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Levels, Sources and Health Risk Assessment of Heavy Metals in Hair Dye Sold in Nigeria

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Abstract: *The presence of heavy metals in hair-dye products is a potential public health concern because repeated dermal exposure may result in cumulative toxicological effects. This study investigated the concentrations, distribution, statistical relationships, and potential health risks associated with selected heavy metals in commercially available hair dyes sold in Nigeria. Seven hair-dye samples were analysed for cobalt (Co), arsenic (As), manganese (Mn), copper (Cu), cadmium (Cd), chromium (Cr), nickel (Ni), and lead (Pb), followed by descriptive, correlation, principal component, hierarchical cluster, and health-risk assessments. Nickel recorded the highest mean concentration (0.04411 mg/kg), followed by Mn (0.02796 mg/kg), Pb (0.02544 mg/kg), and Co (0.02358 mg/kg), while As had the lowest mean concentration (0.00044 mg/kg). The coefficient of variation ranged from 10.78% for Pb to 102.88% for Cu, demonstrating substantial differences in metal concentrations among products. The total measured heavy-metal concentration ranged from 0.1002 mg/kg in HD1 to 0.1735 mg/kg in HD7. Spearman correlation indicated a strong inverse association between As and Cd ($\rho = -0.847$), while Pearson correlation identified significant negative relationships between As–Cd ($r = -0.770$, $p = 0.043$) and As–Ni ($r = -0.789$, $p = 0.035$). Principal component analysis showed that the first three components explained 88.27% of the total variance, indicating substantial multivariate differentiation among the products. Hierarchical clustering further identified similarities between selected samples, particularly HD6–HD7 and HD2–HD3. The findings demonstrate heterogeneous heavy-metal profiles among the investigated hair dyes and underscore the importance of continued monitoring, regulatory control, and improved quality assurance to minimize potential health risks associated with repeated exposure.*

Keywords: *Cadmium, Nickel, Non-carcinogenic risk, Carcinogenic risk, Consumer safety*

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1.0 Introduction

Cosmetics play an important role in modern society, as they are widely used to enhance appearance, maintain hygiene, and boost self-confidence. Among these products, hair dyes have gained increasing popularity across different age groups and social classes due to the desire to change, restore, or improve hair colour (Nohynek *et al*., 2010). Hair dyes can be categorized into natural, metallic, and synthetic hair dyes (Souza *et al*., 2025; Sudhamani *et al*., 2023; Fouladkar, 2024). Natural hair dyes are extracted from plant roots, leaves, seeds, and flowers (Souza *et al*., 2025; Sudhamani *et al*., 2023; Fouladkar, 2024; Cui *et al*., 2022). Metallic hair dyes are produced using metallic salts such as silver nitrate, bismuth citrate, copper salts, and cobalt salts (Souza *et al*., 2025; Sudhamani *et al*., 2023; Fouladkar, 2024). In contrast, synthetic hair dyes are manufactured from a combination of organic and inorganic compounds. The organic compounds include precursor intermediates, usually p-phenylenediamine (PPD) or p-toluenediamine (PTD), and coupling agents such as resorcinol and m-phenylenediamine, while the inorganic compound is typically hydrogen peroxide (He *et al*., 2022). Because hair dyes are applied directly to the hair and scalp, their chemical constituents may come into contact with the skin and, depending on their physicochemical properties and exposure conditions, may contribute to dermal exposure
Natural and metallic hair dyes generally have poor colour fastness, produce less vibrant colours, exhibit slow colour development, and have limited hue ranges (Souza *et al*., 2025; Sudhamani *et al*., 2023; Fouladkar, 2024). In contrast, synthetic hair dyes provide longer-lasting colour, a wider variety of brilliant hues, and faster colour development (Souza *et al*., 2025; Sudhamani *et al*., 2023; Fouladkar, 2024). Due to these properties, synthetic hair dyes are often preferred over other types of hair dyes. However, the widespread use of these products also increases the importance of evaluating potentially hazardous constituents that may be present in their formulations. Although toxicological and epidemiological investigations have largely focused on PPD and other organic constituents of hair dyes, increasing attention has also been directed towards the occurrence of potentially toxic metals in cosmetic formulations (Gago-Dominguez *et al*., 2001; Andrew *et al*., 2004; Harling *et al*., 2010; Chong *et al*., 2016). Although the Food and Drug Administration (FDA) has not reached a definitive conclusion regarding these reports, it has established safety limits for certain compounds used in hair-dye production as a precautionary measure (He *et al*., 2022).

In addition to the potential risks associated with organic hair-dye constituents, the occurrence of potentially toxic elements in hair-dye formulations represents an important but comparatively less investigated consumer-safety concern. Such contamination may occur intentionally when metallic compounds are used as formulation ingredients or pigments, or unintentionally through impurities in raw materials, processing equipment, water, packaging materials, and other manufacturing inputs. Studies using advanced analytical techniques have detected heavy metals in cosmetic products, with contamination potentially originating from raw materials, manufacturing processes, or packaging materials (Lavilla *et al*., 2009; Dang *et al*., 2020; Mostafaii *et al*., 2022). Studies investigating heavy metals in hair dyes have reported varying concentrations of these elements across different products and brands (Mostafaii *et al*., 2022; Ahmed *et al*., 2021; Silva *et al*., 2025; Ojezele & Ojezele, 2026). Through dermal contact with the scalp, these metals may be absorbed into the human body. Collectively, these findings indicate that the occurrence and concentrations of metals can vary considerably among hair-dye formulations, manufacturers, brands, and

geographical markets, highlighting the need for product-specific assessment

Heavy metals such as cadmium, lead, arsenic, and mercury are generally considered non-essential elements because they do not perform essential biological functions in the human body and may be toxic even at relatively low concentrations. In contrast, elements such as iron, zinc, cobalt, nickel, and chromium play important roles in various biochemical processes; however, they may become toxic at elevated concentrations (Tchounwou *et al*., 2012). Non-essential heavy metals and excessive levels of essential metals can induce toxicity by disrupting cellular processes, including reactive oxygen species (ROS) generation, oxidative stress, enzyme inactivation, and impaired antioxidant defence mechanisms (Balali-Mood *et al*., 2021). The potential health implications depend not only on the concentration of a metal in a product but also on its toxicity, exposure frequency, route of exposure, duration of use, and the susceptibility of the exposed individual These effects may result in carcinogenic, mutagenic, and other non-carcinogenic adverse health outcomes (Balali-Mood *et al*., 2021). Owing to the health risks associated with heavy-metal exposure, European Union legislation has restricted the use of certain heavy-metal compounds, including compounds of cadmium, lead, cobalt, chromium, and nickel, in cosmetic products to protect consumers (European Union, 2009). Nevertheless, some of these metals may still occur unintentionally in cosmetic products, including hair dyes, through raw materials, manufacturing processes, packaging materials, or pigments (Lavilla *et al*., 2009; Dang *et al*., 2020; Mostafaii *et al*., 2022). For products marketed in Nigeria, assessment of heavy-metal contamination is particularly relevant to cosmetic product surveillance and consumer protection, given the widespread availability and continued use of locally manufactured and imported hair-dye products.

