



## Assessment of lead levels in traditional eye cosmetic “kohl” frequently used in Morocco and Health hazard

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### Abstract

This study aims to investigate the content of lead in most frequently used brands of cosmetics products in Morocco. 100 samples of kohl (kohl powder, galena, eye pencils and paste) were selected taken from large cosmetic stores in Morocco. And lead of them is analyzed using a Fast Sequential Atomic Absorption Spectrometer (SAAFS 240). The results showed that the concentrations of lead in kohl samples were within the range of 0.01 mg/g and 973.8 mg/g. There were significant differences between the averages of lead content in different brands of the kohl samples. Thus the continuous use of these cosmetics can increase the absorption of heavy metals especially lead into the human body.

**Keywords:** Lead, Lead poisoning, Kohl, AAS, Galena, Health hazard.

### Introduction

Lead (Pb), one of the oldest known metals, it is a pervasive and persistent environmental occupational toxic metal, and lead poisoning remains a health threat [1]. It is not biodegradable and therefore persists in the environment for long periods, its accumulation in the environment can affect the human health [2]. Because it is a naturally occurring element in the earth's crust, lead is widely distributed through the environment, it can also affect the overall quality of the water [3]. Lead exposure mainly arises from contact with Pb based paint in home, fertilizers, cosmetics, automobiles, disposable batteries, etc., especially in developing countries [4].

Lead affects virtually every system in the body [5], several lines of evidence implicate that lead exposure causes many pathological incidence including cardiac [6], hepatic [7], behavioural [8], immunological [10], renal [11], and haematological [12]. More than 90% of the lead body burden is localized in bone with an average half-life of more than 20 years [13]. Bone releases lead during periods of increased bone turnover in women's lives, such as pregnancy, lactation, and menopause [14-15-16]. It is well established that lead can cross the placenta during the pregnancy and has been associated with intrauterine foetal death, premature delivery and low birth weight [17]. There are numerous reports and research papers on the other potential sources for lead exposure that are hidden and need to be addressed, these include ethnic folk remedies and cosmetics, pottery, toys, and certain imported candies and spices [5-18-19-20-21-22-23]. Eye cosmetics could be another source of heavy metals exposure [24-25-26].

The use of leaded eye powder (surma or kohl) has been associated with elevated blood lead levels in children and women [27]. The FDA defined the kohl as a color additive and there is no regulation permitting its use in a cosmetic or in any FDA regulated products [28].

The use of kohl as eyeliner is a popular practice in the Middle East, Asia and Africa. The use of kohl for medical and cosmetic purposes can be traced back over many centuries. Medicinally, it is used to stop bleeding and after circumcision for hygienic measures. All ethnic communities and people belonging to different religions use kohl in these areas of the world [29].

In Morocco, the use of kohl as a traditional eye cosmetic is very common especially between women, children and babies. Some mothers applied kohl to their infants soon after birth, and they blackened the baby's eyes dabbed kohl on their umbilical cord. Some others did this to strengthen the child's eyes and to prevent the child from being attacked by the evil eye [30]. There are several studies around the world that were interested in the kohl and its lead content [29-30-33]. In Morocco, there is insufficient data regarding lead content in cosmetics particularly in the traditional kohl, the only available study is that of Lekouch which was conducted in 2001 on a few samples of kohl collected at the city of Marrakech [20]. Our study included a number of fairly representative samples which involved 23 Moroccan cities, this marks the uniqueness and originality of this research.

In the present study, we investigate lead content in different brands of kohl samples which were collected from various stores in Moroccan markets in order to detect the presence of lead.

## **2. Materials and methods**

### *2.1. Sample collection*

The sampling methodology for kohl is called representative random sample (Center of expertise in environmental analysis of Quebec Centre) [31], it is used when a substance is distributed homogeneously in an environment or a species. Sampling at a point allows obtaining a representative result, for this reason, a thorough survey of markets in different regions was made in order to collect samples of all types of kohl available in Morocco. In total, 100 samples of kohl (67 powder samples), eye pencils (8 samples), kohl in paste (8 samples), and 17 samples of natural unprocessed form of galena were investigated in this study. They were purchased from local herbalist, ethnic shops in addition to local homemade tribal preparations; kohl in Morocco mainly originates from commercial manufactures from Saudi Arabia, India, and Pakistan. All samples were conserved in a polyethylene plastic bag at room temperature. A part of kohl sellers in Morocco claimed that they prepare kohl themselves, and most of kohl is sold without proper labels and packing. Different colors are used (black, silver, white, pink, brown) and different textures (powder, paste, eye pencil). It was also available in natural unprocessed form as pieces of galena or lead sulfide (PbS). There is no selection criterion as to the provenance or texture. Selection of sample is based on their availability in stores at the time of study, most of the tested products were either un-labeled or inadequately labeled.

