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J DENT RES 1995 74: 1521
DOI: 10.1177/00220345950740081401

J Dent Res 74(8): 1521-1527, August, 1995

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Abstract. Concerns exist over the potential release of elevated levels of metal ions such as Ni and Be from Ni-Cr dental casting alloys, due to their susceptibility to accelerated corrosion. In this investigation, we evaluated the release of metal ions from four commercial Ni-Cr alloys, representing a range of compositions, in three-day cell culture tests. Metal ion release, as measured by atomic absorption spectroscopy, was correlated to changes in cellular morphology, viability, and proliferation. The results showed that the test alloys and their corrosion products did not affect cellular morphology or viabilities, but did decrease cellular proliferation. The types and amounts of metal ions released, which corresponded to the alloys' reported surface and corrosion properties, also correlated to observed decreases in cellular proliferation after 72 h. Neptune, which caused the smallest decrease in cellular proliferation as compared with control cells, released the lowest amount of corrosion products, due to its corrosion-resistant, high-Cr-Mo-containing, homogeneous surface oxide. The other test alloys, which were susceptible to accelerated corrosion processes, released higher levels of metal ions that correlated to larger decreases in thymidine incorporation. Metal ion levels increased with test time for all alloys but were not proportional to bulk alloy compositions. Ni ions were released at slightly higher than bulk alloy compositions, while Be was released at from four to six times that of bulk alloy compositions. The elevated release of Be ions was associated with reduced cellular proliferation. Other alloying elements were released at levels similar to or lower than bulk levels. Further research is needed to evaluate possible synergistic effects of released metal ions, especially Ni and Be ions, on cellular activities and functions.

Key words: dental alloys, Ni-Cr alloys, cell culture, fibroblasts, toxicity.

Received December 10, 1993; Accepted April 27, 1995

Introduction

Nickel-based dental casting alloys account for approximately 80% of the prosthodontic restorations used clinically (Morris, 1987). Few cases have been reported indicating adverse side-effects (Moffa, 1982; Eichner, 1983; Hensten-Pettersen, 1992; Morris *et al.*, 1992). Problems reported, though, were related to metal hypersensitivity reactions and were poorly documented. Given the wide range of alloy compositions, corrosion properties, and the concern over potential adverse side-effects, data on the cellular response to the amounts and types of metal ions released are needed.

In vitro cell culture tests have evaluated the cellular response to nickel-chromium dental casting alloys, their constituents, and leachable components. Various investigators have used metal salt solutions to evaluate morphology, viability, proliferation, protein production, sister chromatid exchanges, colony-forming ability, hemolysis of human red blood cells, and the effects on bacterial growth (Rae, 1975, 1978; Bearden and Cooke, 1980; Sen and Costa, 1986; Joshi and Eley, 1988; Wataha *et al.*, 1991a,b). These investigations found that nickel salt solutions were more toxic to the cells than chromium salt solutions. It was also shown that nickel ions may be taken up by fibroblasts *in vitro* and bound to the nuclei, cytosol, mitochondria, and microsomes (Hensten-Pettersen, 1984; Wataha *et al.*, 1993).

When particulates, discs, or thin sheets of pure nickel and chromium were used in cell cultures, nickel was found to interfere with various enzyme systems, disrupt intracellular organelles, alter morphology, decrease cell numbers, and increase hemolysis to a significantly greater extent than chromium (Rae, 1975, 1978; Jacobsen, 1977; Kawahara, 1983; Evans and Thomas, 1986; Craig and Hanks, 1990). Evaluations of solid samples of nickel-chromium alloys revealed a very low cytotoxic response of the cultured cells through morphological and ultrastructural evaluations, viability, synthesis of various

Table 1. Chemical compositions of alloys in weight percent[a]

Alloy	Ni	Cr	Mo	Fe	Be	Other[e]
Neptune[b]	63.36	20.95	8.40	1.73	—	4.1 Nb, Al, Si, Mn, Ti
Rexalloy[b]	67.21	12.88	6.76	5.18	—	7.04 Ga, Si, Mn, Co
Regalloy T[c]	71.20	15.89	4.50	0.10	0.57	3.31 Al, Si, 4.28 Mn
Vera Bond[d]	77.36	12.27	4.84	0.14	1.67	2.76 Al, Co, Ti, Si

