

## Determination of Mercury in Dental Hard Tissue: An in Vitro Study

Nexhmije Ajeti<sup>1</sup>, Violeta Vula<sup>2\*</sup>, Miranda Stavileci<sup>2</sup>, Lindihana Emini<sup>3</sup>

<sup>1</sup>Department of Operative Dentistry and Endodontology, University for Business and Technology, Prishtina, Kosovo

<sup>2</sup>Department of Dental Pathology and Endodontics Faculty of Medicine, University of Prishtina, Prishtina, Kosovo

<sup>3</sup>Department of Dental Pathology and Endodontics, Faculty of Medical Sciences, University of Tetova, Tetova, North Macedonia

### Abstract

**Background:** The purpose of this study was to determine the levels of mercury in hard tissue from dental amalgam fillings under in vitro conditions.

**Methods and Results:** The study included 30 human teeth that were extracted for various clinical reasons. The teeth were stored in a physiological solution until they were used. The teeth were divided into 3 experimental groups: Group 1 (n=10) – occlusal surface cavity preparation (class I according to Black); Group 2 (n=10) – proximal-occlusal surface cavity preparation (class II); and Group 3 (n=10) – mesio-occlusal-distal [MOD] surface cavity preparation. Each of these groups was divided into 2 subgroups: subgroup 1 (n=5) – amalgam fillings were not polished, and subgroup 2 (n=5) amalgam fillings were polished. The teeth were filled with amalgam, and those in subgroups 2 were polished after 24 hours. The amount of mercury released from the amalgam fillings was determined 9 months after the teeth were filled. Before chemical analysis, the teeth were irrigated 4 times over a period of 10 minutes in an ultrasonic bath. From each tooth, 250 mg of the powder was mineralized with royal water (HCl+HNO<sub>3</sub> in a ratio of 1:3) in a microwave oven, for 54 minutes. After mineralization, the samples were filtered and analyzed with inductively coupled plasma optical emission spectrometry. The average mercury level after polishing the amalgam filling was significantly smaller ( $P=0.032$ ) only in Group 1. The average mercury levels in the 3 groups revealed significant differences between both the unpolished samples (one-way ANOVA  $F=69.54$ ,  $P<0.001$ ) and the polished samples (one-way ANOVA  $F=110.54$ ,  $P<0.001$ ). Group 3 with MOD surface cavity preparation was characterized by the highest mercury levels.

**Conclusion:** The more mercury is released from unpolished amalgam fillings than from polished amalgam fillings in teeth with occlusal surface cavity preparation (class I according to Black). The teeth with an MOD amalgam restoration are characterized by the highest mercury levels. (*International Journal of Biomedicine*. 2022;12(4):601-605.).

**Keywords:** dental fillings • amalgam • mercury • polishing

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### Introduction

The use of mercury (Hg) in dentistry has been controversial since at least the middle of the 19th century. This controversy has recently intensified because of techniques that show that mercury is continually released from dental amalgam fillings.<sup>(1)</sup>

Mercury is a naturally occurring element that exists in 3 forms: organic, inorganic, and elemental. Concentrations of mercury in most foodstuffs are often below the detection limit. Fish and marine mammals are the dominant sources, mainly in the form of methylmercury compounds (70–90% of the total)<sup>(2)</sup>

The US Food and Drug Administration (FDA) recently acknowledged several important facts regarding amalgams: (1) they release Hg in the form of Hg vapor; (2) they release amounts of Hg vapor dependent on the number of existing fillings; and (3) they release Hg vapors that may be harmful to certain patients.<sup>(3)</sup>

Amalgam dental fillings with mercury can be a source of human exposure to elemental mercury vapors for many populations. Mercury vapor is released from the surface of amalgam fillings into the mouth and lungs. Depending upon the number of amalgam fillings and other factors, the estimated average daily absorption of mercury vapor from dental fillings

varies between 3 $\mu$ g and 17 $\mu$ g of mercury.<sup>(4)</sup> So ionic mercury leached out from amalgam restorations may present a risk to the dental patient.<sup>(5)</sup>

