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# Impact of exposure to low levels of mercury on the health of dental workers

**Leda Freitas de Jesus*** **and Fátima Ramos Moreira**

*Centro de Estudos da Saúde do Trabalhador e Ecologia Humana, Escola Nacional de Saúde Pública Sérgio Arouca, Fundação Oswaldo Cruz, Rua Leopoldo Bulhões, 1480, 21041-210, Manguinhos, Rio de Janeiro, Rio de Janeiro, Brazil. *Author for correspondence. E-mail: ledafreitas@ensp.fiocruz.br*

**ABSTRACT.** This work evaluated the impact of exposure to mercury on the health of workers comparing dentists and dental assistants exposed to mercury by handling amalgam in a public dental clinic with a reference group which, in private offices, did not make use of the metal in their professional routine. Data collection included mercury levels in urine and air samples determined by cold vapor atomic absorption spectrometry, questionnaires and direct observation. The difference between urine and air samples in both groups was statistically significant while mercury levels in air and urine showed positive associations. Mercury concentration in urine correlated with gender, practice time, and age of workers. Half of those exposed had complaints compatible with mercury contamination. Among the exposed, the most common complaints were cognitive and neurocognitive symptoms. Correlations between symptoms and exposure time and also number of amalgam fillings placed per week were positive. Amalgam handling resulted in environmental and biological contamination by mercury.

**Keywords:** mercury, dental amalgam, occupational exposure, symptoms, urine, air.

## Impacto da exposição a baixos níveis de mercúrio sobre a saúde de trabalhadores da odontologia

**RESUMO.** Este trabalho avaliou o impacto da exposição ao mercúrio sobre a saúde de trabalhadores, comparando dentistas e auxiliares odontológicos expostos ao mercúrio, pela manipulação de amálgama em uma clínica odontológica pública, com um grupo de referência que, em consultórios particulares, não utilizava o metal em sua rotina profissional. A coleta de dados incluiu os níveis de mercúrio na urina e amostras de ar determinados por espectrometria de absorção atômica com vapor frio, questionários e observação direta. A diferença entre urina e amostras de ar em ambos os grupos foi estatisticamente significativa, enquanto os níveis de mercúrio no ar e na urina mostraram associações positivas. Entre os expostos, as queixas mais comuns referiam-se a sintomas cognitivos e neurocognitivos. Havia correlação entre a concentração de mercúrio na urina e sexo, tempo de prática e idade dos trabalhadores. Metade dos expostos apresentou queixas compatíveis com contaminação por mercúrio. Havia correlações positivas entre sintomas e tempo de exposição e, também, com o número de restaurações de amálgama executadas semanalmente. A manipulação de amálgama resultou em contaminação ambiental e biológica por mercúrio.

**Palavras-chave:** mercúrio, amálgama dentário, exposição ocupacional, sintomas, urina, ar.

## Introduction

Mercury (Hg) is a cumulative toxic element and its elemental form is one of the most hazardous. The inhalation of Hg vapor can cause chronic and progressive disorder in the metabolic and cellular functions of individuals exposed. Preclinical disorders in the kidney function and behavioral and cognitive changes associated with effects on the central nervous system are of great concern. The metallic vapor can also produce harmful effects on the digestive, respiratory, immune and reproductive systems, as well as dermal changes. Thus, even at low concentrations, it poses a serious risk for humans and living beings (Agency for Toxic Substances and Disease Registry [ATSDR], 1999; 2011).

Hg exposure in dentistry results from using dental amalgam, an alloy composed of 50% of Hg. Inhalation of vapors is the main route for exposure of dental workers due to improper storage and disposal of amalgam waste and empty capsules, inadequate hygiene and ventilation in the workplace, refrigeration failures during the removal of amalgam restorations, and the accidental spillage of Hg drops in dental clinics (Marshall, Marshall, & Anusavice, 2005).

All safety procedures related to the storage of Hg and amalgam waste should be rigorously followed in dentistry since some studies have shown that no solution completely prevents the release of the mercury vapor to the environment (Moreira & Jesus, 2013). According to the World Health Organization (WHO, 2005), dental amalgam is a greatest non-industrial anthropogenic emission source of Hg to the environment.

On the other hand, the Minamata Convention on Mercury, which addresses the control, restriction and banning of mercury from the workplace, encourages the use of mercury-free dental restoration alternatives and promotes the use of best environmental practices in dental facilities to reduce releases of Hg among other measures (United Nations Environment Programme [Unep], 2013).

Some studies showing the effects of Hg on the health of those workers have shown that the risk of damage occurs even in situations where the level of exposure is considered low (Heyer et al., 2004; Neghab, Choobineh, Zadeh, & Ghaderi, 2011). Nevertheless, values considered as safe for occupational exposure to the metal have not been revised (Neghab et al., 2011).