[a] Chicago Spectr Service Laboratory, Inc., Chicago, IL, USA.
[b] Jeneric/Pentron, Inc., Wallingford, CT, USA.
[c] Dentsply International, Inc., York, PA, USA.
[d] Aalba-Dent, Inc., Cordelia, CA, USA.
[e] Less than 1.0% unless otherwise noted.

proteins, and proliferation (Kawahara *et al.*, 1968; Woody *et al.*, 1977; Hensten-Pettersen and Jacobsen, 1978; Kawahara, 1983; Exbrayat *et al.*, 1987; Craig and Hanks, 1988, 1990). The authors attributed the significant reduction in cytotoxicity of the nickel in the nickel-chromium alloys as well as the relative inertness of the pure chromium samples to a protective chromium oxide layer.

Earlier investigations have suggested that only nickel-chromium alloys with from 16 to 27% chromium develop adequate protective surface oxides, while alloys with less chromium may not (Sarkar and Greener, 1973; Pourbaix, 1984; Brune, 1986; Bumgardner and Lucas, 1993). Furthermore, it was reported that beryllium additions to nickel-chromium alloy chemistries inhibited chromium surface oxide formation and decreased the alloy's corrosion-resistant properties (Baran, 1984; Covington *et al.*,1985; Lee *et al.*, 1985; Herø *et al.*, 1987; Johansson *et al.*, 1989; Bumgardner and Lucas, 1993; Geis-Gerstorfer and Pässler, 1993).

Corrosion studies have shown that nickel-chromium alloys have relatively low breakdown potentials and are susceptible to pitting and crevice corrosion (Lee *et al.*, 1985; Johansson *et al.*, 1989; Lucas *et al.*, 1991; Bumgardner and Lucas, 1993a). Consequently, concerns remain over the potential release of increased levels of metal ions such as nickel, chromium, and beryllium and the development of metal sensitivity, carcinogenicity and/or toxicity. Metal ions released from dental alloys have been shown to alter cellular activities *in vitro* (Bumgardner *et al.*, 1989). In this study, it was hypothesized that the amounts and types of metal ions released from nickel-chromium alloys alter cellular activities, as indicated by proliferation, without altering morphology and viability of cells in culture. Therefore, the aim of this investigation was to evaluate and compare the effects of four commercial nickel-chromium dental casting alloys, representing a range of compositions, on the morphology, viability, and proliferation of cultured cells and to correlate these effects with metal ion release as measured by atomic absorption spectroscopy. These data will also be related to the alloys' bulk and surface chemistries, microstructures, and corrosion properties.

Materials and methods

The compositions of the alloys used in this investigation, as determined by emission spectroscopy, are presented in Table 1. The alloys were chosen to represent a wide range of compositions, including high-and low-Cr-containing alloys, a high-Cr/low-Be alloy, and a low-Cr and high-Be alloy. The alloys were induction-cast according to manufacturers' specifications into discs measuring approximately 15 mm diameter x 3 mm thick. The discs were ground and polished to simulate clinical conditions (Bumgardner and Lucas, 1994). Samples were then sterilized in accordance with current clinical practices for prevention of infectious diseases, as recommended by the University of Alabama at Birmingham (UAB), Department of Prosthodontics. The polished samples were rinsed in distilled water, ultrasonically cleaned in ammonia, cold-solution-sterilized in biocidal solution (Glutaraldehyde 30, HealthCo International, Boston, MA), and then thoroughly rinsed twice with 10 mL sterile distilled water.

Cell culture methods

Human gingival fibroblast cell cultures were used to evaluate the cellular response to the test alloys. Cells were cultured from explants obtained from autopsies through the UAB Office of Tissue Procurement. Cells were grown in a 37°C humidified 5% CO_2 atmosphere in 95% alpha-minimum essential medium (MEM)-5% fetal bovine serum (FBS), known as complete media. All cell culture experiments were conducted on cell passages 3 to 10. Alterations in cellular morphology, viability, and proliferation were used to evaluate the effects of the test alloys on the cultured cells.

