

Electrochemical corrosion and bacterial adhesion study of Two Osteosynthetic Maxillofacial Bone Plates

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Abstract

Objective: To evaluate the novel locally manufactured osteosynthetic titanium bone mini plates used for mandibular fracture fixation, and to compare it with an international brand to have an alternative to expensive plates.

Methods: The study was conducted at National University of Science and Technology from Jan 2013 to June 2013. Local and German brands of osteosynthetic titanium bone mini plates were studied electrochemically through Tafel extrapolation curves using Gamry® electrochemical framework in modified simulated body fluid prepared with pH 7.4 at 37°C. For bacterial adhesion, staphylococcus aureus bacterial culture of 50 µl was used with an OD600 of 1.0 corresponding to approximately 1.79×10⁹ cells ml⁻¹. Data was analysed using SPSS 20.

Results: Corrosion resistance behaviour of local and German plates was not significantly different ($p > 0.05$), but in case of bacterial adhesion the local plates showed significantly low adhesion compared to the imported material ($p < 0.05$). Overall, the biocompatible properties of local plates met international brand qualities.

Conclusion: Electrochemical corrosion and bacterial adhesion of local osteosynthetic maxillofacial bone plates matched the quality of an international brand.

Keywords: Biocompatible, Corrosion, Simulated body fluids, Osteosynthetic plates.
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Introduction

The incidence of trauma involving maxillofacial region is increasing at faster rate nowadays because of a number of reasons, such as scientific progress, large number of vehicles, armed wars and urban lifestyle.¹ The fixation of maxillofacial fractures nowadays is most frequently achieved by utilising bone plates and screws because they offer a number of advantages over other methods used for fracture fixation such as fixation beneath periosteum to maintain enough blood supply, multiple points for fixation, inhibition of rotational movements of fractured fragments, maintaining dimensions of face, rapid healing of fractures, and in the end, the patient does not need to undergo intermaxillary fixation for weeks.²⁻⁴

In Pakistan, osteofixation plates used for maxillofacial

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surgeries are either imported from abroad or are manufactured locally. The plates imported from other countries, such as Germany, are used more than the local plates. Imported bone plates have undergone extensive research and their every aspect is examined intensively. But unfortunately locally manufactured plates are still unexplored in terms of chemical composition, corrosion resistance and bacterial adhesion.

The current study was planned to determine and compare corrosion and bacterial adhesion of plates manufactured in Pakistan (MPP) and imported commercially pure titanium bone plates from Germany (MOG). It was hypothesised that there was no difference between the two.

Materials and Methods

The study was conducted at National University of Science and Technology from Jan 2013 to June 2013. Osteosynthetic titanium bone mini plates were used as MPG (Surgiline Company Germany) and MPP (Moin International Suppliers, Sialkot, Pakistan). The dimension

Table-1: Composition of modified simulated body fluid.

Compounds	Reagents	Amount
Sodium Chloride	NaCl	5.403g
Sodium Bicarbonate	NaHCO ₃	0.504
Sodium carbonate	Na ₂ CO ₃	0.426
Potassium Chloride	KCl	0.225g
Potassium DiHydrogen Phosphate	NaH ₂ PO ₄ .3H ₂ O	0.230g
Magnesium Chloride hexahydrate	MgCl ₂ .6H ₂ O	0.311g
Sodium hydroxide	0.2 M-NaOH	100ml
Calcium Chloride	CaCl ₂	0.293g
Sodium sulphate	Na ₂ SO ₄	0.072g
Sodium hydroxide	1.0 M-NaOH	15ml
2-(4-(2-hydroxyethyl)-1-piperazinyl) ethanesulfonic acid	HEPES	17.892g

for each sample was 6x3x1mm.

Modified simulated body fluid (m-SBF) was prepared as described in literature⁵ by dissolving reagent in deionised water (Table 1), which is almost equal to those of human blood plasma, except bicarbonate (HCO₃⁻) ion. All the reagents and compounds were supplied by the Inorganic Chemistry laboratory of the National University of Science and Technology (NUST), Islamabad, Pakistan.

Strain 8432-D of staphylococcus aureus was used for checking bacterial adhesion of MPG and MPP. The strain was obtained from Atta Ur Rahman School of Applied Biosciences (ASAB), NUST.

A copper wire was soldered to the back of each sample using aluminium solder, and a glass tube covered that wire in order to ensure the electric contact. The samples were mounted in epoxy resin after soldering. Prior to the start of each experiment, the mounted samples were mechanically ground with emery paper up to 600 grit (MetkonGripo 2V Grinder-Polisher, China) and then wet-polished using 0.5u alumina powder (MP-2B Grinder Polisher, China).