Most publications concerning dental amalgam and symptoms of Hg contamination are related to people possessing amalgam restorations (DeRouen et al., 2006; Guzzi et al., 2006). Such situation increases the relevance of research having dental workers as their target population since the amalgam handling leads to an Hg exposure at higher concentrations than those provided by dental fillings, but also inhalation is the main route of exposure. The absorption of Hg vapors is about 80% of the total inhaled (ATSDR, 1999). In this context, this study aimed to evaluate the impact of exposure to Hg on the health of workers. Levels of Hg in the air of a dental clinic and urine of dental workers as well as the prevalence of symptoms consistent with Hg contamination in such workers exposed to vapors of metallic Hg due to the use of dental amalgam were assessed.

## Material and methods

### Study participants

The study was developed with dental workers (n = 29) as follows: dentists (n = 14) and assistants (n = 5) occupationally exposed to Hg and a reference group of non-exposed dentists (n = 10) also called controls.

The exposed professionals worked at the dental clinic (DC) in a public health center from the government of Rio de Janeiro City, Rio de Janeiro State, Brazil. The site consisted of 10 rooms with 2.9 x 2.8 m each, in a corridor of 0.95 m width throughout 29 m in the clinics environment. The average indoor temperature was 24ºC and no windows or even tilting windows allowing natural ventilation. Air circulation was primarily carried out through the entrances of the dental offices, access to the waiting hall and entry of patients into the procedure rooms.

The reference group just worked in their private offices and did not utilize Hg in their procedures. The rooms had different dimensions, no larger than 9 $m^2$. All offices showed inadequate ventilation, mainly performed by air conditioners. In this group, the average temperature of the offices was 24.5ºC. The exclusion criteria were exposure to Hg additional sources and preexistent pathologies with symptoms similar to mercury contamination.

A self-filling questionnaire requested data on socio-demographics, working and medical histories, biosafety and lifestyle, additional sources of mercury exposure and symptoms or pre-existing pathologies consistent with Hg contamination. Other potential influences on psychomotor response such as alcohol consumption, stress and regular intake of medication were also included. The direct observation and conversations provided information on the preparation and disposal of dental amalgam and storage practices and spillage of Hg. Only questions relative to patient care were excluded from the questionnaire applied to the support personnel.

Cage and Q16 tests identified individuals addicted to alcoholic beverages and participants with possible neuropsychiatric symptoms related to neurotoxic substances, respectively (Bühler, Kraus, Augustin, & Kramer, 2004; Bast-Pettersen, 2006). Those tests are not diagnosis tools; they simply help to identify the population of study.

The research was conducted between August and September, 2010. Both Ethics Committee in Research from the National School of Public Health and Bureau of Health and Civil Defense of Rio de Janeiro City gave permissions. All participants have signed a free and informed consent form.

### Sampling and analysis

Urine was sampled after at least two days of continuous exposure in 50 mL decontaminated polyethylene containers. Sampling occurred in the first period of the working day in a bathroom at the DC, away from the risk of Hg contamination. In private offices, their own bathrooms were used. After sampling, all containers were packed in plastic bags in an upright position, transported to the laboratory under refrigeration and kept frozen at -20°C until analysis.

Thirteen samples were collected in the restorative treatment rooms during patient care. Due to the great dispersion of Hg, one air sample was also collected in the dental surgery room, located in the same corridor of the treatment rooms. The storeroom air was also sampled since a large amount of amalgam capsules and liquid mercury were stored there.

Pumps calibrated to a flow rate of 2 L air min$^{-1}$ (model 224 PCXR4 - SKC, Eighty Four, PA, USA) were coupled to two interconnected impingers containing 10 mL of a 5% (w v$^{-1}$) $KMnO_4$ solution each for Hg trapping. The air sampling system was placed on a bench just beside the amalgamator (Hg source) and around 50 cm from the dentist for approximately 90 min. However, they remained turned on for at least one hour in the private offices. After sampling, impingers were taken to the laboratory and kept under refrigeration until analysis.

Hg levels in urine and air samples were determined by cold vapor atomic absorption spectrometry using a Zeeman 5100 atomic absorption spectrometer equipped with FIAS 400. The wavelength of the hollow cathode lamp for mercury was 253.7 nm, current 6 mA and the slit width 0.7 nm (Perkin Elmer, Norwalk, CT, USA).

A mercury standard solution (1 μg mL$^{-1}$) was prepared by appropriate dilution of the stock (1000 μg mL$^{-1}$) from Merck (Elmsford, NY, USA) in 0.2% (v v$^{-1}$) nitric acid (Merck). From this solution, all working standards (5.0, 10.0 and 15.0 μg L$^{-1}$) were made every day by diluting with 0.2% (v v$^{-1}$) nitric acid. The limits of detection (3 $\sigma$) and quantification (10 $\sigma$) of the methodology used are 0.24 and 0.79 $\mu$g L$^{-1}$, respectively.

The accuracy of the results was verified by the analysis of reference urine samples from Lyphocheck (Level I, Lot. 69141 - Biorad) and Interlaboratory Program for Quality Control, supported by the Ministry of Labour and Immigration of the Government of Spain. The same reference materials were used to check the accuracy with air samples since the results for mercury are obtained in mass unit. Therefore, the matrix interference is virtually nonexistent.

