ORIGINAL ARTICLE

Corrosion resistance and cytotoxicity of stainless steels exposed to chloride solutions

Resistência à corrosão e citotoxicidade de aços inoxidáveis expostos a soluções de cloretos

Resistencia a la corrosión y citotoxicidad de los aceros inoxidables expuestos a soluciones de cloruro

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ABSTRACT: Objective: To analyze the pitting corrosion resistance of AISI 304 and AISI 420 stainless steels in chloride-containing medium (0.9 and 3.5% NaCl solution, by weight), as well as their cytotoxicity,*in vitro,* in samples with and without pitting corrosion. **Method:** This is an experimental study. Cyclic potentiodynamic polarization (CPP) techniques were used to characterize the extent and shape of the corrosive attack on the samples. The agar diffusion and viability evaluation method of the NCTC clone 929 cell line (CCIAL 020) was used to evaluate the cytotoxicity of samples of steels with and without pitting. **Results:** The AISI 304 steel showed superior corrosion resistance to the AISI 420 steel. The values of the pitting potentials decreased for both steels when the chloride concentration in the aggressive solution was increased. There was moderate cell toxicity (grade 3 — ISO 10993-5) in all samples. **Conclusions:** The results corroborated the recommendations to avoid unnecessary immersion of the instruments in saline solutions. Moderate cytotoxicity to these steels contraindicates their use in implantable devices, only in surgical instruments.
Keywords: Stainless steel. Corrosion. Chlorides. Toxicity.

RESUMO: Objetivo: Analisar a resistência à corrosão por pites dos aços inoxidáveis AISI 304 e AISI 420 em meio contendo cloretos (solução de NaCl a 0,9 e 3,5%, em massa), assim como sua citotoxicidade,*in vitro,* em amostras com e sem corrosão por pites. **Método:** Estudo experimental. Utilizaram-se técnicas de polarização potenciodinâmica cíclica (PPC) para caracterizar extensão e forma do ataque corrosivo nas amostras. O método de difusão em ágar e avaliação da viabilidade da linhagem celular NCTC clone 929 (CCIAL 020) foi empregado para avaliar a citotoxicidade de amostras dos aços com e sem pites. **Resultados:** O aço AISI 304 apresentou resistência à corrosão superior ao aço AISI 420. Os valores dos potenciais de pite caíram para ambos os aços quando se aumentou a concentração de cloretos na solução agressiva. Houve moderada toxicidade celular (grau 3 — ISO 10993-5) em todas as amostras. **Conclusão:** Os resultados corroboraram as recomendações para evitar a imersão desnecessária dos instrumentais em soluções salinas. A citotoxicidade moderada para esses aços contraindica seu uso em dispositivos implantáveis, apenas em instrumentos cirúrgicos.
Palavras-chave: Aço inoxidável. Corrosão. Cloretos. Toxicidade.

RESUMEN: Objetivo: Analizar la resistencia a la corrosión por picaduras de aceros inoxidables AISI 304 y AISI 420 en un medio que contiene cloruros (solución de NaCl al 0,9 y 3,5%, en masa), así como su citotoxicidad, *in vitro*, en muestras con y sin corrosión por picaduras. **Método:** Estudio experimental. Se utilizaron técnicas de polarización potenciodinámica cíclica (PPC) para caracterizar el alcance y la forma del ataque corrosivo a las muestras. Se utilizó el método de difusión en agar y evaluación de la viabilidad de la línea celular NCTC clon 929 (CCIAL 020) para evaluar la citotoxicidad de las muestras de acero con y sin picaduras. **Resultados:** El acero AISI 304 presentó una resistencia a la corrosión superior al acero AISI 420. Los valores de potencial de picadura disminuyeron para ambos aceros cuando aumentó la concentración de cloruros en la solución agresiva. Hubo toxicidad celular moderada (grado 3 — ISO 10993-5) en todas las muestras. **Conclusión:** Los resultados corroboraron las recomendaciones para evitar la inmersión innecesaria de instrumentos en soluciones salinas. La citotoxicidad moderada de estos aceros desaconseja su uso en dispositivos implantables, reservándolos solo para instrumentos quirúrgicos.
Palabras clave: Acero inoxidable. Corrosión. Cloruros. Toxicidad.

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INTRODUCTION

The materials used in the manufacture of surgical instruments are made of stainless steels, which are metal alloys composed of iron (Fe), with a minimum percentage of 10.50% chromium (Cr), necessary to form a protective film, called layer or passive film, responsible for protecting stainless steels against corrosion, keeping them less reactive to the environment in which they are inserted, i.e., passivated[1].

According to the American Iron and Steel Institute (AISI), stainless steels are classified according to their microstructure and the concentration of the present chemical elements in groups, series or families[2].

Among the existing types of stainless steels, the metal alloys most used in the manufacture of surgical instruments belong to the AISI 300 and 400 series[3].

