STUDY ON CORROSION PRODUCTS FROM EAR PIERCING STUDS

Sizue O. Rogero, Isolda Costa and Mitiko Saiki

Instituto de Pesquisas Energéticas e Nucleares - IPEN-CNEN/SP Caixa Postal 11049- CEP 05422-970, São Paulo, Brasil sorogero@net.ipen.br

ABSTRACT

In this work instrumental neutron activation analysis was applied to analyse elemental composition of commercial gold coated ear piercing substrate and metals present in their corrosion products. The cytotoxic effect was also verified in these corrosion product extracts, probably, due to the lixiviation of Ni present in high quantity in their substrates. The analysis of gold coated ear piercings surfaces by scanning electron microsocpy before and after the corrosion test showed coating defects and the occurrence of corrosion process.

I. INTRODUCTION

Gold coated ear piercing studs have been found to cause contact dermatitis. After perforation of earlobes these studs are kept for at least ten days for cicatrisation and during this period they are closely in contact with body fluids and this might elicit allergic/inflammatory reactions. Consequently the nickel/cobalt sensitization has increased as the habit of piercing the earlobes has extended in popularity[1], particularly in male people.

Stainless steels, specially biomedical AISI 316L stainless steel, are widely used as substrate of ear piercing demonstrated studs because they have good biocompatibility, proper physical and mechanical properties and besides they may be molded into a variety of shapes and sizes. One of the main limitations to their clinical use is the tendency to corrode in the presence of chloride ions, considering that they contain non negligible amounts of chromium (16-18 wt %) and nickel (10-14 wt %). However they still keep an important place in the manufacture of gold coated studs, as gold has been generally accepted as having little or no toxicity[2].

Therefore, the coating should be as defectless as possible and its adherence to the substrate should be appropriate, to avoid the contact between substrate elements and body fluids while the pierced earlobes are healing. The presence of defects in gold coatings, particularly the kinds used for ear piercing, allows the contact between the stud substrate and body fluids. A corrosion reaction might occur as a consequence of this interaction, causing the release of metal ions. Metal ions then bind to tissue and interstitial fluid proteins as soon as they are released from the metallic substrate. Nickel is the major cause of allergic contact dermatitis, this being caused by Ni^{2+} ions, which bind a carrier protein and this nickel-protein complex activates immune reactions[3].

In this study, the corrosion products and the cytotoxicity of commercial gold coated ear piercing studs, with a copper-zinc based alloy and a stainless steel as substrates have been investigated.

II. EXPERIMENTAL

Material. The material studied consisted of two types of commercial gold coated ear piercing studs: St (medical grade stainless steel, Ni free) and Pf (hypoallergenic). In the manufacture gold is deposited in the surface of metallic substrate by an electrolytic process.

Preparation of extract[4]. Twelve pairs of studs were placed in a 120 mL capacity screw capped glass bottle and sterilized by autoclaving at 120 °C for 20 minutes. Subsequently, 60 mL of MEM-FCS [Minimum Eagle's Medium supplemented with 10% fetal calf serum and 1% penicillin-streptomycin solution] medium culture were added. The bottle was incubated at 37 °C for 10 days. When the time given for extraction was over, part of the culture medium was taken for chemical analysis and another part for cytotoxicity assay.

Scanning electron microscopy (SEM). The surface characteristics of the studs were analyzed by scanning electron microscopy (SEM) before and after 10 days immersion in the culture medium. The aim of these analyses was to check the presence of defects which could expose the substrate.

Chemical analysis. The substrate of gold coated ear piercing studs and the extract obtained after immersion of studs in culture medium were analyzed by applying instrumental neutron activation analysis (INAA), described in a previous paper [5]. The gold free substrates were cut in small chips and sample with a mass varying from 15 to 35mg was irradiated for 1h under thermal neutron flux of 10^{12} n cm⁻² s⁻¹ in IEA-R1m nuclear reactor.

Aliquots of the extracts were dried in a polyethylene capsules and irradiated together with elemental standards under a thermal neutron flux of 10^{13} n cm⁻² s⁻¹ for 16h. For gamma ray measurements an hyperpure Ge detector was used and the concentrations of the elements were calculated by comparative method.

Cytotoxicity test. The cytotoxicity assay was carried out with Chinese Hamster Ovary Cells culture (ATCC CHO K1), according to ISO[4] and described in a previous paper[6]. To a CHO cells culture was added serially diluted extracts from the gold coated studs with MEM-FCS. After incubation time the CHO cell colonies were fixed and stained with Giemsa. The number of visible colonies on each Petri dish was counted and compared with the number of colonies in CHO control dish. Phenol solution and titanium extract were used as positive and negative control, respectively.

III. RESULTS AND DISCUSSION

The result of SEM of surface area showed defective regions (Fig.1).

SEM of surface area of studs after immersion in culture medium(MEM-FCS) for 10 days showed presence of corroded areas (Fig.2). If the defects were in the studs place which stay in contact with the pierced earlobes the body fluid could provoke corrosion as culture medium did

The chemical analysis of commercial gold coated substrates showed that there are different kinds of metallic substrates used. Pf have a copper-zinc alloy and St a stainless steel, as indicated in Table 1.



Figure 1. SEM of Surface Area of Commercial Gold Coated Ear Piercing Studs Before Immersion in Cell Culture Medium.



Figure 2. SEM of Surface Area of Commercial Gold Coated Ear Piercing Studs After Immersion in Cell Culture Medium for 10 Days.

TABLE 1. Elemental Composition of Ear Piercing StudsObtained by INAA.