Despite the documented occurrence of potentially toxic metals in cosmetic products and hair dyes, important gaps remain in understanding the extent of heavy-metal contamination in hair dyes available to consumers in Nigeria. Existing studies have generally focused on the occurrence or concentration of selected metals, while comparatively little attention has been given to simultaneously assessing multiple metals, their possible sources, and the associated non-carcinogenic and carcinogenic risks from consumer exposure. Moreover, differences in formulations, brands, raw materials, and manufacturing practices may result in substantial variation in metal concentrations among products. Therefore, updated and integrated information on metal levels, possible contamination sources, and quantitative health risks associated with commercially available hair dyes in Nigeria is needed

Accordingly, this study aimed to determine the concentrations of cadmium (Cd), chromium (Cr), nickel (Ni), lead (Pb), cobalt (Co), arsenic (As), manganese (Mn), and copper (Cu) in commercially available hair dyes sold in Nigeria, assess their potential sources, and evaluate the associated non-carcinogenic and carcinogenic health risks to consumers. The study further assessed variations in metal concentrations among the investigated hair-dye brands and estimated potential exposure risks using established health-risk assessment indices, including the hazard quotient (HQ), hazard index (HI), cancer risk (CR), and total cancer risk (TCR). The findings are expected to provide baseline information on the occurrence of potentially toxic metals in hair-dye products marketed in Nigeria and support evidence-based cosmetic surveillance, regulatory decision-making, product-quality control, and consumer protection. The findings may also provide a reference point for future investigations of metal contamination in cosmetic products and contribute to the development of risk-based strategies for

reducing consumer exposure to potentially hazardous constituents in hair dyes.

2.0 Materials and Methods

2.1 Sampling

Seven commercially available hair-dye products representing different brands and colours (six black and one honey-blonde) were purchased from a supermarket in Asaba, Delta State, Nigeria, in March 2026. The brands were selected based on their availability and popularity in the Nigerian market. The sampling was designed to obtain commercially available products representing different brands and colour formulations. Where available, information on manufacturer, country of manufacture, batch number, manufacturing date, expiry date, and product type was recorded. The selected brands are among the major hair-dye brands sold in Nigeria. . The samples were transported to the laboratory and labelled accordingly as HD1, HD2, HD3, HD4, HD5, HD6, and HD7. The samples were kept in their original sealed containers during transportation and handling to minimize external contamination. All laboratory glassware and sample-contact materials were thoroughly washed, rinsed with deionised water, and dried before use.

2.2 Sample Digestion and Heavy Metal Analysis

The digestion procedure was adapted from Khalili *et al*. (2019) and Pehlić *et al*. (2019), with modifications to the sample mass, reagent volumes, and heating conditions as described below. Briefly, 3 g of each sample was transferred into a 100 mL beaker, followed by the addition of 1 mL of H_2O_2 and 5 mL of HNO_3. The mixture was stirred until a homogeneous mixture was obtained. It was then heated on an electric hot plate in a fume cupboard at 155 °C for 10 min, followed by further heating at 200 °C for 7 min. The mixture was heated until the digest became clear and/or colourless and no visible particulate matter remained, indicating substantial completion of the digestion." After cooling to room temperature, the digest was filtered through pre-cleaned Whatman filter paper into a volumetric flask and diluted to 50 mL with deionised water. The filtrate was diluted to the 50 mL mark with deionised water and transferred into pre-cleaned sample bottles. The samples were stored at 4 °C until analysis. The digested samples were analysed using an Agilent 200 Series atomic absorption spectrophotometer equipped with element-specific hollow-cathode lamps and operated under the manufacturer's recommended conditions. The digested samples were analysed using an Agilent 200 Series atomic absorption spectrophotometer equipped with element-specific hollow-cathode lamps and operated under the manufacturer's recommended conditions, and a UV–Visible spectrophotometer. Lead (Pb), nickel (Ni), cadmium (Cd), copper (Cu), cobalt (Co), manganese (Mn), and chromium (Crwere determined using AAS, while arsenic (As) was determined using UV–Visible spectrophotometry.

The AAS was calibrated using standard solutions of the respective metals. Lead standards were prepared at concentrations of 0.5, 1.0, 1.5, and 2.0 mg/L, while cadmium, chromium, cobalt, copper, manganese and nickel standards were prepared at concentrations of 2, 4, 6, and 8 mg/L. The samples were analysed in triplicate at the respective wavelengths: Pb (217 nm), Cd (228.8 nm), Cr (357.9 nm), Cu (324.7 nm), Co (240.7), Mn (279.5) and Ni (232 nm). The limit of detection (LOD) for Cd, Pb, Ni, Cu, Co, Mn and Cr was 0.0001 mg/kg, while the limit of quantification (LOQ) was 0.00033 mg/kg. A reagent blank was analysed alongside the samples to account for potential contamination from the reagents and analytical procedure. Instrumental operating parameters, including lamp current, spectral slit width, flame composition, and aspiration conditions, were

set according to the manufacturer's recommendations for each element

Arsenic was determined using UV–Visible spectrophotometry based on the methods described by Afkhami *et al*. (2001) and Imran *et al*. (2015). 5 mL of the filtrate was transferred into a 100 mL measuring cylinder, followed by the addition of 2 mL of 0.01% methyl orange solution, 4 mL of 2 M hydrochloric acid, and 4 mL of 2 % potassium iodide solution. The mixture was gently swirled, after which 4 mL of 2% potassium iodate solution was added. The solution was then diluted to the 100 mL mark with distilled water. The mixture was transferred into a 100 mL conical flask and allowed to stand for colour development. The absorbance was measured at 538 nm using a 140S Thermo Scientific visible spectrophotometer. A reagent blank was analysed in parallel. The arsenic concentration in the samples was determined from a calibration curve prepared using arsenic standard solutions. The absorbance response was based on the colour-forming reaction specified in the adopted arsenic spectrophotometric method, and arsenic concentrations were obtained from a calibration curve prepared using certified arsenic standards

2.3 Quality Control and Method Validation

Analytical-grade reagents and deionised water were used throughout the analysis. All glassware and sample-contact materials were thoroughly cleaned before use to minimize contamination. The AAS equipment was flushed with deionised water after each analysis, and all samples were analysed in triplicate. The validity of the analytical method was evaluated in terms of sensitivity, linearity, accuracy, and precision. The linearity of the calibration curves for each metal was assessed using the coefficient of determination (R^2), which ranged from 0.997 to 0.998. The sensitivity of the method was evaluated using the limits of detection (LOD) and quantification (LOQ), calculated as LOD = 3.3σ/S and LOQ = 10σ/S, respectively, where σ is the standard deviation of ten replicate measurements of the reagent blank and S is the slope of the calibration curve. The LOD and LOQ for the analysed metals were 0.0001 mg/kg and 0.00033 mg/kg, respectively. Method blanks, sample duplicates, and certified reference materials were analysed where available to monitor contamination, reproducibility, and analytical accuracy. Accuracy was evaluated using recovery studies. For recovery assessment, 1 g portions of representative hair-dye samples were fortified before digestion with 2 mL of a 1 mg/L mixed standard solution containing the target metals. The fortified samples and corresponding unspiked samples were subsequently subjected to the complete digestion and analytical procedure. The spiked and unspiked samples were digested and analysed as previously described, while a reagent blank was processed and analysed alongside the samples. The percentage recoveries ranged from 97% to 101%. Precision was expressed as relative standard deviation (%RSD) and assessed using three replicate analyses performed within a single day (intra-day precision) and over three consecutive days (inter-day precision. The intra-day precision ranged from 0.51% to 2.94%, while the inter-day precision ranged from 1.21% to 3.53%.

2. 4 Health Risk Assessment

Carcinogenic and non-carcinogenic risks associated with dermal exposure to heavy metals in the hair-dye samples were estimated using equations and exposure parameters derived from applicable US EPA and ATSDR guidance documents.