The AISI 304 steel [18%chromium (Cr)-8%nickel (Ni)], with an austenitic microstructure, is used to manufacture retractors, cannulas, clamps, fasteners, specula, and suction tubes[3].

Materials made with AISI 304 steel, e.g., retractors, are more resistant to corrosion, compared to AISI 420 steels.

The AISI 420 steel [(12%chromium (Cr)-0.35%carbon(C)], with a martensitic microstructure, due to its greater hardness and consequent greater mechanical resistance, related to the higher concentration of carbon in its composition, is intended for the manufacture of rongeurs, bite screws, curettes, scissors, tweezers, retractors, probes, and articulated tweezers[3].

Corrosion can be defined as the deterioration of a metallic material, due to the electrochemical or chemical action of the medium in which it is inserted, whether or not related to mechanical forces[4].

Among the main types of corrosion, pitting is one of the most frequent forms of localized corrosive attack on passive metals such as stainless steels[5-7]. It consists of the formation of small cavities, with variable depths, depending on the thickness of the metal, which can cause, for example, failures and breakdowns in the instruments during the intraoperative period[5,6].

This type of corrosion is associated with low pH values, as well as the presence of chloride anions both in saline solutions (0.9% NaCl) and in body fluids, and in the water used for cleaning and sterilization of surgical instruments at the Sterile Processing Department (SPD)[6-9].

During the corrosive process, corrosion products are released, which are metallic salts of the alloy components capable of posing a risk (cytotoxicity), according to the level of exposure of patients to these chemical compounds, by means of oxidized (corroded) instruments[6-9].

Therefore, the understanding of the corrosive phenomenon is essential for perioperative nurses to act in its prevention and control, thus reducing the risks of adverse events for patients such as inflammatory and allergic processes related to the use of corroded surgical instruments[5,6].

OBJECTIVE

To analyze the pitting corrosion resistance of AISI 304 and AISI 420 stainless steels, in chloride-containing media: 0.9% NaCl solution, by weight (w/w), simulating saline, and 3.5% NaCl, by weight (w/w), this being the international standard concentration (chlorides in seawater), which allows the comparison of corrosion studies as well as to evaluate the *in vitro* cytotoxicity of these metals with and without the presence of pitting corrosion on their surface.

METHODS

This experimental study was conducted at the Electrochemistry and Corrosion Laboratory of the Department of Chemical Engineering of the Polytechnic School of Universidade de São Paulo and at the Cell Culture Center of Instituto Adolfo Lutz. Rolled stainless steel plates of the type AISI 304 (six samples) and AISI 420 (five samples), used in the manufacture of surgical instruments, measuring 30 mm X 50 mm and 2.0 mm thick, with a matte finish, were used as specimens for the electrochemical measurements in order to evaluate their resistance to corrosion.

The difference between the two plates, in addition to their microstructure, lies in the chemical composition described in Chart 1.

Corrosion resistance was evaluated by the electrochemical technique of cyclic potentiodynamic polarization (CPP). All stages of the experiment complied with the protocols established by ASTM G61-86[10], which determines the standard procedures for pitting corrosion analysis.

To perform this technique, an electrochemical cell (Figure 1) was used, positioned inside a Faraday® cage, whose main function is to isolate the entire system from electrical and magnetic interference from the external environment.

The electrochemical cell (Figure 1) is composed of three electrodes:

Chart 1. Specification of the chemical composition of the steels used in the manufacture of the specimens (% by weight) used in this study, according to ASTM F899-12b[3].

Specimen	Carbon (C)	Manganese (Mn)	Phosphorus (P)	Sulfur (S)	Silicon (Si)	Chromium (Cr)	Nickel (Ni)	Other elements
AISI 304	0.07 max.	2.00 max.	0.045 max.	0.030 max.	1.00 max.	17.00–19.00	8.00–11.00	Nitrogen (N) 0.10 max.
AISI 420	0.16–0.25	1.00 max.	0.04 max.	0.030 max.	1.00 max.	12.00–14.00	1.00 max.	-----