The exposed areas of samples were washed in distilled water followed by ultrasonic cleaning in acetone. Electrochemical studies were conducted using G350/700 Potentiostat (USA) interfaced to a computer. The electrolyte used was m-SBF. The potential of hydrogen (pH) of the solution was maintained at 7.4. Freshly prepared solution was used for each experiment. Corrosion polarisation studies were conducted in a bottom polarisation cell. All the potentials were in reference to saturated calomel electrode (SCE). Potentiodynamic polarisation studies were conducted after stabilisation of free corrosion potential. The scan

rate used was 0.166mV/s. The corrosion rate was determined using Tafel extrapolation method in Gamry Potentiostat. The corrosion current (mA) data was plotted against the potential (V).

Staphylococcus aureus 8325-4 was grown in brain heart infusion to an optical density (OD₆₀₀) of around 1 at 37°C. Colony-forming unit (CFU) was assessed to quantitatively measure colonies formed on the plates. For scanning electron microscope (SEM) analyses, each sample with adherent bacterial cells after three hours of incubation was fixed with a 2.5% glutaraldehyde solution in 0.1 M cacodilate buffer for one hour at 4°C, washed with cacodilate buffer and then dehydrated through a graded series of ethanol (25%, 50%, 75% and 100%). Samples were kept in 100% ethanol for 15 minutes and then critical point dried. The specimens, mounted onto aluminium stubs, were sputter coated with gold. Samples with adherent bacteria were observed at X100, X5000 and X10000 magnification respectively. SPSS 20 was used for data analysis.

Results

In MPG samples, the cathodic curve started from -200mV and anodic curve started to build from -1000V. These two curves got united at point E (-600mV) which is the corrosion current (Figure 1).

For MPP the cathodic curve started from -100mv and anodic curve started to form from 0.000mv. Both these curves united at -300mv (point E) (Figure 2).

From this corrosion current the corrosion rate was calculated through Gamry software Echem. The mean corrosion values and standard deviation for MPG and MPP were 0.037±0.031 and 0.021±0.024. The corrosion of MPP was not significantly different from MPG (p=0.45). I corr, E corr and corrosion rate values were noted (Table 2).

In case of bacterial adhesion CFU experiment, the mean values for MPG and MPP were 118± 5.887 and 18± 9.011 respectively. Both CFU and SEM analysis confirmed that there was significantly (p=0.000) more bacterial adhesion on MPG compared to MPP. SEM images of MPG and MPP without inoculums (control) and with inoculums were

Table-2: E Corr, I Corr and Corrosion rate of Pakistani (MPP) and German brands (MPG).

Bone plates	Ecorr(mV)	Icorr(nA)	Corrosion rate (mills per year)
MPG	0.292	10.69	0.035
MPP	0.236	17.80	0.021

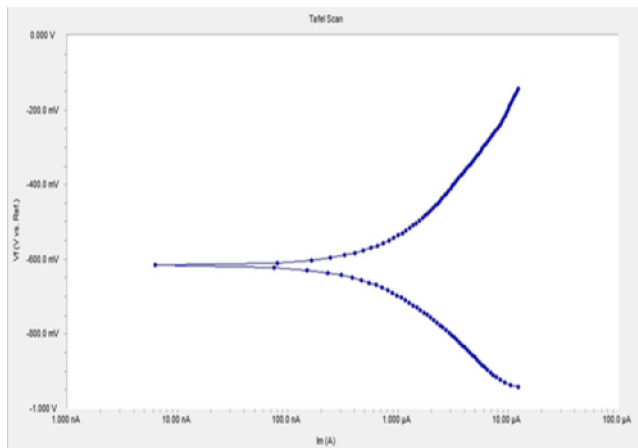


Figure-1: Tafel curve for German brand (MPG).

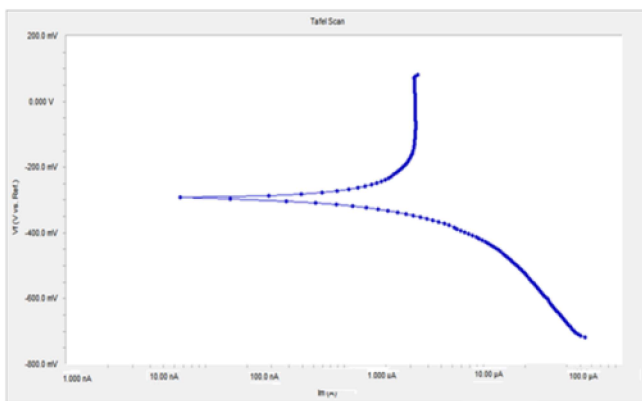


Figure-2: Tafel curve for Pakistani brand (MPP).