### *2.2. Sample treatment*

Weighed kohl samples (0.1g) was placed into a Teflon vessel and reacted with 1ml of regia water (hydrochloric acid and nitric acid 1:3 (v/v)) and 6ml fluoridrique acid, left at room temperature for 1h then placed in the bath sand at 120°C for 3h or in a microwave). After digestion the sample was allowed to cool to room temperature, furthermore, after adding 7g of boric acid, the sample solution were placed at room temperature for another hour. The clear supernatant was transferred to polypropylene tubes and diluted to 50 ml with bidistilled water [32]. Lead content in the eye cosmetic products was reported as milligram per gram (mg/g) on the basis of wet weight. Statistical analysis (SPSS-19, one-way ANOVA) was conducted to determine the relation of lead concentration among the different textures of kohl. The p value of 0.05 was considered as significant.

### *2.3. Apparatus*

Lead analysis was performed using a flame Atomic Absorption Spectrometer type AA240-FS with a hollow cathode lamp, the optimized heating programs followed for the analysis of lead were that described by the instrument manufacturer. Calibration lead standards were prepared each day using a manual standard addition procedure where lipstick or eye shadow samples were divided into six equal portions. Known amounts of aqueous lead solutions were added to these to give final concentrations in the range of 0.25–4.0 ppm. There was a good linear relation between absorbance and standard concentration of lead. Linearity was evaluated by calculating the linear correlation coefficients (r) for 7 runs of kohl, which was  $0.999 \pm 0.0001$ .

Detection limits, calculated following the recommendation of International Atomic Energy Association were 1.0 µg/g for lead [32] and the recovery rate for all solutions was checked between 75 and 80 %.

### *2.4. Quality control*

The certified reference material IAEA- MESL-2013 -02 PT/TM sample from the IAEA for quality control and proficiency test was used for the purpose of method validation, the certified values are  $35.5 \pm 1$  mg/kg, and the result obtained using the analytical procedure was  $35.3 \pm 0.5$  mg/kg.

### 3. Results and discussion

#### 3.1 Results

The samples analyzed showed that lead was detected in all brands of kohl with varying concentration of lead (table1).

**Table 1:** Concentration of lead in different textures of kohl samples

Texture	Number	Min in mg/g	Max in mg/g	St.dev.in mg/g	Average in mg/g
Powder	67	0.31	750.00	177.39	165.26
Galena	17	50.33	973.80	286.10	422.68
Paste	8	0.01	134.58	47.44	17.16
Eye pencil	8	0.09	0.53	0.15	0.30
All	100	0.01	973.80	220.30	180.13

As seen in table 1, the concentration of lead was within the range of 0.01mg/g - 973.8 mg/g. Very high levels of lead were observed in galena samples and kohl in powder that is ordinary composed of galena (PBS) with an average of 440.42mg/g. The content range of lead in the galena samples was higher than that of the other textures. The statistical analysis shows that there was a significant difference between the average of lead content in different textures of kohl samples ( $p < 0.04$ ). The table 2 illustrates the lead content in different colors of studied kohl samples, it shows that kohl in powder with a black color had the highest lead level (750 mg/g), the lowest lead concentration is obtained in kohl in paste with black color (0.01mg/g). There was any significant difference between lead content in all colors ( $p = 0.06$ ).

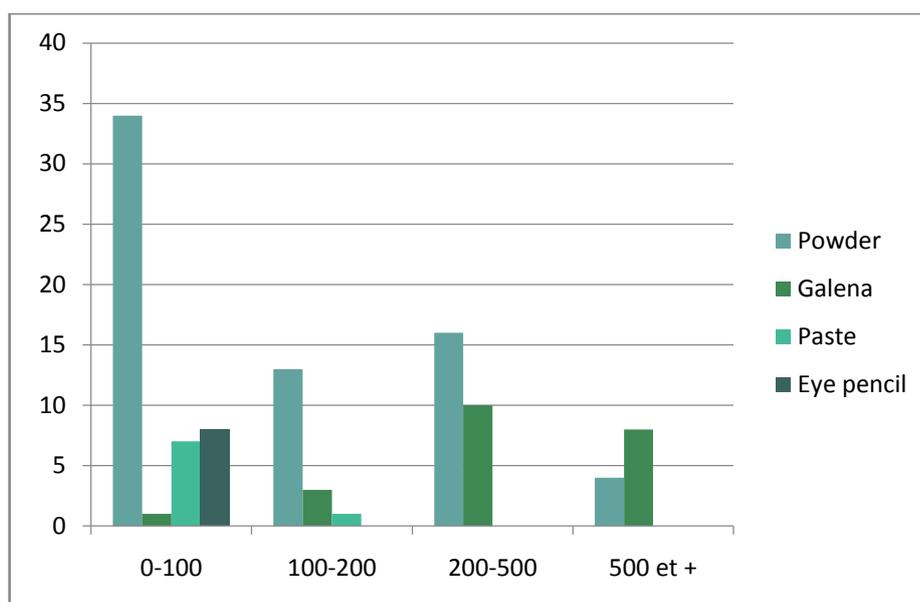
**Table 2:** Concentration of lead in different colors of Kohl products