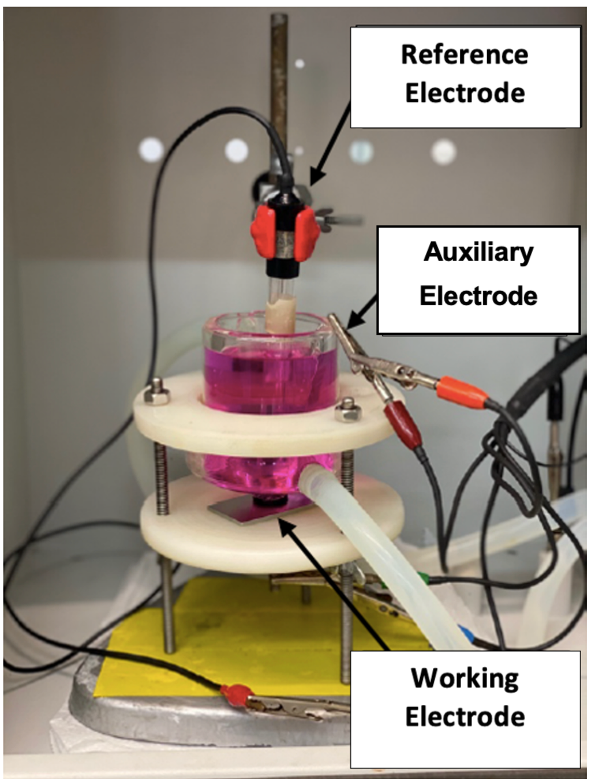

Figure 1. Electrochemical cell used in the study.

- Reference electrode: a known potential that remains constant in relation to the potential of the working electrode to be measured or imposed;
- Auxiliary electrode: it undergoes the opposite reaction to that imposed on the working electrode. If the working electrode is anodically polarized, the auxiliary electrode will be cathodically polarized[4,10,11];
- Working electrode: the material to be studied, in which oxidation or reduction of the material of interest (stainless steel plates) occurs[4,10,11].

Once inside the Faraday® cage, the electrochemical cell was coupled to the potentiostat (Reference 600, Gamry®, USA). The CPP technique is used to evaluate the susceptibility of a metal to localized corrosion (by pitting) and cracks (localized corrosion, which occurs in crack regions, with low aeration, poor in oxygen)[9-12].

The CPP curve starts at the open-circuit potential (OCP); then, by increasing the potential (anodic polarization), it crosses a region of active corrosion (not always present, as in this study); and, subsequently, the corrosion current density (I_{corr}) decreases orders of magnitude to a critical potential, called Flade potential or primary passive potential[11,12].

The time required to obtain the OCP value was 3,600 seconds (1 hour)[10]. As the anodic polarization advances, the potential in the passive region increases and, at a given potential, the current density sharply increases[11].

This increase can result from the evolution of oxygen by the decomposition of water or the breakdown of the passive film (localized corrosion). If the increase in current density results from the decomposition of water and the evolution of oxygen gas, the region and the potential reached are called, respectively, the transpassive region and the transpassive potential (E_{tr})[11,12].

The increase in anodic current density (anodic polarization) at the potential below the oxygen evolution potential marks the beginning of localized corrosion, determined by the pitting potential value (E_{pit})[11,12].

The scanning rates used in this study were 0.167 mV/s (anodic) and 1.0 mV/s (cathodic), limiting the end of cathodic polarization to the potential of 300 mV above the OCP.

The threshold current density (i_s) for reversing the scanning from the anodic (anodic polarization) to the cathodic direction (cathodic polarization) was 5 mA/cm^2, according to ASTM G61-86[10].

All tests were performed in replicate and, in case of non-reproducibility of the first two CPP curves, the test was repeated as many times as necessary, until there was a pattern of similarity and agreement of the obtained curves.

Regarding the total number of tests, in the medium containing 3.5% NaCl, by weight (w/w), for AISI 304 steel, replicates were made (two repetitions) and, for AISI 420 steel, the measurements were performed in triplicate. In the medium containing 0.9% NaCl, by weight (w/w), measurements were performed in replicate for both steels.

For the purposes of graphical presentation of the results, one of the CPP curves was chosen.

All tests were conducted using an electrochemical cell with a test solution at 40°C, a temperature frequently recommended by manufacturers of enzymatic detergents, whose use is the most prevalent in SPDs in Brazil.

Before and after the polarization tests, the pH values of the solutions were measured using a previously calibrated pH meter (PG1800, Gehaka®, BR).

The electrolyte solutions used in the experimental groups of this study, both for AISI 304 and AISI 420 steels, were: saline solution (0.9% NaCl, by weight) and 3.5% NaCl solution, by weight.

At the end of the CPP tests, the specimens were photographed with a digital camera and sent for morphological characterization of the surface, using scanning electron microscopy, SEM (Vega 3, TESCAN, CZ).

In addition to the electrochemical and morphological characterization of stainless steels, this study aimed to analyze the *in vitro* biological response, using cell culture, which were exposed to AISI 304 and AISI 420 stainless steels, with and without pitting corrosion, following the protocols of the international standard ISO 10993-5[13].

For the cytotoxicity tests, 20 AISI 304 and AISI 420 steel specimens were used, distributed as follows:

- Group A — AISI 304 with pitting: five specimens;
- Group B — AISI 304 without pitting: five specimens;
- Group C — AISI 420 with pitting: five specimens;
- Group D — AISI 420 without pitting: five specimens.

All stainless steel specimens with pitting were cut by wire EDM in a square shape, measuring 3 mm, in the internal area of the o-ring, a rubber ring used to prevent leakage and delimit the exposed area of the steel during the electrochemical tests (Figure 2).

