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Research Article

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The Hematological Changes in Dental Staff: Their Relation to Mercury Vapor

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ABSTRACT

Background: Dental staff are exposed to mercury during their work. Aim: To determine the effect of mercury on their hematological parameters. Subjects & Methods: 83 dental staff, 43 use amalgam and 40 not, and 56 healthy persons, 19 have amalgam filling and 37 without it, were enrolled. All were subjected to measurement of mercury in hair, nails and complete blood count. Results: A significant increase of mercury hair and nail was found in the exposed dental staff when compared to both control groups. A significant increase of mercury hair in a non-exposed dental staff was detected when compared to the control group without amalgam. A Significant decrease of hemoglobin and absolute monocytes count was found in both groups of dental staff when compared to the control group without amalgam. 33.3% and 20% of the exposed and non- exposed staff had anemia. Conclusion: Exposed and nonexposed dental staff are liable to complications induced by mercury.

Keywords: *Mercury vapor, Anemia, Dental staff*

INTRODUCTION

Mercury is considered a major environmental toxicant throughout the world. It is harmless in an insoluble form, but in vapor or soluble forms, it can be extremely toxic to humans. Mercury exists in several forms: elemental, organic and inorganic. The elemental mercury, used in dentistry, has no electrical charge (Hg0), inorganic mercury has a positive charge of +1 or +2 (Hg 1+, Hg 2+) and organic mercury is a complex form of mercury with carbon-containing compounds. Elemental mercury is a large component (approximately 50%) of dental amalgam. The manipulation of in situ amalgam results in a short-term mercury vapor exposure to dentists and other dental workers and the inhaling of mercury vapor occurs [1]. Elemental mercury vapor can be inhaled into the lungs, where it moves into the bloodstream and is transported through the body. In plasma, mercury remains in its elemental form (Hg0) and can cross into the brain and into the fetus of a pregnant woman. In red blood cells, elemental mercury is readily metabolized to inorganic mercury (Hg2+), which tends to accumulate in the kidneys and damage sensitive tissues in that organ [2,3]. It was reported that mercury, competing for ligands in the biological systems, can largely change the metabolism and function of the essential trace elements such as copper, zinc, iron, manganese, selenium, and calcium [4]. In addition, the hematopoietic and immune systems are vulnerable to mercury, which could lead to anemia and suppression of humoral and cell-mediated immune responses in male rats [5]. Many studies have demonstrated that the dental workers have higher levels of mercury in their tissues and organs than the members of the control groups [6-8]. Few studies have been done to deal with the effect of mercury on the hematological parameters in dental staff. The aim of this study was to determine the effect of mercury vapor on the hematological parameters in dental staff.

SUBJECTS AND METHODS

Subjects

The analytical methods of this study were carried out in the Medical Biochemistry Department- Faculty of Medicine, UQU, Makkah, Saudi Arabia in accordance with the approved guidelines. The Umm AlQura University ethics committee approved the protocol of this study. All the participants gave informed consent according to the Declaration of Helsinki. This study was carried out from March 2015 to June 2016; it included 83 dental staff working at different polyclinics and private clinics in the Makkah region and 56 healthy persons volunteered as a control group. The dental staff were sub-classified into the exposed dental staff and non-exposed dental staff with regards to mercury use. The exposed dental staff dealt with the amalgam either by removing or replacing it.

Sample Collection

5 ml of blood sample were collected from each participant under complete aseptic conditions. 2 ml was dispensed into a tube containing EDTA as anticoagulant substances for performing complete blood count, while the remaining 3 ml collected in plain tube and the serum was separated for analysis of reduced glutathione. 139 hair and nail samples were collected; hair samples were cut near the scalp area with thin-blade stainless steel scissors. Then, it was accurately weighed and placed inside polyethylene bags and stored at controlled temperature (25°C) and humidity (65% RH). All the participants had been asked in advance not to trim their nails for a couple of weeks or longer, their nails were collected by clipping with stainless steel clipper from the two great toes (or thumbs) and small toes (or another finger), placed in a labelled envelope and it was stored at a room temperature in the driest condition possible.

Inclusion Criteria:

All the dental staff that are more than 18 years, exposed or not exposed to mercury vapor during work were included.

Exclusion Criteria:

We excluded those with chronic or inflammatory disease and those less than 18 years from this study.

