J Mater Sci: Mater Med (2006) 17: 301–305
DOI 10.1007/s10856-006-8225-y

Ion release from gold/platinum dental alloy: could release of other elements be accountable in the contact allergy attributed to the gold?

A. Čelebić · M. Baučić · J. Stipetić · I. Baučić · S. Miko · B. Momčilović

Received: 14 December 2004 / Accepted: 16 June 2005
© Springer Science + Business Media, Inc. 2006

Abstract
Objectives: The release of metal ions (Al, Ag, Au, Ca, Cd, Co, Cr, Cu, Mg, Mo, Ni, Pd, Pt, Ti, and Zn) from the commercial gold/platinum (Au/Pt) dental alloy of declared composition was studied.
Methods: Au/Pt was soaked in pH 6.0 phosphate buffer, 3.5 pH phosphate buffer and pH 3.5 mixture of lactic, formic and acetic acid, and incubated at 37 °C for 1, 2, 3, 4, 5, 6, 7, 14, 21, and 30 days. Six samples (n = 6) of every solution were prepared for any time period. Inductively coupled plasma atomic emission spectroscopy was used for analysis of the released elements.
Results: Results demonstrated release of only Cr, Cu, Fe, and Zn from the tested Au/Pt dental alloy (ANOVA, $p < 0.001$ for buffer, time, and interaction, respectively); however, only Cu and Zn were declared.
Conclusions: The undeclared chromium from Au/Pt dental alloy, or some other element might be responsible for the contact allergy thus far attributed to the gold.

A. Čelebić · M. Baučić · J. Stipetić · I. Baučić · S. Miko · B. Momčilović
School of Dental Medicine, University of Zagreb, Gundulićeva 5, Zagreb, Croatia
e-mail: celebic@sfzg.hr
Tel.: (+3851) 4802125
Fax: (+3851) 4802149

S. Miko
Institute for Geology, University of Zagreb, Sachsova 2, Zagreb, Croatia

B. Momčilović
Institute for Medical Research and Occupational Health, POB 291, Zagreb, Croatia

1. Introduction

High corrosion resistant gold/platinum (Au/Pt) dental alloys are widely used in dental fillings, crowns, and bridges to withstand with the conditions prevailing in the oral cavity [1, 2]. The materials employed in the mouth must be completely tarnish-resistant, they must not react with many alkaline and acid foods, and they cannot be affected by mouth fluids. Indeed, the electrochemical conditions created in oral cavity promote the release of dental alloy metal ions into saliva. Equally, organic and/or inorganic acids from food decrease the local pH and promote dental alloy ion release, as do the low pH values produced by dental plaque [3, 4]. Release of elements is in correlation with adverse effects, such as cytotoxicity [3–6].

Gold is chemically a noble metal and is a valuable constituent of dental alloys, however, it is also considered to be an important etiological factor for the induction of contact allergy [7]. The amount of gold released from an Au/Pt alloy in the oral cavity and subsequently absorbed in the gastrointestinal tract is very small and proportional to the exposed gold surface area in the mouth [8]. Previous *in vitro* experiments have showed that leaching of metal ions from dental alloys depended upon the chemical composition, nature, and acidity of the medium to which the alloy is exposed [8–11].

2. Aim of the study

The aim of this experiment was to "fingerprint" the spectrum of metals leached from an Au/Pt dental alloy of a known declared composition, and under the different conditions of acidity that may be encountered in the oral cavity.

3. Materials and methods

3.1. Clean laboratory

All the samples were prepared and analyzed in the clean laboratory with an air work bench at the Institute for Geology, University of Zagreb.

3.2. Chemicals

All the chemicals used for the analytical ICP-AES work were of the highest grade analytical quality (Suprapur, Merck, Darmstadt, Germany).

3.3. Glassware washing

All the glassware used in the study was soaked in 10% (v/v) nitric acid (Suprapur, Merck, Darmstadt. Germany) for 24 hours and thereafter rinsed five times in the re-distilled deionised water (DDW).