All samples with and without pitting were sent for cleaning and sterilization at the SPD of the University Hospital of Universidade de São Paulo (HU-USP). The material was washed in an ultrasonic washer at 40°C for 20 minutes with 2% enzymatic detergent (Neozime 5Ò), rinsed with reverse osmosis water and manually dried with compressed air.

Subsequently, it was visually inspected and packaged in medical grade paper/film and sterilized in a saturated steam

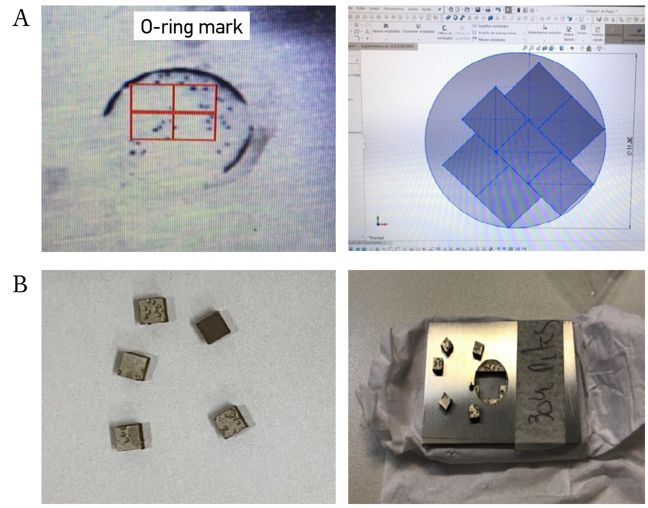

Figure 2. Measurement over the area delimited by the o-ring (A) and wire EDM cuts (B) of the specimens (3 mm x 3 mm) used for the cytotoxicity analysis.

autoclave under pressure validated in a cycle of 134°C for five minutes and monitored by type-6 chemical indicator control and three-hour biological reading indicator.

Then, the samples were sent to the Cell Culture Center of Instituto Adolfo Lutz.

The NCTC clone 929 cell line (CCIAL 020) was cultured in Minimal Essential Medium (MEM), supplemented with 0.1 mM of non-essential amino acids, 1.0 mM of sodium pyruvate, and 10% of fetal bovine serum without antibiotics (MEM with 10%FBS).

The resuspended cells were seeded in volumes of 5 mL in Petri dishes (concentration 3.0x105 cells/mL) and incubated for 48 hours at 37°C in a humid atmosphere, containing 5% carbon dioxide (CO_2).

The monolayer of cells already formed was added to the 5 mL Petri dish, of compound medium, of equal volumes of MEM twice concentrated and 1.8% agar, containing 0.01% neutral red vital dye. The agar was melted and mixed with the MEM at 44±1°C.

Using an aseptic technique, the sections of AISI 304 and AISI 420 stainless steel plates with and without pitting were deposited on the agar in the Petri dishes in order to evaluate possible cytotoxicity.

The Petri dishes were re-incubated in an incubator in an atmosphere with 5% CO_2 at 37°C for 24 hours.

As positive controls, 0.5 cm fragments of latex rubber, a material known to be cytotoxic, were used, and as negative controls, 0.5 cm fragments of non-toxic filter paper.

All tests were performed in quintuplicate and the results were interpreted according to the grades of biological reactivity for the agar diffusion method (ISO 10.993-5)[13], as shown in Chart 2.

RESULTS

The results will be presented in saline solution (0.9% NaCl w/w), 3.5% NaCl w/w solution, and (ci)toxicity tests.

a) Results obtained from the saline solution medium (0.9% NaCl w/w), as shown in Figure 3.

By analyzing the polarization curves (Figure 3), we obtained the mean values of corrosion potential ($E_{corr\ mean}$), pitting ($E_{pit\ mean}$), and the standard deviation (SD) for AISI 304 steel of, respectively, $E_{corr\ mean}$=0.077 V x Ag/AgCl/KCl sat (SD=0.033 V) and $E_{pit\ mean}$=0.567 V x Ag/AgCl/KCl sat (SD=0.047 V).

For the AISI 420 steel, the obtained values were: $E_{corr\ mean}$=0.118 VxAg/AgCl/KCl sat (SD=0.059 V) and $E_{pit\ mean}$=0.331 VxAg/AgCl/KCl sat (SD=0.036 V).

The photographic analysis of the surface of AISI 304 and AISI 420 steels, after polarization tests, is shown in Figure 4.

No pitting was observed on the surface of AISI 304 and AISI 420 steels (Figure 4), but these were detected by electron microscopy (SEM), as shown in Figure 5.

By the analysis of the SEM images (Figure 5), at the same magnification, we can observe pitting in greater amount, apparently deeper and located on the surface of AISI 420 steel. To detect pitting in the AISI 304 steel, a higher magnification was required (Figure 6).

With a higher magnification, there is a significant pitting corrosive attack on the surface of the AISI 420 steel compared to the AISI 304.