For the morphology and viability tests, cells were seeded at approximately 30,000 cells/cm^2 in six-well culture plates (CoStar, Cambridge, MA, USA). Test alloy samples and pure Ni samples were sterilized as above and then rinsed in sterile MEM balanced salt solution (BSS). Pure Ni samples were used as positive controls. One test alloy sample *per* well was placed in direct contact with the monolayer in 3 mL of complete media. Wells without any samples were used as negative controls. Cells were evaluated at one-, two-, and three-day intervals. At the end of each test, cells were visually inspected by light microscopy and qualitatively compared with negative controls for morphological changes. The absorbance of trypan blue at 580 nm (Saijo, 1973) and the absorbance of neutral red at 540 nm (Borenfreund and Puerner, 1984) were used to determine the viability of the cells exposed to the test alloys *via* a Shimadzu UV-160 spectrophotometer (photometric accuracy, ± 0.002 absorbance units at 1.0 absorbance). Duplicate absorbance values were read for each test well in both test procedures. Triplicate samples were run in each test, and each test was

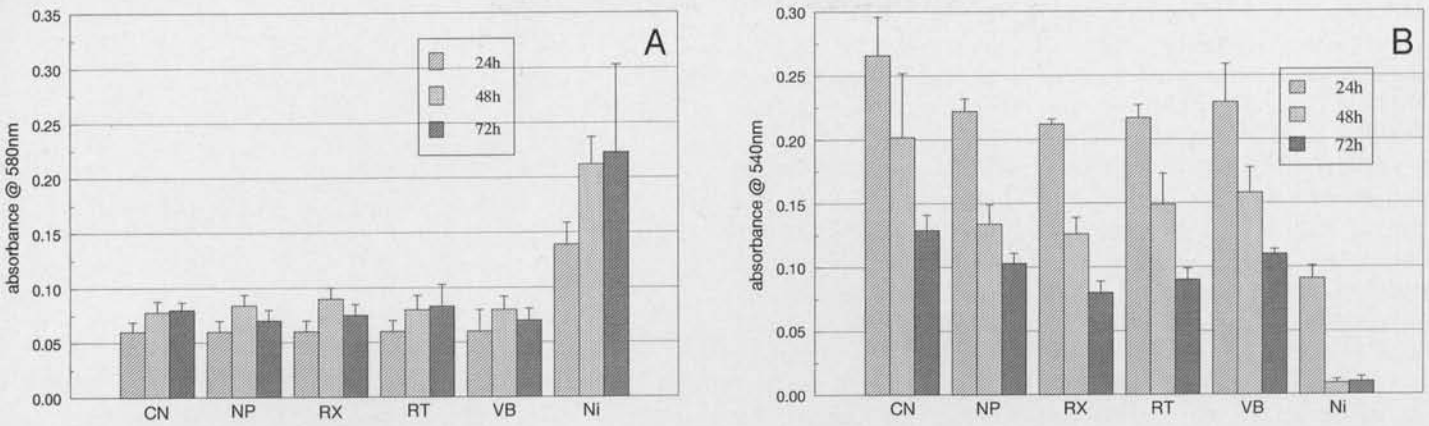

Figure 1. Results of spectrophotometric determinations of viability of cultured human gingival fibroblast cells exposed to Ni-Cr dental casting alloys. (A) Absorbance of trypan blue dye. Trypan blue stains non-viable cells; hence, increased absorbance is proportional to cell death. (B) Absorbance of neutral red dye. Neutral red is taken up by the cells; hence, increased absorbance is proportional to cell viability. Error bars represent standard deviations. CN, control; NP, Neptune; RX, Rexalloy; RT, Regalloy T; VB, Vera Bond; and Ni, pure nickel.

repeated three times.