### Statistical analysis

All values were reported as mean ± standard deviation (SD) unless indicated. Descriptive statistics was used to characterize the participants and show the frequency of Hg contamination in the population of study. The Spearman's correlation was used to examine the relationship between Hg levels in urine and air as well as between biological Hg levels and gender, age, time of exposure to Hg and number of amalgam fillings per week. The Student's *t* test for two independent samples investigated the statistical significance between concentrations of Hg in urine and air samples of both dentist groups.

The presence of signs matching those described in the literature as characteristic of Hg contamination was investigated in those workers based on answers provided in the questionnaire. The Spearman test also established correlations between complaints and time of occupational exposure as well as the number of amalgam fillings placed per week.

A p-value ≤ 0.05 and 95% of confidence were always considered unless otherwise stated. All statistical operations were performed using SPSS v.17 (SPSS Inc., Chicago, USA).

## Results and discussion

### Study population

Two dentists were excluded among all 29 subjects recruited since they have reported additional sources of exposure to Hg. One professional in the exposed group lived near factories with Hg in their manufacturing processes while the other from the controls used to handle amalgam in a course of oral health.

Among those 27 participants, female workers (74%) were the most significant in all groups. Eight women (62%) and five men (38%) constituted the group of dentists exposed while the reference group consisted of eight females (89%) and one male (11%). In the same way, four women (80%) and only one man (20%) were dental assistants.

Although all participants were in the same age range, women were younger than men. The group of exposed dentists had a mean age of 42.4 years (women = 39.0; men = 47.8 years). Controls were 45.2 years old on average (females = 44.5; male = 51.0) while the average was 47.2 years among the assistants, being 42.5 for women and 66.0 years for the man.

The average time of practice as a dentist was 16.2 (range = 10 to 25 years) whereas a mean of 8.2 years of work as a dental assistant (range = 2 to 20 years) was found among auxiliaries. In the reference group, this average was 20.6 years (range = 16 to 27 years). Among those exposed, dentists and assistants worked twenty and forty hours a week, respectively. However, the actual time of mercury exposure was shorter than a working day. On average, exposed dentists used to attend ten patients, while the control group attended eight per day.

Two dentists in the exposed group worked regularly with amalgam, making an average of 12.3 (range = 5 to 30) fillings per week. Another professional did not make any amalgam filling; however she used to open capsules to remove filings and use them to strengthen the glass ionomer cement. Seven of those professionals (53.8%) performed amalgam fillings every day, with an average of 2.4 per dentist a day. All dentists together placed 84 amalgam fillings a week. The correlation between Hg-U and number of amalgam fillings placed per week ($\rho = 0.85$) was statistically significant.

Higher female labor force participation among dentists in this study followed a world trend in dentistry. In an attempt to reconcile career and family obligations, female dentists tend to have a shorter working life and to work fewer hours a week than men. Moreover, they prefer to work in urban areas and as employees, providing a shortage of professionals in rural areas and a lower net income for women. Female dentists have more empathy and communicate better with patients, meaning a less stressful care. At long term, the characteristics of a female work will lead to important changes in the profession, including the increasing number of dentists required to meet patient demand as well as a more humanistic approach to the oral health care (Blanton, 2006; McKay & Quiñonez, 2012).

Unlike our results, significantly more dentists were male and older than the controls in another research (Ritchie et al., 2004). However, the average time of dental practice was similar among dentists between both studies. Although the mean number of hours worked was 32.8 hours a week, those dentists placed an average of 35.5 amalgam fillings a week, an amount smaller than in our research whose dentists placed 84 amalgam fillings in a 20 hours working week.

The current status of exposure to mercury among dental health workers was investigated in Turkey. The average number of amalgam fillings made by the dentists (18 per week) correlated significantly with plasma Hg levels (Yilmaz et al., 2013).

The occupational exposure of dental professionals to metallic mercury in dental offices of a public primary health care in the city of Maringá, Paraná State, Brazil, was evaluated. Regarding to gender, results were similar to this research since women were the majority in the exposed group (84%) and controls (90%). The average age of the exposed group was 38 years old, and in the control group, 36 years old. The dentists and assistants worked 20 and 30 hours a week, respectively. Concerning the time as professionals, the average was 17 ± 7 years for the dentists and 14 ± 8 years for assistants (Oliveira, Nishiyama, Gasparetto, & Machinski Júnior, 2012). Unlike our study, the groups were younger and the assistants worked 10 hours less per week, but they had more time in the profession. However, the working hours a week and average time of dental practice were similar among dentists between both studies.

### Urine

Samples of all participants showed Hg in the urine. A particular dentist from the reference group presented Hg-U, 2.7 ± 0.71 $\mu$g Hg $L^{-1}$, higher than the others since she was exposed to an additional source of Hg according to the questionnaire. Although this professional did not deal directly with patients, she used to teach in a course of oral health. Thus, her Hg-U was not included in the statistics. Results found for mercury concentration in urine of all participants are shown in Table 1.