Element	Pf studs substrate	St studs substrate
As µg g ⁻¹	8.1 ± 0.5	57.4 ± 0.8
$Co \mu g g^{-1}$	27.4 ± 0.4	2203 ± 11
Cr %	0.0070 ± 0.0005	16.1 ± 0.2
Cu %	36.5 ± 1.2	0.35 ± 0.01
Fe %	9.0 ± 0.2	67.9 ± 0.2
Mn %	2.34 ± 0.07	1.81 ± 0.02
Mo %	$\leq 0.2(*)$	0.394 ± 0.002
Ni %	6.80 ± 0.07	7.86 ± 0.07
Ti %	≤ 23	≤23
V µg g ⁻¹	≤77	987 ± 26
Zn %	36.4 ± 3.3	≤ 0.7

* - For the elements not detected, the detection limit values were evaluated according to Currie[7].

The results of the elements determined in the extract of studs in culture medium and in the blank solution composed of the same culture medium are presented in Table 2. In the chemical analysis by INAA, concentrations of Co and Cr found in the extract were in the same magnitude of those obtained in the blank. Results obtained for Fe in St, Zn in Pf and Ni in both St and Pf indicated that these metals were released from the gold coated studs to the culture medium, consequence of corrosion process.

TABLE 2. Elemental Concentrations in Extract of CellCulture Medium and in the Blank, by INAA.

Element		Extract after immersion	
concentration	Blank	Pf	St
Co ng/mL	12.1 ± 1.2	61.6 ± 0.5	99 ± 10
Cr µg/mL	0.72 ± 0.02	0.73 ± 0.04	0.73 ± 0.03
Fe µg/mL	0.60 ± 0.09	0.61 ± 0.08	4.03 ± 0.31
Ni µg/mL	N.D.*	0.96 ± 0.09	0.66 ± 0.06
Zn µg/mL	0.58 ± 0.05	3.84 ± 0.41	0.61 ± 0.01

* N.D. - not detected

The determination of cytotoxicity was performed by quantitative evaluation, based on the cell viability by colony formation assay.



Figure 3. Colony Suppression Curves of Ear Piercing Studs

The cytotoxic potential was evaluated by cytotoxicity index (IC_{50(%)}) plotting the percentage of the colony number and the concentration of the extract on a graphic. IC_{50(%)} is the concentration of the extract which suppress colony formation to 50% of the control value. The negative control should not present any effect as observed with titanium (IC_{50(%)} > 100), and the positive control should present cytotoxic effect, as phenol solution (IC_{50(%)} = 32), as shown in Figure 3. Commercial gold coated ear piercing studs with stainless steel and copperzinc alloy substrates, showed to be toxic in the cytotoxicity assay, presenting IC_{50(%)} = 78 and 44, respectively.

The cytotoxic effect was probably due to high concentration of nickel in the extract, as nickel itself is known to have toxic effects in cell cultures and in tissues[8]. Copper could also be responsible for the cytotoxicity found, since a straight correlation between the amount of copper found in the culture medium and cytotoxicity, has been reported in the literature[9,10]. In this study, copper was not analyzed by INAA, due to the interference of Na present in high concentration in these samples. A high activity of ²⁴Na masks the photopeak of ⁶⁴Cu in the gamma spectrum.

IV. CONCLUSION

The presence of defects in the surface of gold coated ear piercing studs exposes the substrate to body fluids leading the release of metals and consequently allergic/toxic reactions.

ACKNOWLEDGEMENTS

The authors are grateful to Volkmar Ett for the valuable information granted and to José Severo Ramos for providing the commercial gold coated ear piercing studs.

REFERENCES

[1] Meijer, C., Bredberg, M., Fischer, T. and Vidströ, L., Ear piercing and nickel and cobalt sensitization in 520 young Swedish men doing compulsory military service. Contact Dermatitis,vol.32, p. 147-149, 1995.

[2] Wataha, J.C., Craig, R.G., Hanks, C.T., The release of elements of dental casting alloys into cell-culture medium. J.Dent.Res., vol.70(6), p. 1014-1018, 1991.

[3] J. Ryhänen; E. Niemi; W. Serlo; E. Niemelä; P. Sandivik; H. Pernu and T. Salo, **Biocompatibility of nickel-titanium shape memory metal and its corrosion behavior in human cell cultures.** J. Biomed. Mater.Res., vol. 35, p. 451-457, 1997.

[4] ISO document 10993, Biological evaluation of medical devices - Part 5 - Tests for cytotoxicity: *in vitro* methods, 1992.

[5] Saiki, M., Rogero, S.O., Correa, O.V., Costa, I., Higa, O.Z., Neutron activation analysis of corrosion products from gold coated ear piercing studs. Rad.Phys.and Chem., vol. 55, p. 753-756, 1999.

[6] Rogero, S.O., Braga, F.J.C. and Higa, O.Z., **Cytotoxicity test for bioceramics of calcium phosphate.** Mater.Sci.Forum, vol. 299-300, p. 44-47, 1999.

[7] L.A. Currie, Limits for qualitative detection and quantitative determination. Anal. Chem., vol. 40, p. 586-593, 1968.

[8] K. Bordji; J.Y. Jouzeau; D. Mainard; E. Payan; J.P. Delagoutte and P. Netter, Evaluation of the effect of three surface treatments on the biocompatibility of **316L stainless steel using human differentiated cells.** Biomaterials, vol.17, p. 491-495, 1996.

[9] Wright, D.C. and Gallant, F.; **Correlation of corrosion behavior and cytotoxicity in Au-Cu-Ag ternary alloys.** J. Biomed. Mater. Res.,vol.16(4), p. 509-517, 1982.

[10] Craig, R.G. and Hanks, C.T., Cytotoxicity of experimental casting alloys evaluated by cell culture tests. J. Dent. Res., vol. 69 (8), p. 1539-1542, 1990.