We evaluated proliferation of the cells exposed to the test alloys by measuring the uptake of ^{3}H-thymidine. These tests were conducted as above with the exception that CoStar 8.0-µm-pore-size polycarbonate Transwell® inserts were placed in each well. Sterilized alloys and pure Ni samples were rinsed in MEM BSS and placed in the Transwell® inserts. The inserts allowed the cells to be exposed to the samples and their corrosion products without reducing the number of cells and cell growth area, thus avoiding having to correct the level of incorporated thymidine measured due to the area occupied by the samples. Cells and Transwell® inserts containing test samples were incubated in 3 mL of complete media for 12 to 14, 36 to 38, and 60 to 62 h, then pulsed with 3.0 µCi/mL ^{3}H-thymidine (specific activity, 6.7 Ci/mmol) for 10 to 12 h so that proliferation data for one, two, and three days, respectively, could be obtained. The cells were harvested, and disintegrations *per* min (dpm) were determined by means of a Beckman LS 5000TD scintillation counter (Bumgardner *et al.*, 1989). Transwell® inserts without a test sample were used as negative controls. Triplicate samples were run in each test, and each test was repeated three times.

Atomic absorption analyses

Media were saved in quadruplicate from the 24-, 48-, and 72-hour viability tests for atomic absorption analysis (AA). Test media were analyzed for release of Ni, Cr, Mo, Be, and Mn ions on a Perkin-Elmer 3030B Atomic Absorption Spectrophotometer with an HGA 600 Graphite Furnace. Standards for each element were made in MEM BSS to account for matrix effects. Perkin-Elmer-recommended pre-treatment and atomization furnace temperatures were used in furnace programs so that standard linear curves would be generated for each element. Two absorbance readings *per* element analyzed were made for each culture sample. Each reading was used to determine the mean concentration of metal in ppb released from the test alloys *per* day. Standard deviations were also calculated for the means.

Using a Perkin-Elmer 400 Inductively Coupled Plasma (ICP) atomic absorption spectrophotometer, we further analyzed media from cells cultured with either Neptune or Rexalloy to determine the release of niobium (Nb) and gallium (Ga), respectively. Two wavelengths—309 and 316 nm for Nb, and 287 and 294 nm for Ga—were used for each element as verification of any positive results. A mixed standard curve containing from 0.05 to 1.00 ppm of both elements in MEM BSS was used, and concentrations were determined by linear regression analysis.

For the AA evaluations, the percent composition of metal ion corrosion products released from each alloy was calculated based on the means ± standard deviations. Using a personal computer and least-means squares, we conducted analyses of variance of the viability, ^{3}H-thymidine uptake, and atomic absorption data, to determine statistical differences in the effects of the test alloys on the cells and the release of metal ions (SAS®, SAS Institute, Gary, NC, USA). Analyses compared data for the alloys at each test interval as well as data for each alloy over the three-day tests. Statistical differences were defined at $p \leq 0.05$ level of significance.

Results

Cell culture tests

Light microscopic examination of the cell monolayers demonstrated normal spindle-shaped fibroblasts growing adjacent to and surrounding the dental alloys for up to three days. Cells adjacent to and surrounding the pure Ni samples exhibited significantly altered morphologies within 24 h. By day 3 of the three-day tests, the entire monolayer exposed to the pure Ni samples displayed severely altered cellular morphologies, cell rounding, and loss of cell attachment.

The results of the trypan blue and neutral red viability tests are shown in Fig. 1. Analysis of variance of the trypan blue mean absorbances indicated that for each time period the viability of the cells exposed to the alloys Neptune, Rexalloy, Regalloy T, and Vera Bond were statistically similar to the control cells (Fig. 1a). The trypan blue mean absorbances of cells exposed to the pure Ni samples were significantly higher (decreased viability) compared with the controls and cells exposed to the test alloys at all time periods. While there were some statistical differences in the

viabilities of fibroblasts exposed to individual test alloys at 24, 48, and 72 h, there was no distinct trend. There was a significant increase in absorbance for cells exposed to pure Ni samples for from 24 to 48 h but no difference between the absorbance of trypan blue at 48 and 72 h.