2.5 Non–Carcinogenic Risk

The hazard quotient (HQ) and hazard index (HI) are risk assessment measures used to estimate non-carcinogenic health risks (USEPA, 2025a; ATSDR, 2025). The HQ

estimates the potential for a pollutant to cause non-carcinogenic health effects, whereas the HI is the sum of the HQ values and estimates the potential for non-carcinogenic health effects resulting from exposure to multiple pollutants or exposure pathways. Hair-dye users may be exposed to HMs through dermal contact with the dye. The hazard quotient for the dermal pathway is given by equation 1 (USEPA, 2025a; ATSDR, 2025; Silva *et al*., 2025; Khalili *et al*., 2019),

$$HQ_{der} = \frac{CDI_{der}}{RfD_{der}} \quad (1)$$

where

$$CDI_{der} = \frac{C\ X\ SA\ X\ SL\ X\ ABS\ X\ \ ED\ X\ EF}{BW\ X\ AT} \quad (2)$$

where CDI is the chronic daily intake of heavy metals through dermal contact (mg/kg/day); C is the concentration of heavy metals in the hair dye (mg/kg); SA is the scalp skin surface area (cm^2) = 700 cm^2 for adults (USEPA, 2011); SL is the skin adherence factor = 0.07 $mg/cm^2 \cdot h$ for adults (ATSDR, 2023); ABS is the dermal absorption fraction (unitless), = 0.001 for all heavy metals except arsenic, for which an absorption fraction of 0.03 was used (ATSDR, 2023; Zheng, 2010); EF is the exposure frequency (days/year), = 350 days/year; ED is the exposure duration (years), 26 years for adults; BW is body weight (kg), = 80 kg for adults; and AT is the averaging time for non-carcinogenic effects (days), calculated as 9,490 days (26 × 365) for adults (USEPA, 2014).

The United States Environmental Protection Agency (USEPA) and the Agency for ToxicSubstances and Disease Registry (ATSDR) do not provide dermal reference dose (RfDermal) values for the heavy metals assessed in this study. However, the USEPA (2007) guides estimating the dermal reference dose from the oral reference dose (RfDoral) of metals, as follows,

$$RfD_{der} = RFD_{oral} \text{ x } ABSG_I \quad (3)$$

where $ABSG_I$ is the fraction of heavy metals absorbed in gastrointestinal tract (dimensionless) in the critical toxicity study. $ABSG_I$ for As = 95 % (0.95), Cd = 5 % (0.05), Mn = 4 % (0.04), Ni = 4 % (0.04) other metals = 100 % (1). RfD_{der} of the metals is presented in Table 2

$$HI = \Sigma HQ \quad (4)$$

An HQ < 1 indicates that non-carcinogenic health effects are unlikely to occur at the estimated level of exposure, whereas an HQ > 1 indicates the potential for non-carcinogenic health effects. Similarly, an HI < 1 suggests that exposure to the heavy metals is unlikely to pose a significant non-carcinogenic health risk, while an HI > 1 indicates the potential for non-carcinogenic health effects due to exposure to multiple heavy metals (USEPA, 2007).

2. 4. 2 Carcinogenic Risk

The carcinogenic risk associated with exposure to metals in the hair-dye samples was determined using the following equation (USEPA, 2025a; ATSDR, 2025; Silva *et al*., 2025),

$$CR_{der} = CDI_{der} \text{ x } SF \quad (5)$$

AT (averaging time for carcinogens) is 25,550 days (70 × 365) for adults (USEPA, 2014). Similarly, the USEPA, ATSDR, and other relevant regulatory agencies do not provide dermal slope factor values for the heavy metals assessed in this study. However, the USEPA (2007) recommends estimating the dermal slope factor using equation 6,

$$SF_{der} = \frac{SF_O}{ABSG_i} \quad (6)$$

SF_{der} is presented in Table 2.

CR value between 10^{-6} to 10^{-4} is considered acceptable (USEPA, 2021).

2.4 Statistical Analysis

Statistical analysis was performed using the Statistical Package for the Social Sciences (SPSS), version 26.0. One-way analysis of variance (ANOVA), followed by Tukey's honestly significant difference (HSD) post hoc test, was used to determine whether significant differences existed in the mean concentrations of individual heavy metals among the different hair-dye brands. Pearson correlation analysis was used to assess the relationships among the heavy metals within and across the hair-dye

brands and to identify potential common sources.

3.0 Results and Discussion

3.1 Concentrations of Heavy Metals in Hair-Dye Samples

The concentrations of the investigated heavy metals in the seven hair-dye samples are presented in Table 1. Cobalt (Co), arsenic (As), manganese (Mn), copper (Cu), cadmium (Cd), nickel (Ni), and lead (Pb) were detected in varying concentrations across the samples, whereas chromium (Cr) was detected in four of the seven samples. The occurrence of several heavy metals in the investigated products indicates that hair dyes sold in Nigeria may contain trace levels of metallic contaminants, although the concentrations varied considerably among samples and metals.

Table 1. Concentration of Heavy Metals (in mg/kg) in Major Hair-Dye Samples Sold in Nigeria

Sample ID	Colour	Co	As	Mn	Cu	Cd	Cr	Ni	Pb
HD1	Black	BDL	0.0010	0.0318	0.0108	0.0086	0.0142	0.0049	0.0289
HD2	Black	0.0243	0.0008	0.0246	0.0011	0.0072	BDL	0.0183	0.0253
HD3	Black	0.0220	0.0005	0.0368	0.0062	0.0084	BDL	0.0531	0.0275
HD4	Black	0.0370	0.0002	0.0186	0.0021	0.0093	0.0006	0.0566	0.0229
HD5	Black	0.0207	0.0001	0.0292	0.0261	0.0109	BDL	0.0514	0.0280
HD6	Honey Blonde	0.0109	0.0001	0.0251	0.0060	0.0125	0.0049	0.0507	0.0219
HD7	Black	0.0266	0.0004	0.0296	0.0054	0.0104	0.0037	0.0738	0.0236
Mean ± SD	—	0.0202 ± 0.0118	0.00044 ± 0.00035	0.0280 ± 0.0058	0.00824 ± 0.00848	0.00961 ± 0.00178			

Among the metals analysed, nickel recorded the highest overall mean concentration of 0.0441 ± 0.0239 mg/kg, with individual concentrations ranging from 0.0049 to 0.0738 mg/kg. The highest Ni concentration occurred in HD7 (0.0738 mg/kg), followed by HD4 (0.0566 mg/kg), HD3 (0.0531 mg/kg), HD5 (0.0514 mg/kg), and HD6 (0.0507 mg/kg). The relatively higher Ni concentrations in these samples suggest greater variability in the occurrence of Ni among the products. Manganese had the second-highest mean concentration (0.0280 ± 0.0058 mg/kg), with concentrations ranging from 0.0186 to 0.0368 mg/kg. Lead also occurred relatively consistently across the samples, with a mean concentration of 0.0254 ± 0.0027 mg/kg and a narrow range of 0.0219–0.0289 mg/kg.

Cobalt was detected in six of the seven samples, with concentrations ranging from 0.0109 to 0.0370 mg/kg and a mean of 0.0202 ± 0.0118 mg/kg. The highest Co concentration was observed in HD4 (0.0370 mg/kg), while Co was below the detection limit in HD1. Cadmium was detected in all seven samples and showed comparatively low variability, with concentrations ranging from 0.0072 to 0.0125 mg/kg and a mean concentration of 0.00961 ± 0.00178 mg/kg. The relatively narrow range of Cd suggests a more consistent occurrence of this metal across the investigated products.

Copper concentrations ranged from 0.0011 to 0.0261 mg/kg, with a mean of 0.00824 ± 0.00848 mg/kg. The highest Cu concentration occurred in HD5 (0.0261 mg/kg), whereas HD2 recorded the lowest concentration (0.0011 mg/kg). Arsenic was present at the lowest concentrations among the metals investigated, ranging from 0.0001 to 0.0010 mg/kg, with a mean of 0.00044 ± 0.00035 mg/kg. Chromium was detected in HD1, HD4, HD6 and HD7, while it was below the detection limit in HD2,

HD3 and HD5. Its detected concentrations ranged from 0.0006 to 0.0142 mg/kg, with a provisional overall mean of 0.00334 ± 0.00519 mg/kg when BDL values were assigned zero for statistical calculation.

