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Cast Co–Cr alloy and pure chromium in proteinaceous media: an electrochemical characterization

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An electrochemical characterization (by linear sweep cyclic voltammetry and chronoamperometry at constant potential) of a cast Co–Cr alloy and of pure chromium was performed and their behaviour compared using NaCl 0.15 M and albumin-containing solutions as electrolytes. The amount of ions released was determined. The protein detrimental effect over both materials was detected. Metallic surfaces were also investigated by X-ray photoelectron spectroscopy and observed by means of optical and scanning electron microscopy.

1. Introduction

The use of metallic materials in the manufacture of implants and prostheses is very frequent especially for orthopaedic and dental purposes. However, their interaction with the body, while unavoidable, is not always pacific, as is the case when corrosion occurs. Biocompatibility ("the ability of a material to perform with an appropriate host response in a specific application" [1]) may be influenced in a number of ways, for instance, by the inherent pH and pO_2 modification in the microenvironment surrounding the implant [2], by physical and chemical surface changes [3] (topography, elemental composition, oxidation states, etc.) and, of course, by the release and accumulation of ions in the implant site and/or their distribution throughout the organism via systemic circulation [4]. We are far from a full understanding of all the phenomena involved because of the complexity and variability to which they are subjected. Materials behaviour characterization in conjunction with physiological reality is necessary so that, in the near future, suitable biomaterials may be developed for each application.

Some published papers [5–8] show that the inclusion of proteins in standard *in vitro* test solutions is important for the determination of the corrosion resistance of metals and alloys. Nevertheless, further work must be performed in order to answer the questions raised, such as which particular proteins influence the process, at which conditions and concentrations, is corrosion enhanced or inhibited, how differently do materials behave, what happens to the surface, what kind of corrosion products are generated and what is the actual mechanism by which proteins interfere in the degradation of metallic medical devices?

The principal aim of this study is to evaluate, using the appropriate electrochemical techniques [9], the susceptibility to localized corrosion of a cast Co–Cr–Mo orthopaedic alloy when albumin, the most prevalent plasma protein, is present. A qualitative description (e.g. breakdown and repassivation potentials) of the process is obtained by linear sweep cyclic voltammetry. Since the passivation of the mentioned alloy is due to the formation of a spontaneous chromium oxide surface layer, a comparison with the behaviour of pure chromium under the same conditions is made. In addition, quantitative information is provided by chronoamperometry at constant potential, conducted to simulate the ageing of the material over a short period of time. Optical and scanning electron microscopy and X-ray photoelectron spectroscopy (XPS) supply complementary data concerning the resulting surface state. Metallic contents in solution are determined by atomic absorption spectroscopy.

It is also hoped to contribute to the establishment of normalized electrochemical tests that take into account the organic composition of body fluids, allowing a more reliable prediction of the *in vivo* implants performance.

2. Materials and methods

The alloy used in the experiments has the following elemental composition (% by weight): Co(balance), Cr(28), Mo(5.5), Ni(1), Si(0.95), Fe(0.7), Mn(0.65) and C(0.25). Chromium samples have a 99.96% degree of purity.

Both type of materials were cut as discs of 15 mm diameter, and thin enough to be held by vacuum in an

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0957–4530

electrochemical cell of capacity 20 ml (Fig. 1) provided with a SCE (in a separate compartment) and a large-area gold counter electrode. Prior to voltammetric and chronoamperometric essays the surfaces of the samples were polished to a mirror finish using successively finer grades of alumina (Buehler, down to 0.015 μm) and rinsed with distilled water. By polishing the samples (which is a normal practice in electrochemistry) a good, reproducible surface was obtained for use in the corrosion tests.

Bovine serum albumin (Sigma) was added to the basis saline electrolyte (NaCl 0.15 M, p.a. reagent) and the pH was adjusted to the physiological value (7.4). The selected protein concentrations were all inferior to the serum albumin content which is about 50 g dm^{-3} [7].