For the AISI 304 steel, the pH values in the 0.9% NaCl solution, by weight (w/w), were pH=6.05 (before the polarization test) and pH=6.45 (after the polarization test). For AISI 420 steel, the pH values in the 0.9% NaCl solution, by weight (w/w), were pH=6.05 (before the polarization test) and pH=6.46 (after the polarization test).

It should be noted that the emergence of pitting in both steels is expected after surveying the CPP curves, which show the E_{pit}, a potential in which the current density increases sharply.

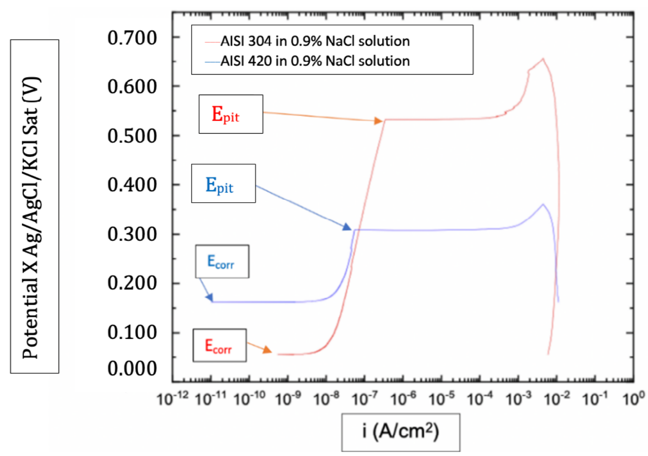

Figure 3. Potentiodynamic polarization curves for AISI 304 and AISI 420 steels in 0.9% NaCl solution, by weight, at 40°C.

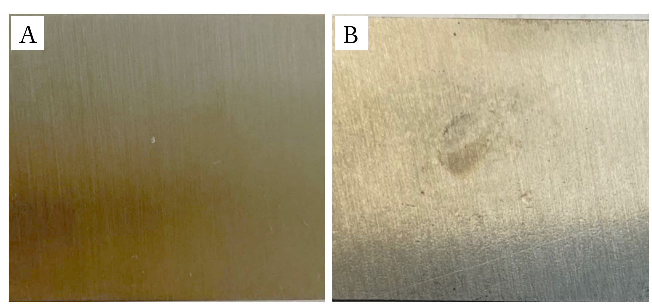

Figure 4. Appearance of the specimens after the potentiodynamic polarization test of stainless steels A) AISI 304 and B) AISI 420 in 0.9% NaCl solution, by weight, at 40°C.

Chart 2. Grades of biological reactivity for the cytotoxicity test by the agar diffusion method according to ISO 10993-5[13].

Grade	Reactivity	Description of the reactivity zone	Cytotoxic effect
0	None	Zone not detectable under or around the specimen.	Negative
1	Low	Some malformed or degenerated cells under the sample.	Negative
2	Mild	Zone limited to the area under the sample.	Negative
3	Moderate	Zone established up to 1 cm from the sample.	Positive
4	High	Zone established more than 1 cm from the sample.	Positive

b) Results obtained from 3.5% NaCl solution. The electrochemical characterization of the behavior of AISI 304 and AISI 420 steels in a medium containing a higher concentration of chlorides (3.5% NaCl, by weight) is shown in Figure 7.

According to the analysis of the polarization curves (Figure 7) for the AISI 304 steel, the mean values of the potentials were: $E_{corr\ mean}$=0.052 VxAg/AgCl/KCl sat (SD=0.006 V) and $E_{pit\ mean}$= 0.543 VxAg/AgCl/KCl sat (SD=0.011 V).

For the AISI 420 steel, the mean values of the potentials were $E_{corr\ mean}$=-0.338 VxAg/AgCl/KCl sat (SD= 0.018 V) and $E_{pit\ mean}$=-0.170 VxAg/AgCl/KCl sat (SD= 0.042 V).

After analyzing the polarization curves, the existing positive hysteresis, and the pitting potentials, we verified that the AISI 304 steel is more resistant to corrosion ($E_{pit\ mean}$=0.543 V, SD=0.011 V) than the AISI 420 steel ($E_{pit\ mean}$=-0.170 V, SD=0.042 V), because it has a more positive value of $E_{pit\ mean}$, demonstrating that the passivation layer is only locally broken at higher potential, better resisting the challenge imposed by the test.

For the AISI 304 steel, the pH values in the 3.5% NaCl solution, by weight (w/w), were pH=6.57 (before the polarization test) and pH=9.99 (after the polarization test).

For the AISI 420 steel, the pH values in the 3.5% NaCl solution, by weight (w/w), were pH=6.99 (before the polarization test) and pH=6.75 (after the polarization test).

