of exogenous adsorption from dust and hand contact with potentially PAH-contaminated surfaces.

Keywords: *PAHs, nail, hair, Vietnam, GC-MS.*

1. INTRODUCTION

As a typical class of organic pollutants, polycyclic aromatic hydrocarbons (PAHs) have attracted considerable research attention over the past decades due to their adverse effects on both human health and the environment.[1] Some PAHs, notably benzo[a]pyrene, have been identified as carcinogenic and mutagenic to humans.[2] In terms of origin, PAHs can arise from both natural and anthropogenic processes.[2] While some natural formation may occur through endogenous synthesis in plants and microorganisms, the majority of PAHs are generated from anthropogenic activities, particularly through incomplete combustion or pyrolysis of organic matter under high temperatures.[3] Humans are exposed to these contaminants mainly through inhalation of polluted air, ingestion of contaminated food and water, dust and soil ingestion, and dermal contact with PAH-containing materials.[4] These accidential exposure events can directly impact public health, especially people affected by extensive industrial, transportation, domestic, and waste processing activities.[5] Exposure assessment and biomonitoring of PAHs in the human body is an essential task.

The exposure assessment can be divided into two approaches: external exposure and internal exposure. The external exposure assessment in humans is the process of predicting or estimating the amounts or doses of pollutants enter the human bodies through contacting with environmental media (such as air, soil, or water), based on contamination degree, exposure frequency, and duration.[6] The internal exposure is performed by determining pollutants and their metabolites in human samples such as urine[7,8], blood[9,10], or breast milk[9]. Considering the toxicology of PAHs, human biomonitoring plays a crucial role in assessing the accumulation of these substances in the body and the potential health risks.[11] However, collection and storage of human samples require critical protocols under strict conditions.[11] To overcome limitations related to conventional biological matrices, alternative approaches using matrices such as hair or nails have been proposed as non-invasive human biomonitoring. These matrices can reflect average exposure concentrations over longer periods and are less influenced by short-term fluctuations.[12] Additionally, these samples are easily collected and stored, showing insignificant impacts on the health and spirits of the donors.[13]

In this study, we developed and applied a rapid, simple, and reliable analytical method to determine 16 priority PAHs in hair and fingernail samples collected from male and female students at several universities in Hanoi. The analytical procedure consisted of four main steps: (1) alkaline digestion of the samples, (2) extraction with an organic solvent, (3) clean-up using solid-phase extraction (SPE) cartridges, and (4) quantitative analysis by gas chromatography-mass spectrometry (GC-MS). This study aimed to provide a preliminary assessment of PAH contamination and accumulation in these two biological matrices, reflecting human exposure degree and tissue-specific accumulation pattern. To the best of our knowledge, this study is among the first studies in Vietnam to simultaneously elucidate PAH exposure in human hair and fingernail samples.

2. MATERIALS AND METHOD

2.1. Samples collection

The hair and fingernail samples were collected in June 2024 from students at universities in Hanoi. Prior to sample collection, all participants were clearly informed about the purpose of the study and agreed to provide samples. The samples were obtained from 10 volunteers, including 5 males and 5 females. The sample categories were assigned as MH (male hair samples), FH (female hair samples), MN (male nail samples), and FN (female nail samples). The hair samples were cut approximately 2–3 cm from the scalp, while the fingernail samples were clipped using stainless steel clippers. The collected samples (about 0.5 g) were stored in polyethylene zip-lock bags at room temperature until analysis.