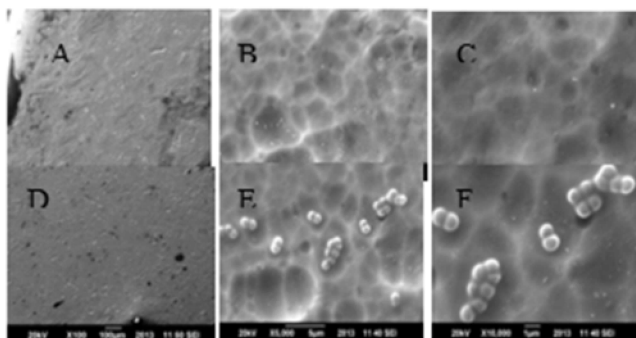


Figure-3: Scanning electron microscope photographs of German brand (MPG) (A, B, C) Scanning electron microscope (SEM) images with no inoculums (control) at X100, X5000 and X10000 magnifications (D) SEM image with inoculums at X100 magnification, surface is clear with no visible staphylococcus aureus growth at this magnification (E) in this image staphylococcus aureus colonies are clearly visible with their typical shape at X5000 magnification while in (F) at X10000 magnification the colonies are much more significantly prominent.

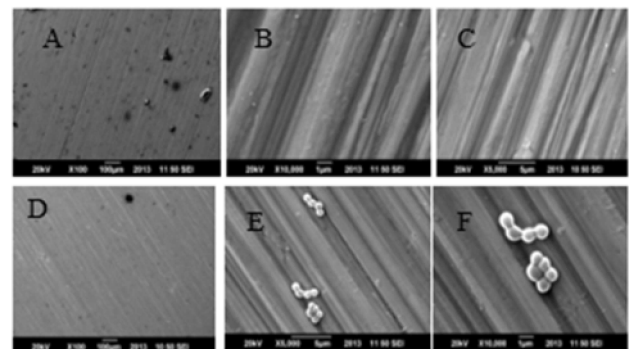


Figure-4: Scanning electron microscope photographs of Pakistani brand (MPP) (A, B, C). Scanning electron microscope (SEM) images with no inoculums (control) at X100, X5000 and X10000 magnifications (D) SEM image with inoculums at X100 magnification, surface is clear with no visible staphylococcus aureus growth at this magnification (E) in this image staphylococcus aureus colonies are clearly visible with their typical shape at X5000 magnification while in (F) at X10000 magnification the colonies are much more significantly prominent.

analysed at magnification X100, X5000 and X10000. The control samples contained no inoculum. No staphylococcus aureus was observed on the surface of samples at magnification of X100, while as the magnification was increased up to X5000, the typical cocci shape of staphylococcus aureus were clearly observed on the samples.

MPG samples showed staphylococcus aureus in grape-like clusters and seemed to be embedded in anodised pores and depressions, the number of cocci adhered were more compared with MPP, while the number of cocci were less. Moreover, the surface of MPP was deprived of anodised pores of oxygen (Figure 3-4).

Discussion

Biomedical safety such as corrosion resistance and antibacterial property are basic requirements of bone plates. In this study locally manufactured and imported from Germany commercially pure titanium mini plates purchased from the local market were compared. The above mentioned properties of these two brands were analysed and compared with each other through various characterisations and standard methods.

All the metals used for fabrication of implants such as bone plates and screws have problem of corrosion. Titanium is usually resistant to the general corrosion, pitting attack, and crevice corrosion that may occur in other metal implants as a result of attack from aggressive organic fluids.⁶

Corrosion was assessed in vitro on MPP and MPG before implantation. The corrosion rates determined were similar to those mentioned in literature.^{7,8}

The free corrosion potential was stabilised in the same way for both plates. On immersion the potential moved towards noble potential and stabilised in a relatively short time. The behaviour of polarisation curves indicated that all the samples immediately passivated on immersion in mSBF.

The breakdown of passive films results in increased corrosion. MPG's passivity was slightly different from that of MPP as its passive film was stable for a little more time than MPP's passive film. This probably seems to be a result of thicker anodisation layer on surface of MPG samples. The potential versus current density curves for corrosion obtained in this study were comparable with previous studies.⁹⁻¹²

The current study also compared MPP and MPG in terms of bacterial adhesion to see which plate had more tendencies for infection. CFU studies showed significantly high adhesion ($p=0.000$) with MPG compared to MPP. SEM analysis also confirmed this finding. It has been established that surface roughness of material is the principle factor responsible for bacterial adhesion, so maybe this more roughness was responsible for more adhesion of bacteria on MPG.¹³

Conclusion

Electrochemical corrosion and bacterial adhesion of local osteosynthetic maxillofacial bone plates matched the quality of an international brand. The finding is beneficial for poor patients of facial fractures who cannot afford expensive imported plates compared to local plates.

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Conflict of Interest: None.

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