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Estimation of heavy metals (Pb, Cd, Zn) in raw, commercial and traditional Eye-khol samples sold in Libya

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Abstract:
Libya and other Arab nations have long used cosmetics, especially traditional Eye-khol, for both cosmetic and therapeutic reasons. The purpose of this study was to determine the levels of heavy metals (Pb, Cd, and Zn) in eyeliner pens, tattoo supplies, and different kinds of eye-khol (raw, commercial, and traditional) that are sold in Libyan markets. The aim was to assess potential health risks and to compare the results with the international legislation safety standards. A total of 27 samples were analysed: 12 eye-liners, 6 tattoo products and 9 raw and commercial eye-khols. Dry digestion method was used and the concentrations of the metals were measured by flame atomic absorption spectrophotometry (FAAS). Lead (Pb) levels in traditional Eye-khol samples were found to be alarmingly high, with some exceeding 10000 µg/g, well above the WHO's safe limits. All product types had detectable levels of cadmium (Cd) and zinc (Zn), with some commercial eyeliners and tattoo products surpassing allowable limits. Results revealed that kohl samples have the highest concentration of most of the studied metals, particularly Pb. The common practice of applying traditional Eye-khol to infants raises serious health concerns because it exposes them to elevated Pb levels. These results highlight how urgently stronger regulatory monitoring and greater public education about the risks of heavy metal pollution in the environment are needed.

Keywords: Heavy metals, Libya, Cosmetics, Eye-Kohl, Eyeliner, Tattoo

تقدير مستويات المعادن الثقيلة (الرصاص، الكادميوم، والزنك) في عينات الكحل الخام والتجاري والتقليدي المباعة في ليبيا

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الملخص
لطالما استخدمت ليبيا وغيرها من الدول العربية مستحضرات التجميل، وخاصة الكحل التقليدي، لأغراض تجميلية وعلاجية. هدفت هذه الدراسة إلى تحديد مستويات المعادن الثقيلة (الرصاص Pb، الكادميوم Cd، والزنك Zn) في أقلام تحديد العين، مستحضرات التاتو وأنواع مختلفة من الكحل (الخام، التجاري، والتقليدي) المتوفرة في الأسواق الليبية. كما سعت الدراسة إلى تقييم المخاطر الصحية المحتملة ومقارنة النتائج بالمعايير الصحية التي وضعتها التشريعات الدولية. تم تحليل 27 عينة: 12 من أقلام تحديد العين، 6 من مستحضرات التاتو، و9 من الكحل الخام والتجاري. تم استخدام طريقة الهضم الجاف وقياس تركيز المعادن باستخدام مطياف الامتصاص الذري باللهب (FAAS). أظهرت النتائج أن مستويات الرصاص في عينات الكحل التقليدي كانت مرتفعة بشكل مقلق، حيث تجاوزت بعض العينات 10000 ميكروغرام/غرام، وهي أعلى بكثير من الحدود الآمنة التي أوصت بها منظمة الصحة العالمية (WHO). كما تم الكشف عن وجود الكادميوم والزنك في جميع أنواع المنتجات، وتجاوزت بعض أقلام التحديد التجارية ومستحضرات التاتو الحدود المسموح بها. أظهرت النتائج أن عينات الكحل احتوت على أعلى تركيز لمعظم المعادن المدروسة، وخاصة الرصاص. تثير الممارسة الشائعة المتمثلة في استخدام الكحل التقليدي للأطفال الرضع مخاوف صحية خطيرة بسبب تعرّضهم لمستويات عالية من الرصاص. كما تسلط هذه النتائج الضوء على الحاجة الملحة لتعزيز الرقابة التنظيمية وزيادة الوعي المجتمعي بمخاطر تلوث البيئة بالمعادن الثقيلة.

الكلمات المفتاحية: المعادن الثقيلة، ليبيا، مستحضرات التجميل، الكحل، محدد العين، التاتو.