Mercury can also be released by electrochemical corrosion from amalgam fillings during chewing.<sup>(6)</sup> Additionally, materials used for tooth bleaching can cause the release of mercury from amalgam fillings. According to Bahar et al.,<sup>(7)</sup> the amount of free mercury released from amalgam fillings depends on the content of silver in the alloy. Alloys with a high percentage of silver (69%) release low levels of mercury from the amalgam, while those with a smaller percentage of silver (45%) release more mercury from the amalgam.<sup>(7)</sup>

Although bleaching gels are commonly applied to the anterior teeth, excess bleaching materials may inadvertently come into contact with amalgam restorations on premolars and molars, and may increase the susceptibility of the amalgam to corrosion and degradation. Bleaching agents such as carbamide peroxide break into free radicals that can theoretically corrode metallic alloys, such as amalgam in close proximity, to release mercury.<sup>(8)</sup>

According to Azarsina et al., polished amalgam fillings release less mercury after the application of carbamide peroxide than do unpolished amalgam fillings.<sup>(9)</sup> Another hazard of mercury is that it is a radioactive element that can be quite toxic, even in small doses.<sup>(10,11)</sup>

To determine levels of mercury in the environment and biological samples, various analytical techniques have been used, such as cold vapor atomic absorption spectrometry, cold vapor fluorescence spectrometry, inductively coupled plasma optical emission spectrometry (ICP-OES), electrothermal atomic absorption spectrometry, anodic stripping voltammetry, and cold vapor inductively coupled plasma mass spectrometry.<sup>(12)</sup> The amount of free mercury released from amalgam fillings can also be determined with nuclear tracking techniques.<sup>(13)</sup> The release of mercury from amalgam fillings can be studied with spectroscopy-induced laser femtosecond,<sup>(14)</sup> and the corrosion of amalgam can also be determined by fluorescent spectroscopy.<sup>(15)</sup>

The influence of pH on the release of mercury from amalgam fillings has also been studied. It has been reported that at variable pH values, mercury dissolves more in amalgam fillings that contain tin at all stages of amalgamation, than in amalgam fillings that do not contain tin.<sup>(16)</sup> According to Rotstein et al.,<sup>(17)</sup> one of the factors influencing the release of mercury from unpolished amalgam is the filling pH.

Researchers agree that amalgam restorations leach mercury into the mouth, but consistent findings are not available to determine whether this poses any significant health risk.<sup>(18)</sup> Mercury does not collect irreversibly in human teeth. The average half-life of mercury is 55 days for transport through the body to the point of excretion. Thus, mercury that came into the body years ago may no longer be present in the body.<sup>(14)</sup> The use of mercury in dental fillings represents approximately 10% of the total global mercury consumption; thus, dentistry is the largest consumer of mercury in the world.<sup>(19)</sup>

The purpose of this study was to determine the levels of mercury in hard tissue from dental amalgam fillings under in vitro conditions.

## Materials and Methods

This in vitro research included 30 human teeth that were extracted for various clinical reasons. The teeth were stored in a physiological solution until they were used. The teeth were divided into 3 experimental groups: Group 1 (n=10) – occlusal surface cavity preparation (class I according to Black); Group 2 (n=10) – proximal-occlusal surface cavity preparation (class II); and Group 3 (n=10) – mesio-occlusal-distal [MOD] surface cavity preparation. Each of these groups was divided into 2 subgroups: subgroup 1 (n=5) – amalgam fillings were not polished, and subgroup 2 (n=5) amalgam fillings were polished.

The teeth were filled with amalgam (Dispersalloy, Johnson and Johnson Inc. Montreal, Canada), and those in subgroups 2 were polished after 24 hours.

