

Assessment of the oral findings, salivary oxidative status and IgA level among group of workers exposed to petroleum pollutants in Al-Daura oil refinery

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ABSTRACT

Background: Oil refinery workers are continuously exposed to numerous hazardous materials. Petroleum contains the heavy metals as a natural constituent or as additives. These metals induce the production of ROS which associated with an oxidative damage to DNA, proteins, and lipids. This study was conducted to assess the salivary levels of heavy metals, salivary oxidative status, oral immunological activity (salivary sIgA) and assessment of the oral findings among the workers of Al-Daura oil refinery in Baghdad city.

Subjects, Materials and Methods: This study was done in Al-Daura oil refinery; samples consist of 60 workers involved in refinery processes (study group) and 20 non-workers (control group). Oral examination and saliva collection was done to assess the oral findings and measurement the level of heavy metals (lead and cadmium), oxidative status (MDA and SOD) and secretary IgA.

Results: salivary lead and cadmium was higher in study group (6.34 µg/dl and 0.56 µg/l) than that of control group (3.3 µg/dl and 0.34 µg/l) with highly significant difference ($p < 0.001$). A significant increase ($p < 0.05$) was found in salivary MDA (15.3 ng/ml) and salivary sIgA (464.36 µg/ml) and significant decrease in salivary SOD (1895.1 pg/ml) among the study group. Lead has shown significant linear correlation with MDA and IgA. A significant reverse correlation was found between heavy metals (lead and cadmium) and SOD. The oral examination revealed no oral lesions of interest.

Conclusion: Workers in Al- Daura oil refinery exposed to pollution with heavy metals (Pb and Cd) which was associated with changes in the biochemical and immunological findings among the oral cavity.

Key words: Petroleum, heavy metals, salivary oxidative stress, sIgA, oral findings. (J Bagh Coll Dentistry 2015; 27(2):48-53).

INTRODUCTION

Iraq is one of the significant countries in oil reservoir, production and exportation. Crude oil fields and oil refineries are present on wide locations of the Iraqi lands. These oil locations distributed from the north to the far south of the country. Some of the fields are present close to or in the nearby cities, therefore its presence has an impact on the environment and health of the residents and workers inside the oil refineries.

Petroleum is perhaps the most substance demanded and consumed all over the world. The structure of petroleum is formed of an extremely complex mixture of hydrocarbon compounds, usually with minor amounts of nitrogen, oxygen and sulfur as well as trace amounts of metal-containing compounds.⁽¹⁾

Oil refinery is an industrial location where the crude oil is processed, decomposed and separated into many usable materials. One of the most important refineries constructed in Iraq is Al-Daura oil refinery. This refinery was constructed in Baghdad and begun to work in 1955. It is composed of many sections that produce different

products to cover the needs of Iraq from fuel such as gasoline, kerosene, jet fuel and other products. Most of these sections are old and many leaks have been noticed in the refinery. These leaks can be seen by soil saturation with oil and emission of different fumes to the air which can be felt at the moment of entry at the main gate.

Oil refinery workers are in continuous exposure to numerous hazardous materials. The work conditions place them at continuous risk of serious pollutants, injury and death. Lead is toxic heavy metal the oil refinery workers are in a daily exposure.^(2, 3) Lead and cadmium emissions have increased dramatically during the 20th century. The petrol was the main source of lead exposure while the reason for cadmium increase was that cadmium-containing products are rarely re-cycled, but often dumped together with household waste. The adverse health effects of cadmium exposure may occur at low exposure levels, Therefore, measures should be taken to reduce the lead and cadmium exposure in the general population in order to minimize the risk of adverse health effects.⁽⁴⁾

Many studies on the petroleum health effects were conducted. The oral health of occupationally exposed workers to petroleum was assessed by some investigators. Dental caries and periodontal

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diseases were increased in petrol filling workers.⁽⁵⁾