Introduction

The use of cosmetics has been widespread throughout history, with their formulations evolving over time. However, the increasing prevalence of these products has raised concerns about their safety, particularly regarding the presence of heavy metals such as lead (Pb), cadmium (Cd), and zinc (Zn). These metals, even in trace amounts, can pose significant health risks, especially when absorbed through the skin or mucous membranes. Eye-kohl, a traditional cosmetic widely used in Arab countries, including Libya, has been identified as a potential source of heavy metal exposure [1-4]. Few people are aware that cosmetics can be absorbed through the skin, and the absorbed dose may be sufficient to cause hazardous effects [5]. There are also no reports specifying the permissible levels of certain heavy metals such as Pb, Cd, Zn in personal care products that have vital effects, and it is therefore difficult to verify the concentration of those metals recorded in the studies is prohibited but the concentrations are lower [6]. Generally, many cosmetic ingredients were not organized under the Food and Drug Administration (FDA) [7]. Many cosmetics producers do not write their ingredients on the product (because it is not tested) and also avoid the results of studies based on those ingredients on the grounds that they are intellectual property of companies [8].

Cosmetic Safety:

Every day the consumer is exposed to about 25 different beauty products. If each product contains 10 different ingredients, it means the consumer is exposed to more than 200 different chemical compounds that directly relate to different parts of the body and some of these parts are sensitive as an area around the eye and oral cavity [3]. Eye-khol is one of the most important cosmetics that is widely used in the Arab countries and in Libya in particular. It is used for the baby after birth to beautify the eyes and as a folk remedy for the location of the umbilical cord, used by women and men of varying ages and women are the most used category. According to the Prophet Mohammed said "It bears sight and springs poetry" [9]. Furthermore, Al-Ethmed, which Prophet Mohammed (Peace be upon him) was using and recommended to use is defined as a black stone hit to the reds that grinds to use its powder to shrink the eye and is often used to beautify and be in the Arabic countries. One of its characteristics is that it strengthens the eyelid follicles and benefits the eyelashes to further prolong, thereby increasing their ability to save eyes from sunlight, dust and dirt and increase visibility [10]. Chemically, Al Ethmed is known as the third antimony sulfide or the fifth antimony sulfide, which is semi-metallic and its shape in the case of sulfide is the main source and is a fragile, rapidly fragmented metal with a laminated composition and has many pharmacological properties, exterminates many parasites, such as lichmanias, bilharsias and filaments, and is used in Britain to treat bilharzia [11].

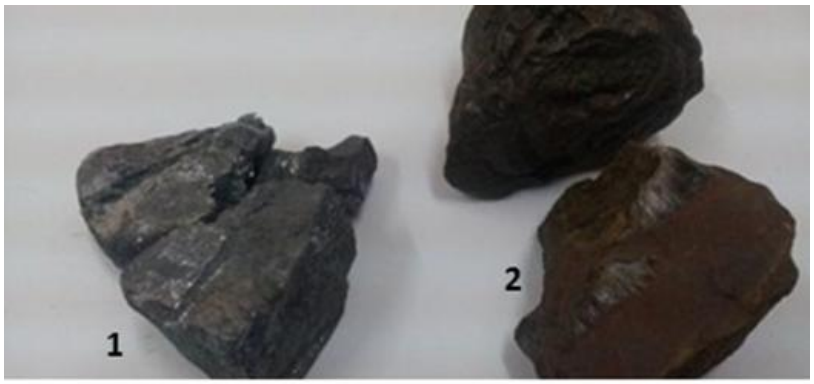

Figure 1 the difference between galena (1) and Al-Ethmed (2).