The amount of mercury released from the amalgam fillings was determined 9 months after the teeth were filled. Before chemical analysis, the teeth were irrigated 4 times over a period of 10 minutes in an ultrasonic bath. The teeth were ground to a particle size of <75 microns with a blinder (Retsch, Grindomix GM 200, Germany). The ground tooth material was dried at a temperature of 105°C for 3 hours. The powder Ber was weighed on a scale with a precision of 4 decimal places (Kern & Sohn GmbH, Germany). From each tooth, 250 mg of the powder was mineralized with royal water (HCl+HNO<sub>3</sub> in a ratio of 1:3) in a microwave oven (Berghof products, Germany), for 54 minutes. After mineralization, the samples were filtered and analyzed with ICP-OES (Perkin Elmer, USA, Optima 2100 DW).

Statistical analysis was performed using statistical software package SPSS version 23.0 (SPSS Inc, Armonk, NY: IBM Corp). For descriptive analysis, results are presented as mean $\pm$ SD. For data with normal distribution, inter-group comparisons were performed using Student's t-test. Multiple comparisons were performed with a one-way ANOVA. A probability value of  $P < 0.05$  was considered statistically significant.

## Results

### Group 1

The average level of mercury in teeth with an occlusal amalgam restoration before the amalgam was polished was (5 teeth, 10 measurements of each tooth from 2 times, n = 10) was 6.53 $\pm$ 0.85 mg/kg (the range of 5.36-8.02). The average level of mercury in teeth with an occlusal amalgam restoration after the amalgam was polished was 5.80 $\pm$ 0.51 mg/kg (the range of 5.12-6.53). The average mercury level after polishing the amalgam filling was smaller, and the difference was significant ( $P=0.032$ ) (Table 1).

### Group 2

The average level of mercury in teeth with a proximal-occlusal amalgam restoration before the amalgam was polished was 10.02 $\pm$ 1.81 mg/kg (the range of 6.72-12.22). The average level of mercury in teeth with a proximal-occlusal amalgam restoration after the amalgam was polished was 8.76 $\pm$ 1.24 mg/kg (the range of 7.74-11.4). The mercury level after polishing the amalgam filling was smaller, but the difference was not significant ( $P=0.086$ ) (Table 1).

**Group 3**

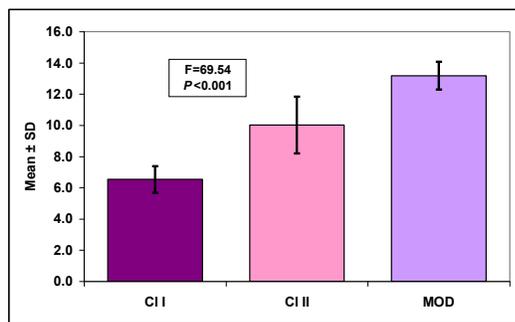
The average level of mercury in teeth with an MOD amalgam restoration before the amalgam was polished was 13.18±0.88 mg/kg (the range of 11.61-14.56). The average level of mercury in teeth with an MOD amalgam restoration after the amalgam was polished was 12.54±1.14 mg/kg (the range of 10.53-14.07). The mercury level after polishing the amalgam filling was smaller, but this difference was not significant ( $P=0.177$ ).

**Table 1.**

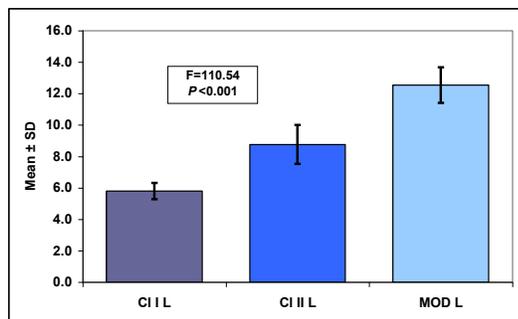
**Mean value (mg/kg) of mercury in the study groups.**