Analysis of variance of the neutral red absorbance data demonstrated that at 24-, 48-, and 72-hour time periods, the control cells had the statistically highest mean absorbance values (highest viability), while cells exposed to the pure Ni samples had the lowest (Fig. 1b). Neutral red mean absorbance values for cells exposed to the dental casting alloys were similar to each other but significantly lower than the controls for all test periods. Comparison of the neutral red absorbance of the control cells and cells exposed to the individual test alloys over the one-, two-, and three-day test periods showed that the absorbance of neutral red was statistically different and decreasing at each test interval (Fig. 1b). There was no difference in the absorbance of cells exposed to pure Ni samples at 48 and 72 h.

The results of the ^{3}H-thymidine uptake tests are presented in Fig. 2. Analysis of variance of the ^{3}H-thymidine data indicated that the control cells had the statistically highest levels of ^{3}H-thymidine incorporation at all time intervals, while cells exposed to the pure Ni samples had the lowest. Cells exposed to the dental alloys incorporated significantly lower levels of ^{3}H-thymidine than the control cells at all test periods. Cells exposed to the high-Cr-Mo alloy, Neptune, consistently incorporated higher levels of ^{3}H-thymidine than cells exposed to the other test alloys. Only in the 72-hour tests was the incorporation of ^{3}H-thymidine by cells exposed to Neptune statistically higher than that of cells exposed to the other test alloys. Rexalloy, Regalloy T, and Vera Bond caused similar reductions in cellular proliferation, as measured by the uptake of ^{3}H-thymidine. Individual analyses of the control cells and cells exposed to the test alloys indicated that the uptake of ^{3}H-thymidine was statistically different and decreasing at each successive test period. For cells exposed to the pure Ni samples, only ^{3}H-thymidine levels at 24 h were different.

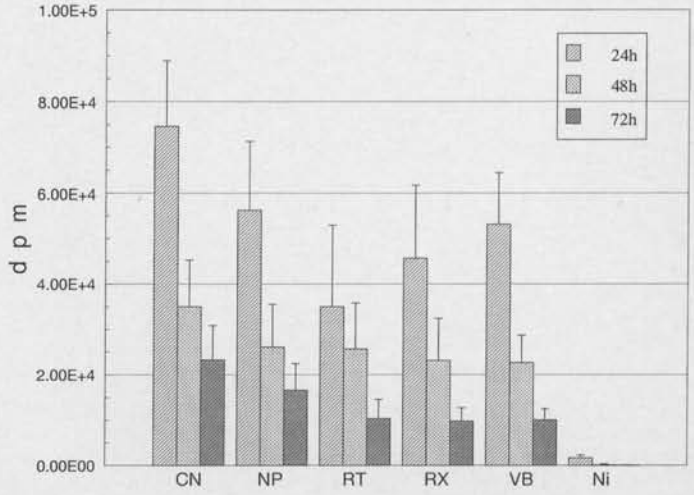

Figure 2. Results of the incorporation of radiolabeled thymidine by human gingival fibroblasts exposed to Ni-Cr dental casting alloys for up to 72 h. Error bars represent standard deviations. Abbreviations as in Fig. 1.

Atomic absorption analyses

The results of graphite AA analysis of the media from cell cultures exposed to the dental alloys are presented in Fig. 3. Analysis of Ni levels showed that, of the dental alloys tested at all three time intervals, Neptune released the statistically lowest amount of Ni. There were some differences in the release of Ni ions from Rexalloy, Regalloy T, and Vera Bond at 24- and 48-hour test periods. However, by 72 h, there was no difference in the amount of Ni released. Pure Ni samples released greater than 324.1 ppm Ni over the 24- to 72-hour tests. These levels were 1000x greater than those from the commercial alloys. All alloys released statistically increasing amounts of Ni ions at successive test intervals.

Analysis of released Cr ions indicated that Rexalloy released the statistically highest levels at all time intervals. There were no differences in the amount of Cr released from the remaining three alloys over the three test periods. Only the Rexalloy alloy released statistically different and increasing Cr ions at each test time.