METHODS

All participants were subjected to the followings:

1. A full clinical history.
2. Complete blood count on Sysmex XT-2000i, Siemens diagnostic- Germany, which included the following parameters; red blood cells count, hemoglobin concentration, packed cell volume, mean cell volume (MCV), mean corpuscular hemoglobin (MCH), red cell distribution width (RDW), platelets count , total white blood cell count (WBC) and differential counts.
3. Analysis of hair and nail mercury: Hair samples were first washed with acetone – water-water- water – acetone as recommended by the International Atomic energy agency [9]. The washed samples were placed in glass beakers individually and allowed to dry at 50C overnight in a drying oven. For nails, any visible dirt on the surface of nails has been removed. Nails were thoroughly washed using an ultrasonic bath with distilled water, followed by MilliQ water, then acetone. Approximately, 0.1 -0.5 g of dry sample was weighed into dry, clean Teflon digestion vessel. 3 ml of concentrated nitric acid and 1 ml of hydrogen peroxide were added and kept overnight. The vessel was placed in a microwave digestion (Henan Brand, model no. APEX-LJ91). An Efficiency of 600 W was applied in the process for 30 minutes. Then cooling for 30 minutes was applied. Each digested solution was quantitatively transferred to a 10ml volumetric flask, and 100 µl of an internal standard solution was added. Samples were analyzed three times using a Perkin Elmer (ICP-MS 7300). The detection limits of Hg were higher than 95% with detection value $\leq 3 ng/m^3$. The maximal value of the relative standard deviation for the three [3] replicates' analyses of every individual sample was less than 4%.
4. Reduced glutathione was determined by coloremitry utilizing of QuantiChrom Glutathione Assay Kit [9].

Statistical Analysis

The statistical analysis of this study was done using the SPSS program version 20. Quantitative data were described in the form of mean ± SD for the normally distributed data. The median and range were used for the data that were not normally distributed. The comparison between the groups was performed by using Student t-test and ANOVA test. The Mann–Whitney U test and the Kruskal–Wallis test was used for the data that were not normally distributed or if the number was small. The chi-square test or Fisher exact test was used for comparison between qualitative data.

RESULTS

The results of this study are summarized in tables from 1 to 5 and figure 1

This study included 83 dental staff and 56 healthy subjects as a control group. The dental staff were divided into 2 groups, exposed and non exposed according to the use of mercury in their work. The exposed dental staff were 43 in number; 13 were males and 30 females and their mean age was 34.65±6.9, and the duration of exposure was 10.01±6.63. The non-exposed dental staff were 40 in number; 16 males and 24 females and their mean age was 33.85±6.64. The duration of occupation was 8.9±5.34. The control group was categorized into 2 groups according to the availability of amalgam filling; those that have amalgam filling were 19; they were 7 males and 12 females, their mean age was 39.21± 4.1 and the control without amalgam filling were 37; they were 21 males and 16 females, and their mean age was 33.97±11.01. The number of amalgam was varying from 1 to 3.

The comparison between different groups with regards to demographic, clinical mercury levels in hair and nails and anti- oxidants are shown in Table 1.

A Significant increase of both mercury hair and nail in exposed dental staff, when compared to the control with and without amalgam was found. A Significant increase of mercury hair in the non-exposed dental staff was also found when compared to the control group without amalgam. No significant difference was found between the non-exposed dental staff with regards to mercury nail and both control groups. No significant difference was found between exposed and non-exposed dental staff with regards to mercury hair and nail.

Table 1. Comparison between different groups with regards to demographic, clinical and biochemical data

Variables	Dental staff		Control		P value
	Exposed 43	non-exposed 40	Amalgam filling+ve 19	amalgam filling¯ve 37	
Age	34.65±6.9	33.85±6.64	39.21±4.1	33.97±11.01	0.08
Gender Male Female	 13 (30.2%) 30 (69.8%)	 16 (40%) 24 (60%)	 7 (36.8%) 12 (63.2%)	 21 (56.8%) 16(43.2%)	0.11
Smoking	7 (16.3%)	6 (15%)	2 (10.5%)	8 (21.6%)	0.74
Position Doctor Dental staff	 21 (48.8%) 22 (51.2%)	 16 (40%) 24 (60%)	 -	 -	0.42
SBP	117.3±11.43	118.6±10.73	118.42±4.73	119.59±5.94	0.75
DBP	79.05±7.53	76.6±8.28	80.53±3.29	79.05±4.22	0.14
HTN	2 (4.7%)	2 (5%)	0%	0%	
Hair mercury Median Min-max	3.62±2.02 2.82 1.01-9.28	3.31±2.15 2.81 0.77-9.4	2.04±1.61 1.46 0.82-8.15	1.95±1.43 1.35 0.76-6.52	0.0001*•
Nail mercury Median Min-max	2.91±1.97 2.1 0.78-8.13	2.67±2.18 1.8 0.71-7.93	1.52±1.32 0.96 0.54-4.91	1.61±1.52 1.03 0.65-6.36	0.003*