Texture/color	Number	Min in mg/g	Max in mg/g	St.dev. in mg/g	Average in mg/g
Powder Black	38	0.69	750	194.61	178.20
Silver	29	0.31	568	150.00	146.77
Galena Silver	17	50.33	973.80	264.45	440.42
Paste Black	5	0.01	134.52	60.11	27.05
Others	3	0.62	0.74	0.66	0.68
Eye pencil Black	5	0.28	0.53	0.10	0.39
Others	3	0.09	0.24	0.08	0.14
All	100	0.01	973.80	220.30	180.13

The table 3 illustrates the concentration range of lead in different textures of kohl samples, it shows that all eye pencils had a concentration range within 0- 100 mg/g, which corresponds to 8% of the entire sample. In the 8 samples of paste, 7 samples of them had also the same concentration range (0-100 mg/g). We can conclude that these two textures had the lowest lead levels in comparison with the texture in powder and galena which contained a concentration range exceeding 500 mg/g. The figure 1 illustrates clearly the frequency of lead content according to each texture.

**Table 3:** the concentration range of lead in different textures of kohl samples

range (mg/g)	Frequency percent (%)				
	Powder	Galena	Paste	Eye pencil	total
0-100	34	1	7	8	50
100-200	13	3	1	0	17
200-500	16	10	0	0	26
500 et +	4	3	0	0	7
Total	67	17	8	8	100



**Figure 1:** Distribution frequency of lead concentration range according to texture

Table 4 summarizes the concentration range content in different studied brands of kohl samples, as shown 18% and 32% of the galena and powder, respectively, had a lead levels more than 100 mg/g. Branded kohl samples obtained from different regions of Morocco were classed in two different groups: those containing low lead level, and those with high level.

**Table4:** Concentration range of lead (mg/g) in the different brands

range in mg/g	Frequency percent (%)				
	Unbranded	Kohl Achifaayne	Hashmi kajal	Others	Total
0-100	27	5	4	14	50
100-200	9	2	1	5	17
200-500	23	0	0	2	25
500 et +	8	0	0	0	8
Total	67	7	5	21	100

Unbranded samples represents 67% of all samples, 40.3% of them had a lead content within the range 0-100mg/g, 13.4% contained lead between 100 and 200mg/g, 34.3% within 200 and 500mg/g and 12% had a lead levels more than 500mg/g. While 33% were branded, 23% of them have a lead concentration ranged with 0-

100mg/g, 8% have a lead content ranged with 100-200mg/g, and 2% have a lead concentration between 200-500mg/g (figure2). Based on these findings, it may be concluded the first group (unbranded) seemed to be prepared mainly from a lead ore. The minimum of lead concentrations were detected in samples of kohl in paste with a value of 0.01mg/g, the observed traces of lead could be due to the contamination or intentional mixing up of other materials. Generally, lead content in branded samples is lower than the unbranded samples ( $p < 0.02$ ). It seems that most of unbranded kohl that were over the counter (OTC) contains, in most cases, significant proportions of galena. Especially those prepared in home by women when no control is applied and the rules of cleanliness and packaging are poorly respected. Indeed, the majority of samples included in this study are improperly packaged or unpackaged.