**Table 1.** Levels of Hg in urine (Hg-U).

| Hg-U ($\mu$g $L^{-1}$) | Non-exposed Group (n = 8) | Exposed Group | |
|---|---|---|---|
| | | Dentists (n = 13) | Assistants (n =5) |
| Mean ± SD | 0.92 ± 0.33 | 1.79 ± 0.68 | 1.70 ± 0.66 |
| Median | 0.90 | 1.07 | 1.07 |
| Minimum | ≤ 0.79* | 1.07 | 1.07 |
| Maximum | 1.07 | 5.80 | 4.22 |

*Limit of quantification; SD: standard deviation.

Although at lower levels, the presence of Hg was also detected in the urine of the non-exposed group. Three dentists (37.5%) in this group showed Hg-U equal or lower than the quantification limit of the methodology adopted in the study (0.79 $\mu$g $L^{-1}$).

Hg concentrations in the urine of the dentists exposed were significantly different from the non-exposed group ($p = 0.038$), indicating that Hg-U in the DC dentists was 1.93 fold higher than in the non-exposed group. Positive associations were found between Hg concentration in urine and gender of exposed ($\rho = 0.754$) and control groups ($\rho = 1$), time of practice in the profession of participants exposed ($\rho = 0.629$) and controls ($\rho = 0.354$) as well as age of exposed ($\rho = 0.552$) and non-exposed ($\rho = 0.940$) groups.

Hg levels in urine of all participants were lower than the Biological Exposure Indices (BEI) of 35 $\mu$g $g^{-1}$ creatinine (approximately 50 $\mu$g $L^{-1}$) established by the American Conference of Governmental Industrial Hygienists (ACGIH, 2014), and also of 36 $\mu$g $L^{-1}$ proposed by the European Commission (Pan American Health

Organization [Paho], 2011). Those findings corroborated the statement of the World Health Organization (WHO, 2003) in which all living organisms are subject to a certain degree of Hg exposure.

Results found in urine samples were similar to those reported by a Scottish study developed among 180 dentists, in which all but one professional presented a urinary Hg level below the Health and Safety Executive Health guidance value of 20 μmol $mmol^{-1}$ creatinine (approximately 36.72 $\mu g\ L^{-1}$) in the United Kingdom. The research also revealed an average concentration of mercury in the urine of dentists exposed (4.74 $\mu g\ L^{-1}$) four times higher than that of controls (1.23 $\mu g\ L^{-1}$) (Ritchie et al., 2004).

In a work developed in Turkey, Hg levels in the urine of the dentists exposed (6.29 ± 3.5 $\mu g\ L^{-1}$) were three fold those found in the control group (1.98 ± 0.9 $\mu g\ L^{-1}$) (Karahalil, Rahravi, & Ertas, 2005). However, in Tunisia, the group exposed presented Hg-U significantly higher than controls (about 29.1 ± 60.6 and 0.06 ± 0.4 $\mu g\ L^{-1}$, respectively) (Chaari et al., 2009). The highest levels of contamination among professionals from developing countries suggest an ordinary negligence of these workers regarding to occupational safety.

The average Hg-U of exposed group in Maringá, Paraná State, Brazil (Oliveira et al., 2012), was 2.08 ± 2.11 $\mu g\ g^{-1}$ creatinine (2.97 ± 3.01 $\mu g\ L^{-1}$) while the controls was 0.36 ± 0.62 $\mu g\ g^{-1}$ (0.51 ± 0.89 $\mu g\ L^{-1}$), level over five times that in the last group and higher than that found among the exposed in Rio de Janeiro.

Those findings published by Trzcinka-Ochocka, Gazewski, and Brodzka (2007) corroborated our results since a positive correlation was observed between Hg concentration in the urine of participants and time of occupational exposure in both studies. The variable 'time of professional practice' may have influenced our results since most professionals belonged to only two ranges.

The four oldest dentists of the exposed group (between 55 and 58 years) presented the lowest concentrations of Hg in urine in this research. In the reference group, eight dentists (88.88%) were between 40 and 51 years old, which may have influenced the positive correlation between Hg-U and age in this group. Unlike this research, another study found no correlation between age of dentists and Hg-U (Ritchie et al., 2004). The most sensitive target following inhalation exposure to metallic Hg is the central nervous system (CNS). Although the metallic mercury is readily oxidized to mercuric ion ($Hg^{2+}$) after absorption and its transport into the brain is limited, once in the CNS, the metallic Hg is oxidized to $Hg^{2+}$ and then trapped in the central nervous system due to its limited ability to cross the blood-brain barrier. Moreover, it was demonstrated that the ratio of Hg in the brain to that in the kidneys increased significantly with age in mice (ATSDR, 1999). Thus, the increased Hg in the brain probably reduced the renal content in older workers, lowering the Hg levels in urine.