As for the eye-kohl used in Libya, the lead sulfide PbS (galena) is the most important raw metal for lead [12], where lead is 86.6% and sulfur is 13.4%, the metal crystallizes in the cube family and its atoms are sequenced so that each lead atom is surrounded by six sulfur atoms, In the form of eight, as each sulphur atom is surrounded by six atoms of lead. Eye-kohl is an example of traditional cosmetics used in the Middle East and North Africa region. In fact, eye-kohl (Surma) and eye cosmetics were identified as sources of exposure and lead poisoning on a visual system in a number of adults and children [13-15]. Eye-kohl by the US Food and Drug Administration (FDA) is considered unsafe for use and an illegal substance so that it is not imported or sold in the US [7-15], while in other countries such as Libya, it is still sold widely without any control.

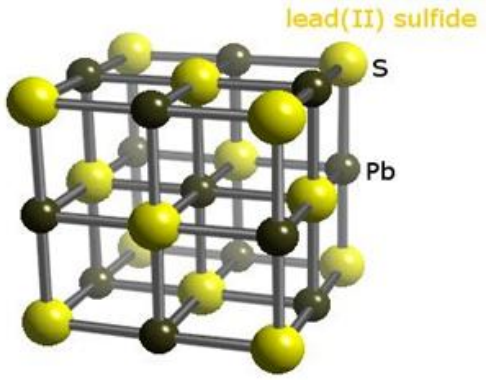

Figure 2 Galena Element (PbS) structure (16).

Health risk of heavy metals

Cadmium Element (Cd): A non-essential element of the life of organisms, it is very toxic to terrestrial and aquatic organisms even when a small concentration and is very toxic to human life than other elements, carcinogenic and disease-inducing (17,18). The accumulation of Cd in the body and its effect on it biologically, cumulatively and toxically depends on the body's nutritional condition and the level of iron in the body, Cd is a devastating nerve poison to the kidney and bones and must minimize exposure to it. The estimated dose of Cd showing destruction of the kidney is (0.3 - 3 µg/g) as well as destroying the bone (19). It is forbidden to add cadmium to cosmetics in any quantity and long use of cosmetics containing these elements give a threat to human health. Unfortunately, there are no reports of such ingredient levels in cosmetics and the permissible limits in Libya.

Lead Element (Pb): It has a harmful effect in the body as it deposits in the liver and bones and expel calcium from bones and replace it with osteoporosis, especially in pregnant women which increases the child's absorption of lead and increases his or her harm (20). The amount of Pb in the blood and tissue is commensurate with the time and duration of exposure to it and therefore toxicity is determined by the acute or minor toxicity, and to know the amount of toxicity measured lead element µg/dl blood, as the U.S. Centers for Disease Control and Prevention (CDC) and WHO reported the World Health Organization (WHO) that the level of Pb in the blood higher than 10 micrograms per deciliter (µg/dl) causes cancer. CDC uses a blood lead reference value (BLRV) of 3.5 (µg/dl) to identify children with blood lead levels that are higher than most children's levels, that means even at lower levels causes risk to human health and that the safe level is unknown (21,22). There are studies that guess that eye-kohl is a cause of lead poisoning, where eye-kohl is used with great popularity in the Arab world and Libya in particular.

Zinc Element (Zn): The zinc element is an important element needed by the human body but in small quantities where it is involved in many important processes such as maintaining blood sugar and strengthening immunity, but its increase in blood leads to obvious symptoms such as feeling bitter in the mouth, vomiting, nausea, stomach pain and these symptoms are similar to poisoning symptoms.

Increasing the zinc element in the blood leads to a lack of copper element in the body and the speed of stones and kidney failure (23). Daily application of products and cosmetics with large amounts of lead, cadmium, zinc, on the skin causes health problems. Heavy metals have binding with cell proteins by forming complexes with carboxylated acids (- COOH), amine (-NH2), and thiol (- SH), impeding their functions and causing cell death leading to multiple diseases (24).