2.2. Standards and chemicals

The native PAH standard mixture (H-QME-01 Quebec PAH Mix; AccuStandard, USA) contained naphthalene (Nap), acenaphthylene (Acy), acenaphthene (Ace), fluorene (Flu), phenanthrene (Phe), anthracene (Ant), fluoranthene (Flt), pyrene (Pyr), benz[a]anthracene (BaA), chrysene (Chr), benzo[b]fluoranthene (BbF), benzo[k]fluoranthene (BkF), benzo[a]pyrene (BaP), dibenz[a,h]anthracene (DA), indeno[1,2,3-cd]pyrene (IP), and benzo[ghi]perylene (BP) at a concentration of 500 µg/mL each compound. An deuterated surrogate standard mixture (ES-2044 Surrogate Cocktail; Cambridge Isotope Laboratories, USA) contained Nap-d_8, Acy-d_8, Phe-d_{10}, Pyr-d_{10}, BaP-d_{12}, and BP-d_{12} at a concentration of 200 µg/mL each compound. The internal standard solution contained Chr d_{12} (DLM-261-1.2; Cambridge Isotope Laboratories, USA) at a concentration of 200 µg/mL. Working standard solutions for native, surrogate, and internal standards at a concentration of 100 ng/mL were prepared from the stock standards using hexane as the solvent. The calibration standards were prepared in hexane over a concentration range of 10–500 ng/mL.

Chromatographic-grade solvents used in this study included acetone (99.5%; Macron Fine Chemicals™, Germany), hexane (96%; Daejung, South Korea), dichloromethane (DCM) (99.5%; Daejung, South Korea), and ethyl acetate (EA, 99.98%; Fisher Chemical, Mexico). Sodium hydroxide (>98.5%) and sodium sulfate (>99%) were obtained from Sharlau (Spain). A 1 M sodium hydroxide solution for sample digestion was prepared by dissolving 2 g of NaOH in 50 mL of deionized water. Bond Elut SI silica gel SPE cartridges (500 mg, 3 mL, 120 µm) were purchased from Agilent Technologies (USA).

2.3. Chemical analysis

The nail and hair samples (0.2–0.4 g) were cut into small pieces (<0.2 mm), weighed into 10 mL glass tubes, and spiked with 200 µL of the surrogate solution (100 ng/mL). The samples were then digested with 5 mL of 1 M NaOH solution at room temperature, vortexed for 1 min, ultrasonicated for 10 min, and left to stand for 24 h for compete digestion. The sample mixture was then added with 2 mL of hexane, vortexed for 1 min, and centrifuged at 3500 rpm for 5 min. The upper organic phase was collected and transferred to a clean 10 mL glass tube. The extraction process was repeated two more times. The combined organic extract (approximately 6 mL) was dehydrated with anhydrous sodium sulfate, vortexed for 1 min, and centrifuged at 3500 rpm for 5 min. The dried extract was transferred to a concentrator tube and evaporated under a gentle nitrogen stream to 500 µL. The sample cleanup was performed using a silica gel SPE cartridge. The cartridge was preconditioned with 6 mL DCM and 6 mL hexane. The concentrated extract (500 µL) and rinsate (500 µL) were loaded onto the cartridge, and this loading sample was discarded. PAHs were eluted with 3 mL of DCM/hexane (1:3, v/v) mixture. The eluate was

spiked with 200 µL of internal standard solution (100 ng/mL) and concentrated to 200 µL before GC-MS analysis.

2.4. Instrumental analysis

PAHs were separated and quantified using a gas chromatograph (GC 8890, Agilent Technologies) coupled with a triple quadrupole mass spectrometer (7010B GC/TQ, Agilent Technologies) equipped with a DB-5ms capillary column (30 m × 0.25 mm × 0.25 µm; Agilent Technologies). The column oven temperature was programmed as follows: initial temperature 80 °C (held for 1 min), to 170 °C (20 °C/min), to 220 °C (4 °C/min), to 270 °C (3 °C/min), to 310 °C (20 °C/min, held for 20 min). Helium was used as the carrier gas at a flow rate of 1.2 mL/min. Samples (1 µL) were injected in splitless mode. The interface and ion source temperatures were set at 310 °C and 230 °C, respectively. The mass spectrometer operated in electron impact (EI) ionization mode at 70 eV, and data acquisition was performed in selected ion monitoring (SIM) mode.