This investigation was conducted to explore some scientific information and to add data for the more in depth researches about the impact of the petroleum and its derivatives on salivary constituents, Up to our knowledge there are insufficient data relevant to this study, therefore the purpose of this investigation was considered to detect the salivary malondialdehyde and superoxide dismutase (as an indicator of oral oxidative status) in addition, Lead and cadmium heavy metals were assessed as an important constituents of petroleum and its possible association with tissue damage or any oral findings. Since lead- and cadmium-induced tissue damages have been attributed, at least in part, to toxicant-induced oxidative stress.^(6,7)

SUBJECTS, MATERIALS AND METHODS

After approvals were obtained from the scientific committee and local authorities, a total of eighty participants were enrolled in this study and they were divided into 2 groups:-

1. Study group: Sixty non-smoker male workers in Al-Daura oil refinery were involved as a study group, with no signs, symptoms and history of any systemic disease. They were Baghdad city residents.
2. Control group: Twenty non-smoker male subjects of Baghdad city resident were involved in this study, with no signs and symptoms of any systemic disease.

All the participated subjects have answered questionnaire form included information regarding their name, gender, age, smoking, residence, type of work and duration of employment.

The oral examination for each individual was done using the disposable dental diagnostic tools and artificial light to detect the oral condition according to the sequence of WHO.⁽⁸⁾

Sample collection

All saliva samples were collected at morning time between 9 a.m. and 1p.m. Before collection of saliva, the subjects were instructed not to eat or drink (except water) for 1 hour.⁽⁹⁾

Mouth washing with pure water was carried out before sampling. All participants were

instructed to collect saliva in their mouths for 5 minutes and to spit into a clean plastic container.

Saliva samples were kept in ice during the collection. In order to reduce bubble and foam, samples were centrifuged and stored at -23 C freezer for analysis.

Biochemical and Immunological Analysis; The salivary malondialdehyde, superoxide dismutase and immunoglobulin A were measured by the use of Enzyme-linked Immunosorbent Assay Kits.

The heavy metals in saliva were analyzed at the poisoning consultation center\specialized surgeries hospital by using the Atomic Absorption Spectrophotometer. Salivary lead level was measured by the flame atomic absorption spectrophotometer. Working Pb standards (0,5,10,15 µg/dl) were prepared.. The standards, samples and quality control specimens were aspirated for measurement of lead. Hollow cathode lamps were used for lead. Absorption was measured at 283.2 nm wavelength.

The measurement of salivary cadmium concentration was done by the flameless atomic absorption spectrophotometer. The samples were analyzed and atomic absorption was done with optical beam at 228.9 nm wavelength.

RESULTS

The mean of salivary lead and cadmium was higher in study group (6.34 µg/dl and 0.56 µg/l respectively) than that of control group (3.3µg/dl and 0.34 µg/l respectively) with highly significant difference (p<0.001). The mean of the salivary MDA in study group (15.39 ng/ml) was higher than that of control group (7.96 ng/ml) with significant difference (p<0.05). Unlike the MDA, the mean of salivary SOD of study group (1895.1 pg/ml) was lower than that of control group (2506.6 pg/ml) with significant difference (p<0.05)

A significant difference (p<0.05) was found between the mean of sIgA of the study group (464.36 µg/ml) and the mean of the control group (344.3 µg/ml), the sIgA in workers group was higher (p<0.05) compared with the control group.

The increase in the level of salivary lead was correlated with an increase in the level of salivary MDA, IgA with significant correlation (p<0.05).

The statistical analysis has shown a significant inverse correlation between the heavy metals (Lead and cadmium) and SOD (p<0.05).

Table 1: The mean and SD of the parameters in study and control group

| Parameters | Patients | | | Control | | | Sig. |
|------------|----------|--------|----|---------|------|----|----------|
| | Mean | SD | N | Mean | SD | N | |
| Age | 43.93 | 10.95 | 60 | 43.7 | 8.28 | 20 | >0.05 |
| Pb µg/dl | 6.348 | 1.321 | 60 | 3.3 | 0.76 | 20 | <0.001** |
| Cd µg/L | 0.56 | 0.103 | 60 | 0.34 | 0.11 | 20 | <0.001** |
| MDA ng/ml | 15.39 | 16.54 | 60 | 7.96 | 10.9 | 20 | <0.05* |
| SOD pg/ml | 1895.1 | 1392.7 | 60 | 2506.6 | 1034 | 20 | <0.05* |
| sIgA µg/ml | 464.36 | 186.7 | 60 | 344.3 | 122 | 20 | <0.05* |