*Post hoc test : the significance is between the exposed dental staff and both control subgroups.
•Posthoc test: the significance is between the non-exposed dental staff and control do not have amalgam filling.
SBP= systolic blood pressure, DBP= diastolic blood pressure, HTN= hypertension

The comparison between different groups with regards to hematological parameters Table2.

A Significant decrease of the hemoglobin, red blood cell counts and the absolute monocytes count was found in the dental staff exposed to amalgam when compared to the control group without amalgam $p<0.000$, $p<0.009$ and $p<0.003$ respectively. In addition, when comparing the exposed dental staff to the control with amalgam, it also showed a significant decrease in hemoglobin and red blood cells count $p<0.019$ and $p<0.02$ respectively. A Significant decrease of hemoglobin was found in the exposed dental staff when compared to the non-exposed staff $p=0.05$. No other significant differences were found with regards to the white blood cell count, platelets, absolute neutrophils count , and absolute lymphocytes count. The non-exposed dental staff showed a significant decrease of hemoglobin, and absolute monocytes count when compared to the control group without amalgam $p<0.012$, and $p<0.001$ respectively. Moreover, no significant differences were found when the non-exposed dental staff were compared to the control group with amalgam with regards to all hematological parameters studied.

Table 2. Comparison between different groups with regards to hematological parameters

Parameters studied	Control with amalgam (N=19)	Control without amalgam (N= 37)	Exposed Dental staff (N=43)	Not exposed Dental staff (N=40)	Significance
Hemoglobin g/dl Mean ± SD	14.0± 1.6	14.7±1.5	12.57 ±2.09	13.44±1.92	. 000 HS*#
Median	13.7	14.6	12.5	13.55	
Min-Max	11.7- 16.8	11.7- 17.6	8.3-16.6	7.9-17.1	
RBC $X10^{12}$ Mean ± SD	5.1± 0.6	5.2± 0.8	4.73±0.49	4.91±0.67	. 034 S*
Median	5.3	5.0	4.7	4.95	
Min-Max	4.1- 6.1	4.1-6.6	3.7-5.6	3.65-6.57	
Platelets Mean ± SD	252.4± 47.0	260.1± 63.7	288.9± 95.5	279.88±79.11	. 376 NS
Median	251	259.5	260	266	
Min-Max	162-331	165.0-411.0	160-651	64-429	
WBC Mean ± SD	7.7± 1.2	8.0 ± 1.6	7.4± 2.4	7.39±2.05	.351 NS
Median	7.5	8.0	6.9	7.17	
Min-Max	5.2-9.4	4.5-10.7	3.7-15.02	4.76-13.97	
Polymorph Mean ± SD	3.9± 0.73	3.9± 1.2	3.9± 1.85	4.02±1.85	.941NS
Median	4.0	4.0	3.7	3.71	
Min-Max	2.9-4.9	1.9-6.2	1.04-8.7	1.69-10.68	
Lymphocytes Mean ± SD	2.8± 0.8	2.8±0.8	2.9± 0.99	2.6±0.75	.474 NS
Median	2.9	2.9	2.61	2.58	
Min-Max	1.4-4.3	1.2-4.3	1.3-5.4	1.39-4.78	
Monocytes Mean ± SD	0.6± 0.2	0.64±.16	0.53±0.16	0.53±0.18	.005 S†
Median	0.5	0.65	0.54	0.53	
Min-Max	0.4-1.08	0.2-0.94	0.16-0.9	0.16-1.08	

* Post Hoc test: The significance is between control with and without amalgam versus exposed dental staff. #Post Hoc test: The significance is between control without amalgam versus non- exposed dental staff. †Post Hoc test: The significance is between exposed and non- exposed dental staff versus control without amalgam RBC= red blood cells, WBC= white blood cells

The Comparison between control with amalgam and without amalgam with regards to hematological parameter table 3.

A significant decrease of hemoglobin and the absolute monocyte count was found in the control group with amalgam when compared to the control without amalgam p=0.016 and 0.039 respectively. No other significant differences were found in red blood cell count, platelets count, white blood cells count, absolute neutrophil count, absolute lymphocyte count and absolute eosinophil count.