2.5. QA/QC

Method blanks (n = 5) were analyzed in parallel with real samples to control for potential PAH contamination during the analytical procedure. The method detection limit (MDL) was determined using the formula: MDL = C_B + 3 × S_B, where C_B is the mean concentration of PAHs in the blanks and S_B is the corresponding standard deviation. The MDLs for PAHs ranged from 0.04 to 10 ng/g. The recoveries of PAHs in spiked samples (67–102%) and surrogate compounds in real samples (60–121%) satisfied the acceptance criteria of the AOAC International and the US Environmental Protection Agency (US EPA) for analytes at ppb levels. Method repeatability, evaluated using spiked samples at 20 and 50 ng/g (n = 3 for each level), showed relative standard deviations (RSD) of less than 25%.

3. RESULTS AND DISCUSSIONS

3.1. Concentrations of PAHs in hair samples

Total concentrations of 16 PAHs in the hair samples ranged from 258 to 811 (mean 493) ng/g, as presented in Figure 1. The highest level was observed in sample FH1 (811 ng/g), while the lowest was found in sample FH4 (258 ng/g). The mean PAH concentration in female hair samples (544 ng/g) was higher than that in the male hair samples (441 ng/g). However, this difference was not statistically significant (Mann-Whitney U test, $p > 0.05$), probably due to high variation within each group and the limited sample size. One possible explanation for higher PAH levels in female hair than in male hair is the higher frequency of haircuts in men compared to women, resulting in longer accumulation duration for PAHs in female hair.

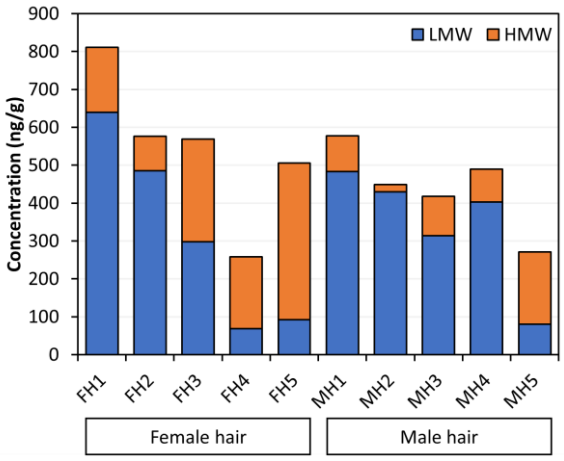

Figure 1. Concentrations of PAHs in hair samples

The PAH concentrations detected in our study were in line with those previously reported for hair samples collected from Vietnamese people aged 16 to 71, ranging from 217 to 1173 ng/g.[14] Lin et al. (2020) reported the concentrations of 15 PAHs from 6.24 to 692 ng/g in several exposure groups in China, including electronic waste recycling (EW) workers, non-EW workers, adults, and children in China.[15] They indicated that PAH levels in the hair of EW workers were markedly higher than those of the other groups, and that the PAH concentrations in children's hair were approximately 2.5–3 times lower than in adults.[15] This difference can be explained by the higher exposure of adults to PAH-emitting activities such as smoking, cooking, and traffic.[15,16] Tobacco smoke is recognized as a major source of PAHs that can accumulate in human hair.[17,18] Previous studies have reported significantly higher PAH concentrations in the hair of smokers (mean 310 ng/g) compared to non-smokers (mean 183 ng/g).[18] Elevated PAH levels have been observed in the blood and breast milk of smokers.[19,20] It is estimated that more than 70% of total human exposure to PAHs was resulted from inhalation of tobacco smoke and consumption of contaminated food.[17]

In addition, residential locations play an important role in the PAH accumulation in hair. Wang et al. (2020) examined PAH levels in adult hair samples collected from two regions in