* p<0.05 significant, ** p<0.001 Highly significant

Table 2: Pearson's Correlations (r) between the study parameters

| r | Age | Lead µg/dl | Cadmium µg/L | MDA ng/ml | IGA µg/ml |
|--------------|--------|------------|--------------|-----------|-----------|
| P-value | | | | | |
| Lead µg/dl | 0.263 | | | | |
| | 0.042* | | | | |
| Cadmium µg/L | 0.262 | 0.333 | | | |
| | 0.043* | 0.009* | | | |
| MDA ng/ml | 0.247 | 0.304 | 0.185 | | |
| | 0.057 | 0.018* | 0.158 | | |
| IGA µg/ml | 0.236 | 0.377 | 0.263 | 0.310 | |
| | 0.069 | 0.003* | 0.052 | 0.09 | |
| SOD pg/ml | -0.17 | -0.346 | -0.294 | -0.222 | -0.055 |
| | 0.194 | 0.007* | 0.023* | 0.088 | 0.676 |

* p <0.05 significant, ** p <0.001 Highly significant

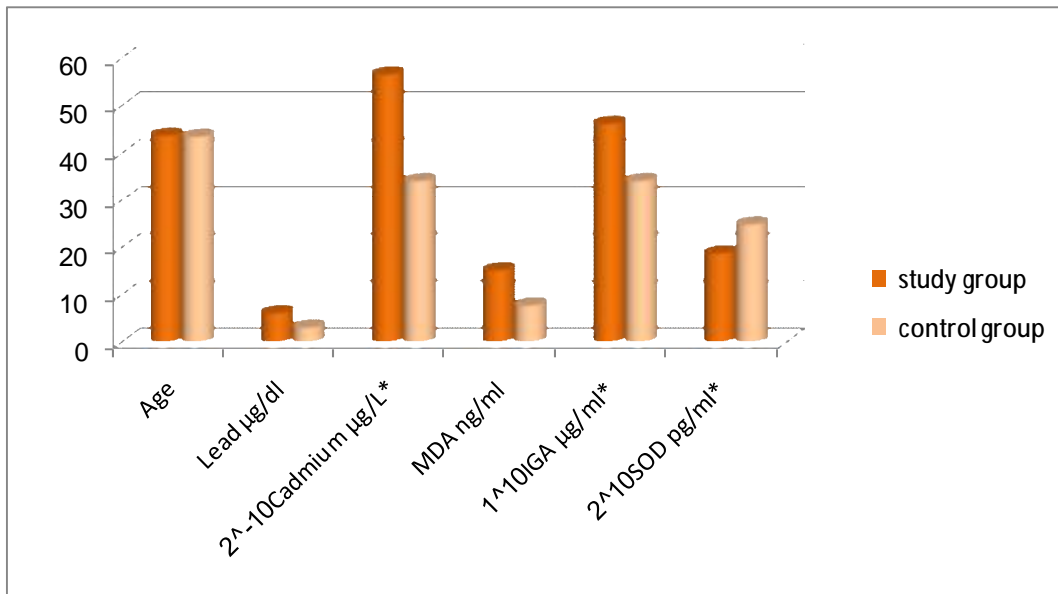


Figure 1: Difference in levels of the parameters between study and control groups.