Group	Subgroup	N	Mean ± SD	Rank	Statistics
Group 1	Subgroup 1	10	6.53 ± 0.85	5.36 - 8.02	T=2.329 P=0.032
	Subgroup 2	10	5.80 ± 0.51	5.12 - 6.53	
Group 2	Subgroup 1	10	10.02 ± 1.81	6.72 - 12.22	T=1.816 P=0.086
	Subgroup 2	10	8.76 ± 1.24	7.74 - 11.4	
Group 3	Subgroup 1	10	13.18 ± 0.88	11.61 - 14.56	T=1.405 P=0.177
	Subgroup 2	10	12.54 ± 1.14	10.53 - 14.07	
Total		60	9.47 ± 3.00	5.12 - 14.56	

The average mercury levels in the 3 groups revealed significant differences between both the unpolished samples (one-way ANOVA  $F=69.54$ ,  $P<0.001$ ) and the polished samples (one-way ANOVA  $F=110.54$ ,  $P<0.001$ ) (Figures 1 and 2). The teeth with an MOD amalgam restoration were characterized by the highest mercury levels.



**Fig. 1.** The average mercury levels in the study groups (unpolished samples)



**Fig. 2.** The average mercury levels in the study groups (polished samples).

**Discussion**

Exposure to mercury from dental amalgams, with possible adverse health effects, has generally been considered to occur via either erosion or evaporation directly from the surface of fillings, followed by ingestion.<sup>(20)</sup>

Numerous epidemiological studies have assessed the impact of mercury exposure from oral dental amalgam. In a recent study, males with high mercury levels in their hair (>1ppm) had a 50% higher probability of having periodontitis than females with normal mercury levels (<1ppm). The results suggest that mercury exposure, regardless of sex, is associated with periodontitis.<sup>(21)</sup> In 1994, it was shown that the amount of tin in the  $\gamma 1$ -phase is related to the emission of mercury vapor. Based on this paper, it is possible to identify the brands tested: conventional amalgams, amalgams with a reduced amount of  $\gamma 2$ , and non- $\gamma 2$ -amalgams. The result is clear: non- $\gamma 2$ -amalgams emit substantially more mercury vapor than the old conventional amalgams. Using the highest emitter of the low copper amalgams as a baseline, the high copper amalgams emit 3–4 times as much mercury vapor, depending on the brand.<sup>(22)</sup>

In an investigation measuring differences in mercury vapor emissions in corroded and uncorroded samples, only one non- $\gamma 2$ -amalgam and one low copper amalgam were used. The pattern is once again confirmed, with the non- $\gamma 2$ -amalgam emitting substantially more mercury vapor than the conventional one, and corroded samples emitting more mercury vapor than those not corroded.<sup>(23)</sup> In another investigation, using the same brands of amalgam as Mahler et al.,<sup>(24)</sup> Marek studied the average values of mercury solubility in phase  $\gamma 1$  (Ag-Hg) and phase  $\gamma 2$  (Ag-Hg-Sn) with atomic absorption spectroscopy. The results showed that solubility is better in phase  $\gamma 1$  than in phase  $\gamma 2$ . It was noted that the release of mercury from amalgam is smaller in phase  $\gamma 2$  than in phase  $\gamma 1$ .

The release of Hg from amalgam fillings depends on the type and age of the amalgam filling.<sup>(25)</sup> Harris et al.<sup>(20)</sup> determined the migration of Hg, Ca, Zn, and Cu in teeth with amalgam fillings more than 20 years ago. They concluded that Hg (up to approximately 10 mg g(-1)) and Zn (>100 mg g(-1)) were detected in the teeth several millimeters from the location of the amalgams.