Mo ions released from either Vera Bond or Regalloy T were measured below the determined detection limit of 10 ppb for all test periods, and hence, their release levels are not reported. Neptune and Rexalloy released levels above the detection limit, and these levels were statistically different at all test intervals. Over the three-day tests, Mo analyses showed a trend of increasing Mo ion levels, which were statistically higher at 72 h than at 24 h for both alloys.

Analysis of Be ion levels indicated that Vera Bond released statistically higher levels than Regalloy T at both 24 and 48 h. However, there was no difference between Be ion release of the alloys at 72 h. Comparison of Be release showed statistically different and increasing levels at each time interval only for Regalloy T. Additionally, for Regalloy T, there was no difference in the amount of Mn released at 24 and 48 h, but significantly higher levels were measured at 72 h. Nb and Ga ions were not measured in media above the detection limit of 50 ppb, and thus their release levels are not reported.

As seen in Table 2, Ni ions were released in greater quantities, while Cr was released at lower levels than indicated by the alloy bulk compositions. For the non-Be alloys, Neptune and Rexalloy, Mo ions were released at slightly greater levels than indicated by bulk composition, while for the Be-containing alloys, Vera Bond and Regalloy T, Mo was not measured above the detection limit. Comparison of the bulk alloy composition and the composition of released metal ions from the Be-containing alloys, Vera Bond and Regalloy T, indicated that Be ions were released at from four to six times the bulk alloy composition. The metal ion composition of the corrosion products did not change appreciably over the three-day tests, even though individual metal ion levels did increase.

Discussion

Previous investigations have reported no adverse changes in morphology and viability of cultured cells exposed to Ni-Cr alloys (Kawahara *et al.*, 1968; Woody *et al.*, 1977; Hensten-Pettersen and Jacobsen, 1978; Kawahara, 1983; Craig and

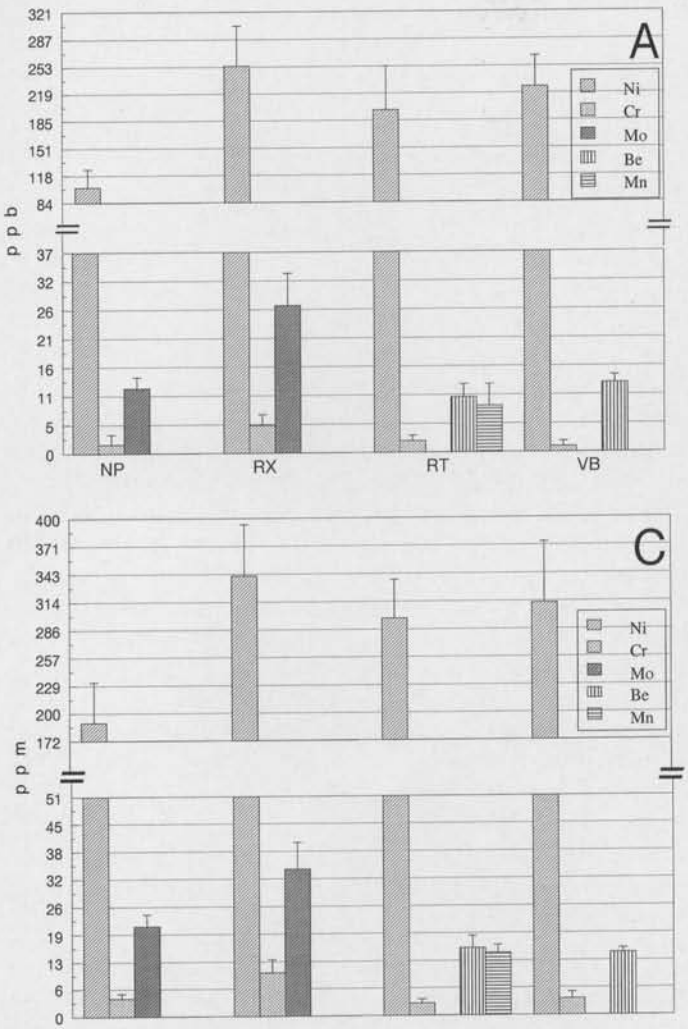

Figure 3. Results of graphite atomic absorption analyses of cell culture media for metal ions released from Ni-Cr dental casting alloys. (A) Metal ion levels at 24 ± 1 h. (B) Metal ion levels at 48 ± 1 h. (C) Metal ion levels at 72 ± 1 h. Error bars represent standard deviations. Abbreviations as in Fig. 1. Detection limits for the elements were 12.5 ppb Ni, 2.5 ppb Cr, 10 ppb Mo, 0.5 ppb Be, and 2.5 ppb Mn.