During the electrochemical studies, the system was maintained at 37°C (by circulating water through the double wall of the cell) and was agitated at 200 rpm. A Bank Electronic Wenking POS73 potentioscan was used to control the working electrode potential and the response was monitored by a Houston Omnigraphic 2000 X–Y–t recorder. Voltammetric measurements were carried out at 5 mV s^{-1} scan rate and were begun at the rest potential (determined after 30 min of sample–solution contact). The scans were reversed at 700 mV which is a potential higher than, but reasonably close to the breakdown value. A 700 mV potential was also applied during the chronoamperometric essays for the same reasons. All electrochemical experiments were done at least three times.

Sample surfaces were examined using a Zeiss ICM405 optical microscope and a Jeol JSM-T 300 scanning electronic microscope.

X-ray photoelectron spectroscopy, XPS, was performed in a Kratos XSAM 800 spectrometer using Mg K_α exciting radiation (1253.7 eV) at 1.33×10^{-7} Pa vacuum. The angle between the normal to the sample surface and the entrance to the electron energy analyser was 60°. The area analysed was 1 mm^2. All metallic surfaces were washed with distilled water and acetone and dried in a flow of nitrogen before XPS essays.

Atomic absorption spectrometry (AAS) analysis of the solutions containing the corrosion products was made with a Pye Unicam SP9 spectrometer connected to a Philips 910 Computer. An air–acetylene flame and specific lamps for each element were used in the analysis. In the calibration procedure; BDH spectrosol standard solutions were used. The blank (consisting of the electrolyte) and all the solutions were nitric acid digested. The AAS detectable limit for both Co and Cr was 0.05 ppm.

All data (potential values and Co and Cr contents) are presented as mean values with 95% confidence intervals calculated by Student's t test.

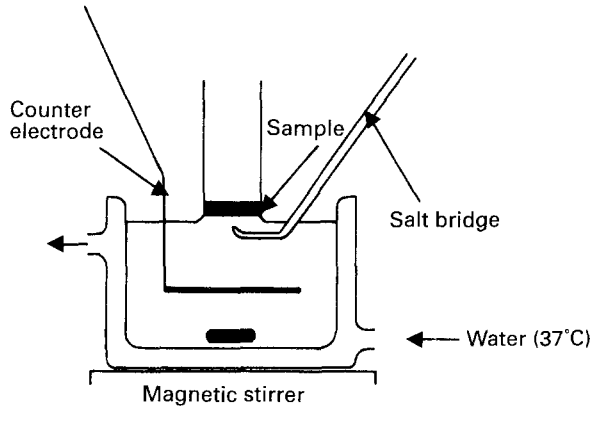

Figure 1 Electrochemical cell.

3. Results and discussion

3.1. Open-circuit potentials and cyclic voltammetric studies

Table I shows the 30 min open-circuit potentials of Co–Cr–Mo alloy and of pure Cr for the different albumin concentrations used. They are very much more negative than the breakdown potentials that will be presented later and so corrosion is not occurring at the beginning of the electrochemical tests.

Analysis of the cyclic voltammograms displayed in Fig. 2 and Fig. 3 for Co–Cr–Mo alloy and for pure chromium, respectively, reveals the detrimental effect of the albumin on those materials.

The presence of the protein modifies the range of potentials corresponding to a passivated surface state, for which electrochemical degradation may be disregarded. The anodic potential limit of the passivation domain (for which the Faradaic current is no longer null), also known as the breakdown potential, is for both cases, about 0.40 ± 0.02 V versus SCE when the electrolyte is albumin free. However, the loss of integrity of the thin chromium oxide film starts at 0.25

TABLE I Open-circuit potentials

Albumin (g dm^{-3})	Cr	Co–Cr–Mo
	(V versus SCE)	
0.0	− 0.59 ± 0.01	− 0.42 ± 0.03
2.5	− 0.62 ± 0.05	− 0.40 ± 0.04
5.0	− 0.66 ± 0.02	− 0.42 ± 0.01
10.0	− 0.65 ± 0.04	− 0.40 ± 0.03
17.5	− 0.68 ± 0.03	− 0.41 ± 0.03
25.0	− 0.69 ± 0.03	− 0.38 ± 0.04