China (Nanjing and Ningbo) and reported that hair-bound PAH concentrations in Nanjing (317–1580 ng/g, mean 753 ng/g) were several times higher than those in Ningbo (60–440 ng/g, mean 222 ng/g).[21] This discrepancy was attributed to the higher population density, traffic intensity, and presence of chemical industries in Nanjing, whereas Ningbo is a rural area with mainly agricultural activities and insignificant industrial emissions.[21] These findings highlight that the exposure to direct emission sources of PAHs significantly influences the accumulation of these contaminants in the human body.[15,18,21]

3.2. Concentrations of PAHs in nail samples

Concentrations of PAHs in the nail samples ranged from 29 to 667 (mean 335) ng/g (Figure 2). Overall, PAH levels in nails were lower than those observed in hair from the same individuals; however, a pronounced inter-individual variability was observed. The female nail sample FN2 exhibited the highest concentration of 667 ng/g, whereas the male nail sample MN2 showed the lowest level of 29 ng/g, suggesting substantial differences in exposure degree and living habits among these participants.

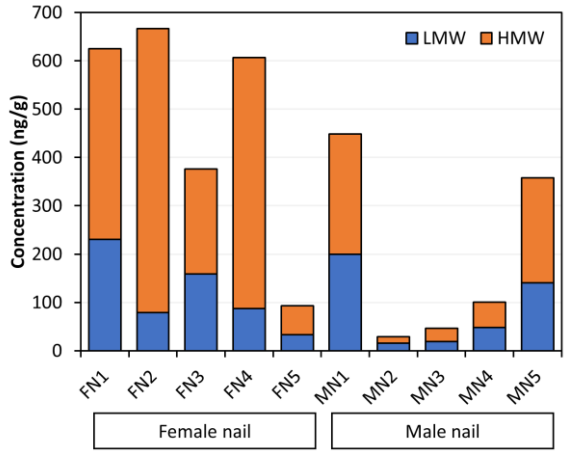

Figure 2. Concentrations of PAHs in nail samples

A clear sex-dependent pattern was found, with female students showing a considerably higher mean PAH concentration in nails (474 ng/g) than male students (197 ng/g), approximately 2.4 fold higher. This difference can be attributed to both morphological and behavioral factors: (i) longer nails provide a greater surface area for the retention of hydrophobic contaminants and (ii) females generally trim their nails less frequently, prolonging the contact time with contaminated dust and household surfaces.[22,23] Unlike hair, nail contamination is strongly influenced by direct external contact, indicating that nails predominantly capture external exposure pathways (hand–object–dust transfer) rather than endogenous accumulation.[24,25] The primary mechanism behind PAH accumulation in nails is surface adsorption, where hydrophobic aromatic rings interact with lipids, sebum residues, or particulate matter lodged in the subungual gap through hydrophobic and π-π interactions.[26] Since fingernails consist of avascular hard keratin, endogenous transfer through the bloodstream is negligible, in contrast to hair, which may incorporate contaminants during its growth phase through systemic circulation.[26,27] Thus, fingernails represent a biomarker for external exposure, whereas hair reflects both internal and external exposure.[26-27]

The PAH levels measured in the fingernail samples of our study were within the ranges reported for similar samples from China. Ma et al. (2021) measured PAH concentrations in nail samples from four different exposure groups, ranging from 7.97–551 (mean 127), 7.05–431 (63.2), 7.93–289 (70.5), and 8.88–1280 (509) ng/g for EW workers, non-EW workers, adults, and children, respectivelty.[24] Tang (2020) revealed that children had higher nail concentrations than adults, attributed to frequent hand contact with indoor dust.[25] Another study conducted in Guangdong urban areas reported substantially higher PAH levels (98–3690 ng/g; mean 831 ng/g), which may be associated with intense urban–industrial emissions in the region.[28] The sex-related trend of higher PAH concentrations in female nail samples is consistent with international observations, reinforcing the role of nail morphology and hand-contact behavior in determining the level of PAH accumulation.[24,28]