Table 3. Comparison between control with amalgam and without amalgam with regards to hematological parameters

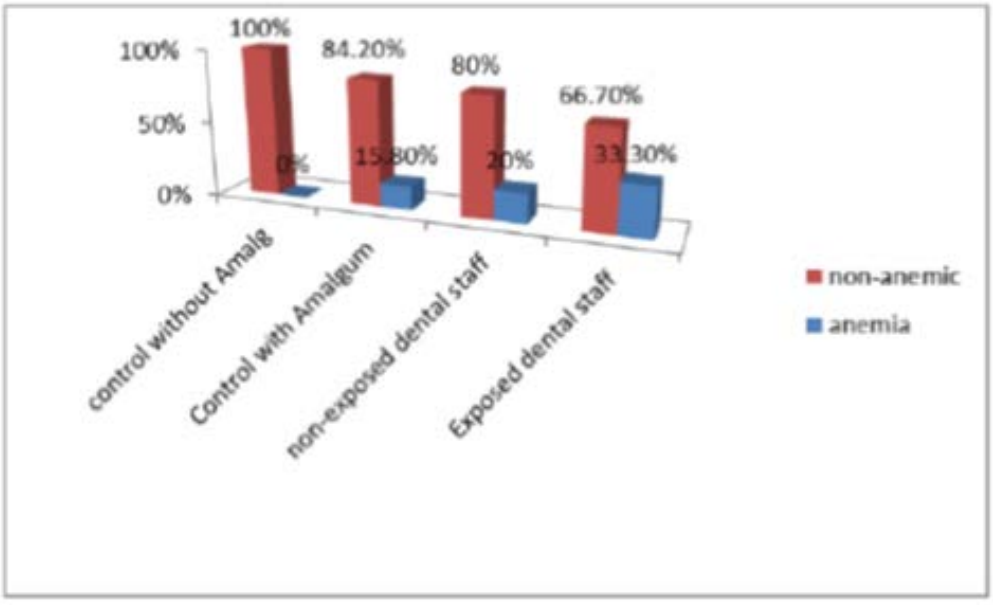
Parameters studied	Control with amalgam (N=19)	Control without amalgam (N= 37)	Significance
Hemoglobin g/dl Mean ± SD	14.0± 1.6	14.7±1.5	.016 S
Median	13.7	14.9	
Min-Max	11.7- 16.8	12.5- 17.6	
RBC $X10^{12}$ Mean ± SD	5.1± 0.6	5.2± 0.8	.859
Median	5.3	5.0	
Min-Max	4.1- 6.1	4.1-6.6	
Platelets Mean ± SD	252.4± 47.0	260.1± 63.7	.518
Median	251	259.5	

Min-Max	162-331	165.0-411.0	
WBC Mean ± SD	7.7± 1.2	8.0 ± 1.6	.723
Median	7.5	8.0	
Min-Max	5.2-9.4	4.5-10.7	
Polymorph Mean ± SD	3.9± 0.73	3.9± 1.2	.626
Median	4.0	4.0	
Min-Max	2.9-4.9	1.9-6.2	
Lymphocytes Mean ± SD	2.8± 0.8	2.8±0.8	.501
Median	2.9	2.9	
Min-Max	1.4-4.3	1.2-4.3	
Monocytes Mean ± SD	0.6± 0.2	0.64±0.16	.039
Median	0.5	.65	
Min-Max	0.4-1.08	0.2-0.94	

RBC= Red Blood Cells, WBC= White Blood Cells

The comparison of the frequency of anemia in the different group studied in Figure 1.

33.3 % (14/42) of the dental staff who were exposed to amalgam had anemia and 66.7% (28/42) had no anemia. The MCV and MCH of the anemic group decreased significantly when compared to the non-anemic group and the type of anemia was microcytic hypochromic anemia. The MCV was 77.06 ± 9.3 versus 83.8±7.05 and the MCH was 23.8± 3.8 versus 27.8±2.2, the p was 0.012 and 0.002 respectively. In the non-exposed staff, 20 % (8/40) had anemia and 80% (32/40) had no anemia. The MCV and MCH of the anemic group were not significantly different from the non-anemic group. The MCV was 78.8 ± 6.95 versus 83.5±7.05 and the MCH was 26.1±2.6 versus 27.9±2.3, the p was 0.097 and 0.11 respectively. In the control group with amalgam, 15.8 % (3/19) had anemia and 84.2% (16/19) had no anemia. The MCV of the anemic group was not significantly different from the non-anemic group. The MCV was 81.3± 1.0 versus 83.2±4.5 p=0.49. The MCH was significantly decreased in the anemic group 25.3± 0.8 versus 27.7± 1.6, the p was 0.02. In the control without amalgam, non had anemia. The comparison between the four-group studied with regards to the presence of anemia showed a significant difference p=0.005 (Figure 1).