A study was conducted in Saudi Arabia in 2004 on eye-kohl samples (25), where lead levels of up to 53% were detected in some eye-kohl samples. From a blood analysis of users of ordinary eye-kohl, high lead concentration and relatively low hemoglobin levels were revealed and this leads to health risks. Lead concentrations were high in eye-kohl samples studied in Morocco in 2001 (26), where the study showed that lead concentrations in eye-kohl were very high but low in mixed eye-kohl with some products where lead concentration lower however, in both types of eye-kohl, concentrations were very high and therefore posed a risk to public health, especially for children (In a study assessing the health risks of heavy metals in traditional cosmetics sold in Tunisian domestic markets, published in 2016 (27), the results of this study revealed that the contents of lead, cadmium, copper and zinc in the investigative samples were high, making the prolonged use of these products a potential risk to human health.

Pb, Cd and Zn can also be estimated in cosmetics products using several technics such as X-ray fluorescence (XR-F), Gas Chromatography Mass (GC-MS) and Atomic Flame Absorption Spectrophotometer (FAAS), after digesting the sample, precise, fixed method for preparations containing significant overlap of their contents of items under detection limits in devices (21). It should be noted that, in Libya there is no regulation relating to

the heavy metals content in cosmetics, our data were compared with the similar studies and the international standards available in literature. This study aims to estimate heavy metals (Pb, Cd and Zn) in several types of crude, commercial and home-made eye-kohl prepared in traditional ways, as well as to estimate the same metals in different samples of tattoo and eyeliner manufactured by international companies with a traded mark sells in Libyan local markets using atomic absorption spectrophotometer (AAS). The concentrations of these metals have also been compared with those allowed in the European Union legislation (EU) (28), World Health Organization (WHO) (29), and Canadian authorities (30). In addition, this study aims to highlight the toxicity and risk of heavy metals studied to public health. The study also aims to raise women's awareness of the purchase and use of commercial products of unknown composition and industry and to limit the use of some types of eye-kohl, and to renounce the habits that oblige the use of traditional eye-kohl for infants whether for make-up or treatment.

Materials and Methods

Materials

Table 1 and Figure 3 provide detailed information regarding to the code and origin of the samples, which were categorized into 3 groups: 12 eyeliner pencils, 6 different commercially popular tattoo samples, and 8 samples of both raw and commercial eye-kohl purchased from markets in Sebha City. The brands were carefully chosen to represent the types that people in different economic brackets use. Additionally, one more sample was obtained through a personal connection and consisted of a homemade preparation made from date stones.

Hydrochloric Acid Concentration 20%, Distilled water, Burning oven from German company Naber, Balance Sensitive Mettler Chinese Company. Pb, Cd and Zn were estimated in cosmetic samples using Atomic Absorption Spectrophotometer (AAS), a brand ThermoElement with the use of a special cathode bulb for each item and adjusting the device using pre-equipped standard solutions for each metal to be estimated for use in calibrating the device and making standard curves.

Table 1: List of samples, their codes and origin of the collected samples

Brand name	Sample code	Color	Origin
Eyeliner pencil			
Bellaoggi	E1	Black	Italy
Last touch 24H	E2	Black	P.R.C.
Last touch kajal	E3	Black	P.R.C.
Expression kajal (Maybelline NewYork)	E4	Black	USA
Colossal kajal (Maybelline NewYork)	E5	Black	USA
Essence (kajal pencil)	E6	Black	Germany
Reserve Naturelle (Crayon khol)	E7	Black	France
Deborah (Extra Eyepencil)	E8	Black	Italy
Last touch	E9	Black	Unknown
Fatena (Eyeliner pencil)	E10	Black	P.R.C.
Deborah Kajal	E11	Black	Pakistan
Max Factor	E12	Black	Italy
Tattoo			
Peacock dye	T1	Black	Japan
Penakie	T2	Black	Spain
Last touch tattoo	T3	Black	P.R.C.
Forever	T4	Brown	Germany
Ever Beauty	T5	Black	USA
Kajal	T6	Black	Turky
Eye-Kohl (powder)			
Al-Hashimi paste	K1	Black	Pakistan
Sharifeen	K2	Black	India
Date stones	K3	Black	Pakistan
Al-Hashimi (coppery)	K4	Black	Pakistan
Al-hashimi (Golden)	K5	Black	Pakistan
Al-hashimi (Silver)	K6	Black	Pakistan
Traditional Eye kohl (1)	K7	Black	Home-made
Traditional Eye kohl (2)	K8	Black	Home-made
Raw Eye-kohl	K9	Black	Home-made