Overall, the mean concentration followed the order, Ni > Mn > Pb > Co > Cu > Cd > Cr > As. The relatively high mean concentration of Ni compared with the other metals is noteworthy, whereas As occurred at substantially lower concentrations. The variation in metal concentrations among the products may reflect differences in raw materials, pigments, precursor chemicals, additives, manufacturing processes, or contamination introduced during production and packaging. However, concentration data alone cannot establish the precise source of the metals; source attribution would require additional information on the composition of raw materials, manufacturing processes, and product formulations.

The observed concentrations were generally low when compared with the concentrations reported in several previous studies of heavy metals in hair dyes. For example, Ahmed *et al*. (2019) reported Cd concentrations of 0.0015–0.0035 ppm and Pb concentrations of 0.003–0.014 ppm in hair dyes used in Riyadh, Saudi Arabia. Fonseka *et al*. (2024) reported Pb concentrations of 0.04–0.28 ppm and Cd concentrations from below the detection limit to 2.33 ppm in hair dyes from Sri Lanka. Mostafaii *et al*. (2022) also reported considerably higher concentrations of several metals in hair-dye samples from Iran, including Pb, Cd, Cr, Ni and Co. These comparisons indicate that the concentrations measured in the present study are generally lower than those reported in some international studies, although differences in analytical procedures, detection limits, product formulations, sampling strategies, and units of reporting should be considered when making direct comparisons.

The presence of heavy metals at relatively low concentrations does not necessarily imply an absence of health concern. Hair dyes are applied directly to the scalp and hair, and repeated use may result in repeated dermal exposure. The potential health significance therefore depends not only on the measured concentration but also on exposure frequency, duration, dermal absorption, the chemical form of each metal, and individual susceptibility. Consequently, the concentration data provide an important basis for the subsequent assessment of non-carcinogenic and carcinogenic risks.

It is also important to distinguish detection from toxicological significance. The detection of a metal in a cosmetic product does not by itself demonstrate that the product poses a health risk. Risk characterization requires integration of the measured concentrations with exposure assumptions and toxicological reference values. Accordingly, the subsequent hazard quotient (HQ), hazard index (HI), and cancer-risk assessments should be calculated from the corrected concentration dataset and clearly stated dermal exposure parameters.

4.2 Health Risk Assessment

The potential non-carcinogenic and carcinogenic health risks associated with dermal exposure to heavy metals in the investigated hair-dye samples were evaluated using the exposure parameters and equations described in Section 2.4. The assessment considered cobalt (Co), arsenic (As), manganese (Mn), copper (Cu), cadmium (Cd), chromium (Cr), nickel (Ni), and lead (Pb). The estimated chronic daily intake (CDI), hazard quotient (HQ), hazard index (HI), and carcinogenic risk (CR) were calculated based on the measured metal concentrations and the adult exposure assumptions described in the methodology.

4.2.1 Reference Dose and Slope Factor Parameters

The reference dose and slope-factor parameters used for estimating non-carcinogenic and carcinogenic risks are presented in Table 2. Because dermal reference doses and dermal

slope factors are not directly available for all the metals considered, the corresponding dermal values were derived from the oral toxicity values using the procedures described in Section 2.4, following USEPA guidance. The gastrointestinal absorption fractions were incorporated into the derivation of the dermal reference doses and slope factors.

Table 2: Toxicological parameters used for the health-risk assessment of heavy metals in hair dyes

Heavy metal	Oral RfD (mg/kg/day)	GI absorption fraction	Derived dermal RfD (mg/kg/day)	Oral slope factor $(mg/kg/day)^{-1}$	Derived dermal slope factor $(mg/kg/day)^{-1}$
Co)	0.03*	1.00	0.03*	—	—
As	0.0003	0.95	0.000285	1.5	1.425
Mn	0.14	0.04	0.0056	—	—
Cu	0.04	1.00	0.04	—	—
Cd	0.0005	0.05	0.000025	6.1	0.305
Cr	0.003	1.00	0.003	0.5**	0.5**
Ni	0.02	0.04	0.0008	—	—
Pb	0.0035	1.00	0.0035	0.0085***	0.0085***

The results in Table 2 demonstrate that the toxicological parameters differ substantially among the investigated metals. In particular, arsenic and cadmium have considerably lower dermal reference doses than the other metals, indicating that relatively small absorbed doses can contribute appreciably to their non-carcinogenic risk. The derived dermal reference dose for arsenic was obtained by accounting for its gastrointestinal absorption fraction of 0.95, while the substantially lower absorption fraction assigned to cadmium (0.05) resulted in a correspondingly lower derived dermal reference dose. Similarly, the low gastrointestinal absorption fraction adopted for manganese and nickel affected the derivation of their dermal reference doses.

For carcinogenic assessment, arsenic and cadmium are particularly important because of their established carcinogenic potential and the availability of corresponding slope-factor information. Their relatively high toxicity means that even when their concentrations in the hair-dye products are comparatively low, their contribution to overall carcinogenic risk may be significant. Consequently, interpretation of the carcinogenic-risk results should consider both the measured concentrations and the toxicity-weighting parameters rather than concentration alone.

4.2.2 Non-Carcinogenic Risk

The calculated chronic daily intake and hazard quotients for individual metals were obtained using the dermal exposure equations presented in Section 2.4. The calculations incorporated the measured concentrations in Table 1 together with the assumed scalp surface area, adherence factor, dermal absorption fraction, exposure frequency, exposure duration, body weight, and averaging time.

The HQ values provide an indication of the potential for individual metals to produce non-carcinogenic effects through dermal exposure. An HQ below 1 indicates that the estimated exposure is below the corresponding reference dose, whereas an HQ above 1 indicates that the estimated exposure exceeds the reference dose and may therefore warrant concern. The combined contribution of the metals was evaluated using the hazard index (HI), calculated as the sum of the individual HQ values, as described by equation (4) in Section 2.4.

Given the relatively low concentrations of most of the metals measured in the hair-dye samples, the calculated HQ values are expected to remain below unity for most individual metals. However, metals with substantially lower reference doses, particularly arsenic and cadmium, require greater attention because their risk contribution is not determined solely by their measured concentration. The HQ results should therefore be compared across metals using their respective toxicological reference values rather than by directly comparing concentrations.

The HI provides a more comprehensive assessment because simultaneous exposure to several metals may produce a cumulative non-carcinogenic risk. An HI below 1 indicates that the combined estimated exposure is unlikely to produce appreciable non-carcinogenic effects under the assumed exposure conditions, whereas an HI above 1 would indicate a potential concern requiring further evaluation.

Important correction: before this section is finalized, the numerical HQ and HI values should be recalculated directly from the original concentrations in Table 1 and the verified toxicological parameters in Table 2. The standard deviations in Table 1 should describe analytical variability and should not be inserted into the HQ/HI equations as if they were additional exposure concentrations. This is particularly important because some of the originally reported standard deviations are larger than the corresponding means and would produce physically questionable analytical uncertainty.

4.2.2 Non-Carcinogenic Risk

The calculated non-carcinogenic health risks associated with dermal exposure to heavy metals in the investigated hair-dye samples are presented in Table 3, while the overall summary of the calculated chronic daily intake (CDI), hazard index (HI), and carcinogenic risk (CR) is presented in Table 4. The results show that the individual hazard quotient (HQ) values were below 1 for all metals and samples, indicating that the estimated exposure to individual metals did not exceed their respective reference doses under the exposure conditions adopted in this study.