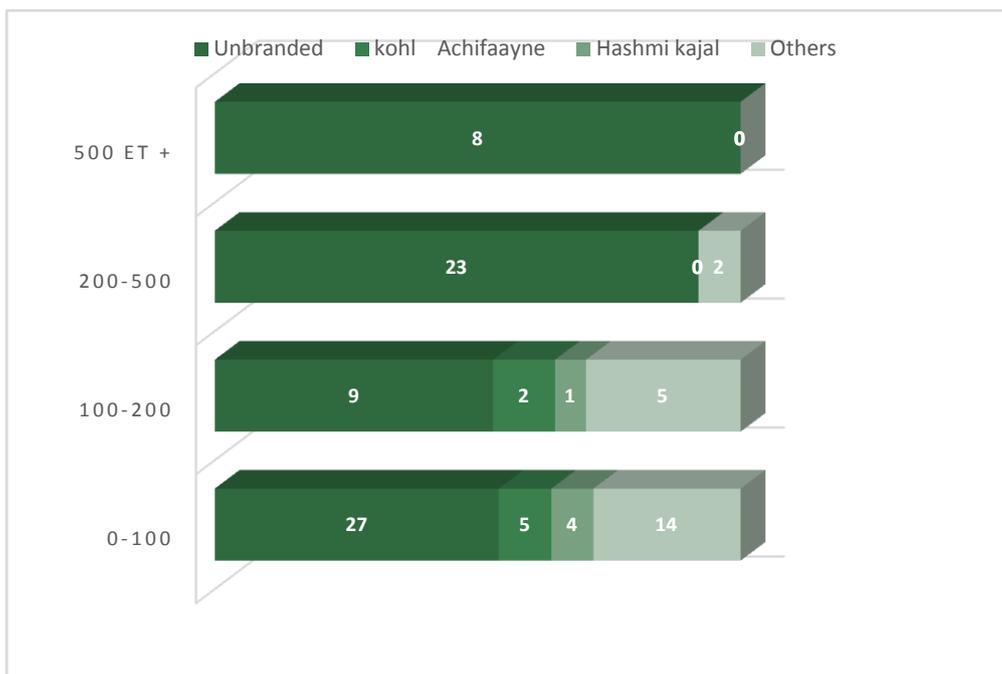


Figure2: Distribution frequency of lead concentration range according to brand

The results of the present study showed that lead was detected in all studied brands of kohl samples with a varying concentration. These results show that lead content is significantly higher than that reported in the study of Lekouch (up to 89%) [20] and Al Ashban that obtained 52.4% of lead [29]. However, our results are consistent with those of Catherine S (up to 95%) [30]. In Morocco, there is no regulation relating to the lead content in cosmetics, particularly in traditional kohl. Our data were compared with similar studies and international standards available in literature. The lead content was generally higher in the unbranded samples which are prepared mainly from a lead ore. Local kohl are particularly charged with the lead, especially the homemade form of kohl, whereas the concentration of lead was higher than the other form made by an artificial way or imported from other countries such as India and Pakistan. However, this type of kohl is the most available in Moroccan markets for their accessibility and low price, exposing the regular users of this product to chronic lead poisoning. It has been shown that the blood analysis of regular kohl users revealed a high lead concentration and relatively low hemoglobin levels [29].

In Europe, lead and its compounds are specifically listed as prohibited ingredients in cosmetics [34]. Whereas, all samples investigated in the current study contained lead in their composition even at trace concentration, indeed the analysed kohl samples are not in accordance with law, and they are clearly identified as a source of lead exposure. Our findings are supported by earlier studies where kohl was reported to contain high levels of lead, causing serious health hazard and death [35-36].

The Food and Drug Administration (FDA), has set a limit for the content of lead that is considered to be safe in colour additives which is typically between 10 and 20 ppm [37], and Canada uses a limit of 10 ppm that is considered to be safe for the consumers [38]. World Health Organization (WHO) in 2006, has estimated that the

level of tolerable weekly intake of lead from all sources is 25 $\mu$ g / kg, the acceptable daily intake (ADI) corresponding to a 70 kg adult is 250  $\mu$ g per day.

Applying on the eye kohl 10-50 mg (about 5.7 mg Pb / 100 mg kohl) per day, it would file thus about 2850 $\mu$ g of lead, which exceeds the ADI[39]. A reconsideration of health risk is necessary to this cosmetic. Therefore all the samples investigated in this study are not consistent with international standards, so they are a source of lead exposure especially for pregnant women and young children because lead can easily cross the placenta and produce congenital lead poisoning [40]. The way in which Moroccan women use the kohl by coating the **miroued** that is a traditional instrument for kohl application 3 at 4 times a week may also increase the risk for lead toxicity [20]. The continuous use of this cosmetic could have adverse effects on the ocular system [41], finally the health authorities must informed the population from the potential dangers of prolonged use of this traditional eye cosmetic and promote the use of lead-free kohl in order to ensure less lead exposure in the general population.

## Conclusion

In this study, 100 samples of traditional kohl are investigated, the results shows that lead was detected in all samples with a lead content varied within

0.01 mg/g and 973.8 mg/g, the concentration of lead content in all samples under study was higher than that of the international standards. Traditional Kohl subject of this research appears to be a substance that may be a source of lead exposure in Morocco, and no way of protecting the consumer from those that are hazardous. The continuous use of this leaded eye cosmetic may affect the health of the regular consumer especially pregnant women and younger children.

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