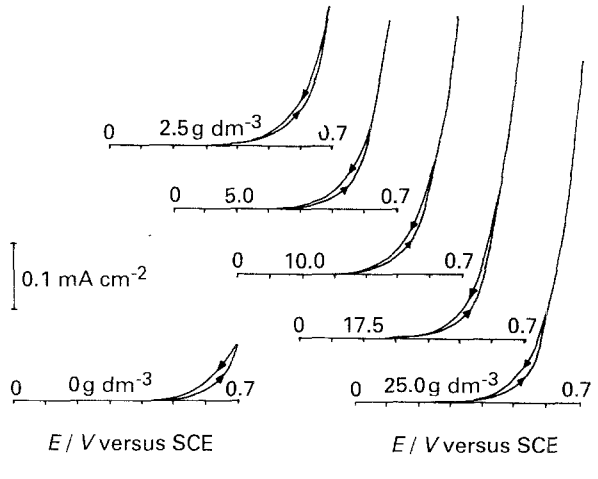

Figure 2 Voltammetric responses of Co–Cr–Mo for different albumin concentrations.

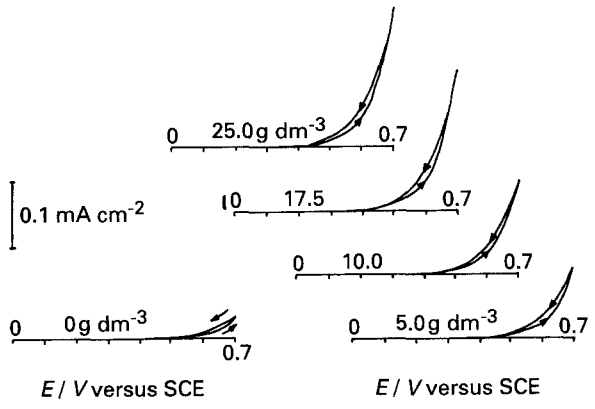

Figure 3 Voltammetric responses of pure chromium for different albumin concentrations.

$\pm$ 0.06 V versus SCE for the alloy electrode and at 0.35 $\pm$ 0.05 V versus SCE for the pure metal sample when the protein is added to the electrolyte (there is no clear distinction among the breakdown potentials determined for the different albumin contents).

Anodic current values are also enhanced when proteinaceous solutions are used, more markedly for the Co–Cr alloy, probably due to the dissolution of other species along with chromium (we shall see later that cobalt is dissolved in important quantities). The anodic corrosion rates recorded by voltammetric means increase to some extent with the protein concentration (Fig. 4). Elsewhere [10] we have pointed out that this could be due to a spatial saturation of albumin molecules at the working electrode surface; actually it is known that this protein strongly adsorbs to the Co–Cr alloy surface [11]. This interpretation is reinforced by the present data because the corrosion rate plateau is attained for nearly the same albumin concentration (17.5 g dm^{-3}) for both samples presenting identical geometric areas.

In spite of these latter features, albumin does not interfere in the hysteresis behaviour shown by the different materials, which is very slight (current values are almost the same in direct and reverse scans) and similar in all the responses recorded; on the other hand, the repassivation potentials, defined as the values where the current reaches the zero value ($\pm$ 0.001 mA cm^{-2}) in the reverse scan, coincide with breakdown potentials.

The voltammetric experiments illustrate the importance of the presence of chromium in the alloy composition in order to achieve a better corrosion resistance.

3.2. Chronoamperometric essays at constant potential

With the aim of performing a simulation of the ageing of the materials, four chronoamperometric essays were conducted at 700 mV versus SCE under the conditions depicted in Table II. The current was recorded and its evolution with time is plotted in Fig. 5. As expected, the adverse effect of the presence of albumin with resulting larger corrosion rates is noticeable. In terms of the oxide film damage, the action of the protein is time dependent and corrosion propagation occurs until an equilibrium state is achieved, which results from a compromise between metal/metal oxide dissolution and formation due to anodic polarization. Probably, the adsorbed albumin molecules which may be somehow responsible for the increase in corrosion, suffer rearrangements with time (in order to accommodate more molecules and/or to achieve a certain conformation) until a steady-state adsorption profile is reached, corresponding to a constant corrosion rate.