3.4. Dental alloy

Commercially available gold/platinum dental alloy (18 + 8, Noble Metal Refinery, Zagreb, Croatia) came as a standard 8.0 × 6.5 × 1.0 mm leaf with a 133 mm^2 effective exposure surface. The declared metal composition of the alloy is showed in Table 1. To preclude the bacterial growth contamination, the Au/Pt leafs were washed with Alconox soap (Alconox Inc, Alconox, NY), rinsed with the DDW, further soaked for 20 min in alcohol, again rinsed twice with DDW in a laminar flow hood, and dried at 60 °C for 24 h.

3.5. Immersion (extraction) solutions

We prepared three different immersion solutions for metal extraction: (1) pH 6.0 phosphate buffer, to resemble pH of saliva (*saliva)* [12]; (2) pH 3.5 phosphate buffer to resemble extreme conditions (*acid)*, and (3) pH 3.5 mixture of 0.1 M lactic acid, 0.1 M sodium chloride, 0.1% acetic acid, and 1.0% formic acid; (*pro analysis,* Kemika, Zagreb, Croatia) to resemble conditions under dentobacterial plaque (*plaque)*. Natural fresh saliva has a pH 5.7–7.0. Bacteria in the active dental plaque generate a powerful pH 4, or even lower, tooth mineral destroying mixture of lactic, formic, acetic and other metabolic acids [13].

Table 1 The declared and detected Au/Pt dental alloy composition

Metal	Declared (%)	Detected
Gold (Au)	75.0	Not detected (ND)
Platinum (Pt)	8.0	ND
Silver (Ag)	9.5	ND
Copper(Cu)	5.1	Detected
Brass (Cu+Zn)	1.5	Detected
Zinc (Zn)	Declared in brass	Detected
Other elements	0.9	–
Iron (Fe)	Undeclared	Detected
Chromium (Cr)	Undeclared	Detected

3.6. Metal soaking

Individual 15 ml glass tubes with plastic stopper (culture tubes with GL thread, AR-Glass®, Brand Gmbh, Wertheim, Germany) were used. Six replicates of the Au/Pt leafs were immersed in 7 ml of *saliva*, *acid,* and *plaque* extraction solution, respectively, and incubated at 37 °C for 1, 2, 3, 4, 5, 6, 7, 14, 21, or 30 days.

3.7. Metal analysis

The amount of metal leached into the immersion solutions was assessed with the inductively coupled plasma atomic emission spectrometer (ICP-AES, Jobin Yvon 50 P, Horiba group, Longjunean, France). We used certified multielement stock standards for the quality control and to avoid matrix induced interference (Spex, Metushem, NY, USA). The multielement calibration standards of 0, 1, 5, 10, 25, 50, 500, 1000 and 5000 μg/L in every immersion (extraction) solution were prepared from the above stock standard (100 mg/L).

3.8. Reagent blanks

The blank immersion solutions of *saliva*, *acid*, and *plaque* for all the analyzed elements were prepared in the same glassware, kept at the same temperature for the same period of time as were the immersion solutions in which the Au/Pt dental alloy were soaked. No metal was detected in the reagent blanks.

3.9. Detection limits

The detection limits were determined from the matrix-matched multielement calibration standards prepared from the certified stock standard (see above). The accurate detection limits for Al, Ca, Cd, Co, Cr, Cu, Mg, Ni, V, Ti, and Zn were 10 μg/L, and that for Ag, Au, Mo, Pd, and Pt were 100 μg/L. Some signal may be percepted even below the already stated accurate detection level (perceptible).

3.10. Statistics

The results were expressed as a Mean ± standard error (SE). The overall difference between the bufferes and exposure time was assessed with ANOVA, whereas the pairwise