As shown in Table 4, the calculated HI values ranged from 0.23936 to 0.34464. The lowest HI was recorded for HD2 (0.23936), whereas the highest was observed for HD6 (0.34464). The samples followed the general order HD6 > HD7 > HD5 > HD4 > HD1 > HD3 > HD2 with respect to overall non-carcinogenic risk. Importantly, all HI values were below the critical value of 1, suggesting that the combined exposure to the investigated heavy metals through dermal contact with the hair dyes is unlikely to produce significant non-carcinogenic health effects under the assumed exposure conditions.

The total estimated CDI values presented in Table 4 ranged from 1.43×10^{-4} to 2.06×10^{-4} mg/kg/day. HD2 recorded the lowest total estimated CDI, whereas HD6 had the highest. This pattern generally corresponds with the variation observed in the HI values, although the relationship is influenced by the different toxicity thresholds assigned to individual metals. Thus, total exposure alone does not fully determine the magnitude of health risk; the toxicity of each metal must also be considered.

Cadmium was the major contributor to the non-carcinogenic risk in all the samples. Its HQ values ranged from 0.16915 in HD2 to 0.29366 in HD6. The relatively high contribution of Cd is associated with its low derived dermal reference dose and demonstrates that even relatively low concentrations of highly toxic metals can make substantial contributions to overall health risk. Nickel was another important contributor, particularly in HD7, where its HQ reached 0.05418. Arsenic also made a noticeable contribution, with the highest HQ of 0.06182 occurring in HD1.

The remaining metals generally produced substantially lower HQ values. Therefore, the

results indicate that Cd was the principal determinant of non-carcinogenic risk, while the differences among the samples were largely associated with variations in Cd, Ni and As concentrations. Nevertheless, the HI values remained below unity for every sample, indicating that the combined estimated exposure was within the non-carcinogenic risk threshold.

4.2.3 Carcinogenic Risk

The carcinogenic-risk estimates obtained for the investigated hair dyes are presented in Table 3, with the sample-level summary provided in Table 4. The total estimated carcinogenic risk ranged from 1.66×10^{-6} to 1.14×10^{-5}. The highest value was obtained for HD1 (1.14×10^{-5}), followed by HD2 (7.94×10^{-6}) and HD3 (5.22×10^{-6}), while the lowest value was recorded for HD5 (1.66×10^{-6}).

The comparatively high carcinogenic risk observed for HD1 was primarily attributable to arsenic. As shown in Table 3, the arsenic-associated CR for HD1 was 9.33×10^{-6}, accounting for the majority of its total estimated carcinogenic risk. This result is consistent with the relatively higher arsenic concentration measured in HD1. Arsenic also remained the principal contributor to carcinogenic risk in the other samples, although its contribution varied according to the measured concentration.

Cadmium made a smaller but consistent contribution to the total carcinogenic risk, with values ranging from approximately 4.79×10^{-7} to 8.32×10^{-7}. Where chromium was included in the carcinogenic assessment, its contribution was considerably lower than those of arsenic and cadmium. The results therefore demonstrate that arsenic was the dominant contributor to the estimated carcinogenic risk, despite not being the dominant contributor to the non-carcinogenic HI.

Table 3. Recalculated non-carcinogenic and carcinogenic risks associated with dermal exposure to heavy metals in hair-dye samples

Parameter	HD1	HD2	HD3	HD4	HD5	HD6	HD7
HQ-Co	–	0.00048	0.00043	0.00072	0.00041	0.00021	0.00052
HQ-As	0.06182	0.04946	0.03091	0.01236	0.00618	0.00618	0.02473
HQ-Mn	0.00334	0.00258	0.00386	0.00195	0.00306	0.00263	0.00310
HQ-Cu	0.00016	0.00002	0.00009	0.00003	0.00038	0.00009	0.00008
HQ-Cd	0.20204	0.16915	0.19734	0.21849	0.25608	0.29366	0.24433
HQ-Cr	0.00278	–	–	0.00012	–	0.00096	0.00072
HQ-Ni	0.00360	0.01344	0.03898	0.04155	0.03774	0.03722	0.05418
HQ-Pb	0.00485	0.00425	0.00461	0.00384	0.00470	0.00368	0.00396
HI	0.27859	0.23936	0.27623	0.27907	0.30854	0.34464	0.33163
CR-As	9.33×10^{-6}	7.46×10^{-6}	4.66×10^{-6}	1.87×0^{-6}	9.33×0^{-7}	9.33×10^{-7}	3.73×10^{-6}
CR-Cd	5.72×10^{-7}	4.79×0^{-7}	5.59×10^{-7}	6.19×10^{-7}	7.25×10^{-7}	8.32×10^{-7}	6.92×10^{-7}
CR-Cr	1.55×10^{-6}	–	–	6.54×10^{-8}	–	5.34×10^{-7}	4.04×10^{-7}
Total CR	1.14×10^{-5}	7.94×10^{-6}	5.22×10^{-6}	2.55×0^{-6}	1.66×10^{-6}	2.30×10^{-6}	

As shown in Table 4, all the calculated total CR values were below 1×10^{-4}, indicating that the estimated risks remained below the upper boundary commonly applied for acceptable environmental carcinogenic risk. However, the CR for HD1 (1.14×10^{-5}) slightly exceeded the more conservative 1×10^{-5} benchmark. Consequently, HD1 deserves comparatively greater attention, particularly because of its arsenic content. The other samples had CR values below 1×10^{-5}.

Table 4. Summary of recalculated CDI and risk indices

Sample	Total estimated CDI, mg/kg/day*	HI	Total CR	Non-carcinogenic interpretation	Carcinogenic interpretation
HD1	1.68×10^{-4}	0.27859	1.14×10^{-5}	HI < 1	Within conventional acceptable range
HD2	1.43×10^{-4}	0.23936	7.94×10^{-6}	HI < 1	Within conventional acceptable range
HD3	1.65×10^{-4}	0.27623	5.22×10^{-6}	HI < 1	Within conventional acceptable range
HD4	1.66×10^{-4}	0.27907	2.55×10^{-6}	HI < 1	Within conventional acceptable range
HD5	1.84×10^{-4}	0.30854	1.66×10^{-6}	HI < 1	Within conventional acceptable range
HD6	2.06×10^{-4}	0.34464	2.30×10^{-6}	HI < 1	Within conventional acceptable range
HD7	1.98×10^{-4}	0.33163	4.83×10^{-6}	HI < 1	Within conventional acceptable range

4.2.4 Comparative Interpretation of CDI, HI and CR

The summary presented in Table 4 provides an integrated comparison of exposure and health-risk indicators among the seven hair-dye samples. The highest estimated CDI was observed in HD6 (2.06×10^{-4} mg/kg/day), which also had the highest HI (0.34464). This indicates that HD6 represents the sample with the greatest estimated non-carcinogenic exposure burden among the products examined.

However, HD6 did not have the highest carcinogenic risk. Instead, HD1 recorded the highest CR (1.14×10^{-5}) despite having a lower total estimated CDI (1.68×10^{-4} mg/kg/day) than HD6. This difference illustrates an important feature of health-risk assessment: a higher total metal exposure does not necessarily correspond to a higher carcinogenic risk, because the final risk is strongly influenced by the toxicity and carcinogenic potency of the individual metals.

Similarly, HD2 had the lowest estimated CDI (1.43×10^{-4} mg/kg/day) and the lowest HI (0.23936), suggesting the lowest overall non-carcinogenic exposure burden. Its total CR of 7.94×10^{-6}, however, was higher than those of HD3–HD7 except HD1, demonstrating again that risk depends on the specific composition of the metal mixture rather than total metal concentration alone.

Overall, the results summarized in Table 4 indicate that the investigated hair dyes did not present a significant non-carcinogenic risk based on the HI criterion, since all HI values were less than 1. The carcinogenic-risk estimates were also below the upper commonly applied acceptable-risk limit of 1×10^{-4}. Nevertheless, the elevated contribution of Cd to non-carcinogenic risk and As to carcinogenic risk warrants continued monitoring of these metals in commercially available hair-dye products.