(c) Cytotoxicity tests. Regarding cytotoxicity, the results of the tests showed a halo of cell death greater than 0.1 mm, corresponding to grade number 3, moderate reactivity[13], in all samples of AISI 304 steel with and without pitting (Figure 8).

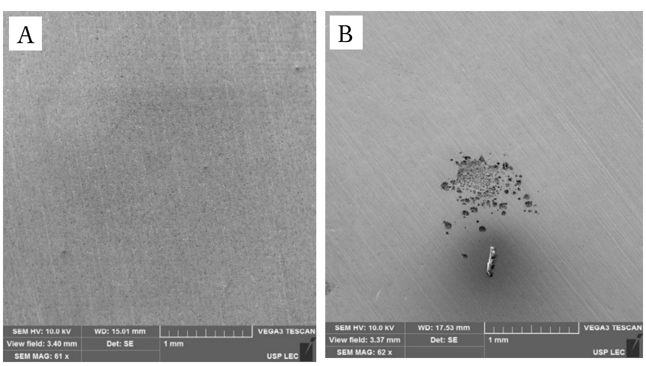

Figure 5. SEM images of the surface of stainless steel samples after the potentiodynamic polarization test, being A) AISI 304 and B) AISI 420, in 0.9% sodium chloride (NaCl) solution, by weight.

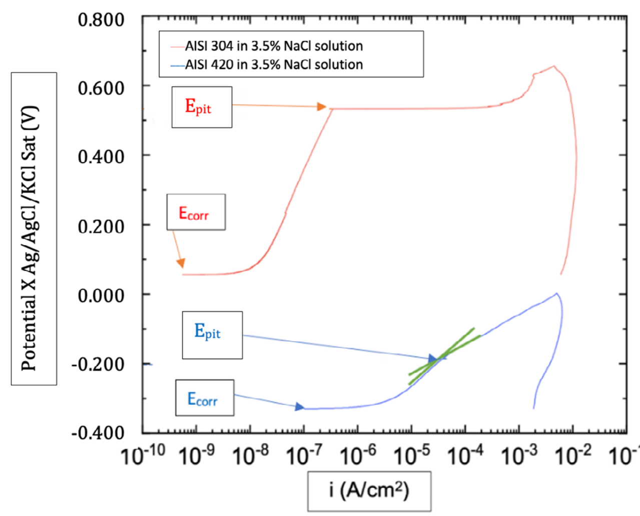

Figure 7. Cyclic potentiodynamic polarization curves for AISI 304 and AISI 420 steels in 3.5% NaCl solution, by weight, at 40°C.

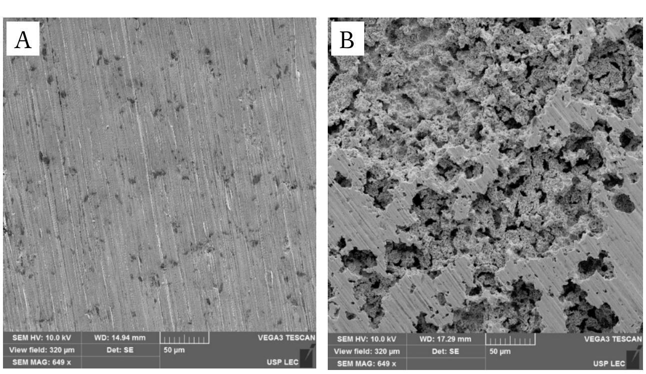

Figure 6. SEM images, after a potentiodynamic polarization test in stainless steels, being A) AISI 304 and B) AISI 420, in 0.9% sodium chloride (NaCl) solution, by weight, at 40°C.

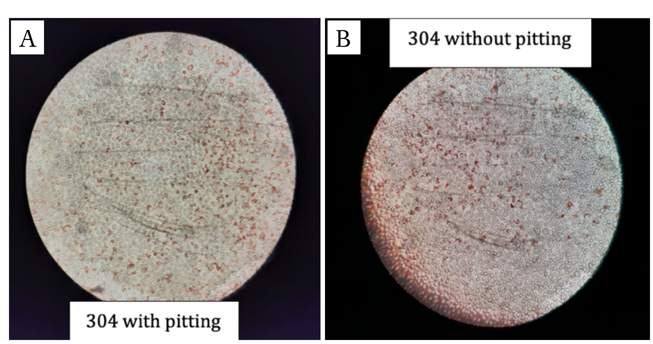

Figure 8. *In vitro* cytotoxicity test of AISI 304 steel samples, being A) with pitting and B) without pitting, showing grade-3 biological reactivity (cytotoxicity) in NCTC clone 929 cells, by agar diffusion method.

By analyzing the samples (Figures 8 and 9), we can observe living cells, stained with neutral red and cells from light areas, formed by dead cells, which did not incorporate the vital dye. This halo, due to its size (from 0.1 cm to 1.0 cm), was classified as grade 3 (moderate cytotoxicity) for both steels.