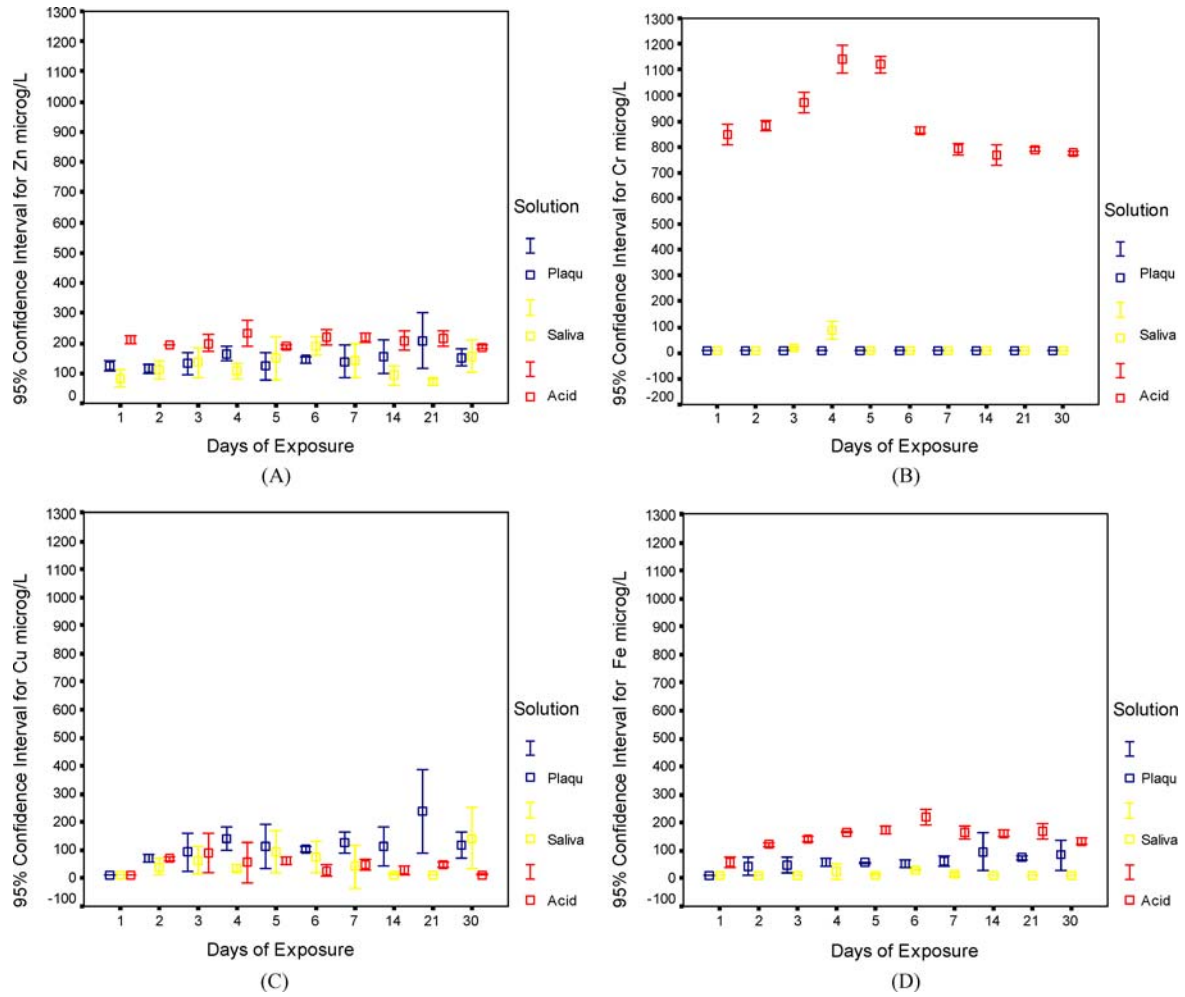

Fig. 1 A–*D* Legend: 95% confidence intervals of Zn, Cu, Fe, and Cr ions released from the Au/Pt dental alloy immersed into the solution of *saliva*, *acid*, and *plaque* over the period of thirty days. (see Materials and methods for the details). 1A. 95% confidence intervals of Zn; 1B 95% confidence intervals of Cr; 1C 95% confidence intervals of Cu; 1D 95% confidence intervals of Fe

comparisons were made using Scheffe post-hoc with the significance set at $p < 0.05$ [14]. All analyses were run on SAS version 6.11 (SAS Institute Inc., Cary, NC, USA).