4.2.5 Non-Carcinogenic and Carcinogenic Health-Risk Assessment

The estimated health-risk indices for the investigated hair-dye samples indicate that dermal exposure to the analysed heavy metals should be considered in evaluating the potential health implications of prolonged hair-dye use. The hazard quotient (HQ) values calculated for individual metals were generally below the

benchmark value of 1, indicating that exposure to each metal through the dermal pathway is unlikely to result in appreciable non-carcinogenic effects at the estimated exposure conditions. The corresponding hazard index (HI), obtained from the summation of the individual HQ values, provides an integrated measure of the potential risk associated with simultaneous exposure to multiple metals. The HI values remained below unity for the evaluated samples, suggesting that the combined non-carcinogenic risk from the metals investigated was within the acceptable range.

Although the calculated HQ and HI values were below unity, differences were observed among the samples. These variations can be attributed primarily to differences in the concentrations of individual metals and their respective dermal absorption fractions and toxicity reference values. Metals occurring at relatively higher concentrations do not necessarily produce the highest HQ because the magnitude of HQ also depends on the reference dose and the fraction absorbed through the skin. Consequently, risk interpretation should be based on the calculated HQ rather than concentration alone.

Among the analysed metals, arsenic requires particular attention because a higher dermal absorption fraction was adopted for As than for most of the other metals. Thus, even at relatively low measured concentrations, arsenic may make a comparatively important contribution to the overall non-carcinogenic risk. Similarly, metals with lower dermal reference doses may contribute disproportionately to the total HI despite having concentrations comparable to or lower than those of other metals. This demonstrates the importance of integrating both exposure concentration and toxicity parameters when evaluating the health implications of heavy metals in cosmetic products.

The carcinogenic-risk estimates provide additional information concerning the potential long-term consequences of exposure. The calculated dermal cancer-risk values were within the generally accepted regulatory risk range, indicating that the estimated lifetime cancer risk associated with the investigated exposure scenario was low. However, the presence of carcinogenic metals in cosmetic products remains relevant from a public-health perspective because hair dyes may be used repeatedly over extended periods. The calculated values therefore represent estimated risks under the specified exposure assumptions rather than evidence that the products are completely free of carcinogenic concern.

The observed risk levels should also be interpreted in relation to the exposure assumptions adopted in this study. The assessment was based on adult exposure parameters, including an exposure frequency of 350 days/year, an exposure duration of 26 years and an adult body weight of 80 kg. These assumptions provide a standardized basis for comparison among samples but may not represent the actual behaviour of every consumer. Frequent users, individuals with prolonged contact between the dye and scalp, or persons with damaged skin may experience different exposure levels. In addition, cumulative exposure from simultaneous use of different cosmetic products containing the same metals was not incorporated into the present assessment.

The results suggest that the investigated hair dyes did not present an appreciable non-carcinogenic health risk through the dermal pathway under the assumed exposure conditions, while the estimated carcinogenic risks remained within the generally acceptable range. Nevertheless, the detection of heavy metals in products intended for direct application to the hair and scalp highlights the need for continued monitoring, improved manufacturing quality control and regulatory surveillance of cosmetic products. Particular attention should be given to samples containing relatively elevated concentrations of Pb, Cr, Ni,

Mn and As because repeated exposure could increase cumulative risk over time.

The findings further demonstrate that chemical concentration alone is insufficient for evaluating the health safety of hair-dye products. A comprehensive assessment should combine measured metal concentrations with dermal absorption, exposure frequency, exposure duration, body weight and metal-specific toxicity parameters. Such an integrated approach provides a more meaningful estimation of the potential health implications of prolonged use of hair dyes and supports evidence-based regulatory control of heavy-metal contamination in cosmetic products.

4.3 Statistical Analysis of Heavy-Metal Concentrations

Descriptive and multivariate statistical analyses were performed to further characterize the distribution, variability, inter-metal relationships and overall patterns of heavy metals detected in the hair-dye samples. The statistical analyses complement the concentration data presented in Table 1 and provide a quantitative basis for evaluating differences among the products. Mean, standard deviation (SD), minimum, maximum and coefficient of variation (CV) were used to describe the distribution of individual metals. Correlation analysis was subsequently applied to investigate relationships among the metals, while principal component analysis (PCA) and hierarchical cluster analysis (HCA) were used as exploratory multivariate techniques for identifying patterns among the samples.

4.3.1 Descriptive Statistics

The mean concentration was calculated using equation 7

$$\bar{X} = \frac{\sum_{i=1}^{m} X_i}{n} \quad (7)$$

where $\bar{X}$ is the mean concentration, X_i is the concentration measured in sample i, and n is the number of quantified observations. The standard deviation was used to describe the dispersion of individual measurements around the mean, while the coefficient of variation was calculated using equation 2 and the results obtained are presented in Table 5

$$CV(\%) = \frac{SD}{\bar{X}} \times \frac{100}{1} \quad (8)$$

Table 5. Descriptive statistics of heavy-metal concentrations in hair-dye samples

Metal	n	Mean (mg/kg)	SD (mg/kg)	Minimum (mg/kg)	Maximum (mg/kg)	CV (%)
Co	6	0.02358	0.00850	0.01090	0.03700	36.05
As	7	0.00044	0.00035	0.00010	0.00100	79.15
Mn	7	0.02796	0.00583	0.01860	0.03680	20.87
Cu	7	0.00824	0.00848	0.00110	0.02610	102.88
Cd	7	0.00961	0.00178	0.00720	0.01250	18.52
Cr	4	0.00585	0.00585	0.00060	0.01420	100.07
Ni	7	0.04411	0.02387	0.00490	0.07380	54.11
Pb	7	0.02544	0.00274	0.02190	0.02890	10.78

As shown in Table 5, nickel (Ni) exhibited the highest mean concentration (0.04411 mg/kg), followed by manganese (Mn; 0.02796 mg/kg), lead (Pb; 0.02544 mg/kg) and cobalt (Co; 0.02358 mg/kg). In contrast, arsenic (As) had the lowest mean concentration of 0.00044 mg/kg. The relatively higher mean concentration of Ni indicates that nickel constituted the most prominent metal in the analysed products, although concentration alone does not determine toxicological risk because toxicity and exposure parameters also differ among metals.

The variability of the metals differed substantially. Lead exhibited the lowest coefficient of variation (CV = 10.78%), indicating relatively consistent Pb concentrations among the quantified samples. Cadmium and manganese also showed comparatively low variability, with CV values of 18.52 and 20.87%, respectively. Conversely, copper had the highest CV (102.88%), followed by chromium (100.07%) and arsenic (79.15%). Such high CV values indicate considerable variation in the concentrations of these metals among the products. The large variability in Cu and Cr suggests that their occurrence may be strongly influenced by differences in product formulation, raw materials or manufacturing processes.

The comparatively high CV for Ni (54.11%) also indicates appreciable variation among products despite its highest overall mean concentration. This observation is important because the risk posed by individual products may differ considerably even when the products belong to the same general category of hair dye.

However, interpretation of the variability of Cr requires particular caution because only four quantified observations were available, with the remaining measurements reported as below detection limit (BDL). Consequently, the calculated Cr mean and CV represent only the quantified observations and should not be interpreted as a complete representation of chromium occurrence across all seven samples.

4.3.2 Ranking of Metals According to Mean Concentration

To determine the relative contribution of each metal to the overall measured metal burden, the metals were ranked according to their mean concentrations. The percentage contribution was calculated using equation 9 and the calculated values are shown in Table 6

$$Contributions\ (\%) = \frac{\overline{X_i}}{\sum \overline{C_i}} \qquad (9)$$

The resulting ranking is presented in Table 6.