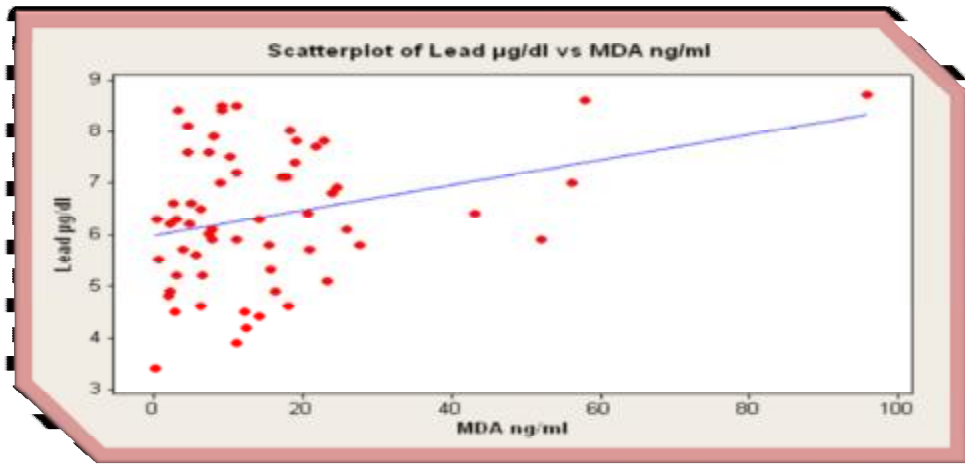


Figure 2: Positive correlation between Lead and MDA

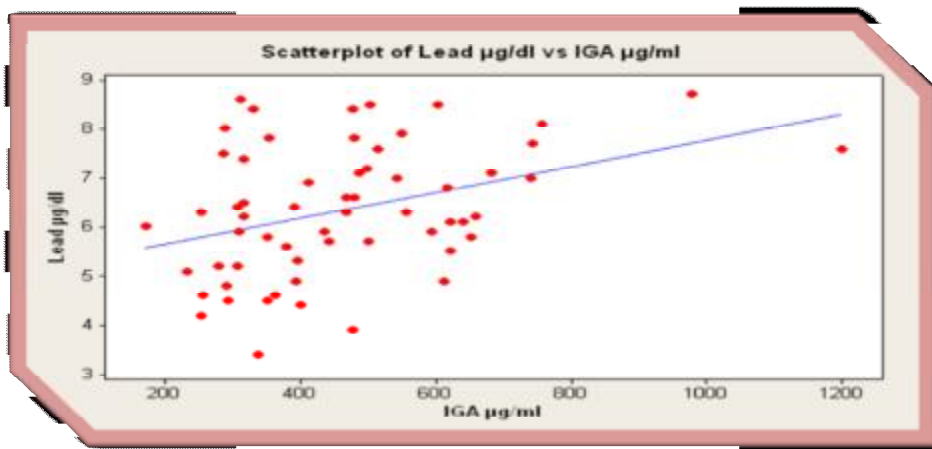


Figure 3: Positive correlation between Lead and sIgA.

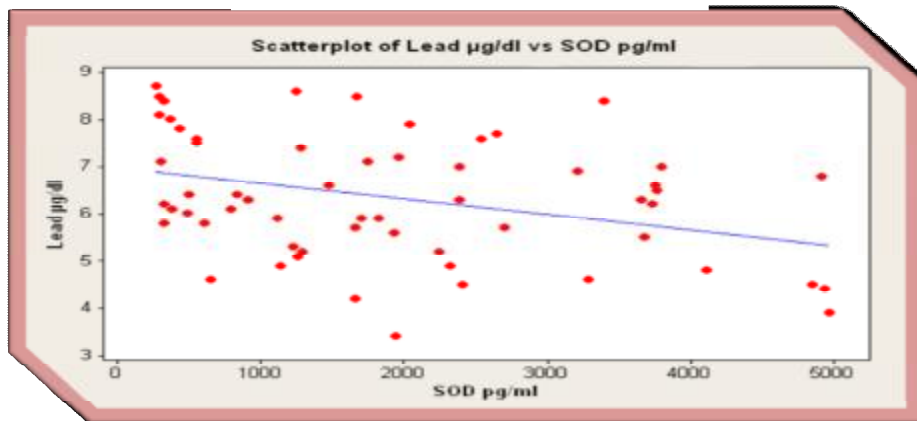


Figure 4: Inverse correlation between lead and SOD.