Indeed, the same features were observed for saline electrolytes containing 10% (v/v) complete serum [10]. Although the existence in serum of many proteins other than albumin that may interact with metals

TABLE II Conditions for chronoamperometric essays

Experiment	Material	Albumin (g dm^{-3})	Time (h)
1	Co–Cr–Mo	0	1
2	Co–Cr–Mo	5	1
3	Cr	0	2
4	Cr	5	2

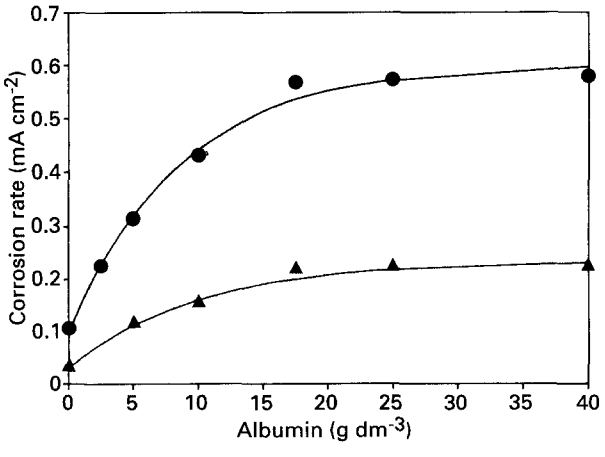

Figure 4 Corrosion rate dependence with albumin concentration at 700 mV versus SCE: ● Co–Cr–Mo: $Y = (0.50 \pm 0.02)\ (1 - \exp(-(0.12 \pm 0.01)\ X)) + (0.10 \pm 0.02)$; ▲ pure Cr: $Y = (0.20 \pm 0.02)\ (1 - \exp(-(0.10 \pm 0.02)\ X)) + (0.03 \pm 0.01)$.

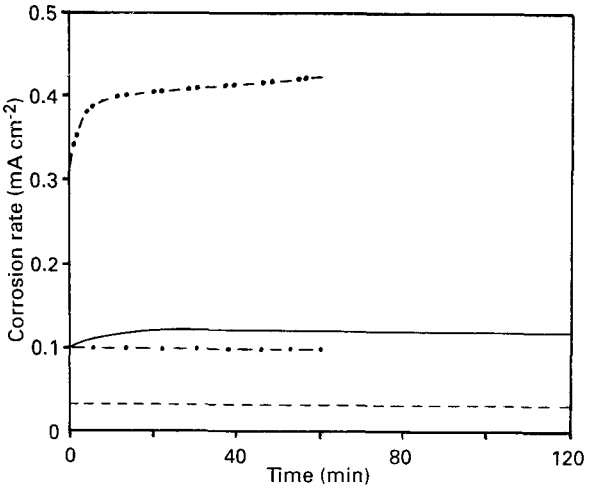

Figure 5 Current evolution during the chronoamperometric experiments (– · – essay 1, (– ·· – essay 2, – – – essay 3, —— essay 4).

in a different and more specific way, corrosion was also enhanced and to a larger extent than in the present case where we use 5 g dm^{-3} of albumin (concentration predicted in 10% (v/v) serum). In fact, because of its high concentration in serum, we think that albumin has an important role in implants degradation.

3.3. Metallic solution contents

The consequent metallic contents in solution are shown in Table III. For the alloy, the Cr/Co ratio is enhanced in the albumin-containing extract (from 0.33 $\pm$ 0.02 to 0.39 $\pm$ 0.03). The same effect was previously observed for gamma-globulin and serum media [10]; for instance, the referred ratio was 0.41 $\pm$ 0.03 for a 2.5 g dm^{-3} gamma-globulin solution and 0.38 $\pm$ 0.03 for a 10% (v/v) serum electrolyte. The protein has a stronger action on the dissolution of chromium for the alloy than for pure chromium; extract concentrations are, respectively, 5.4 and 3.9 times larger than the determined values in simple saline solutions.