Frequency of anemia in different groups studied

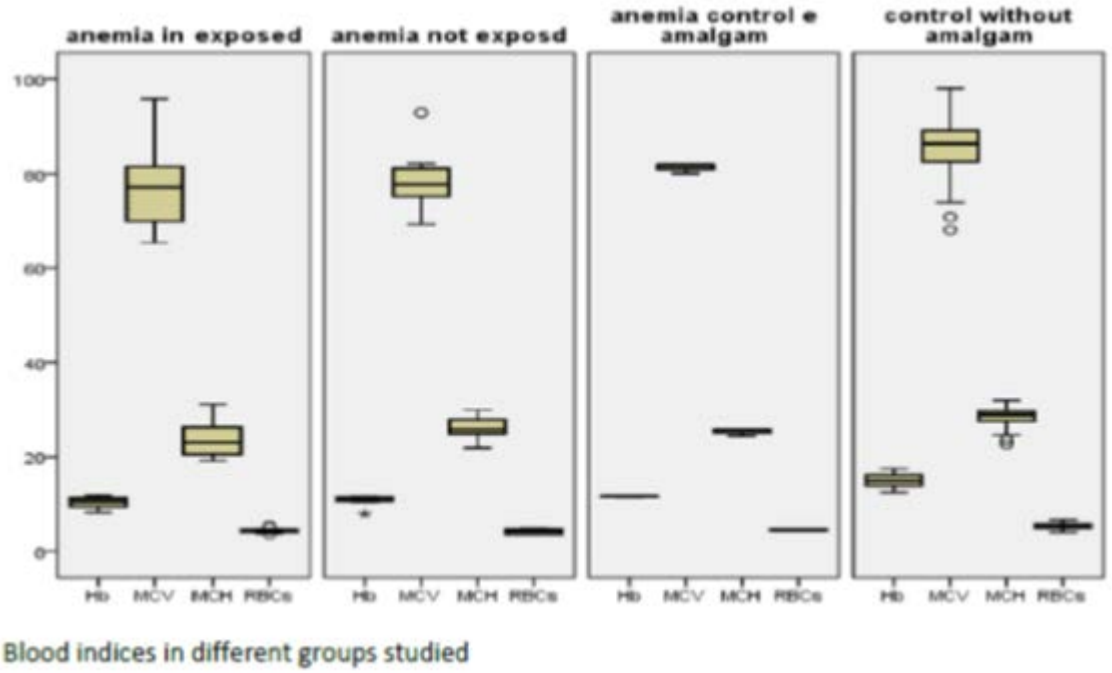


Figure 1. Comparison between different groups studied with regards to the frequency of anemia and red blood cells indices

The comparison between anemic and non-anemic with regards to mercury hair and nails in the dental staff are shown in Table4.

No significant differences were found between anemic and non-anemic in the exposed and non-exposed dental staff with regards to mercury hair and nail. As shown in Table 4, the median of mercury hair and nail in anemic exposed dental staff was higher than the non-anemic in the same group but without significant difference. In the exposed dental staff, a significant increase of both mercury hair and nail in the anemic and non-anemic group was found when compared to the control without amalgam. *In the non-exposed anemic staff*, there was a significant increase of mercury nail when compared to the control without amalgam. Whereas, in the non-exposed non-anemic dental staff, there was a significant increase of both mercury hair and nail when compared to the control without amalgam (Table 4).