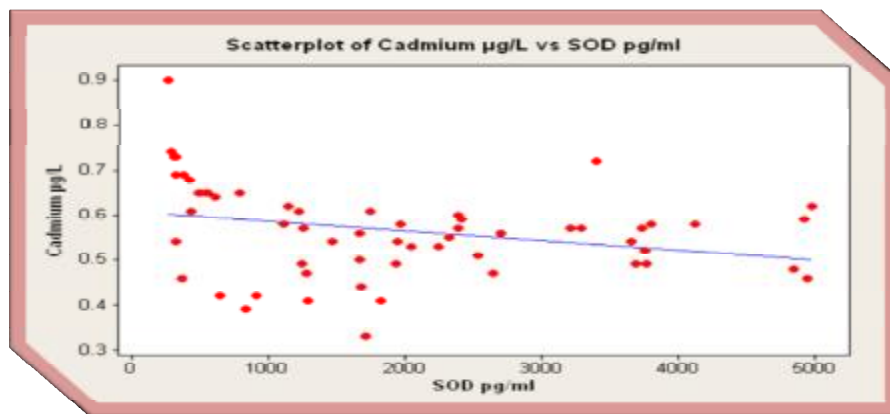


Figure 5: Inverse correlation between cadmium and SOD.

DISCUSSION

In this study the mean age of the study group and control was approximately equal to avoid the effect of age on the clinical, biochemical and immunological findings.

In Iraq the Tetraethyl lead (TEL) is used widely till now to increase the octane rating of gasoline (petrol), although this phase has been canceled and even the production of it was considered globally illegal.

There is no strict regulation in management and maintenance of the constructions and pipelines. Increase in lead and cadmium levels may be associated with presence of many leaks of these pipelines. Poor education and concern about the effects of heavy metals on the health of the workers lead to absence of the protective measures which include the use of masks and gloves.

In this study, the increase in MDA and decrease in SOD was agreed with study on occupationally exposed workers to petrol in petrol stations.⁽¹⁰⁾ A significant positive relation was found between lead and the oxidative stress marker MDA, this result was agreed with a studies on painters⁽¹¹⁾ and battery manufacturing workers.⁽¹²⁾

The significant correlation between lead and MDA (lipid peroxidation) can be explained by the lead-induced oxidative damage to membranes which is associated with changes in the fatty acid composition.⁽¹³⁾ In addition, the fatty acid chain length and unsaturation are the determinant for membrane susceptibility to peroxidation, and lead induced arachidonic acid elongation which might be responsible for the enhanced lipid peroxidation of the membrane.⁽¹⁴⁾

The reverse correlation between the heavy metals (lead and cadmium) and SOD was agreed with study on rats exposed to combined effect of lead and cadmium,⁽¹⁵⁾ however, it was disagreed with study on inhabitants of polluted area.⁽¹⁶⁾ The

decreased SOD activity in workers group is probably due to interaction of lead with copper molecule. As SOD is a Zn-Cu containing enzyme, hence lead exposure induced copper deficiency resulted in decreased SOD activity.⁽¹⁷⁾

It has been demonstrated that cadmium can replace Zn to reduce SOD activity.^(18,19) Given the complex composition of petroleum, the difference in oxidative status among Al-Daura oil refinery workers may be associated with other constituent of the petroleum. Benzene is one of these components that can increase the oxidative damage.^(20, 21) Toluene, ethylbenzene and xylene can induce oxidative stress.⁽²²⁾

The correlation between the lead and IgA in this study was agreed with a study on workers exposed to lead which found an increase in level of serum immunoglobulin A.⁽²³⁾ The increase in serum IgA level may be due to the influence of lead on the differentiation of B cells into antibody producing cells, thereby amplifying B-cell expansion to secrete IgA antibody.⁽²⁴⁾

In this study, the oral examination among workers revealed two cases of line and four cases of pigmentations in different sites in the oral cavity, some of these pigments may be physiological pigments which may need more investigations to determine if there is any association with systemic signs and symptoms of heavy metals toxicity.