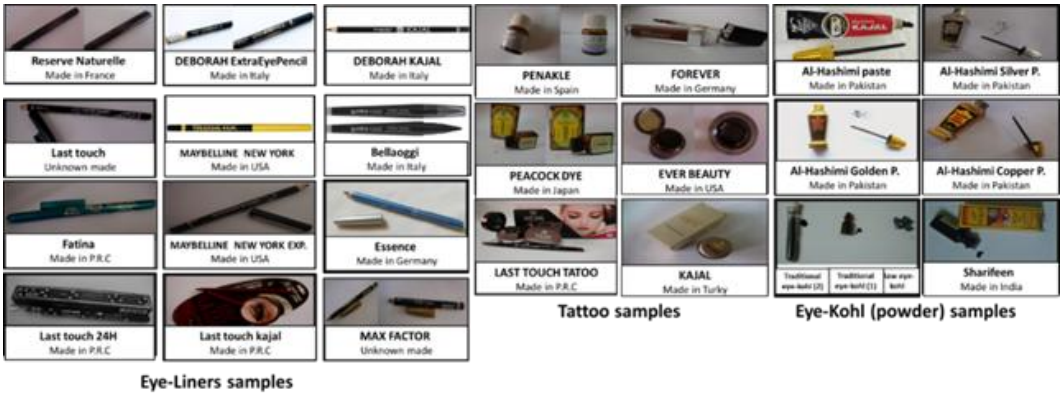

Figure 3 Trade names and origins of Eyeliner, Tattoo and Eye-kohl samples

Method used

The pulp from the eyeliner pencils was meticulously separated to prevent contamination from the surrounding wood or plastic materials. Both the tattoo and eye-kohl samples were prepared using the same method [31]. All samples were processed using the dry digestion method (dry ashing), where 1g of each sample, weighted with a high-precision balance (four decimal places), was placed in a flame-resistant container and inserted into a combustion oven at 100°C. The temperature was gradually raised to 500°C over 5 hours until the combustion process was complete and the samples turned to ash. The samples were then allowed to cool to room temperature. A 20% hydrochloric acid (HCl) solution was prepared, and 5 ml of this solution was added to the ash to dissolve it. The mixture was filtered using Watman No. 4 filter paper to remove any undissolved particles, and the filtrate was collected in a 50 ml standard volumetric flask, which was then filled to the mark with distilled water. Every digested sample was analysed in triplicate for lead (Pb), cadmium (Cd), and zinc (Zn). Standard solutions were prepared and analyzed for selected metals using an atomic absorption spectrophotometer in the laboratories of the Chemistry Department at the Faculty of Science, University of Sebha.

Results And Discussion

The concentration of three heavy metals (Pb, Cd, Zn) was studied in 12 samples of eyeliner pens, 6 tattoo and 9 traditional, raw and commercial eye-kohl using FAAS.

Lead (Pb)

The concentrations of selected heavy metals were assessed in different brands of eyeliner. The findings revealed that the average concentration of Pb in the eyeliner samples was 6.92 µg/g, which was higher than that detected in the tattoo samples (0.903 µg/g) while, both lower than the EU permissible limit (20µg/g). However, The highest concentration was reported in natural and commercial traditional kohl samples, reaching 5328.43 µg/g. This elevated level is likely attributed to the composition of raw kohl, which consists of 86.60% Pb and 13.40% sulfur. Figure 4 shows some eyeliner samples (E1, E2, E3, E4, E5, E6 and E7) had Pb concentration below the detection limit (0.01 µg/g). Pb concentration of the rest of eyeliner samples (E8, E9, E10, E11 and E12) was (0.295, 0.760, 3.545, 11.895, 66.530 µg/g) respectively, while the only last value was higher than the EU permissible limit (20µg/g) may be due to the additives added.