Table 6:Overall ranking of heavy metals according to mean concentration

Rank	Metal	Mean concentration (mg/kg)	Contribution to summed mean concentrations (%)
1	Ni	0.04411	30.37
2	Mn	0.02796	19.25
3	Pb	0.02544	17.52
4	Co	0.02358	16.24
5	Cd	0.00961	6.62
6	Cu	0.00824	5.68
7	Cr	0.00585	4.03
8	As	0.00044	0.30

The ranking in Table 6 confirms that Ni was the dominant metal based on average concentration, accounting for approximately 30.37% of the summed mean concentrations. Mn, Pb and Co contributed 19.25, 17.52 and 16.24%, respectively. Together, these four metals accounted for approximately 83.38% of the summed mean concentrations, demonstrating that they constituted the major components of the measured heavy-metal profile.

Arsenic contributed only 0.30% and therefore represented the least abundant metal based on concentration. Nevertheless, its low concentration should not automatically be interpreted as indicating negligible health significance because risk assessment depends on both exposure concentration and metal-specific toxicological parameters. This

distinction is particularly relevant to the subsequent carcinogenic and non-carcinogenic risk assessment.

4.3.3 Total Heavy-Metal Burden of Individual Hair-Dye Samples

The total measured heavy-metal concentration for each hair-dye sample was estimated by summing the quantified concentrations of the individual metals. This was evaluated using equation 10

$$C_{total} = \sum_{j=1}^{m} C_j \quad (10)$$

where C_{total} is the total concentration of detected metals in a sample, C_j is the concentration of individual metal j, and mm is the number of metals quantified in that sample. The results are presented in Table 7.

Table 7: Total measured heavy-metal concentration by hair-dye sample

Sample	Total concentration of detected metals (mg/kg)	Mean concentration of detected metals (mg/kg)	Number detected
HD1	0.1002	0.01431	7
HD2	0.1016	0.01451	7
HD3	0.1545	0.02207	7
HD4	0.1473	0.01841	8
HD5	0.1664	0.02377	7
HD6	0.1321	0.01651	8
HD7	0.1735	0.02169	8

The results in Table 7 demonstrate substantial differences in the overall metal burden among the hair-dye products. HD7 recorded the highest total measured concentration (0.1735 mg/kg), followed by HD5 (0.1664 mg/kg) and HD3 (0.1545 mg/kg). HD1 exhibited the lowest total concentration (0.1002 mg/kg). Thus, the total measured metal burden of HD7 was approximately 73% higher than that of HD1.

The differences among samples indicate that the products did not have uniform heavy-metal profiles. This variability may arise from differences in pigments, stabilizers, raw materials, manufacturing processes or other formulation ingredients. Importantly, the total concentration should be considered a screening indicator rather than a direct measure of health risk because it assigns equal importance to metals with very different toxicological characteristics.

4.3.4 Correlation Analysis

Correlation analysis was performed to investigate whether variations in one metal were associated with variations in another. Because the number of samples was small and several measurements were BDL, Spearman's rank correlation coefficient (ρ\rho) provides a useful exploratory measure because it does not require the variables to follow a normal distribution.

Spearman's coefficient is calculated from the ranked observations according to equation 11

$$\rho = \frac{6\sum d_i^2}{n(n^2-1)} \quad (11)$$

where d_i is the difference between the ranks of paired observations and nn is the number of paired observations. The resulting correlation coefficients are presented in Table 8.

The strongest interpretable Spearman association was observed between As and Cd ($\rho = -0.847$), indicating a strong inverse relationship in the available data. Positive relationships were also observed between Mn and Cu ($\rho = 0.643$), Cu and Pb ($\rho=0.643$) and Mn and Pb ($\rho = 0.607$). These positive associations indicate that samples with

relatively high concentrations of one metal tended, in this small dataset, to have relatively high concentrations of the other.

Nevertheless, correlation does not establish a common source. The observed associations may reflect similarities or differences in product formulation and should therefore be regarded as exploratory. The correlations involving Cr are particularly unreliable because they are based on only four quantified observations.

4.3.5 Pearson Correlation and Statistical Significance

Pearson's correlation coefficient was additionally used to assess the linear association between selected pairs of metals. The coefficient was calculated based on equation 12

$$r = \frac{\sum(X_i-\bar{X})((Y_i-\bar{Y}))}{(X_i-\bar{X})^2(Y_i-\bar{U})^2} \quad (12)$$

The statistical significance of the correlation was evaluated at α = 0.05. The results are presented in Table 9.

Table 8: Spearman rank-correlation coefficients among heavy metals

Metal	Co	As	Mn	Cu	Cd	Cr	Ni	Pb
Co	1.000							
As	0.464	1.000						
Mn	-0.314	0.378	1.000					
Cu	-0.657	-0.162	0.643	1.000				
Cd	-0.543	**-0.847**	-0.143	0.357	1.000			
Cr	-1.000*	0.400	0.800	1.000*	-0.200	1.000		
Ni	0.600	-0.414	-0.036	-0.214	0.250	-0.800	1.000	
Pb	-0.143	0.523	0.607	0.643	-0.393	0.400	-0.429	1.000

Table 9. Selected Pearson correlation significance tests

Metal pair	Pearson r	p-value	Interpretation at α = 0.05
Co–As	0.182	0.730	Not significant
Co–Mn	-0.401	0.430	Not significant
Co–Cu	-0.296	0.569	Not significant
Co–Cd	-0.523	0.287	Not significant
Co–Ni	0.156	0.767	Not significant
Co–Pb	-0.052	0.923	Not significant
As–Mn	0.343	0.452	Not significant
As–Cu	-0.279	0.544	Not significant
As–Cd	-0.770	0.043	Significant
As–Ni	-0.789	0.035	Significant
Mn–Cu	0.327	0.474	Not significant
Mn–Pb	0.688	0.088	Not significant
Cu–Pb	0.574	0.178	Not significant
Cd–Ni	0.545	0.206	Not significant
Cd–Pb	-0.452	0.309	Not significant
Ni–Pb	-0.521	0.231	Not significant

The Pearson analysis identified statistically significant negative relationships between As and Cd ($r = -0.770$, $p = 0.043$) and between As and Ni ($r = -0.789$, $p = 0.035$) at the 5% significance level. The negative correlations indicate that samples with higher concentrations of As tended to have lower concentrations of Cd and Ni within the dataset. However, these findings should be interpreted conservatively. The analysis is based on only seven products, resulting in low statistical power. Furthermore, a relatively large number of pairwise correlations were tested, increasing the possibility of obtaining apparently significant relationships by chance. Therefore, the observed relationships should not be interpreted as definitive evidence of common or contrasting sources. Additional independent hair-dye samples would be required to confirm these associations.

4.X.6 Principal Component Analysis

Principal component analysis was employed as an exploratory multivariate technique to reduce the dimensionality of the heavy-metal dataset and identify combinations of metals contributing to the observed variation among products. Prior to PCA, the variables were standardized to prevent metals with relatively larger numerical concentrations from dominating the analysis.

The principal components were obtained from the standardized correlation matrix, and the proportion of variance explained by each component was determined from its eigenvalue (equation 13)

$$Variance\ explained(\%) = \frac{\lambda_i}{\sum \lambda_i} \times \frac{100}{1} \quad (13)$$

where λ_i is the eigenvalue associated with principal component i. The reported PCA variance structure is presented in Table 10. The first three principal components accounted for 88.27% of the total variance, indicating that most of the variation in the dataset could be represented by three principal dimensions. PC1 explained 46.27% of the variance, while PC2 and PC3 explained 24.76% and 17.24%, respectively. Consequently, the loading matrix is presented in Table 11.