3.3. Profiles of PAHs in nail and hair samples

The compositional characteristics of PAHs in nail and hair samples are presented in Figure 3. A clear difference in PAH accumulation patterns was observed between the two biological matrices, despite being collected from the same individuals. In the nail samples, Phe accounted for the highest proportion (average 23%), followed by Pyr (14%), BbF (13%), Flt (10%), BaP (8%), DA (7%) and Chr (3%). The remaining PAHs, including BkF, IP, Ant, BaA and Flu, contributed approximately 2–3%, while Acy and Ace represented less than 2% of the total PAHs. High-molecular-weight PAHs (HMW-PAHs, 4–6 rings) dominated the profile (70%), substantially exceeding low-molecular-

weight PAHs (LMW-PAHs, 2–3 rings; 30.2%), suggesting that the nail largely reflects external accumulation through particulate adhesion. This is consistent with the strong hydrophobicity and environmental persistence of HMW-PAHs, which readily adsorb and remain stable on the highly cross-linked keratin surface of nails. The overall profile is comparable to previous studies conducted in China, where Phe and Pyr were also found to be predominant constituents.[24,25] However, Zeng et al. (2022) reported high levels of Nap, indicating potential differences in emission sources or site-specific exposure.[28]

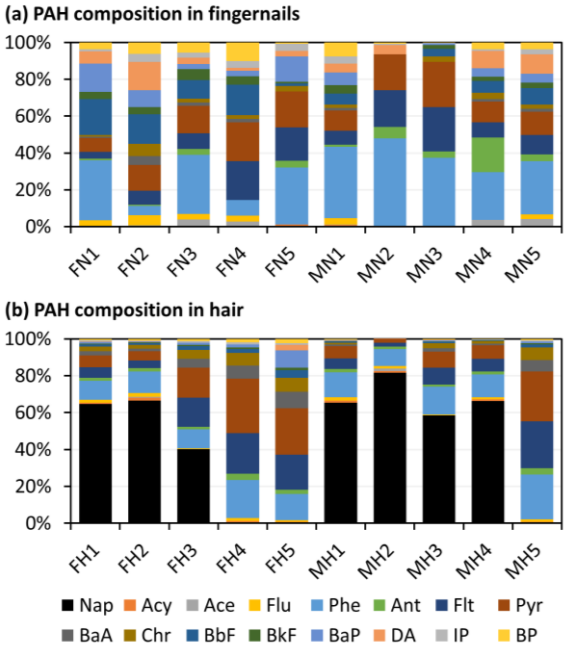

Figure 3. Profile of PAHs in fingernail and hair samples

In contrast, the hair samples exhibited a markedly different composition, with Nap contributing the largest proportion (50%), followed by Phe (13%), Pyr (12%) and Flt (10%), while HMW-PAHs such as BaP, DA, IP and BP each accounted for only 1–3%. Overall, LMW-PAHs (67%) were dominant over HMW-PAHs (33%). This distribution pattern reflects the dual accumulation pathway of hair, in which LMW-PAHs more readily penetrate the hair shaft during growth or are re-adsorbed to the hair surface via lipid-associated mechanisms, while HMW-PAHs exhibit more limited incorporation unless the exposure source is particularly intense or localized.[26] The large inter-individual variability in Nap levels, with some samples (e.g., FH1) exceeding 60% of total PAHs whereas others showed non-detectable concentrations, also indicates differences in microenvironmental contact such as indoor air, fuel combustion, or personal activity pattern. An exceptional sample, such as FH5, showed higher BaP, DA and BP concentrations than the remaining samples, implying potential exposure from specific sources such as industrial emissions or vehicular exhaust. The compositional patterns observed in our hair samples are consistent with international literature, where Nap, Flt, Phe and Pyr frequently dominate the PAH profiles in human hair.[15,18,21]

3.3. Estimation of BaP equivalents

The benzo[a]pyrene equivalents (BaP-EQ) of the PAH mixture were calculated from the measured concentrations with the corresponding toxic equivalency factors (TEFs).[29] The TEF approach, originally proposed by Nisbet and LaGoy (1992), allows individual PAHs to be expressed relative to the toxicity of BaP, the most critical PAH substance in terms of toxicity and carcinogenicity.[29] The BaP-EQ levels in the samples of this study are shown in Figure 4.

