Chapter 8
Surface Characteristics and In Vitro Corrosion Behavior of HAp-coated 316L Stainless Steel for Biomedical Applications

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1 Introduction

The study of biomaterials is going on since many decades. The need for safe and efficient biomaterials is rising tremendously because of increase in number of accidents causing fractured human bones, joint replacement, etc. The basic requirement of biomedical implants is the existences of proper physiological environment inside the body, strength, wear resistance, biological fixation, cell growth and corrosion resistance [1]. The surface interface and its properties determine the acceptance or rejection of the biomedical implants on the body [2–4].

The quality of machined surface highly affects the bioactive properties like corrosion resistance, wear resistance and cell proliferation of the metallic bio-implants [5, 6]. Success of biomaterials is based on the characteristics such as topography, chemical composition, mechanical strength, corrosion resistance and surface roughness.

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Hence, many surface modification and surface coating techniques are being used nowadays to enhance the required properties of biomedical implants. Modifications of biomaterials also overcome the risk of rejection due to loosening of joint, toxicity, infection within the individual providing proper cell adhesion, cell proliferations and antibacterial properties [7–10].

Among all of the biomaterials, i.e., polymeric, ceramic, natural, metallic and composites, metallic biomaterials are widely acceptable due to their mechanical properties and compatibility with the human bone. Titanium alloys, 316L stainless steel, chromium and cobalt are the metallic biomaterials; amid all, 316L stainless steel because of their easy availability, low cost, better adhesion and cell growth preferred much more [11–15]. But the presence of sodium, chlorine, water, saliva and amino acids within the human body disturbs the equilibrium state and consumes the metallic implant by various anodic and/or cathodic reactions [16–18]. As a result, metallic biomaterial implant corrodes after certain years of implantation. In order to overcome such issue, coating of bioactive layer on surface is best solution which not only evades early corrosion but also avoids release of harmful ions [19].

Electric discharge coating (EDC) is an application of well-known electrical discharge machine (EDM) by reversing the polarity during the machining process. Reverse polarity alters the flow of electrons compared to conventional process and deposits the material mixed in dielectric to modify the surface characteristics of the workpiece [20–23]. The intermetallic compounds and carbides thus formed enhance the properties of substrate biomaterial offering better adhesion and proliferation of human osteoblast-like MG-63 cells.

2 Literature Review

Among numerous studies on metallic biomaterials, Mahajan and Sidhu [24] reviewed the need for surface modification of biomaterials for the enhancement of their functionality. The surface modification of metallic biomaterials, i.e., stainless steel, titanium using EDM, helps to obtain the biocompatible surfaces, increases bond ability, etc. EDM is the method that has the ability to replicate the architecture and characteristics of the natural bones. It had great applications in biomedical fields for the enrichment of required surface characteristics. Other researchers like Prakash et al. [25] also expressed EDM as a potential and new innovative way for surface modification of various metallic implants for orthopedic applications. According to the in vitro bioactivity analysis, powder-mixed dielectric machined surface offers better cell growth than substrate and surface machined without the addition of powder in dielectric [26].

The surface of 316L stainless steel was treated by Mazur et al. [27] utilizing sol–gel technique to deposit ceramic layer of SiO_2–Y_2O_3. Newly formed ceramic layer helped to enhance the bioactivity and corrosion resistance of 316L which was confirmed by EDS and Raman spectra analysis. Compounds like calcium (Ca) and phosphorous (P) were observed confirming the formation of apatite ceramic layer on stainless steel 316L with improved biocompatibility. Electron beam physical vapor deposition (EBPVD) was employed by Kaliaraj et al. [28] to modify 316L stainless steel surface with monoclinic (m-ZrO_2) and tetragonal (t-ZrO_2) phase of zirconium dioxide. They found improved cell viability during the cell culture analysis of coated samples. Superior corrosion resistance was shown by coated samples of 316L stainless steel in artificial blood plasma (ABP) as electrolyte using electrochemical test.

Abbas et al. [29] investigated the present research scenario of electrical discharge machining, suggesting PMEDM as dominating non-conventional machining process for the improvement of surface characteristics with the addition of powder in dielectric medium. Here, high MMR, better surface finish and low TWR can be obtained. The surface characteristics of aluminum powder-mixed ED machined Ti-6Al-4V was examined by Abdul-Rani et al. [30] for the formation of intermetallic compounds and carbon enriched layer on the substrate surface which helps in osseointegration and bone proliferations. Furthermore, modified surface exhibited enhanced corrosion resistance of titanium alloy.