4. Results

Most of the principal metal constituents declared to be present in the Au/Pt dental alloy did not leach into either of the immersion solutions whereas, some of the undeclared metals did leach in the amounts well above their detection limit (Table 1). Thus, copper, zinc, iron and chromium leached from the Au/Pt alloy, whereas no traces of gold, platinum, or silver were detected, regardless of how long the alloy was immersed in either of the three extraction solutions. Some zinc might have been expected due to the declared presence of brass, an alloy of copper and zinc, but the presence of iron, and especially chromium, could not be envisaged from the declared Au/Pt alloy composition.

As evident from the Fig. 1, the within sample variability of results over the time line of 30 days could be quite remarkable. Zinc and copper could be accurately quantified in all the immersion solutions since day 1 and day 2, respectively; the copper signal during day 1 was perceptible at the low limit of detection. Iron became perceptible in both acid and plaque immersion solution already on day 1, but fully detectable (precisely quantified) on day 2 and thereafter. However, *in saliva* iron built up from a barely perceptible level at day 1 to a quantifiable level by day 4, then remained well detectable through day 14 to fall down again to a barely perceptible level on days 21 and 30, respectively. Chromium, on the other hand, would stay at the low level of detection limit in the *plaque* solution during the entire immersion period of 30 days, it was well detected in only a few samples on days 3 and 4 in *saliva* and thereafter only perceptible for the rest of the immersion periods, but it was profusely abundant in *acid* ever since the day 1. In some cases the concentration of metal in the immersion solution would slowly go up for

Table 2 Cumulative (all time interval) *in vitro* release of zinc, copper, iron, and chromium from the Au/Pt dental alloy

Element	Electropotential (V_0 mV)	Saliva[A] (μg/L)	Acid[B] (mg/L)	Plaque[C] (mg/L)
Zn	−0.76	$124 \pm 6.63^{a‡}$	207 ± 3.27^{c}	146 ± 6.02^{b}
Cu	+0.34	53.1 ± 8.15^{a}	45.1 ± 4.93^{a}	113.1 ± 10.2^{b}
Fe	−0.44	14.6 ± 1.43^{a}	150 ± 5.54^{c}	58.5 ± 4.64^{b}
Cr	−0.76	18.3 ± 3.22^{b}	895 ± 7.4^{c}	$\leq 10.0^{a}$

[†]H/H = 0.00 mV.[‡]All the samples showed some perceptable presence of the metal;

[A,B,C]Columns bearing different superscripts differ significantly (ANOVA, $p < 0.001$),

[a,b,c]Means bearing different superscript in the same Zn, Cu, Fe, and Cr row differ significantly (Scheffe's post-hoc test, $p < 0.05$).

the first few days, only to drop down almost to the day 1 level thereafter; presumably due to the surface re-adsorption of the already leached metal and/or the unhomogeniety of the analyzed alloy sample. In summary, there were no general rule on how the particular metal will respond, as their release into the solution depended both upon the nature of the solution and the exposure time. Indeed, leaching of Zn, Cu, Fe and Cr, in the solution was dependent upon the nature of the immersion solution and duration of the immersion. Phosphate acid at a low of 3.5 pH would extract considerable amount of chromium, iron and zinc, while organic acid (plaque) at pH 3.5 would extract considerable amounts of copper. There was a strong interaction between these variables (ANOVA, $p < 0.0001$ for the immersion solution, soaking time, and interaction for the every detected metal, respectively).

The sum of all the results for the same element and the same immersion solution regardless of the immersion time is shown in Table 2. Evidently, zinc, iron and chromium leached the most when in the a*cid* (phosphate buffer pH 3.5) whereas copper leached the most when under the p*laque* conditions (organic acid mixture, pH 3.5). In contrast, zinc and iron leached the least in the s*aliva* (phosphate buffer, pH 6.0); copper in the *acid* (pH 3.5 phosphate buffer), and chromium leach out in the *plaque* immersion solution. The presence of undeclared electronegative zinc, iron, and chromium in the presence of the electropositive copper would generate a high electrical potential within the oral cavity, promote the corrosion of the Au/Pt dental alloy and increase the leach out of the metals.