Some controversy exists around the oxidation state of chromium released from implants [12, 13]. In its trivalent state, chromium appears to be essential for biological systems (forming part of the glucose tolerance factor) whereas as hexavalent ion it has cytotoxic and carcinogenic effects [14]. The total charge transferred during the corrosion process can be calculated from the integration of the current versus time plots of Fig. 5, and compared with the values estimated from the metallic contents in solution given by atomic absorption spectrometry assuming the presence in the extracts of Co(II) and Cr(III) or Cr(VI) (Table III). It appears that the degradation of the pure metal in extracts with and without albumin gives rise to hexavalent chromium. The same results are more difficult to interpret for the alloy because of its complex composition. Nevertheless, and at least for the experiment performed in the saline solution, a mixture of both valences is possible as the average amount of charge transferred obtained from the current plot (0.38 $\pm$ 0.02 C) is an intermediate value between the other two calculated from atomic absorption data (0.31 $\pm$ 0.03 C for Cr(III) and 0.42 $\pm$ 0.04 C for Cr(VI)); however the hypothesis of having only Cr(VI) in solution is not absurd if we take into consideration the experimental data errors. Indeed, after corrosion, all solutions presented the soft yellow colour characteristic of chromates corresponding to an absorbance at 372 nm [15] and measurements of Cr(VI) content will be performed in future studies using photometric analytical techniques.

TABLE III Metallic contents in solution and charge transferred during corrosion

Experiment	Co (ppm)	Cr (ppm)	Charge transferred (C) A	B	C
1	3.0	1.0	0.38	0.31	0.42
	$\pm$ 0.3	$\pm$ 0.1	$\pm$ 0.02	$\pm$ 0.03	$\pm$ 0.04
2	13.9	5.4	2.42	1.51	2.11
	$\pm$ 0.6	$\pm$ 0.4	$\pm$ 0.02	$\pm$ 0.07	$\pm$ 0.09
3	–	1.8	0.43	0.20	0.40
		$\pm$ 0.1	$\pm$ 0.01	$\pm$ 0.01	$\pm$ 0.02
4	–	7.0	1.61	0.78	1.56
		$\pm$ 0.4	$\pm$ 0.08	$\pm$ 0.04	$\pm$ 0.08

A – from the integral of current versus time plots.
B – from atomic absorption spectroscopy data assuming the presence of Co(II) and Cr(III) in solution.
C – as B, assuming the presence of Cr(VI) in solution.

3.4. Surface analysis

The Co–Cr–Mo surface before and after chronoamperometric essays was examined by XPS analysis. The metallic contents (atomic percentages) detected in the oxide film are shown in Table IV (oxygen is omitted) for a sample without electrochemical treatment (polished and immediately introduced in the vacuum chamber of the spectrometer) and for the other two samples resulting from the experiments with and without protein. Apart from the usual composition differences among surface and bulk material [16] it must be noted that the surface of the sample submitted to corrosion in the saline solution shows a strong enrichment in chromium and a slight enrichment in nickel and molybdenum in comparison with the polished surface. It is also the lowest cobalt content case. The surface produced in proteinaceous medium has an oxide layer composition with just a little more chromium and nickel and with a little less cobalt than the one from the polished sample; additionally, the almost nonexistence of molybdenum in the surface is a very interesting fact.

Cobalt and chromium were our main concern in this work due to the alloy composition and to their inherent biocompatibility problems [17]. However, this last feature has given rise to the possibility of there being a preferential dissolution of the molybdenum from the alloy by the action of the protein, and further work is in progress to clarify these aspects.

The deconvolution of the chromium peak from the XPS essays (Table IV) also indicates the existence of hexavalent chromium in the oxide film under all circumstances.

The metallic surfaces were observed by optical (Fig. 6) and scanning electron microscopy (Fig. 7). The intergranular corrosion in the Co–Cr–Mo alloy can readily be seen, with a strong attack at the grain boundaries adjacent areas corresponding to

TABLE IV XPS analysis for the Co–Cr–Mo alloy

	Experiment 1	Experiment 2	Polished sample
	(atomic percentages)		
Co	5.4	19.6	21.3
Cr	74.7	69.2	61.2
Mo	8.7	1.0	8.2
Ni	11.2	10 2	9.3
	(percentages from total chromium)		
Cr (0)	6	4	13
Cr (III)	66	78	75
Cr (VI)	28	18	12