The removal of amalgam from the cavity also causes Hg release. The amount of free amalgam released from fillings depends on whether it is removed with water or through absorption. It has been reported that the amount of free mercury is greater (34.0–796 $\mu\text{g}/\text{m}^3$ ) if water or suction is not used during its elimination, compared with the use of water rush (4.09-19.0 796 $\mu\text{g}/\text{m}^3$ ) and the use of suction (14.0-19.0 $\mu\text{g}/\text{m}^3$ ).<sup>(26)</sup>

Derand et al. reported that there is a large difference in mercury levels between polished fillings stored in a room environment and corroded fillings stored in saliva. Additionally, the simple composition frees up more mercury than do fabricated fillings.<sup>(27)</sup>

In our in vitro research, the teeth were stored in saliva, and it was found that the average values of mercury released

from teeth with class I, II, and MOD amalgam restorations were significantly different, similar to the polished and unpolished groups.

Bolsoni et al.<sup>(28)</sup> investigated the corrosion of polished and unpolished amalgam fillings over various time intervals. The corrosion products were measured with ICP-OES. They concluded that the greatest concentration of mercury occurred in the first 24 hours of the experiment, and that the corrosion of the amalgam was caused by the degradation of gamma phase 2 during amalgamation. Our research, in contrast with the findings of Bolson et al., has shown that mercury release is greater 9 months after amalgam placement; this determination of mercury release was made by ICP-OES.

Pleva et al.<sup>(29)</sup> found that the chewing surface of a 5-year-old amalgam had lost almost half of its mercury, while a 20-year-old amalgam had no mercury left on the chewing surface.

More mercury is released after chewing, and newer fillings release more mercury than older fillings.<sup>(30)</sup> The results of a study conducted by Canay et al.<sup>(31)</sup> showed that unpolished amalgam has a higher corrosion level than does polished amalgam.

Bjorkman & Lind<sup>(32)</sup> studied the influence of various factors on the evaporation of Hg from amalgam fillings. They found that rinsing the mouth with warm water for 1 minute increases the level of mercury vapor by a factor of 1.7 when the water temperature rises from 35°C to 45°C.

Loto et al.<sup>(33)</sup> reported greater release of mercury vapor in the environment of the Restorative Clinic than in other dental clinics.

The corrosion behavior and dissolution of such a structure as a set amalgam depend on the characteristics of each individual phase and on the electrochemical interaction between these phases in a special environment, such as the oral cavity during mouthguard bleaching.<sup>(34)</sup>

Fredin et al.<sup>(35)</sup> studied various aspects of mercury release from amalgam fillings under in vivo and in vitro conditions using light microscopy mercury globules. They concluded that amalgam fillings should not be considered as an appropriate filling given the long-lasting exposure to mercury vapors, which have a toxic effect on the human body.

Mercury vapors can cause various diseases in the human body. Anaerobic bacteria from periodontal disease produce H<sub>2</sub>S and CH<sub>3</sub>SH, which are responsible for gingivitis.<sup>(36)</sup>

According to Sibirund et al.,<sup>(37)</sup> people with amalgam fillings showed more health problems than did people without amalgam fillings. These sulfur compounds react with the mercury amalgam to produce a black pigment on the gum tissue (amalgam tattoo), consisting of HgS, which is extremely toxic and can cause oral and systemic diseases.<sup>(38)</sup>

According to Paknahad et al.,<sup>(39)</sup> amalgam fillings in patients who are exposed to Wi-Fi wave emissions release more mercury than those in patients not exposed. Leszek et al.<sup>(40)</sup> reported the deposition of mercury in various parts of the body within 29 days after the placement of amalgam restorations. They concluded that high concentrations of mercury from amalgam fillings are found in the kidneys and liver.

## Conclusion

The teeth with an MOD amalgam restoration are characterized by the highest mercury levels. The more mercury is released from unpolished amalgam fillings than from polished amalgam fillings in teeth with occlusal surface cavity preparation (class I according to Black).

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## Competing Interests

The authors declare that they have no competing interests.

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\*Corresponding author: Prof. Violeta Vula. Department of Dental Pathology and Endodontics, Faculty of Medicine, University of Prishtina, Clinical Centre N.N. Prishtina, Kosovo. E-mail: [violeta.vula@uni-pr.edu](mailto:violeta.vula@uni-pr.edu)

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