Table 4. Comparison between anemic and non-anemic with regards to mercury hair and nails in the dental staff

Parameter studied	Exposed		Non-exposed		Control without amalgam
	Anemic	Non-anemic	Anemic	Non-anemic	
Mercury hair					
Mean ± SD	4.3±2.9	3.2±1.4	2.6±1.9	3.4±2.2	
Median	3.2	2.8	2.1	3.2	1.45
Min-Max	1-9.28	1-6.89	1.2-6.8	0.77-9.4	0.76-6.52
P1		0.002		0.246	
P2		0.000		0.002	
P3		0.468		0.321	
Mercury nail					
Mean ± SD	3.5±2.4	2.6±1.7	2.3±2.0	2.8±2.2	
Median	3	2	1.8	1.8	1.07
Min-Max	0.98-8.13	0.78-7.68	0.78-7.13	0.71-7.93	0.65-7.01
P1		0.000		0.037	
P2		0.000		0.000	
P3		0.263		0.607	

P1= anemic versus control**, P2=** non-anemic versus control**, P3=** anemic versus non-anemic

The comparison between different groups studied (anemic and non-anemic) with regards to reduced glutathione, white blood cell differential count, and the occupation duration.

In anemic patients, with regards to reduced glutathione, no significant differences were found between both the exposed dental staff and the control with amalgam versus the control group without amalgam p>0.05. A Significant decrease of reduced glutathione was detected in the non-exposed dental staff when compared to the control without amalgam table 5. In non-anemic patients, there were no significant differences between different groups studied and the control without amalgam (the data are not shown).

Table 5. Comparison of the concentration of reduced glutathione and monocytes count in anemic patients in different group studied versus control without amalgam

Parameters studied	Exposed dental staff	Non-exposed dental staff	Control with amalgam	Control without amalgam
*glutathione (anemic)				
Mean ± SD	419.8±170.6	303.3±159.8	387.0±35	526.2±213.7
median	388.5	309.9	406	514.6
min-max	190.7- 673.0	120.5-599.6	347.1-404.9	35.5-927.4
p	0.086	0.008	0.137	
Monocytes $X10^9$/l				
Mean ± SD (anemic)	0.5 ± 0.15	0.62± 0.2	0.45±0.08	0.62±0.2
P	0.009	0.565	0.034	
Mean ± SD (non-anemic)	0.53± 0.17	0.51±0.18	0.599±0.2	
p	0.008	0.001	0.124	

* The data of the non-anemic is not shown (no significant difference was found between the different groups studied.)

With regards to the absolute monocytes count in anemic patients, a significant decrease of monocytes count was found in exposed dental staff and the control with amalgam when compared to the control without amalgam. Whereas, in the non-anemic patients a significant decrease of the absolute monocytes count was found in both the exposed and non-exposed when compared to the control without amalgam. In addition, a significant decrease of absolute lymphocyte count was observed in the non-anemic non- exposed dental staff when compared to the control without amalgam, 2.5 ± 0.8 and 2.9 ± 0.8 respectively, P=0.030.

With regards to the duration of occupation 10.1±6.63 vs. 8.9±5.334 and its effect on the occurrence of anemia, no significant differences were found between anemic and non-anemic in the different group studied p>0.05. In anemic exposed dental staff, the median of the duration of occupation was 9 years with a range from 1 year to 17 years. In the non-anemic exposed dental staff, the median was 9 years with a range from 1 year to 30 years. In anemic non- exposed dental staff, the median was 10 years with a range from 2 years to 16 years. In the non-anemic exposed dental staff, the median was 7 years with a range from 3 years to 30 years.

The correlation between reduced glutathione, mercury and hematological parameters in different studied groups

In exposed dental staff: in anemic, a significant positive correlation between the mercury hair and nail was found. r= 0.603, p=0.022. In non-anemic, a significant positive correlation of reduced glutathione with each of hemoglobin, MCV, and MCH. r= 0.400, 0.499,0.504, p=0.035,0.007, 0.006 respectively. In the non-exposed dental staff: in anemic a significant negative correlation of mercury hair with monocytes, r=-0.778 p= 0.023. In non-anemic, a significant negative correlation of mercury nail with hemoglobin r= - 0.461, p=0.008.

DISCUSSION

Nowadays, 24% of diseases and 23% of the deaths of human beings can be attributed to environmental factors, based on the world health organization (WHO) report [10] in which environmental pollutants emerge as the greatest danger to public health. The Dental staff are chronically exposed to mercury during their work. The aim of this study was to determine the effect of the mercury vapor on the hematological parameters in dental staff both exposed and not exposed to it.