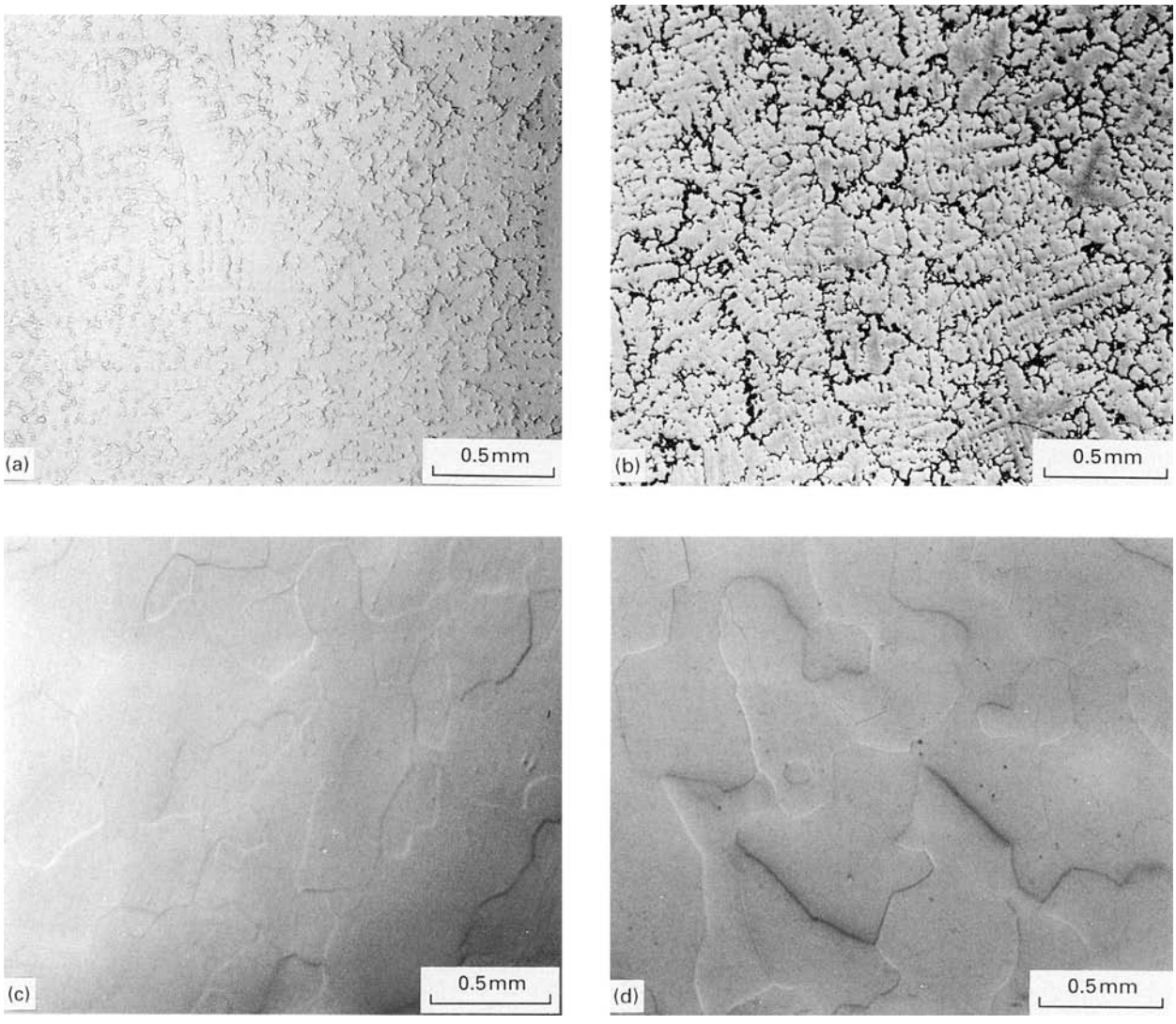

Figure 6 Optical micrographs: (a) alloy polished surface; (b) alloy surface after essay 2; (c) pure chromium polished surface; (d) pure chromium surface after essay 4.