The positive association between the number of amalgam fillings placed and levels of Hg in urine found in our study was similar to a research, whose dentists also presented Hg-U within limits considered safe (Naleway et al., 1985). Another study compared mercury contamination in dentists and dental assistants in Monastir (Tunisia) to a population not exposed occupationally. Similar results were reached since the exposed group had levels of Hg-U below the safe limit and a correlation was found between Hg-U and the number of dental fillings made per week (Chaari et al., 2009).

Similarly to this work, a Norwegian research also observed a positive association between the variable 'gender' and Hg-U in the dentists evaluated. The variation according to gender may be due to differences in the pulmonary absorption or kinetics of urinary excretion of Hg as well as daily fluctuation in the metal concentration in urine (Paho, 2011). However, the higher prevalence of women among the participants in our study may have affected the outcome.

In contrast to all works already undertaken on the subject, this research found the highest levels of Hg in two professionals (a dentist and an assistant) who did not directly deal with amalgam. Those individuals worked at opposite ends of the clinic and away from the offices where such an alloy was used. In the questionnaire, both positively marked the two questions related to the usual symptoms of Hg contamination. The ability of Hg vapors to spread easily over the entire environment could explain the situation described (Unep, 2013).

The biological response to a particular chemical is independent of the concentration of that substance in the environment (American Conference of Governmental Industrial Hygienists [ACGIH], 2014). However, some individual characteristics can increase the susceptibility to Hg effects (Echeverria et al., 2010), explaining an increased susceptibility to the metal in those two participants since they were not subject to additional sources and special health conditions to explain higher body burdens of Hg. All other dentists and assistants presented similar levels of urinary Hg, indicating that people environmentally exposed to similar concentrations had different absorption of the metal (Ritchie et al., 2004; ACGIH, 2014).

**Air**

Although only seven dentists have reported the use of dental amalgam in their professional routine, Hg was present in all fifteen air samples from the dental care unit, even where the material was not manipulated.

Table 2 shows the concentrations found for Hg in air samples. The calculation of averages excluded outliers' values. In order to compare the results found with existing reference values, concentrations in air samples were estimated for an exposure time of 8 hours a day and 40 hours a week. Hg concentrations in air samples collected in the DC and reference offices ($p = 0.038$) were significantly different. Also, the average concentration in the DC was 1.9 times higher than offices of the controls. Hg levels in air and urine showed positive associations between exposed ($\rho = 0.129$) and control groups ($\rho = 0.761$).

Hg levels found in air samples were well below the Threshold Limit Value (TLV = 8 hours a day and 40 hours a week) of 25 μg $m^{-3}$ (ACGIH, 2014). An Iranian research reported a mean concentration of atmospheric Hg (3.97 ± 6.28 μg $m^{-3}$) higher than that in this study (0.34 ± 0.10 μg $m^{-3}$), but still within the TLV (Neghab et al., 2011). Neghab, Choobineh, Zadeh, and Ghaderi (2011) also observed an association between airborne and urinary concentrations of Hg in dentists ($p = 0.02$), corroborating our result ($p \leq 0.05$). The evaluation of 180 dental offices in Glasgow, Scotland, found 122 environments (68%) with Hg concentrations (range = 6.5 to 38.9 μg $m^{-3}$) higher than 25 μg $m^{-3}$ (ACGIH, 2014). The Scottish study found a significant correlation between levels of Hg in urine ($p \leq 0.01$) and the majority of the environmental samples ($p = 0.02$) (Ritchie et al., 2004).

Although Hg has been found in all air samples from the dental care unit, levels were below that limit considered as safe for the occupational health (ACGIH, 2014). According to publications on the subject, the environmental concentration of Hg in a dental office varies throughout the day depending on factors such as ventilation (Karahalil et al., 2005), temperature (Occupational Safety and Health Administration [Osha], 2010), procedure being performed (Bains, Loomba, Loomba, & Bains, 2008) and place for sample collection (Ritchie et al., 2004). Thus, it is difficult to determine the actual concentration of Hg to which dental workers are exposed during their professional routine.

Low concentrations of urinary and environmental Hg found in this research are opposite to that published over three decades ago (Kelman, 1978). Reductions observed may reflect an increased awareness among dentists related to the risks arising from the use of Hg, automated methods for preparing the amalgam, satisfactory implementation of biosecurity measures, and primarily due to the replacement of amalgam by another restoring material. All of these situations that, hypothetically, prevent contamination by Hg were present in the environments evaluated. In addition, hot sterilizing equipment (oven and autoclave) identified as contributing factors to the vaporization of Hg were located in another room, away from the dental offices.

According to the National Institute for Occupational Safety and Health (Niosh, 2011), the limit value for occupational exposure to metallic Hg would be 0.05 μg $m^{-3}$ for 8 hours. When compared, all of those estimated values exceed the limit proposed by Niosh (2011). In fact, only with the actual sampling time, 87.5% of the samples had concentrations above this threshold while the mercury level was equal to the proposed limit in the remaining 12.5%. On the other hand, for a period of 40 hours, samples would present environmental concentrations considered as safe when compared to the limits established by regulatory agencies for occupational health such as ACGIH (2014) and Occupational Safety and Health Administration (Osha, 2010). However, according to the World Health Organization (WHO, 2003), no level of exposure to Hg vapor is 100% safe. Such a statement shows the need to reduce the limits of exposure to Hg mercury. Thus, the development of alternative restorative materials offering qualities such as the strength, durability and low cost of the amalgam will contribute to decrease the exposure.