Hanks, 1988). Indeed, Craig and Hanks (1988) indicated that in cell culture tests, Ni-based alloys were biocompatible in the polished condition. The results of the morphology evaluations and trypan blue viability tests used in this study were consistent with these earlier observations. However, the results of the neutral red tests appeared to differ with those earlier reports, in that the viability of cells exposed to the test alloys was lower than that of control cells in the three-day tests. In both tests, the test samples were in direct contact with the monolayer, which decreased the number of cells and cell growth area as compared with controls. The uptake of neutral red by cells has been shown to be sensitive to cell numbers (Borenfreund and Puerner, 1984). Trypan blue staining, on the other hand, is not as sensitive to cell numbers (Walum *et al.*, 1990). It was believed that the reduction in the uptake of neutral red by cells exposed to the test alloys as compared with controls was attributable to a decrease in the number of cells and cell growth area, rather than to the test alloys and their corrosion products. Indeed, in follow-up tests exposing the fibroblasts to the test samples by means of Transwell® inserts, only cells exposed to pure Ni samples showed decreased uptake of neutral red, compared with cells not exposed to any test sample. Hence, the results of both viability tests did in fact agree and supported the results of previous investigations reporting tolerance of nickel-chromium alloys in cell culture viability tests.

The decrease in the uptake of neutral red by control cells over the three-day tests also suggested that this test depends on an active cellular process which, as the cells neared confluency, decreased and reduced the amount of neutral red incorporated. Hence, the decrease in the uptake of neutral red by the negative control cells and the cells exposed to the test alloys during the three-day tests was attributed to decreased cellular activity as the monolayer neared confluence rather than to decreases in cellular viabilities during culture. A similar pattern was observed for the cells in the thymidine uptake tests. In future studies, the use of a lower initial cell inoculum would avoid the confounding effects of decreasing cellular activities due to the monolayer approaching confluency.

The results of the AA analyses showed that metal ion release was not proportional to alloy composition. However, metal ion release was found to correlate with previous investigations of the alloys' surface and corrosion properties (Bumgardner and Lucas, 1993). For the non-Be-containing alloys, increased levels of released metal ions were associated with the corrosion-susceptible, low-Cr-containing alloy Rexalloy, compared with the corrosion-resistant, high-Cr-containing alloy Neptune. Preferential release of Ni and Be ions was observed for the corrosion-susceptible Be-containing alloys, Regalloy T and Vera Bond. The preferential release of Ni and Be ions from Regalloy T and Vera Bond corresponded with previous reports of preferential pitting in the alloys' Cr- and O-deficient surface oxide and the underlying Ni-Be eutectic (Herø *et al.*, 1987; Johansson *et al.*, 1989; Bumgardner and Lucas, 1993, 1994). The higher levels of metal ions released from Rexalloy,

Table 2. Percent composition of metal ion corrosion products released from Ni-Cr dental casting alloys (mass%) based on atomic absorption measurements

Alloy	24 h					48 h					72 h				
	Ni	Cr	Mo	Be	Mn	Ni	Cr	Mo	Be	Mn	Ni	Cr	Mo	Be	Mn
Neptune	89-88	2.4-0	11-10	—	—	91-90	3-2	8-7	—	—	89-87	2-1.7	11-9	—	—
Rexalloy	89-88	2-1	10-9	—	—	90-87	2.4-2	10-7	—	—	89-88	3-2	9	—	—
Regalloy T	92-90	1.1-0.7	—	5-4.4	4.4-3	90-91	1.2-0.8	—	5-5.3	3.3	90	1-0.6	—	5-4.6	4.5
Vera Bond	94	≤1	—	5.7-5	—	95-93	1.5-0.4	—	5-5.5	—	95-94	1.3-0.8	—	5.2-4	—

Regalloy T, and Vera Bond resulted in larger reductions in cellular proliferation compared with metal ions released and cells exposed to Neptune samples. However, these levels were below those necessary to cause changes in morphology and viability within the time periods evaluated. Therefore, released metal ion profiles of these alloys were predictable, based on surface and corrosion data, and correlated with the observed changes in proliferation of the cultured cells.