Singh et al. [31] investigated the effect of input process parameters on MRR by machining AISI 316L stainless steel using EDM process. Current was found as the most significant factor influencing MRR. Highest MRR of 6.25 mg/min was obtained at the combination of current 28 A, T_{on} 90 μs, T_{off} 60 μs and voltage 80 V. The performance of traditional EDM process with the addition of Al powder using reverse polarity was studied by Sharma et al. [32]. Their investigation showed that various characteristics of powder like concentration, size, etc., had its dominating effect on the performance of electric discharge machine.

Karamian et al. [33] coated HAp/zircon on stainless steel 316L and concluded that powder coating helps to improve adhesion properties of metallic biomaterial. The investigation of HAp/TiO_2 coating on the titanium substrate was performed by Ramires et al. [34] for improving the bioactive properties such as osseointegration and cell proliferations. Manam et al. [35] intensely reviewed the biocompatibility of metallic biomaterials, i.e., titanium alloys, stainless steel and chromium–cobalt alloys. They studied that corrosion resistance of biomaterial was an important factor. Moreover, corrosive nature causes the failure of implantation and leads to surgery. However, surface modification of biomaterials with bioactive layer offered better corrosion resistance and evaded the harmful ions to release. They concluded that metallic biomaterials could not be replaced with polymers and ceramics due to the required mechanical properties for a material to be biomaterial.

Bains et al. [36] and Long et al. [37] optimized the MRR by employing PMEDM on using copper and graphite tool with titanium and SiC-mixed dielectric. They concluded that powder concentration increases metal erosion as compared to no powder in dielectric with discharge current as one of the most significant factors followed by other variables.

In this research, EDC was employed on 316L stainless steel with hydroxyapatite nanoparticles-mixed dielectric to examine improved surface characteristics in terms of surface roughness and formation of bioactive compounds on 316L surface. Additionally, in vitro corrosion analysis was performed on machined province to scrutinize the improved corrosion resistance of HAp-coated 316L stainless steel surface.

3 Methodology

3.1 Materials

Medical grade 316L stainless steel was procured from Metline Industries, Mumbai, for the experimentation in the form of rectangular plate (size: 50 mm × 100 mm × 5 mm). Electrolytic copper tool because of its conductivity with dia. 900 μm was chosen as electrode for the ED machining of 316L SS. The compositional analysis of 316L stainless steel is listed in Table 1.

Hydroxyapatite nanopowder with average particle size 20–45 nm and purity of 99.5% mixed in dielectric medium for the surface modification of 316L stainless steel.

3.2 Design of Experiment

Taguchi's orthogonal arrays are generally executed to reduce the number of experimental trials according to selected machining parameters. In the present work, L_{18} $(2^1 \times 3^4)$ was used for conducting the experiments selecting five input parameters, i.e., dielectric medium, discharge current, pulse-on-time, pulse-off-time and voltage. Minitab-17 was utilized to generate design of experiments with three levels of input parameter as shown in Table 2.

Table 1 Chemical composition of 316L stainless steel

Element	C	Mn	Si	Cr	Mo	Ni	S	N	P	Fe
%	0.03	2.00	0.75	16.00	2.00	10.00	0.03	0.10	0.045	Balance

Table 2 Parameters and their levels

Input parameters	Units	Level 1	Level 2	Level 3
Dielectric medium	–	EDM oil	EDM oil + HAp	–
Discharge current	Ampere	20	24	28
Pulse-on-time	μ-seconds	60	90	120
Pulse-off-time	μ-seconds	60	90	120
Voltage	Volts	40	60	80