**Table 2.** Hg concentration in air (Hg-Air), according to the actual sampling time and time-weighted average (TLV – TWA).

| Hg–Air (μg $m^{-3}$) | Private Offices (n = 9) | | | Public Dental Clinic (n = 15) | | |
|---|---|---|---|---|---|---|
| | Actual Time (1 hour) | Estimate 8 hours | Estimate 40 hours | Actual Time (1.5 hours) | Estimate 8 hours | Estimate 40 hours |
| Mean ± SD | 0.10 ± 0.06 | 0.59 ± 0.30 | 2.94 ± 1.50 | 0.34 ± 0.10 | 1.38 ± 0.35 | 6.89 ± 1.75 |
| Median | 0.08 | 0.54 | 2.71 | 0.31 | 1.32 | 6.60 |
| Minimum | 0.06 | 0.31 | 1.58 | 0.13 | 58 | 2.87 |
| Maximum | 0.58 | 3.70 | 18.51 | 0.84 | 3.66 | 18.51 |

The Minamata Convention on Mercury addresses the control, restriction and, in some cases, the banning of mercury from the workplace. One of its articles establishes control measures for the use of dental amalgam. Promoting and encouraging the use of mercury-free alternatives for dental restoration as well as promoting the use of best environmental practices in dental facilities to reduce releases of mercury are some of those measures. According to the Convention, governments that ratified the treaty are required to eliminate the Hg use and promote mercury-free gold processing alternatives. The critical role of the health sector in addressing the Hg exposure was also recognized (Unep, 2013).

### Symptoms of mercury contamination

Besides the two professionals already excluded, another dentist from the reference group was also excluded since she marked eleven neuropsychological symptoms and reported going under menopause treatment.

Fifty percent of workers exposed presented some signs similar to those of Hg contamination. Among professionals reporting those symptoms, five (two dentists and three assistants) did not handle amalgam directly; they only worked in a place where Hg was disperse. Regarding dentists manipulating amalgam routinely, three did not present any symptom. Three dentists exposed who were positive in the Cage test had their answers related to neuropsychological disorders excluded from the calculation of morbidity (Table 3).

**Table 3.** General morbidity in the studied population.

| Morbidity | Dentists | | Assistants (n = 5) |
|---|---|---|---|
| | Non-exposed (n = 8) | Exposed (n = 13) | |
| | n (%) | n (%) | n (%) |
| Complaints Presented | 2 (25) | 6 (46.2) | 3 (60) |
| CAGE Test | - | 3 (23) | - |
| Cognitive/Neurocognitive | 2 (25) | 6 (46.2) | 4 (80) |
| Gastrointestinal | 1 (12.5) | 2 (15.4) | 1 (20) |
| Dermatological | - | 2 (15.4) | - |
| Fem. Reprod. System[a] | - | 3 (37.5)[b] | 1 (25)[c] |

n: total number of participants in each group; n: number of participants for each morbidity. [a]Fem. Reprod. System: Female Reproductive System; [b]n = 8 women; [c]n = 4 women.

Memory loss was the only signal reported by all three groups, being two cases in the reference group. Among the exposed, the most common complaints were neurocognitive. Table 4 shows the cognitive and neurocognitive symptoms presented by both exposed populations. There were also two dentists complaining of excessive tiredness. An assistant was the only person to complain of muscle weakness as well as 'pressure' on the chest and reduced sex drive. The recurrent tremor in eyelids of a dentist and an assistant stood out since both individuals were the youngest in their groups.

**Table 4.** Description of the cognitive and neurocognitive symptoms presented by both exposed populations.

| Symptoms | Exposed Dentists (n = 13) | Assistants (n = 5) |
|---|---|---|
| | n (%) | n (%) |
| Memory loss | 3 (23.1) | 2 (40.0) |
| Insomnia | 4 (30.8) | 1 (20.0) |
| Tingling and numbness | 3 (23.1) | 1 (20.0) |
| Poor concentration | 1 (7.7) | 2 (40.0) |
| Tremor | 1 (7.7) | 1 (20.0) |
| Sensitivity loss | 1 (7.7) | 1 (20.0) |
| Lethargy | 1 (7.7) | 1 (20.0) |
| Irritability | 1 (7.7) | 1 (20.0) |
| Unfounded sadness | 1 (7.7) | 1 (20.0) |

n: total number of participants in each group; n: number of participants presenting each symptom.

The exposed group also reported three cases of nausea (2 dentists and 1 assistant). However, different dentists presented each one of the following symptoms itching on hands and arms, skin peeling, miscarriage, ectopic pregnancy and endometriosis. Only one assistant reported uterine polyp. In the reference group, there was only one complaint of nausea.