The oral lesions including ulcers, white and red lesions, infections and other mucosal problems were absent among workers and they didn't have any history of pronounced oral lesions that can be associated with occupational exposure to pollutants. This absence of lesions in the presence of pollution may be due to increased level of mucosal immunity (\uparrow sIgA) among workers since the level of secretory IgA in saliva has an important role in the protection of the oral tissues against diseases.⁽²⁵⁾

REFERENCES

1. Speight G. The chemistry and technology of petroleum. 3rd ed. New York/Basel: Marcel Dekker; 1998.
2. Engler R. Oil refinery health and safety hazards: their causes and the struggle to end them. Philadelphia, U.S.A: Philadelphia Area Project on Occupational Safety and Health, 1975.
3. Gennaro V, Ceppi M, Boffeta P, Fontana V, Perrotta A. Pleural mesothelioma and asbestos exposure among Italian oil workers. Scand J Work Environ Health 1994; 20(3): 213-5.
4. Jarup L. Hazards of heavy metal contamination. British Medical Bulletin 2003; 68: 167-82.
5. Ammar F, Ahlam T. Oral health status in relation to selected salivary elements among a group of gasoline stations workers. Iraqi Academic Sci J 2013; 25(3): 125-9.
6. Fu H, Ye XB, Zhu JL. et al. Oxidative stress in lead exposed workers. in IARC Gargnano Conference 1999; 2-3.
7. Patra RC, Swarup D, Dwivedi SK. Antioxidant effects of α tocopherol, ascorbic acid and L-methionine on lead induced oxidative stress to the liver, kidney and brain in rats. Toxicology 2001; 162(2): 81-8.
8. WHO. Basic methods of oral health survey. 5th ed. World Health Organization, Geneva, Switzerland, 2013.
9. Martinez KO, Mendes LL, Alves JB. Secretory A immunoglobulin, total proteins and salivary flow in recurrent aphthous ulceration. Rev. Bras Otorrinolaringol 2007; 73: 323-8.
10. Odewabi AO, Ogundahunsi OA, Oyalowo M. Effect of Exposure to Petroleum Fumes on Plasma Antioxidant Defense System in Petrol Attendants. Bri J Pharmacology and Toxicology 2014; 5(2): 83-7.
11. Mohammad IK, Mahdi AA, Raviraja A, Najmul I, Iqbal A, Thuppil V. Oxidative stress in painters exposed to low lead levels. Arh Hig Rada Toksikol 2008; 59(3):161-9.
12. Singh Z, Pooja Chadha, and Suman Sharma. Evaluation of Oxidative Stress and Genotoxicity in Battery Manufacturing Workers Occupationally Exposed to Lead. Toxicology International 2013; 20(1): 95-100.
13. Knowles SO, Donaldson WE. Dietary modification of lead toxicity: effects on fatty acid and eicosanoid metabolism in chicks. Comparative Biochemistry and Physiology C 1990; 95(1) 99-104.
14. Lawton LJ, Donaldson WE. Lead-induced tissue fatty acid alterations and lipid peroxidation, Biological Trace Element Research 1991; 28(2): 83-97.
15. Wang X, Yin X, Bai X. Combined effect of lead and cadmium on lipid peroxidation in renal tubular epithelial cells of rats. Wei Sheng Yan Jiu 2002; 31(4): 232-4.
16. Wieloch M, Kamiński P, Ossowska A, et al. Do toxic heavy metals affect antioxidant defense mechanisms in humans? Ecotoxicology Environmental Safety 2012; 78:195-205.
17. Mytroie, AA. Erythrocyte SOD activity and other parameter of copper status in rats ingesting lead acetate. Toxicol Applied Pharmacol 1986; 82: 512-20.
18. Bauer R, Demeter I, Hasemann V, Johansen JT. Structural properties of the zinc site in Cu,Zn-superoxide dismutase; perturbed angular correlation of gamma ray spectroscopy on the Cu, 111Cd-superoxide dismutase derivative. Biochem Biophys Res Commun 1980; 94:1296-1302.
19. Kofod P, Bauer R, Danielsen E, Larsen E, Bjerrem MJ. 113Cd-NMR investigation of a cadmium-substituted copper, zinc-containing superoxide dismutase from yeast. Eur J Biochem 1991; 198: 607-11.
20. Uzma N, Kumar BS, Hazari MA. Exposure to benzene induces oxidative stress, alters the immune response and expression of p53 in gasoline filling workers. American J Industrial Medicine 2010; 53(12):1264-70.
21. Moro AM, Charão MF, Brucker N, Durgante J et al. Genotoxicity and oxidative stress in gasoline station attendants. Mutation Research 2013; 14: 754(1-2): 63-70.
22. Kim JH, Moon JY, Park EY, Lee KH, Hong YC. Changes in oxidative stress biomarker and gene expression levels in workers exposed to volatile organic compounds. Industrial Health 2011; 49(1): 8-14.
23. Mishra KP, Singh VK, Rani R, Yadav VS, Chandran V, Srivastava SP, Seth PK. Effect of lead exposure on the immune response of some occupationally exposed individuals. Toxicology 2003; 188: 251-59.
24. McCabe MJ Jr, Lawrence DA. The heavy metal lead exhibits B cell stimulatory factor activity by enhancing B cell IgA expression and differentiation. J Immunol 1990; 145: 671-77.
25. Martin SG, Michael G. Burkett's Oral Medicine Diagnosis and Treatment. 10th ed. Hamilton Ontario: BC Decker Inc; 2003.