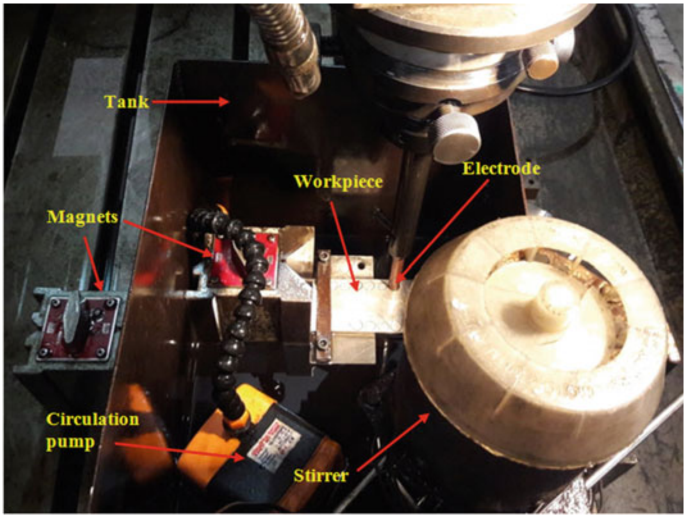


Fig. 1 Experimental setup

3.3 Experimentation

Die sinker-type electrical discharge machine with reverse polarity {workpiece as cathode (−) and electrode as anode (+)} was employed for conducting the experimentation. Out of 18 experimental runs, nine were performed in pure dielectric, i.e., EDM oil, whereas for powder-mixed experimentation, indigenously developed setup (shown in Fig. 1) consisting of stirrer and pump for proper and homogeneous mixing of HAp particles was used. Machining time for each run was fixed for 40 min with powder concentration of 15 g/l. After the ED machining, Mitutoyo SJ-400 surface roughness tester was used to measure roughness (R_a) of machined surface. Each sample measured diametrically from three locations and was averaged for further analysis.

In this experimental investigation, dielectric medium, discharge current (I), pulse-on-time (T_{on}), pulse-off-time (T_{off}) and Voltage (V) were selected as input parameters being the most common and influencing parameters during the machining process. The selected output response was surface roughness (R_a) in µm.

3.4 In Vitro Corrosion Analysis

Electrochemical corrosion test was performed to validate the enhanced corrosion resistance and subsequently low corrosion rate of modified 316L stainless steel surface with HAp particles. Ringer solution at 37 °C was used as simulated body fluid (SBF) to replicate the human body environment as electrolyte immersing 316L sample as working electrode (WE), whereas platinum wire as counter electrode (CE) and Ag/AgCl employed as reference electrode (RE). Prior to analysis, specimen was bathed in acetone and insulated with tape leaving an exposed machined area of 0.32 cm^2. The insulation restricts the contribution of base metal during the electrochemical testing.

4 Results and Discussion

The experimental design matrix and surface roughness of machined specimen are shown in Table 3. The surface response table is represented according to signal-to-noise ratio (S/N) methodology—a ratio of strength of signal to the magnitude of error. S/N ratios depend on the type of responses measured such as "Larger is better" type which is given by the equation below:

$$(\mathrm{S}/\mathrm{N})_{\mathrm{Larger}} = -10\log\left\{\frac{1}{R}\sum_{i=1}^{R}\left(\frac{1}{y_i^2}\right)\right\}$$

where R = number of repetitions of responses; y_i = value of response at ith trial.

4.1 Analysis of Variance for S/N Ratios of Surface Roughness

The surface response of machined workpiece was analyzed using analysis of variance (ANOVA) of S/N ratios as shown in Table 4 with the aid of Minitab-17 software. Momentous process parameters affecting the roughness of machined 316L stainless steel were recognized using p-value and subsequently their percentage contribution. Pulse-on-time was the most dominating parameter affecting surface roughness (contribution 33.40%) followed by discharge current (contribution 24.52%) and dielectric medium (contribution 16.81%). Consequently, the desired porous surface for proper adhesion, cell growth validating the surface modification of selected biomaterial examined in HAp powder-mixed dielectric at higher value of on-time (i.e., 90 µs) and peak current (i.e., 28 A) which concurrently endow with the desired surface topography and phase transformation of 316L stainless steel for requisite bioactivity. Figure 2 illustrates the S/N ratio plot for surface roughness.

Further, powder-mixed ED machined 316L stainless steel surface was examined for changed morphology, formation of intermetallic compounds and in vitro corrosion analysis to validate the surface modification with bioactive powder and enhanced bioactivity.

Table 3 Experimental design and surface roughness response

Exp. No.	Dielectric medium	I (A)	T_{on} (μs)	T_{off} (μs)	Voltage (V)	Surface roughness (μm)			
						$R1$	$R2$	$R3$	S/N ratio
1	EDM oil	20	60	60	40	0.46	0.15	0.72	−12.3129
2	EDM oil	20	90	90	60	0.27	0.41	0.73	−8.5618
3	EDM oil	20	120	120	80	0.75	0.87	0.86	−1.7134
4	EDM oil	24	60	60	60	0.44	0.62	0.66	−5.2558
5	EDM oil	24	90	90	80	0.43	0.24	0.19	−12.2591
6	EDM oil	24	120	120	40	1.12	1.01	0.96	0.2037
7	EDM oil	28	60	90	40	0.59	0.76	0.82	−3.0759
8	EDM oil	28	90	120	60	0.51	0.49	0.64	−5.4204
9	EDM oil	28	120	60	80	1.09	1.18	1.22	1.2848
10	EDM oil + HAp powder	20	60	120	80	0.58	0.50	0.78	−4.5820
11	EDM oil + HAp powder	20	90	60	40	0.26	1.08	0.93	−7.4835
12	EDM oil + HAp powder	20	120	90	60	1.04	1.24	0.87	0.1531
13	EDM oil + HAp powder	24	60	90	80	0.69	0.94	0.54	−3.4645
14	EDM oil + HAp powder	24	90	120	40	0.56	0.68	0.75	−3.7592
15	EDM oil + HAp powder	24	120	60	60	0.89	1.13	1.19	0.3751
16	EDM oil + HAp powder	28	60	120	60	1.37	1.26	0.77	0.2238
17	EDM oil + HAp powder	28	90	60	80	1.76	1.29	1.52	3.4468
18	EDM oil + HAp powder	28	120	90	40	1.25	0.87	1.21	0.5483