Manifestations of poisoning occurring after some time of exposure to mercury vapors have been called erethism syndrome, characterized by irritability, anxiety, mood lability and impaired sociability, shyness, lack of interest in life, and low self-confidence followed by depression, delirium, hallucinations, fatigue, depression and memory loss (Carocci, Rovito, Sinicropi, & Genchi, 2014).

Occupational exposure has resulted in erethism and a fine tremor develops with continuing exposure. Subclinical manifestations of sensorimotor polyneuritis and the presence of polyneuropathy in dentists have been demonstrated. Long-term, low-level exposure has been found to be associated with less pronounced symptoms of erethism, characterized by fatigue, irritability, loss of memory, vivid dreams, and depression (ATSDR, 1999)

Excessive tiredness, itching on the hands and arms, skin peeling on the thumbs, abortion, ectopic pregnancy and endometriosis were only reported by the exposed dentists, while the assistants reported feeling muscle weakness, chest pressure, reduced sex drive, nausea and uterine polyps. However, both dentists and assistants had insomnia, tingling and numbness, difficulty in concentration, tremors, lethargy, irritability and unfounded sadness. All complaints mentioned by both exposed groups are compatible with the erethism syndrome and other manifestations that occur after continuous exposure to mercury.

In general, the erethism syndrome is associated to elevation of Hg-U, gum bleeding, softening of the teeth and sharp tremors. However, several authors (Rojas, Guevara, Rincón, Rodríguez, & Olivet, 2000; Li et al., 2008; Moen, Hollund, & Riise, 2008; Neghab et al., 2011) have found symptoms of the same syndrome in workers exposed to values considered safe as 0.1 mg $m^{-3}$ in the air and below 50 $\mu g$ Hg $g^{-1}$ creatinine in urine (Paho, 2011).

Differences between frequencies of neurocognitive ($p < 0.01$), gastrointestinal ($p < 0.001$) and dermatological ($p < 0.001$) symptoms as well as those related to the female reproductive system ($p < 0.05$) in both groups were statistically significant. The presence of symptoms and number of amalgam fillings placed per week among dentists exposed presented a positive correlation ($\rho = 0.43$). However, the relationship between the presence of those symptoms and time of occupational exposure was only slight ($\rho = 0.02$).

Our study found significantly higher Hg levels in the exposed group than controls. A higher prevalence of symptoms was achieved although both averages of Hg-Air and Hg-U had been lower than the threshold limit value of 25 μg $m^{-3}$ and biological exposure index of 42 $\mu g$ Hg $L^{-1}$ (ACGIH, 2014), respectively. An Iranian study reported similar findings, with low Hg-Air (mean = 3.35 $\mu g$ $m^{-3}$) and Hg-U (mean = 2.86 $\mu g$ $L^{-1}$) and a high prevalence of symptoms observed in exposed dentists (Neghab et al., 2011). On the other hand, our results differed from those found in a Norwegian research which investigated cognitive and neurological symptoms in dentists with relatively low Hg-U (10.50 $\mu g$ $L^{-1}$ for females and 10.70 $\mu g$ $L^{-1}$ for males) and the prevalence of symptoms was the same in both groups. However, the authors reported that uncontrolled confounding variables such as drug abuse, mental illness, cardiovascular disease and injuries in the head, among others, could have influenced their result. In addition, Norway has forbidden the use of mercury in dental fillings since 2008 (Hilt, Svendsen, Syversen, Aas, & Qvenild, 2011).

According to the World Health Organization (WHO, 2005), exposure levels of approximately 20 $\mu g$ $m^{-3}$ or higher induced subtle effects on the central nervous system in long-term occupational exposures to Hg vapor. However, the Iranian research found a low Hg-Air (mean = 3.35 $\mu g$ $m^{-3}$) and dentists exposed presented a higher prevalence of neurological symptoms ($p \leq 0.05$) than the control group (Neghab et al., 2011).

The DC environment showed good biosafety conditions. Nevertheless, half of dental workers presented disorders matching those described in the literature as characteristic of mercury contamination (ATSDR, 2011), including some who did not use amalgam regularly in their professional routine. So, despite the small number of participants, dentists exposed were more likely to suffer from disorders related to Hg contamination than those non-exposed in the present study.

The high prevalence of cognitive and neurocognitive symptoms among DC dentists (46.2%) and assistants (80%) drew our attention. Some studies also reported the occurrence of neuropsychological symptoms in dentists and dental assistants while others found no relationship between occupational exposure to Hg in dentistry and neurological and cognitive symptoms in dental workers (Moen et al., 2008; Hilt et al., 2011; Thyghesen, Flachs, Hanehøj, Kjuus, & Juel, 2011). However, the reliability of self-report is limited in the context of occupational and environmental health (Bast-Pettersen, 2006)

A dentist and an assistant in the exposed group and two dentists from the controls presented neuropsychological symptoms. Meanwhile, they were all going through menopause, which can cause neuropsychiatric disorders among others (Benediktsdóttir, Tómasson, & Gíslason, 2000). Similarly, complaints of excessive tiredness may also be a result of their double working day as all of them performed their profession elsewhere. On the other hand, tingling and numbness are also characteristic responses of the 'Work-Related Musculoskeletal Disorder', a highly prevalent pathology in dentistry (Hayes, Cockrell, & Smith, 2009).