5. Discussion

The principal finding of our study is the presence of undeclared chromium in the Au/Pt dental alloy. Chromium was released even under the normal pH conditions (phosphate buffer, pH 6.0). It may be argued that the detection limit of 10 μg/L for chromium is much lower then that of 100 μg/L for gold, so that the appreciable amount of gold might be present undetected under the normal pH 6.0 conditions in the oral cavity. However, the amount of chromium released in the *acid* (pH 3.5 phosphate buffer) was almost 900 μg/L, implying that the maximum undetected concentration of gold could be under 100 μg/L in the worst possible case and what is certainly far below of the already presented amount of chromium. Our observation is indirectly supported by Liden *et al*. [15] who also did not found any gold release from 13 different gold-containing jewellery materials by using both flameless AAS and ICP-MS; the later is much more sensitive instrument then our ICP-AES.

Contact allergy to metals is a fascinating subject as many of the trace elements, including chromium, are at the same time allergenic and essential for the human health and well being [16]. Cronin [17] in his seminal book on contact dermatitis mentioned only four cases of contact dermatitis which, perhaps, may be attributed to the gold. Only one of them was presented in some detail and he was allergic to both gold and chromium (+++ patch test) but, unfortunately, the author did not proceed further with the differential diagnosis to elucidate that confounding evidence. Recently, Issakson and Bruze [18] summarized the problems accompanying the preparation of adequate gold patch standard material for assessing sensitivity to the gold, but did not consider the possibility that gold may be contaminated with some other, more common, allergenic material. Most recently, Muller [7] reviewed the subject and suggested that contact allergy to dental gold alloy may be as high as 15%; the same to the highly allergenic nickel. Chromium was neither mentioned, nor tested, in their gold sensitive patients. The uncertainty about the gold allergy may reach its peak in just published paper by Nonaka *et al*. [19] stating that "gold allergy has changed from being clinically overt to becoming occult in Japan".

In contrast to the prevailing opinion, the results of our study demonstrated that the presence of highly allergenic, but undeclared chromium from an Au/Pt dental alloy, is much more likely to be the principal culprit in the induction of contact allergy thus far attributed to the gold.

Wataha and his group [8–11] studied the release of metal ions from different Au/Pt dental alloys with atomic absorption spectrophotometry (AAS) under even more severe corrosion conditions than reported here. However, they accepted the declared dental alloy composition face value and did

not "fingerprint" their solutions for chromium or any other metal/s; indeed, when performing an AAS analysis it is necessary to use a special lamp for every element you may have anticipated. The allergenic potential of Cr [20] and, in the decreasing order that of Ni and Co, was shown to exceed by far the allergogenic potential of gold [21].

Indeed, an oxide protective layer on the surface of the dental alloy will be only re-exposed to the surrounding saliva after every tooth brushing and chewing of the food and thus provide conditions for the new release [22, 23]. The difference in the electrochemical potential of the metals within the alloy may only help in the further life shortening of the custom made prosthodontic appliance. Technological procedures can also act undesirably on the crystal structure of the alloy, or surface may not be properly polished, etc. In 1984 the ADA workshop on the biocompatibility of metals in dentistry stated that sensitivity to chromium results from contact with chromate salts, which result from the corrosion of such alloys [24].

Since Zn, Cu, Fe and Cr leach differently in different immersion (extraction) solution, their relative oral cytotoxicity (Zn > Cu > Fe > Cr) [6, 25, 26] may also change relative to the changes in their mutual proportions.

Fortunately, all four detected elements, Zn, Cu, Fe, and Cr, are essential nutrients and, more often then not, lacking in the diet of the older people, [27] who usually wear such prosthetic appliance. However, the safe limit for dietary chromium is set at 250 μg per day [28], so that fixed partial dentures should be always assessed for the unadwarent toxicity and relative to the other associated metal contaminants of the food. Today, knowledge of biocompatibility of different metals in numerous dental alloys available is fundamentally important to ensure the health of patients, dental staff members, and practitioners [29].

Further 'fingerprinting" investigations are necessary to assess which elements and how much of them may be released from an Au/Pt alloy from different manufacturers before any contact allergy may be clearly attributed to gold in the presence of other metal ions.

6. Conclusion

"Fingerprinting" of metal ions released from each alloy should be helpful in all cases of possible contact allergies.

Acknowledgements This work was supported by the grants 0065014 and 0022013 from the Croatian Ministry of Sciences and Technology, Zagreb, CROATIA.