Cr released from the alloys was very low, especially compared with the release of Mo and Mn ions. Similarly low Cr release data have been reported for Ni-Cr alloys in artificial saliva, lactic acid, and saline solutions (Geis-Gerstorfer and Weber, 1987; Tai *et al.*, 1992). The reason for the increased release of Mo and Mn ions from the alloys, compared with Cr, is not known at this time. Despite the increased release, Mo and Mn ions, which were detected at relatively low levels, are regarded as having low toxicities (Rae, 1978; Kawahara, 1983; Saric, 1986). Nb and Ga ions, which were released at levels below the detection limits of this study, also have been reported as having low toxicities (Kawahara, 1983; Hanawa *et al.*, 1992; Scansetti, 1992). However, Be ions, which were released at levels from four to six times those of the bulk alloy compositions, are regarded as having high toxicities (Covington *et al.*, 1985). The elevated release of Be ions found in this study agreed well with previous reports of elevated Be ion release from other Be-containing alloys in other physiological solutions (Geis-Gerstorfer and Weber, 1987; Covington *et al.*, 1985; Tai *et al.*, 1992).

Even though information on the different types and amounts of metal ions released from the alloys may be obtained from 24-hour tests, statistical differences were not observed in the cellular response to the alloys and their corrosion products in that time period. However, by the end of the 72-hour tests, statistical differences in the incorporation of thymidine by the cells exposed to the different alloys began to emerge. Indeed, Hensten-Pettersen and Jacobsen (1978) indicated that cytotoxic reactions to nickel-chromium alloys in cell cultures were unlikely, and that the more sensitive cell proliferation test was required to detect effects of the alloys on the cells. Additionally, possible synergistic effects of released metal ions, due to their various individual toxicities, remain to be investigated. More sensitive cell culture tests and/or longer *in vitro* test periods are needed for evaluation of the responses of cultured cells to the test alloys and the amounts and types of metal ions they release.

In summary, it is important not only to evaluate the cellular response to Ni-Cr dental casting alloys, but also to determine the amounts and types of metal ions released and their effects on cellular viabilites and functions. The results of this investigation supported the hypothesis that Ni-Cr alloys and their corrosion products affect activities, as measured by proliferation, without affecting viability or morphology of cells in culture. Small decreases in proliferation were observed in cells exposed to the high-Cr-Mo-containing alloy, Neptune. This alloy released the lowest level of metal ions of the alloys tested. Larger decreases in proliferation were observed in cells exposed to the low-Cr and Mo alloy Rexalloy and to the Be-containing alloys, Regalloy T and Vera Bond. Rexalloy released significantly elevated levels of Ni, Cr, and Mo ions compared with the Neptune alloy, while the Be-containing alloys released elevated levels of Ni and Be ions as well as Cr and, if present, Mn ions. The release of metal ions, especially from low-Cr and Mo alloys and the Be-containing alloys, raises questions about the continued use of these alloys.

Acknowledgments

The authors would like to express their gratitude to Dr. A. Prasad of Jeneric/Pentron, Inc., for supplying the Rexalloy discs. The authors are indebted to Ms. Debbie Bradshaw and Ms. Martha Cole at Perkin-Elmer for their invaluable advice and assistance in using graphite furnace atomic absorption spectrophotometry. The authors are also indebted to Ms. Ann Lyons of the Environmental Analytical Chemistry Section at Southern Research Institute, Birmingham, AL, for the ICP-AA analyses. This work was supported by the National Institutes of Dental Research, grant #1 RO3 DE10275-01.

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