Studies have showed that the appearance of signs and symptoms of Hg contamination depended on the exposure from other sources, number of amalgam fillings placed and time of exposure (Rojas et al., 2000; Hilt et al., 2011; Thyghesen et al., 2011). Additionally, the absorption of Hg also varies substantially among individuals exposed to similar concentrations and some people are more susceptible to the metal toxicity than others (ATSDR, 1999).

None of the three DC workers who presented the highest rates of positive responses in the questionnaire was subject to Hg additional sources and neither did they present any special conditions such as pregnancy or illness associated with increased susceptibility to the toxicity of the metal. Thus, individual or genetic factors could have facilitated the toxicity of Hg in those people (Heyer et al., 2004; Echeverria et al., 2010).

Hg contamination is mainly characterized by neuropsychological effects and tremor is the most typical sign of chronic exposure (Paho, 2011). Similarly to the present study, cases of eyelid tremor were reported among Chinese workers of artisanal mercury mines (Li et al., 2008). Evaluations with dentists exposed to Hg through amalgam handling mentioned neurological symptoms (cephalea, irritability, cramps, paresthesia, tremors, dizziness, muscle weakness, forgetfulness and difficulty concentrating, memory loss, muscle fiber atrophy) as well as psychic disorders (irritability, mood swings, nervousness, depression and insomnia) (Rojas et al., 2000; Heyer et al., 2004; Ritchie et al., 2004; Moen et al., 2008).

Significantly higher concentrations of Hg in brain tissues collected from dentists during autopsies compared to the control group proved the risks posed to the nervous system through exposure to the metal (ATSDR, 1999). Mood swings caused by Hg contamination had a significant effect on the life quality of dentists (Heyer et al., 2004). The neuropsychological effects related to mercury exposure persisted despite the recovery of functions has occurred (Zachi, Taub, Faria, & Ventura, 2008). Rojas and his coworkers observed the need to monitor the environmental mercury and conduct clinical evaluation and neurobehavioral tests to detect early effects on health of dental workers (Rojas et al., 2000).

The long-term inhalation of Hg vapor can cause nausea (Paho, 2011) as reported in in the exposed group, although some studies have found no gastrointestinal symptoms on dentists who presented Hg contamination symptoms (Ritchie et al., 2004; Neghab et al., 2011)

The chronic inhalation of Hg vapor can also cause skin reactions (Paho, 2011). Dentists investigated in the Iranian study showed such symptoms (Neghab et al., 2011), corroborating our findings.

An investigation found a prevalence of abdominal pain (OR = 1.470) and dysmenorrhea (OR = 1.66) significantly higher among female workers exposed to Hg in food processing plants than in the control group (Yang, Chen, & Jiang, 2002). However, dental workers who handled amalgam during pregnancy had no increased risks of miscarriage when compared to pregnant women not exposed to the metal (ATSDR, 2011). Elemental Hg inhaled can cross the placental barrier although significant fetotoxic or developmental effects have not been well studied in humans (ATSDR, 1999).

Few researches investigated the relationship between the number of amalgam fillings placed and symptoms of Hg contamination in dental professionals. Two of them found a positive association between the number of amalgam fillings placed and the prevalence of neuropsychological symptoms and muscular disorders ($p \leq 0.001$) in dentists as well as fertility of female dental assistants ($p < 0.05$) (ATSDR, 1999). However, a positive correlation was not achieved in another investigation concerning neurological symptoms in dental assistants (Moen et al., 2008). The lack of information on this subject shows the need to conduct further studies on such association in dental professionals.

Studies found a positive association between the time of dental practice and symptoms of contamination such as those related to the female reproductive tract ($p < 0.05$) and long-term visual memory ($p = .01$) (Lindbohm et al., 2007). Such results are similar to those found in our research. However, they differ from another one which found no positive correlation among those variables (Hilt et al., 2011). As the biological half-life of Hg in brain tissues is about one year, individuals occupationally exposed to mercury for many years remain a longer time under the neurotoxic action of the metal after the exposure has ceased (ATSDR, 1999).

## Conclusion

Although the small number of participants has been the major limitation of this study, our findings were corroborated by the literature. Despite the good working conditions, amalgam handling resulted in an environmental and biological contamination by mercury. Therefore, the use of Hg amalgam may be harmful to the health of dental workers even when levels in urine and air of the workplace are within reference ranges. Minimizing the exposure to mercury and preventing the environmental contamination require measures of hygiene. Simultaneously, campaigns should promote the economic and clinically effective use of mercury-free alternatives to dental restorations.

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