References

1. R. D. HARBISON, in: "Platinum, in Hamilton's and Hardy's industrial toxicology" (Mosby, Sant Louis, 1998) p. 167.
2. R. V. LEA, in: "Gold, in Hamilton's and Hardy's industrial toxicology" (Mosby, Sant Louis, 1998) p. 148.
3. J. C. WATAHA, *J. Prosth. Dent*. **83** (2000) 223.
4. P. R. MEZGER, A. L. H. STOLS, M. VRIJHOEF and E. H. GREENER, *Dent. Mater.* **5** (1989) 350.
5. J. C. WATAHA, C. T. HANKS and Z. SUN, *Dent. Mater.* **10** (1994) 156.
6. A. YAMAMOTO, R. HONMA and R. SUMITA, *J. Biomed. Mater. Res.* **39** (1998) 331.
7. H. MULLER, *Contact Dermatitis* **47** (2002) 63.
8. J. C. WATAHA, R. G. CRAIG and C. T. HANKS, *J. Dent. Res.* **70** (1991) 1014.
9. J. C. WATAHA and P. E. LOCKWOOD, *Dent. Mater.* **14** (1998) 158.
10. J. C. WATAHA, P. E. LOCKWOOD, S. S. KHAJOTIA and R. TURNER, *J. Prosth. Dent.* **80** (1998) 691.
11. J. C. WATAHA, P. E. LOCKWOOD and S. K. NELSON, *J. Oral Rehab.* **26** (1999) 798.
12. I. W. SCOPP, in: "Oral medicine. A clinical approach with basic science correlation" (The CV Mosby Co., St Louis, 1969) p. 54.
13. A. AMADAL-SCHEIE, W. M. LUAN, G. DAHLEN and O. FEJERSKOV, *J. Dent. Res.* **75** (1996) 1901.
14. G. W. SNEDECOR and W. G. COCHRAN, in: "Statistical methods" (Iowa State University Press, Ames, 1980).
15. C. LINDEN, M. NORDENADLER and L. SKARE. Contact Dermatitis 41 (1998) 57.
16. B. MOMČILOVIĆ B, in "Trace Elements in Man and Animals 6" (Plenum Press, New York, 1987) p. 173.
17. E. CRONIN, in "Metals in Contact Dermatitis" (Churchill and Livingstone, Edinburgh, 1980) p. 270.
18. M. ISAKSSON and M. BRUZE, in "Handbook of Dermatology" (Springer, 2000), p. 544.
19. H. NONAKA, T. NAKADA and M. IIJIMA. *Contact Dermatitis* **48** (2003) 112.
20. D. BASKETTER, L. HOREV, D. SLODOVNIK, S. MERIMES, A. TRATTNER and A. INGBER, *Contact Dermatitis* **44** (2001) 70.
21. A. DOOMS-GOOSSENS, A. CEUTERICK, N. VANMAELE and H. DEGREEF, *Dermatologica* **160** (1980) 249.
22. J. C. WATAHA, R. G. CRAIG and C. T. HANKS, *J. Dent. Res.* **71** (1992) 1417.
23. J. C. WATAHA, P. E. LOCKWOOD, M. NODA, S. K. NELSON and D. J. METTENBURG, *J. Prosth. Dent.* **87** (2002) 94.
24. WORKSHOP, *J. Am. Dent. Assoc.* **109** (1984) 469.
25. M. P. SAUVANT, D. PEPIN, J. BOHATIER, C. A. GROLIERE and J. GUILLOT, *Ecotox. Environ. Safety* **37** (1997) 131.
26. B CZARNECKA, H. LIMANOWSKA-SHAW and J. W. NICHOLSON, *J. Mater. Sci. Mater. Med.* **14** (2003): 601.
27. W.H.O. in "Environmental Health Criteria" (World Health Organization, Geneva, Switzerland, 2002) p. 208.
28. C. REILLY, in: "Metal contaminants of food. Its significance for food quality and human health" (Blackwell Science Ltd, Oxford, 2002) p. 135.
29. J. C. WATAHA, *J. Prosth. Dent.* **86** (2001) 203.