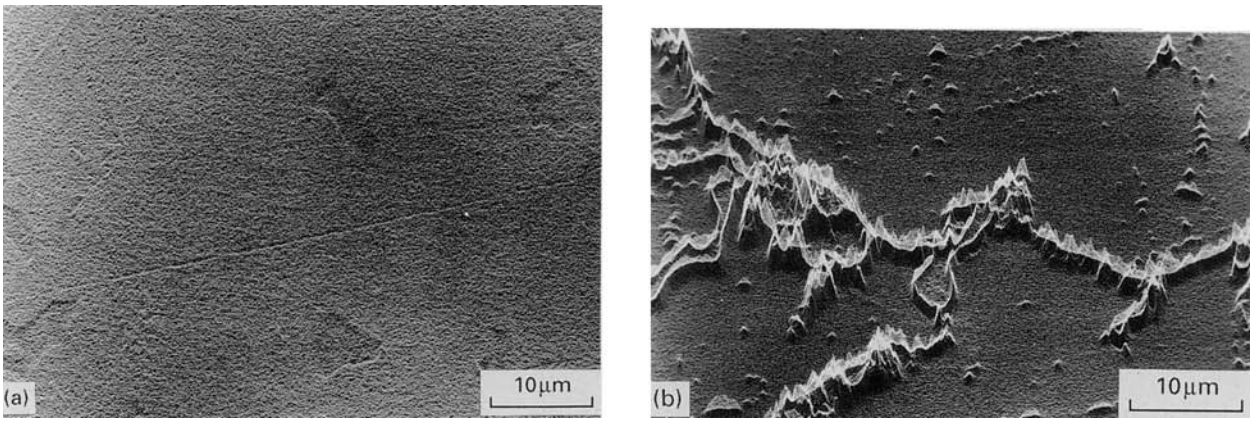

Figure 7 Y-modulated SEM micrographs (A-alloy polished surface; B-alloy surface after essay 2).

chromium depleted zones surrounding carbide precipitates. The consequences of the localized corrosion on the surface of pure chromium were not so noticeable.

4. Conclusions

The effect of proteins on the degradation of metals and alloys must not be neglected. The present results have shown, for both tested materials, that the incorporation of albumin in the electrolyte has a negative influence over the corrosion resistance.

For the *in vitro* experiments, under all corrosion conditions studied, hexavalent chromium has been detected in solutions. Cr(VI) is also always present on the Co–Cr–Mo alloy surface and the disappearance of molybdenum from surfaces subject to accelerated ageing in a proteinaceous electrolyte is a noteworthy aspect.

This study reinforces the need for, and the advantages gained by, testing metallic biomaterials under adequate conditions prior to their use inside organisms.

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References

1. D. F WILLIAMS, *J. Mater. Sci.* **22** (1987) 3421
2. L L. HENCH and E. C ETHRIDGE, in "Biomaterials. an interfacial approach", Biophysics and Bioengineering Series (Academic Press, New York, 1982) p. 21.
3. A. NAJI and M F HARMAND, *J. Biomed. Mater. Res* **24** (1990) 861
4. A BARTOLOZZI and J BLACK, *Biomaterials* **6** (1985) 2.
5. S. R. SOUSA and M. A BARBOSA, *J. Mater. Sci Mater. Med.* **2** (1991) 19.
6. S K. CHAWLA, S A BROWN, K MERRITT and J H PAYER. *Corrosion* **46** (1990) 147.
7. K. MERRITT and S. A BROWN, *J Biomed. Mater. Res.* **22** (1988) 111.
8. D F WILLIAMS and G C. F CLARK, *J. Mater. Sci.* **17** (1982) 1675.
9. M F VIEGAS, L M ABRANTES and J LECOEUR, *J. Mater. Sci. Mater. Med.* **1** (1990) 105.
10. H. TOMÁS, A PONCES FREIRE and L M. ABRANTES, *Mater. Chem. Phys.*, submitted for publication.
11. *Idem.* presented at Biointeractions '93, Noordwijkerhout, August, 1993.
12. S A. BROWN, L. J. FARNSWORTH, K MERRITT and T D CROWE. *J. Biomed. Mater Res.* **22** (1988) 321.
13. K. MERRITT, R. S. WORTMAN, M. MILLARD and S. A BROWN, *Biomat. Med. Dev. Art. Org.* **11** (1983) 115.
14. G. T. ROGERS. *Biomaterials* **5** (1984) 244.
15. H TOMÁS, A PONCES FREIRE and L. M. ABRANTES, presented at the Trace Elements in Health and Disease Conference, Lisboa, September, 1992.
16. B. D RATNER, A B. JOHNSTON and T J LENK, *J. Biomed. Mater. Res: Appl. Biomater.* **21-A1** (1987) 59.
17. J. BLACK, *Biomaterials* **5** (1984) 11.