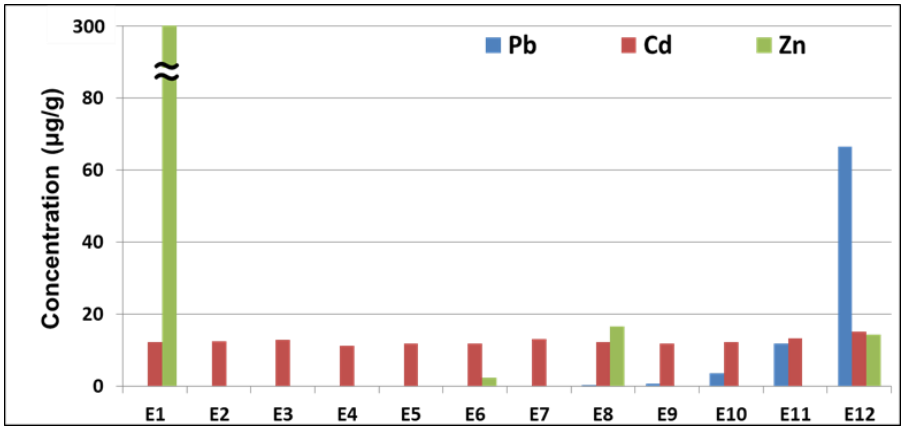

Figure 4 shows the concentration of Pb, Cd and Zn in eyeliner pen samples.

In tattoo samples, the Pb concentration for all samples (T2, T3, T4, T5, T6) was below the detection limit (0.01 µg/g), except for the T1, where the concentration of the lead is reached (5.420 µg/g) that is below the EU permissible limit, Fig. 5. Notwithstanding the warning, is written in the paper attached to the box of the dye which is strongly cautioned to be used on the cilia, the peripheral area of the eye or on the eyebrows.

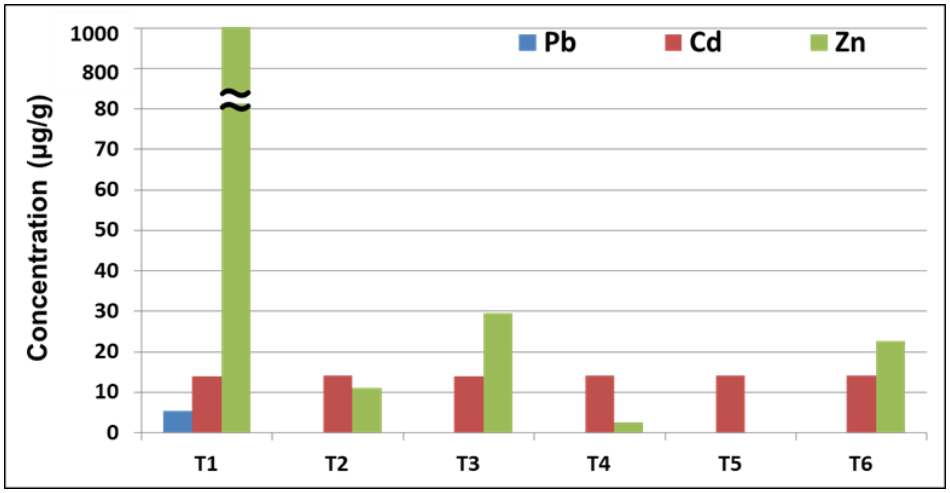

Figure 5 shows the concentration of Pb, Cd and Zn in Tattoo samples.