DISCUSSION

Pitting corrosion is often linked to defects in the metal surface, failures in the oxide layer, and the presence of aggressive anions in the media such as chlorides in the saline solutions used intraoperatively, organic matter, and body fluids[14,15]. These anions rupture the oxide layer located at preexisting weak points in the passive layer, thus initiating localized corrosion[14-20].

Therefore, maintaining good surgical instrumentation practices is essential. Healthcare professionals should keep surgical instruments clean, removing excess organic matter with sterile distilled water intraoperatively, avoiding leaving these materials immersed in saline solution when unnecessary.

By analyzing the most positive values of the pitting potentials of the AISI 304 steel, both in medium containing 0.9% NaCl solution, by weight (w/w), and in more concentrated solution, 3.5% NaCl, by weight (w/w), these demonstrate greater resistance to pitting corrosion when compared to the AISI 420 steel. The higher and the more positive the E_{pit} value, the more resistant the stainless steel will be to pitting corrosion in the analyzed medium.

The localized breakdown of the passive layer in more positive E_{pit} values, for the AISI 304 steel, corroborates findings of the literature, which show greater resistance of austenitic steel (AISI 304), which has, in its chemical composition, higher percentages of chromium (12.00–14.00%) and nickel (8.00–11.00%), which are protective alloying elements against corrosion, when compared to martensitic steel (AISI 420), whose concentration of alloying elements is lower: chromium (12.00–14.00%) and nickel (1.00%)[3].

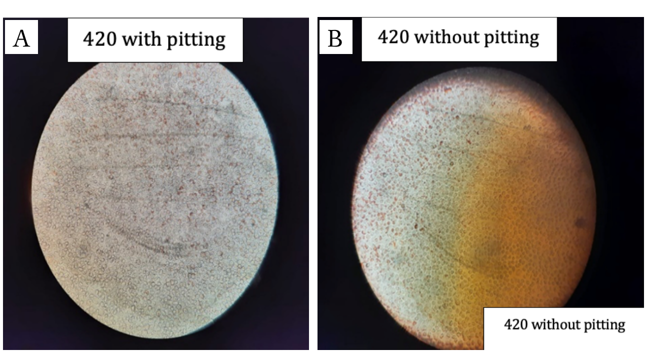

Figure 9. *In vitro* cytotoxicity test of AISI 420 steel samples, being A) with pitting and B) without pitting, showing grade-3 biological reactivity (cytotoxicity) in NCTC clone 929 cells, by agar diffusion method.

For the AISI 304 steel, the analysis of the evidence also demonstrated, during the immersion test, a reduction in the values of the pitting potentials, from E_{pit}=400 mV, at the concentration of 1 M of NaCl at 30°C, to E_{pit}=100 mV, when the concentration of the solution increased to 5 M of NaCl at 30°C[17].

The effect of saline solutions (0.9% NaCl) and fluorides (0.9% NaCl + 0.05% NaF) on the corrosive process of an AISI 316 L stainless steel was analyzed[19]. After 42-day immersion tests, the CPP curves were performed at a temperature of 37°C, with a scanning rate of 1 mV/s and a value of E_{pit}=0.300 mVxE_{ref} SCE (saturated calomel electrode) in the medium containing 0.9% NaCl + 0.05% NaF[19].

Although the stainless steel analyzed is more resistant to corrosion (AISI 316 L) compared to AISI 304, the presence of fluorides, in addition to 0.9% NaCl, justifies the low pitting potential found by the authors (E_{pit}=0.300 mVxE_{ref} SCE)[19].

Overall, when it comes to surface finish, the roughness of matte materials is greater than that of polished materials, which increases the possibility of pitting corrosion[20]. The steels analyzed in this study had a matte finish, a factor that could lead to lower pitting potential values.

The corrosion resistance of a Ni-Cr alloy, in a medium containing sodium chloride (100 ppm) and artificial tears (0.5% sodium salt of carboxymethylcellulose), at 37°C, was analyzed by Indian researchers, by means of polarization curve survey and electrochemical impedance spectroscopy[9]. In the medium containing artificial tears, the corrosion potential (E_{corr}) presented more negative values (E_{corr}=-269 mVxE_{ref} SCE) than the solution containing sodium chloride (E_{corr}=-94 mVxE_{ref} SCE)[9]. However, when analyzing the i_{corr}, which represents the corrosion rate, a lower value in the medium containing artificial tears (i_{corr}=1.306x10^{-8} A/cm^2) was verified compared to the medium containing sodium chloride (i_{corr}=1.617x10^{-8} A/cm^2), corroborating the destructive power of chlorides in metal alloys[9].

The degradation of surgical instruments versus water quality was the object of a study conducted by Japanese researchers[8]. They analyzed pitting corrosion in 279 surgical instruments and measured the concentration of chloride (Cl) and silica (SiO_2) in the two types of water used in the hospital:

reverse osmosis water and tap water[8]. The concentration of Cl in tap water was higher (0.7 mg/L) than in reverse osmosis water (0.1 mg/L). The concentration of SiO_2 was the same in both types (0.3 mg/L). Pitting corrosion was observed in 71% of the 279 analyzed instruments and it was directly associated with the highest concentration of chlorides[8].