In this study, the exposed dental staff (all, anemic and non -anemic) had significantly higher values of mercury hair and nails when compared to both groups of the control. As mentioned previously, mercury can be absorbed into the body through numerous routes, including inhalation, ingestion, absorption through the skin, and injection-both subcutaneous and intravenous [8-11]. In our exposed dental staff, the inhalation of mercury vapor during their work is the cause of its elevation. In addition, all the non-exposed dental staff had significantly higher values of mercury hair when compared to control without amalgam. Moreover, the anemic non- exposed had a significant increase of mercury nail. Also, the non -exposed non -anemic, had significantly higher values of the mercury hair and nail. The significant increase of both mercury hair and nail or one of them in the non-exposed dental staff may indicate that

mercury vapors reach the non-exposed workers through the air. It was previously reported that high mercury levels were found in the breathing zone, which includes the amalgam preparation area, autoclave, and amalgam storage. In contrast, areas of greater mercury contamination at floor level (outside the breathing zone) appeared to have a little biological impact [12-13]. This may explain the high mercury levels present in the non-exposed dental staff as they are present in an environment rich in mercury vapor although they do not work with it. The absence of the significance between exposed and non-exposed dental staff with regards to mercury hair and nails indicates that they inhale the mercury vapor directly during their work and indirectly from the air in the clinic.

In this study, the significant decrease of hemoglobin and red blood cells count in exposed dental staff versus control with and without amalgam was observed. Also, the significant decrease of hemoglobin in non -exposed dental staff when compared to control without amalgam confirms the previous findings about the effect of mercury on the hematopoietic system [5]. The significant decrease of hemoglobin in exposed dental staff when compared to non-exposed dental staff indicates that there is more affection on the exposed dental staff than the non- exposed persons. The higher percentage of anemia present in the exposed staff 33.3% versus non exposed 20% is confirmed in our work.

In this study, 33.3 %, 20% and 15.8 % of the exposed, non-exposed dental staff and control with amalgam were anemic respectively. The highest frequency of anemia in the exposed dental staff may be explained by the higher dose of mercury present in those staff. The significant increase of mercury in their hair and nails is confirmed in our work. In addition, the significant positive correlation between mercury hair and nails in the anemic exposed dental staff was observed. Also, the significant negative correlation of mercury with hemoglobin was resulted. The higher the mercury, the lower the hemoglobin. The type of anemia in the exposed dental staff was microcytic hypochromic anemia. Whereas, it was normocytic normochromic in non-exposed dental staff and in the control with amalgam. Heavy metals (e.g. Cd and Hg) could affect iron homeostasis through two major pathways, namely directly competing with iron ions in biomolecules and indirectly generating oxidative stress [14].

It was reported that mercury alters the metabolism and function of iron by competing for ligands in biological systems [4]. Iron is essential for the formation of hemoglobin and its deficiency leads to iron deficiency anemia which is microcytic hypochromic anemia.

The mechanism of development of iron deficiency anemia in exposed dental staff was not completely understood. It was demonstrated that, following exposure to environmental pollutants which includes mercury, the hepcidin-FPN axis is misregulated in administrated animals along with disordered systemic iron homeostasis. i.e. decrease iron absorption with subsequent iron deficiency anemia. Hepcidin-FPN signaling is one of the key mechanisms responsible for iron supply, utilization, recycling, and storage [14]. In addition, in another work, the authors gave the rat a low and high dose of drinking mercury and they recorded a decrease in the absorption of iron and it was accompanied by a decrease in serum iron and a development of iron deficiency anemia [15]. As previously recognized, iron is mostly bonded in the center of a large heterocyclic organic ring called porphyrin. However, according to the Irving–Williams series, Fe^{2+} in the heme group could be replaced by other metal ions with a higher affinity. In this way, the heavy metals can disturb the balance of the intracellular iron pool by competing with iron transporters or iron-regulated enzymes, such as transferrin, divalent metal transporter 1 and iron-regulatory proteins [16]. All these lead to a deficiency of serum iron and the development of iron deficiency anemia, which is characterized by decreased hemoglobin, decreased red blood cells count (RBC), decreased mean cell volume and decreased mean corpuscular hemoglobin. All are present in our anemic exposed dental staff cases. In contrast, to our results, previous authors reported an increase in RBC count [17].

In this study, the normocytic normochromic anemia that was present in non-exposed dental staff, may be explained by the reduction in the reduced glutathione that was found in this group. The extracellular glutathione is a reflect of intracellular glutathione and it is released into plasma as exportation. Glutathione is an antioxidant and can prevent damage to the important cellular components caused by reactive oxygen species such as free radicals, peroxides, lipid peroxides and heavy metals such as mercury [18]. As reported earlier, mercury vapor oxidized to inorganic mercury by catalase and can attach to the thiol groups in most proteins – enzymes, glutathione, or almost any structural protein [18]. Because of the binding of mercury to glutathione and the subsequent elimination of intracellular glutathione, levels of reduced glutathione are lowered in several specific types of cells on exposure to all forms of mercury [19, 20]. In our work, the significant positive correlation of glutathione with hemoglobin, MCV and MCH and the significant negative correlation of mercury nail with hemoglobin confirm our explanation.