R Repetitions

Table 4 Analysis of variance for S/N ratios of surface roughness

Source	DF	Seq SS	Adj SS	Adj MS	*p*-value	% contribution
Dielectric medium	1	58.929	58.929	58.929	0.032*	16.81
Discharge current (A), *I*	2	85.986	85.986	42.993	0.041*	24.52
Pulse-on-time (µs), T_{on}	2	117.103	117.103	58.551	0.020*	33.40
Pulse-off-time (µs), T_{off}	2	11.329	11.329	5.665	0.549	3.24
Voltage (V)	2	7.218	7.218	3.609	0.676	2.05
Residual error	8	70.083	70.083	8.760		19.98
Total	17	350.648				100

*Significant at 99% confidence level, Rank 1: pulse-on, Rank 2: current, Rank 3: dielectric

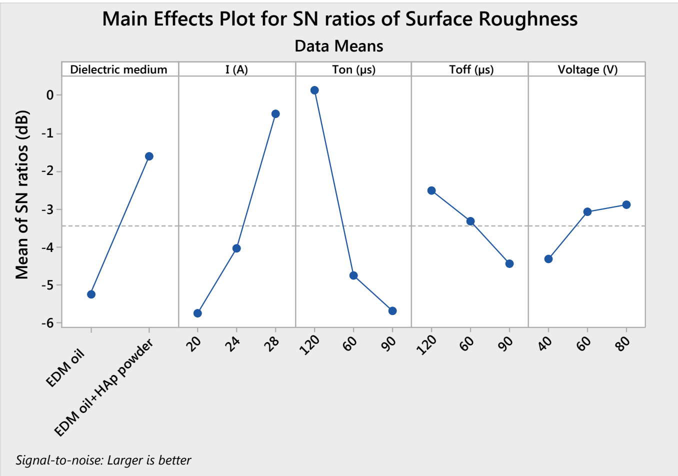


Fig. 2 Main effects plot for S/N ratios of surface roughness

4.2 *Surface Morphology of Machined Surface*

Scanning electron microscopy (JSM-6610 LV Joel, Japan) was used to inspect the morphology of machined 316L specimen in hydroxyapatite nanopowder-mixed dielectric medium. Deposition of powder particles and presence of homogeneous porous structure surface encouraging cell growth, adhesion between bone and implant, and cell proliferation were revealed by SEM (Fig. 3) analysis.

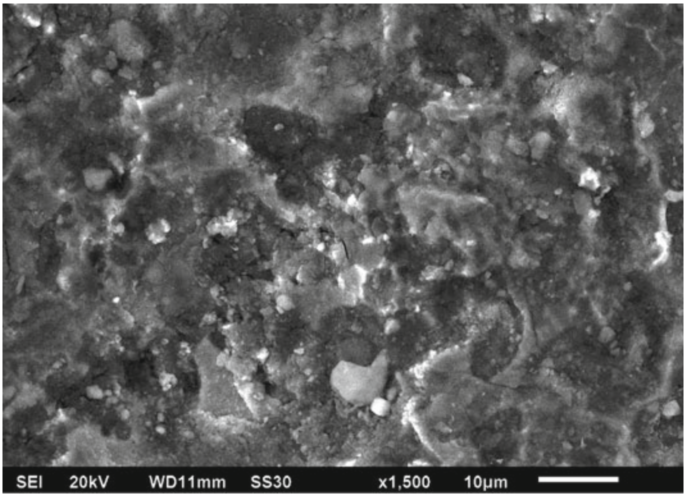


Fig. 3 Surface representing porous structure at $I = 28$ A; $T_{on} = 90$ µs; $T_{off} = 60$ µs; voltage = 80 V