Hence, we highlight the need to control the quality of the water used in healthcare services regarding the concentration of chlorides for cleaning, disinfection, and sterilization of surgical instruments.

By analyzing the polarization curves in the medium containing 3.5% NaCl, by weight (w/w), the curve obtained for the AISI 420 steel shows that it is not very easy to determine the pitting potential, as the current density does not increase so sharply ($E_{pit\ mean}$=-0.170 V X Ag/AgCl/KCl sat, SD=0.042 V). Taking this into consideration, we used the intersection of the line defined by the polarization curve (E x log i) before the sharp increase in current density, with the line defined by the curve after this increase — green lines (Figure 7).

In addition, in the medium containing 3.5% NaCl, by weight, there was a tendency to corrosion in cracks; therefore, considering the existence of cracks, pitting and cracking corrosion can occur simultaneously and, instead of classifying the potential found as pitting (E_{pit}), we prefer to call it as breakdown potential.

One of the hypotheses for the emergence of the cracks may be related to the long period in which the specimen remained polarized during the tests.

For the AISI 304 and AISI 420 steels, in the medium containing 0.9% NaCl, by weight (w/w), the determination of the E_{pit} was very clear, because the sharp increase in current indicated, unequivocally, the breakdown potential of the passive layer, a concentration of greater interest for the SPDs.

Regarding the greater pH variation in the AISI 304 test (initial pH=6.57/final pH=9.99), one of the hypotheses raised was the longer test time under polarization because, during the oxidation of the metal, the oxygen in the medium is reduced (cathodic reaction) to hydroxyl (OH^-), which increases the pH of the medium.

As for cytotoxicity, the grade 3 (moderate) results reinforce the employability of the AISI 304 and AISI 420 steels only as surgical instruments, as they were designed, and not as permanent implants.

Even though the cytotoxicity tests showed similar values in the samples with and without pitting, the use of corroded instruments can cause serious adverse events to patients, considering that corroded metal alloys are mechanically weakened, and may have cracks that first emerged inside the pitting, resulting in breakdown.

Therefore, if prolonged contact with human tissue is necessary, as in the case of orthoses and prostheses, the indication is to use the AISI 316L steel [16% chromium-0.03% carbon and 2% molybdenum], which has greater biocompatibility and resistance to corrosion[18].

One limitation of our study was the impossibility of testing stainless steels from different manufacturers and temperature degrees.

CONCLUSIONS

The austenitic steel AISI 304 showed higher corrosion resistance than the steel AISI 420 in the two studied media.

In the presence of chlorides, the mean values of pitting potentials in saline solution at 0.9% NaCl, by weight (w/w), for the AISI 304 steel were higher than for the AISI 420 steel.

By increasing the concentration of the NaCl solution from 0.9%, by weight, to 3.5% NaCl, by weight, a decrease in the mean value of the pitting potential for both steels was observed, evidencing a greater corrosive attack in this medium.

In the more concentrated saline solution (3.5% NaCl, by weight), there was a tendency to crack corrosion for both AISI 304 and AISI 420 steel, corroborating the greater aggressiveness of this medium.

The results of the electrochemical and morphological characterization tests corroborate the recommendations to avoid prolonged immersion, when unnecessary, of surgical instruments in saline solutions.

Intraoperatively, it is recommended to use sterile distilled water, instead of saline solution, to remove abrasions and dirt from the instruments. In addition, surgical instruments should be sent to the cleaning process in the SPD quickly in order to remove chlorides.

The analysis of the *in vitro* cellular cytotoxicity results showed that AISI 304 and AISI 420 steels present moderate cellular toxicity (grade 3), regardless of the presence of pitting corrosion.

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CONFLICT OF INTERESTS

The authors declare no conflicts of interest.

AUTHORS' CONTRIBUTIONS

WAJ: Project management, Formal analysis, Conceptualization, Data curation, Investigation, Methodology, Resources, Writing — original draft, Writing — review & editing, Software, Supervision, Validation, Visualization. BPS: Project Administration, Formal analysis, Conceptualization, Data curation, Investigation, Methodology, Writing — original draft, Writing — review & editing, Software. ASC: Project administration, Formal analysis, Conceptualization, Data curation, Investigation, Methodology, Resources, Writing – original draft, Writing – review & editing. KUG: Formal analysis, Conceptualization, Data curation, Investigation, Methodology, Writing — original draft, Writing — review & editing, Supervision, Validation. IVA: Project administration, Formal analysis, Conceptualization, Data curation, Investigation, Methodology, Resources, Writing — original draft, Writing — review & editing, Software, Supervision, Validation, Visualization.

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