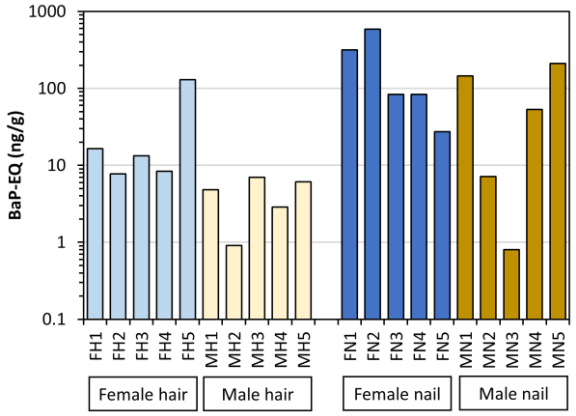

Figure 4. BaP-equivalent toxicity (BaP-EQ) levels in hair and nail samples

Levels of BaP-EQ ranged from 0.90 to 130 (average 20) ng BaP-EQ/g in the hair samples and from 0.80 to 591 (average 150) ng BaP-EQ/g in the nail samples. In addition to the differences in total PAH concentrations, a clear contrast was also observed in the BaP-EQ levels between nails and hair. Among females, nail samples such as FN1 and FN2 exhibited remarkably high BaP-EQ values (318 and 591 ng/g, respectively), far exceeding those found in the corresponding hair samples, even though the overall PAH levels in hair were not significantly lower. The hair samples showed relatively low BaP-EQs even in cases where the total PAH concentrations are high. Relatively high BaP-EQ levels were found in male nail samples (e.g., 145 and 210 ng BaP-EQ/g in MN1 and MN5), although the levels in corresponding hair samples were quite low. The BaP-EQ profiles in

the hair samples were dominated by BaP (average 36%), DA (28%), BaA (13%), BbF (5%), BkF (2%), and IP (2%). Meanwhile, the BaP-EQs in the nail samples showed the highest contributions of DA (69%), followed by BaP (21%), BbF (5%), and BkF (2%). These results suggest that the accumulation of PAHs in general and highly toxic compounds in particular depends on the physicochemical properties of pollutants, emission source patterns, and tissue-specific distribution.

4. CONCLUSION

This study provides preliminary data on the occurrence of 16 PAHs, including concentrations, accumulation patterns, and BaP-EQ levels in hair and fingernail samples collected from university students in Vietnam. Overall, hair samples exhibited higher total PAH concentrations than nail samples, and female participants generally showed higher levels than males. The ranges observed in this dataset are comparable with those reported in previous studies carried out in residential areas and informal e-waste recycling sites in China. Regarding compositional profiles, nails were dominated by Phe, Pyr, BbF, BaP, and DA, whereas hair was dominated by Nap, Phe, and Pyr. This distribution pattern is consistent with earlier findings in the literature, supporting the view that nails mainly reflect externally deposited PAHs originating from dust and surface contact, while hair captures a mixture of endogenous and external exposure. As a result, BaP-EQ values were markedly higher in nails than in hair, indicating that the nail matrix represents not only the presence of PAHs but also the toxic relevance of exposure, whereas hair better reflects the overall exposure burden. To the best of our knowledge, this is the first study in Vietnam to simultaneously assess PAHs in both hair and nail matrices from the same individuals, thereby offering a new perspective on how these two keratin-based tissues complement each other in exposure assessment. Further studies should be performed to characterize potential health impacts of PAHs and to clarify how these exposure patterns translate into toxicological or epidemiological outcomes.

Acknowledgments

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