Table 10. PCA explained variance

Princip/	Eigenvalue	Variance explained (%)	Cumul variance (%)
PC1	3.701	46.27	46.27
PC2	1.981	24.76	71.03
PC3	1.379	17.24	**88.27**
PC4	0.688	8.60	96.87
PC5	0.212	2.64	99.51
PC6	0.040	0.49	100.00

**** Princip = Principal, Cumul = Cumulative**

Table 11. PCA loading matrix

Metal	PC1	PC2	PC3
Co	-0.395	-0.294	-0.381
As	0.445	-0.340	0.001
Mn	0.296	0.290	-0.356
Cu	0.087	0.596	-0.258
Cd	-0.260	0.519	0.390
Cr	0.359	0.113	0.533
Ni	-0.432	0.200	-0.160
Pb	0.409	0.191	-0.446

The PCA loading pattern suggests that PC1 was primarily influenced by As, Ni and Pb, whereas Cu and Cd contributed more strongly to PC2. PC3 showed relatively strong contributions from Cr, Cd and Pb. These results suggest that different groups of metals may contribute to different dimensions of variation among the hair-dye products.

However, the PCA should be interpreted strictly as exploratory because only seven samples were available. In addition, the presence of BDL values, particularly for Cr, can substantially influence multivariate analyses depending on how censored observations are treated.

4.3.7 PCA Scores of Individual Hair-Dye Samples

The sample scores associated with the first three principal components are presented in Table 12.

Table 12:PCA scores for individual hair-dye samples

Sample	PC1	PC2	PC3
HD1	4.029	0.201	1.009
HD2	0.691	-2.290	-0.415
HD3	0.572	-0.013	-1.663
HD4	-2.329	-1.407	0.211
HD5	-0.405	2.353	-1.256
HD6	-1.353	1.024	2.017
HD7	-1.205	0.132	0.097

HD1 showed the most pronounced positive PC1 score (4.029), distinguishing it from the other products. HD5 and HD6 displayed relatively high positive PC2 scores, whereas HD4 had a strongly negative PC1 score. These differences indicate that the overall elemental profiles of the hair-dye products were not homogeneous. The separation of HD1 is particularly noteworthy because PCA considers the combined pattern of all metals rather than their individual concentrations. Therefore, a sample can be separated in PCA space because of its distinctive combination of metals even when it does not necessarily have the highest total concentration.

4.3.8 Hierarchical Cluster Analysis

Hierarchical cluster analysis was further used to investigate similarities among the hair-dye products based on their standardized heavy-metal profiles. Ward's method was applied to identify groups of samples exhibiting relatively similar multielement characteristics. The clustering pattern in Table 13 indicates that HD6 and HD7 had relatively similar multimetal profiles, while HD2 and HD3 also showed similarities. HD1 was the most distinctive sample in the final grouping. The clustering results are broadly consistent with the PCA, which also indicated separation of individual products according to their combined metal profiles.

The agreement between PCA and HCA strengthens the interpretation that the hair-dye samples exhibited measurable differences in their overall elemental composition. Nevertheless, because the analysis involves only seven samples, the clusters should be considered exploratory and should not be used to infer definitive manufacturing or geographical sources.

Table 13: Exploratory hierarchical clustering of hair-dye samples

Cluster level	Samples grouped
Initial cluster	HD6–HD7
Initial cluster	HD2–HD3
Subsequent grouping	HD4 with HD5/HD6/HD7 group
Final grouping	HD1 separated from the remaining samples

4.3.9 Overall Statistical Interpretation

The major statistical findings are summarized in Table 14.

The statistical analysis demonstrates considerable heterogeneity in the heavy-metal composition of the analysed hair dyes. Ni was the predominant metal based on mean concentration, while Pb exhibited the greatest consistency among products. In contrast, Cu and Cr displayed very high relative variability, indicating substantial differences in their concentrations between products. The correlation analysis revealed some potentially meaningful relationships, particularly the inverse As–Cd and As–Ni relationships, but the small sample size prevents definitive source attribution.

The PCA and HCA further demonstrated that the products differed in their overall multielement composition. The first three principal components accounted for 88.27% of the total variance, while the clustering analysis identified groups of relatively similar products, particularly HD6–HD7 and HD2–HD3. These findings indicate that evaluating individual metals separately may not fully describe differences between hair-dye products; multivariate approaches provide additional information regarding their overall elemental fingerprints.

Table 14. Summary of major statistical findings

Parameter	Result	Major observation
Highest mean metal	Ni = 0.04411 mg/kg	Ni was dominant by concentration
Lowest mean metal	As = 0.00044 mg/kg	As had the lowest concentration
Highest CV	Cu = 102.88%	Greatest relative variability
Lowest CV	Pb = 10.78%	Most consistent concentration
Highest total sample concentration	HD7 = 0.1735 mg/kg	Highest combined detected-metal burden
Lowest total sample concentration	HD1 = 0.1002 mg/kg	Lowest combined detected-metal burden
Strongest Spearman association	As–Cd, ρ = -0.847	Strong inverse association
Significant Pearson relationship	As–Cd, p = 0.043	Significant at α = 0.05
Significant Pearson relationship	As–Ni, p = 0.035	Significant at α = 0.05
PC1 variance	46.27%	Largest component
PC2 variance	24.76%	Second-largest component
PC3 variance	17.24%	Third-largest component
PC1–PC3 cumulative variance	88.27%	Most variation represented
Closest exploratory cluster	HD6–HD7	Similar overall profiles

4.0 Conclusion

This study evaluated the concentrations, distribution, and potential health risks associated with heavy metals in commercially available hair dyes sold in Nigeria. The results demonstrated that Ni had the highest mean concentration (0.04411 mg/kg), followed by Mn (0.02796 mg/kg), Pb (0.02544 mg/kg), and Co (0.02358 mg/kg), whereas As recorded the lowest mean concentration (0.00044 mg/kg). Considerable variability was observed among the products, with Cu showing the highest coefficient of variation (102.88%), while Pb exhibited the lowest (10.78%), indicating substantial differences in the elemental composition of individual products. At the product level, HD7 had the highest total measured heavy-metal concentration (0.1735 mg/kg), whereas HD1 had the lowest (0.1002 mg/kg).

The statistical analyses further demonstrated that the hair-dye products had distinct multimetal profiles. Spearman correlation revealed a strong inverse association between As and Cd ($\rho = -0.847$), while Pearson analysis identified significant negative relationships between As–Cd ($r = -0.770$, $p = 0.043$) and As–Ni ($r = -0.789$, $p = 0.035$). Exploratory PCA showed that the first three principal components accounted for approximately 88.27% of the total variance, while hierarchical clustering indicated similarities between selected products, particularly HD6–HD7 and HD2–HD3. These findings suggest that differences in product formulation and raw materials may contribute to the observed variation in heavy-metal composition.

The health-risk assessment indicates that the potential significance of the detected metals cannot be determined solely from their concentrations, because dermal absorption,

exposure duration, toxicity reference values, and carcinogenic potency differ among metals. Consequently, the calculated hazard quotients, hazard indices, and carcinogenic risks provide a more appropriate basis for evaluating potential consumer health implications. Overall, the findings demonstrate the presence of multiple heavy metals in Nigerian hair-dye products and highlight the need for continued surveillance and effective regulatory control of their elemental composition. Regular monitoring, improved quality control by manufacturers, transparent disclosure of metal-containing ingredients, and enforcement of permissible contaminant limits are recommended to minimize potential long-term exposure of consumers to hazardous metals.

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Author Contributions

Henrietta I. Kelle conceived and coordinated the study, designed the methodology, performed data analysis, and prepared the manuscript. Festus J. Ehijene and Emeka C. Ogoko contributed to sample collection, laboratory investigations, and data interpretation. Adebisi A. Fagbohun supported analytical procedures and quality control. Bethel O. Ekute and Chinyere V. Ogbodo contributed to statistical analysis, literature

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