In eye-kohl samples, the concentration of Pb in all samples was above the EU permissible limit, while, K1 (2.115 µg/g), K2 (36.520 µg/g) and K4 (47.205 µg/g) were lower than other samples may be due to adding some products to the kohl which reduces Pb concentration. High concentration was recorded for K9 (10324.4 µg/g), K5 (1017.4µg/g) and K6 (10392.9 µg/g), this rise may be due to the fact that no other products have cleaned the galena metal (the main component of the eye-kohl). The study found that Pb concentration in K7 and K8 samples (9683.7 µg/g and 10384.5 µg/g respectively) containing additives such as pond beads, sea tongue had a Pb concentration. This is consistent with a study conducted in the State of Morocco [26]. In the case of K3, the concentration of Pb was found to be 6067.2 µg/g, as illustrated in Figure 6. This notable increase could be attributed to contamination from irrigation water, agricultural soil, or the possibility that the samples were mixed with galena at the source.

Cadmium (Cd)

Figures 4,5,6 show the results of average concentration of cadmium in eyeliner (12.546 µg/g), tattoo (14.051 µg/g), raw, traditional and commercial eye-kohl (14.017 µg/g), respectively. In general, all samples had close Cd concentrations ranging from 11.35 to 17.01 µg/g may be due to the colored substance containing this element. The concentration of cadmium in all samples were above the maximum allowed limits set by the EU (5.0 µg/g).

Zinc (Zn)

The results showed that the average concentration of the zinc element in the eyeliner samples was (27.979µg/g) which is lower than the average concentration in the tattoo samples (177.133µg/g). However, the highest concentration was recorded in raw and commercial kohl samples (604.590µg/g). The total concentrations of Zn in eyeliner samples were below the permissible food limit (50 µg/g) as follows: (E2, E3, E4, E5, E7, E10, E11) where, Zn concentration was lower than the detection limit (0.01 µg/g). However, was 14.275, 16.635, 2.300 µg/g for E12, E8, E6 samples, respectively. E1 sample was exception while, Zn concentration was 302.540 µg/g, Fig 3.

Figure 4 shows that all Tattoo samples (T2, T3, T4, T6) have Zn concentration of 11.125, 29.495, 2.500, 22.595 respectively, which are below the permissible limit (0.01 µg/g). The highest value was detected in T1 sample (997.085 µg/g).

Figure 5 shows the concentrations of eye-kohl samples, while Zn concentration was above the permissible food limit with the exception of K8 which was (27.306 µg/g) and K9 (42.660 µg/g). However, for K4 was 1030.660 µg/g, K6 was 817.670 µg/g, K5 was 827.100 µg/g, K1 was 1051.907 µg/g, K7 was 540.071 µg/g and K3 was 159.313 µg/g. It should be noted that there is no permissible limit for cosmetic products was found.

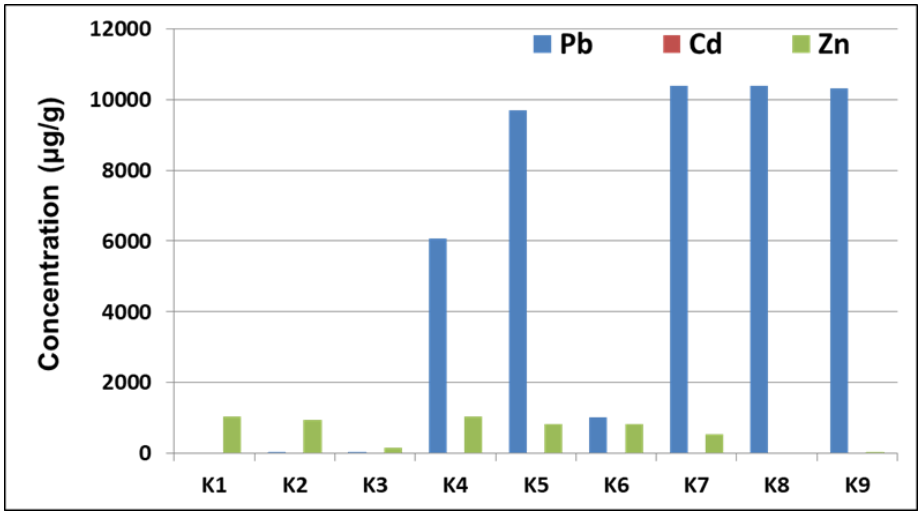

Figure 6 shows the concentration of Pb, Cd and Zn in raw, traditional and commercial eye-kohl