4.3 Phase Transformation of Machined Surface

It was described in the previous research [38, 39] that sparks produced during ED machining changed the chemical composition of the machined surface. X-ray diffraction (XRD) technique was used to investigate the formation of intermetallic and bioactive compounds on ED machined 316L stainless steel surface (trial 17). Figure 4 witnesses the XRD pattern confirming the formation and deposition of intermetallic compounds, carbides as well as silicides on the EDMed surface. Newly formed

Fig. 4 XRD pattern showing formation of new compounds

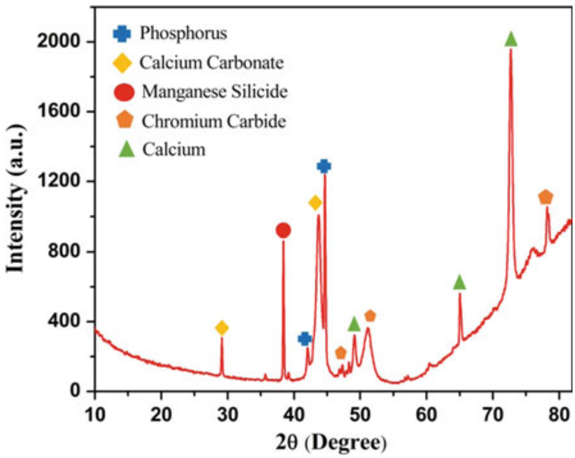
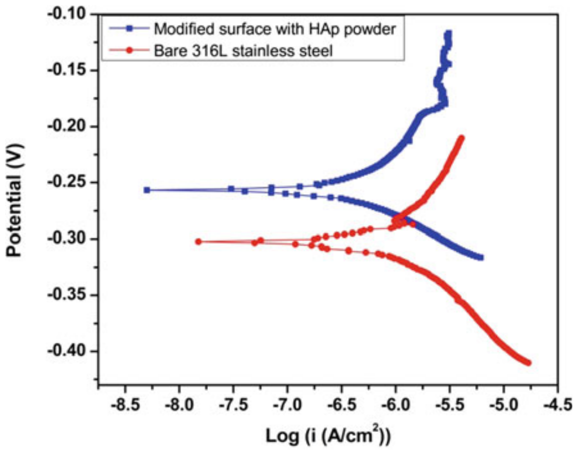


Fig. 5 Tafel plot showing polarization curve of specimens (trial 17)



bioactive compounds contribute for enhanced bioactivity in terms of improved corrosion resistance, proper adhesion between bone and implant, and cell growth.

4.4 In Vitro Corrosion Analysis

After the examination of porous structure and compositional analysis using SEM and XRD, respectively, electrochemical corrosion analysis was carried out to validate the response of modified surface in terms of enhanced corrosion resistance. Ringer's solution with pH value 7.2 at 37 °C was used as simulated body fluid (SBF) to reproduce the human body environment for the current investigation.

It was evident from the Tafel plot (Fig. 5) that surface modified with HAp powder-mixed dielectric depicts higher value of corrosion potential ($E_{corr} = -257.810$ mV) compared to bare metal possess −308.490 mV. Consequently, modified surface of 316L stainless steel showed improved corrosion rate of 0.0972 mm/year compared to substrate material with corrosion rate of 1.79 mm/year. The newly modified ED machined 316L surface exhibited improved corrosion resistance that is necessitated for better and proper bone–implant adhesion. It will avoid the release of metallic ions due to the presence of enzymes and reacting environment within the individual causing poor osseointegration and cytotoxicity.

5 Conclusions

Present work analyzed the surface modification of medical grade 316L stainless steel using electric discharge coating (EDC) in HAp powder-mixed dielectric. Based on the surface characteristics and in vitro corrosion analysis, following conclusions were drawn:

1. Surface roughness directly increased with the increase in pulse-on and current applied. Desired porous surface (1.523 μm) was observed at 28A; T_{on} 90 μs; T_{off} 60 μs; and 80 V in the presence of HAp in the dielectric medium.
2. SEM revealed porosity and deposition of powder particles on HAp powder-mixed dielectric machined surface.
3. XRD confirmed the phase transformation of surface (trial 17) with the formation of bioactive compounds (calcium, phosphorus, calcium carbonate) and several intermetallic compounds.
4. Surface machined at higher discharge current, i.e., 28 A sounds more crowded with intermetallic compounds and carbides compared to surface machined at lower value of discharge current.
5. In vitro corrosion analysis exhibited improved corrosion resistance and accordingly low corrosion rate of HAp powder modified 316L stainless steel surface (0.0972 mm/year) compared to bare metal (1.79 mm/year).
6. Enhanced surface characteristics and corrosion resistance validate the surface modification of 316L with HAp powder using EDC for biomedical applications.

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