Moreover, it was reported that glucose 6 phosphate dehydrogenase (G6PD) enzyme was reduced in chronic mercury exposure [17, 21]. The G6PD enzyme is responsible for donating H molecule via NADPH to the oxidized form of glutathione to be returned to the reduced form. In its deficiency, the process stops with consequent hemolysis of the red blood cell on exposure to an oxidizing agent. So, we suggested that the normocytic normochromic anemia present in the anemic non- exposed dental staff is due to hemolysis of red blood cells. Also, it was reported that, chronic mercury exposure leads to a reduction in the erythropoietin release from the kidney. The reduction in erythropoietin leads to normocytic normochromic anemia. Unfortunately, we did not measure reticulocytes in our cases which is an indicator of hemolysis and can differentiate the cause of anemia either due to hemolysis or a reduction in the erythropoietin. The normocytic normochromic anemia present in the control with amalgam filling may explained by

a reduction in the erythropoietin release from the kidney, as the glutathione in this group was normal. We suggest that the difference in the type of anemia between exposed, non-exposed workers and control with amalgam was related to the dose or to the amount of mercury exposure. High dose of mercury leads to affection of iron and iron deficiency anemia, whereas low doses lead to mild hemolysis of red blood cells or decrease in erythropoietin secretion with subsequent normocytic normochromic anemia. In other words, high dose affects iron metabolism, whereas, the low dose affects glutathione, glucose 6 phosphate dehydrogenase enzyme and the release of erythropoietin from the kidney, separated or in combination. These findings need more studies to prove our suggestion.

The absence of significant differences between anemic and the non- anemic dental staff with regards to the duration of occupation makes us suggest that the appearance of anemia depends on the amount of mercury they exposed to and not to, in the prolonged duration.

In this work, a significant decrease of the absolute monocytes count was found in the exposed dental staff (all, anemic and non-anemic), in the non- exposed dental staff (all and non-anemic) and in the control with amalgam (anemic) when compared to the control without amalgam. Mercuric compound was found to inhibit monocytes function by inducing apoptosis via a reduction in reduced glutathione [19-23]. This reduction predisposes cells to reactive oxygen species damage and at the same time activates death-signaling pathways. This could explain the decrease in the absolute count of monocytes in our study [23]. Moreover, the significant negative correlation that was found between mercury hair and monocytes in anemic non-exposed dental staff could confirm our suggestion. In addition, to the significant decrease in monocytes count, lymphocyte count was also decreased in the non-exposed non-anemic. The explanation could be the same. The decrease in the absolute monocytes counts in the different studied groups (anemic and non- anemic) and the decrease in the lymphocyte count in the non- anemic non- exposed dental staff indicate no effect of anemia on monocytes and lymphocyte count. The reduction in the count is due to the apoptotic effect of mercury on the cells.

The absence of the significant difference between both groups of dental staff and the control with amalgam with regards to the absolute monocytes count indicate that the mercury present in the control with amalgam influences the absolute monocytes count causing a decrease in the count.

LIMITATION OF THE STUDY

No measurement of the iron profile which includes serum iron, total iron binding capacity, and serum ferritin was done. In addition, there is no measurement of reticulocytes and erythropoietin in anemic cases.

CONCLUSION

Exposed and nonexposed dental staff were liable to anemia and there was a decrease in mononuclear cell count because of inhalation of mercury vapor during their work. These changes predispose them to different complications. More studies are needed to confirm our results.

Geolocation Information

This study was carried on Makkah, Saudi Arabia

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Authors' Contribution

Zaghloul A, Al-Amodi H and Alrefai A share in the design of the study, researched literature and conceived the study. AL-Amodi H and Alrefai A and Adly H gained ethical approval, and contributed to data collection and lab analysis. Zaghloul A analysed the data, preparing tables and figures, wrote the first draft of the manuscript. Zaghloul A and Alrefai A reviewed and edited the manuscript and all authors approved the final version of this manuscript

Disclosure of Interest :

The authors report no conflict of interest

Ethics Approval

The Umm AlQura University ethics committee approved the protocol of this study.

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