Comparison the results with previous studies

When comparing the results of this study with findings from previous research, there appears to be a noticeable variation in the levels of lead (Pb), cadmium (Cd), and zinc (Zn) found in Kohl products from different regions. Several international studies have consistently detected these metals in all tested Kohl samples, although the concentrations vary widely. For instance, research conducted in Tunisia [27], Algeria [32], Spanish and German [33], Europe [34], Nigeria [35] and Pakistan [36] also found lead present in most of the samples they tested, but the amounts differed significantly from one study to another as shown in Table 2. However, all of the kohl (powder) samples described in Table 2 had excessive levels of lead.

Table 2. A comparison of selected metal concentrations (µg g^{-1}) in eyeliner pencil and kohl samples of this study with some published data samples reported in previous studies.

Type of sample	Ref.	Origin	No. Sample	Method	Analytical method	content (µg/g)		
						Pb	Cd	Zn
Kohl (surma)	(27)	Tunis	11	Acid digestion	AAS	51.1 – 4839.5	1.0 – 158.6	0.7 – 185.0
Local kohl	(32)	Algeria	16	NM	AAS	3890 - 85570	-	-
Kohl	(33)	Germany and Spain	12	Microwave	ICP-OES	1.7 - 410000	0.0006 – 20.7	2.0 - 360306
(Kohl+paste+pencils)	(34)	Europe	23	NM	XRF	8.0 - 467632	14.5 - 369	94 - 380827
Eyeliners	(35)	Nigeria	20	Acid digestion	FAAS	9.60 – 322.5	2.80 – 13.5	9.2 – 33.0
Eyepencil	(35)	Nigeria	20	Acid digestion	FAAS	3.30 – 33.8	0.70 – 5.20	107 – 456.2
Kohl (surma)	(36)	Pakistan	15	Dry-ashed digestion	AAS	2.77 - 1071	0.095 – 0.94	1.36 – 508.8
Eyeliners	This study	Libya	12	Dry-ashed digestion	FAAS	1.0 – 66.53	11.35 – 15.13	0.1 – 302.54
Tattoo	This study	Libya	6	Dry-ashed digestion	FAAS	1.0 – 5.42	13.82 – 14.18	001.0 – 997.08
Kohl	This study	Libya	9	A Dry-ashed digestion	FAAS	2.1 - 10392.9	12.22 – 17.01	27.31 – 1051.9
EU limit	(28)					20	5	NM
WHO limit	(29)					0.5	0.5	NM
Canadian Limit	(30)					10	3	NM

NM: Not Mentioned, (F)AAS: (Flame) Atomic Absorption Spectrometry, ICP-MS: Inductively Coupled Plasma- Optical Emission Spectroscopy, XRF: X-ray fluorescence.

Recommendations

- We advise more research on additional heavy metals in different kinds of eye kohl.
- Women should limit their daily use of cosmetics and refrain from using eyeliner pens of unknown origin.
- Because traditional eye kohl contains high levels of heavy metals, especially lead, which can have major negative health effects, mothers should avoid using it on their babies.
- Regular inspections of cosmetic products by regulatory authorities are essential to detect and prevent the sale of hazardous products, especially those without quality tags.

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Disclaimer

The article has not been previously presented or published, and is not part of a thesis project.

Conflict of Interest

There are no financial, personal, or professional conflicts of interest to declare.

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