الخلاصة

مقدمة: عمال مصفاة النفط يتعرضون بشكل مستمر إلى العديد من المواد الخطرة. يحتوي النفط على المعادن الثقيلة مكون طبيعى أو كمادة مضافة. تحفز هذه المعادن إنتاج الجذور الحرة التي ترتبط بضرر تأكسدي في الحمض النووي، البروتين، والدهون. هذه الدراسة أجرت لتقييم المستويات اللعابية للمعادن الثقيلة، الحالة التأكسدية في اللعاب، الحالة المناعية في اللعاب (الغلوبيولين المناعي نوع-أ) وتقييم الحالات المرضية الفموية لدى عمال مصفى الدورة النفطية. المواضيع، المواد والطرق: هذه الدراسة أجريت في مصفى الدورة النفطية، العينات شملت 60 عاملاً يمثلون مجموعة الدراسة و20 شخصاً كمجموعة ضابطة. أجري فحص الفم وجمع عينات اللعاب للتحري عن الحالات المرضية الفموية وقياس المستوى اللعابي للمعادن الثقيلة (الرصاص والكاديوم)، الحالة التأكسدية (المالوندايلديهيد والسوبراوكسايد دسميوتيز) والحالة المناعية الفموية (الغلوبيولين المناعي أ).

النتائج: المستوى اللعابي للمعادن الثقيلة (الرصاص والكاديوم) في مجموعة الدراسة (العمال) (6.34 $\mu\text{g}/\text{dl}$ and 0.56 $\mu\text{g}/\text{l}$) أعلى إحصائياً من المستوى لدى المجموعة الضابطة (3.3 $\mu\text{g}/\text{dl}$ and 0.34 $\mu\text{g}/\text{l}$) نتائج هذه الدراسة أظهرت أيضاً زيادة إحصائية في المالوندايلديهيد (15.3 ng/ml) والغلوبيولين المناعي نوع (أ) (464.3 $\mu\text{g}/\text{ml}$) و إنخفاض في السوبراوكسايد دسميوتيز (1895.1 pg/ml) لدى مجموعة الدراسة. أظهر الرصاص علاقة طردية ($P < 0.05$) مع المالوندايلديهيد والغلوبيولين المناعي الإفرازي نوع (أ). وجدت علاقة عكسية ($P < 0.05$) بين المعادن الثقيلة (الرصاص والكاديوم) و السوبراوكسايد دسميوتيز. لم يظهر الفحص الفموي أي حالة مرضية ذات أهمية.

الاستنتاج: عمال مصفى الدورة النفطية يتعرضون إلى تلوث ناتج من المعادن الثقيلة (الرصاص والكاديوم)، وهذا التلوث مرتبط بتغيرات كيميائية حيوية ومناعية على مستوى الفم.