

Article

# Experimental Investigation on Bio-Machining of Nickel, Titanium and Nitinol (Shape Memory Alloys) Using *Acidithiobacillus ferrooxidans* Microorganisms

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**Abstract:** Micromachining plays a vital role in the manufacturing industry in producing micro-components with high sensitivity and fine dimensional tolerances for implant materials in medical applications. Micro-machining can be carried out through various machining processes like physical, chemical and biological processes, although the use of biological machining is limited. In biological machining, microorganisms are used as a source of energy to machine the components, and machining with microorganism brings a lot of advantages in the machining process like the production of components with lower energy resources, low cost, no heat-affected zone and fine dimensional tolerances, which makes it suitable for machining implant materials. In other machining process like conventional and unconventional machining processes, the heat-affected zone, dimensional tolerances and environmental-related problems are the major issues, as these processes generate more heat while machining. This damages the material, which will not be able to be used for certain applications, and this issue can be overcome by bio-machining. In this present work, nickel, titanium and nitinol are manufactured using the powder metallurgy technique. They are manufactured as a 10 mm diameter and 5 mm thick pellet. The fabricated nickel, titanium and nitinol shape memory alloys are machined with *Acidithiobacillus ferrooxidans* microorganisms to obtain a better material removal rate and surface roughness and to check the bio-machining performance by considering various parameters such as shaking speed, temperature, pH and percentage of ferric content for the future scope of biomedical applications. Considering these parameters, microorganisms play a vital role in the temperature, shaking speed and time of the bio-machining process, and it was observed that a better material removal rate and surface roughness are achieved at a temperature of 30 °C, shaking speed of 140 rpm and machining time of 72 h.

**Keywords:** shape memory alloys (Ni, Ti, NiTi); *Acidithiobacillus ferrooxidans*; shaking speed; temperature; pH; ferric content (%)



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## 1. Introduction

The machining of a metal workpiece is usually carried out through a conventional or non-conventional machining process. From components to other devices to joint implants, many of them require the machining of medical-grade materials. This includes the unique challenges of machining titanium to extreme levels of accuracy [1,2]. Titanium has many

useful features including excellent corrosion resistance and strength, high biocompatibility and non-magnetic properties, making it useful for magnetic resonance imaging (MRI) application. With these advantageous features, titanium is an optimum material for medical devices [3,4].

Nowadays, orthopaedics is one of the fastest-growing sectors in medical device manufacturing, and it includes reconstructive devices like spinal implants, arthroscopy and knee replacement [5,6]. These devices use manufacturing processes such as machining, casting, polishing, metal injection moulding and rapid manufacturing. For these reasons, medical devices continue to become smaller and smaller [6,7]. Features and components of a few microns require specialist processes of micromanufacturing, which include micromachining and micromoulding. The use of nanotechnology in medical devices and medicine is expected to grow. The powder metallurgy technique seems to be particularly advantageous as pores can originate from the particle compacting arrangement. The porous structure also presents adequate mechanical strength, as large pores have a deleterious effect on the mechanical properties of biomedical implants. The gradient of maximum porosity must be adjusted with respect to porosity and pore size in order to ensure an implant's suitable mechanical strength. Powder metallurgy is also used for the production of titanium parts for medical applications that are close to the final size, resulting in reduced machining operations and fabrication costs [8,9]. It is also used for the forming of complex shapes/composites with uniform microstructure and requires a few or no secondary operations, making it cost- and time-efficient. Dimensional deviations are low and tolerances are quite high with this method. A high production rate is another advantage of this method. Titanium-based alloys can be produced with infiltration and impregnation of other materials with different physical and mechanical properties such as hardness, strength, density and porosity, producing parts that have compatibility with human organs with a low scrap rate [3,9].

Non-conventional machining processes are fast and efficient methods, but they also have some disadvantages, such as heat generation, substantial amount of waste, dimensional tolerances and major environmental and health problems while machining. Generally, titanium reduces the strength of the yield, so less energy and force are needed when the heat is generated. It is also easier to perform and results in increases in ductility and the removal of or reduction in chemical inhomogeneities due to the elevated temperature and diffusion involved. Controlling this size has a positive effect on cell adhesion and biocompatibility. The hot work of titanium is performed at a slow rate, which increases the production time and cost [3,10].

By using wire electrical discharge machining (EDM), we can achieve tolerances of  $\pm 0.0001''$  on parts up to 12'' in length, 10'' in width and 8'' in height. This type of work is typically low-volume, made from hardened materials and requires a high degree of precision. In terms of the environmental issues of one of the non-conventional machining processes like EDM, there are several hazard potentials like hazardous smoke and vapours, electromagnetic radiation, hydrocarbon dielectrics and sharp-edge metallic particles that damage the skin.

The machining of NiTi shape memory alloys is very difficult because of the rapid work hardening behaviour of the material. Machining in a milling machine would be especially tedious because of the frequent failure of cutting tool materials. In the case of medical applications, the necessity of the closer dimensional tolerance is high, and achieving this closer tolerance through the conventional machining process is difficult. Machining with non-conventional machining process like EDM and laser machining can yield better results like closer dimensional tolerance and surface finish, but the heat-affected zone cannot be eliminated and, therefore, secondary processes are required to overcome the issues. To overcome these issues from various machining processes, bio-machining is a novel machining process that finds its place owing to its excellent characteristics, such as the production of components with lower energy resources, low cost and no heat-affected zone. Although bio-machining processes used to machine metals using microorganisms have

been reported [11,12], the major issue found is the heat-affected zone in the case of physical and chemical micromachining processes, and the use of microorganisms as a tool to remove metal is a relatively new manufacturing technique to overcome the issues that occur in other traditional machining processes [13,14]. The machining of grooves on pure iron and pure copper using *Acidithiobacillus ferrooxidans* as investigated experimentally shows that the depth of the grooves generated with respect to the machining time and the removal rate for copper was larger than for iron [15]. Surface changes and material removal rate (MRR) occurred for copper blocks with *Acidithiobacillus ferrooxidans* microorganisms, which have a slow material removal rate, and also with respect to machining time [15], and a flat removal instead of grooves. This will be applied for precision machining applications [16].

The current work reveals the manufacturing of nickel, titanium and nitinol (shape memory alloys) using powder metallurgy, and these were machined with *Acidithiobacillus ferromagnetes*. MRR and surface roughness (Ra) are measured. It should be noted that this kind of research, with titanium or nitinol involvement, has not previously been described in the literature. Previous experiments were conducted for metals like copper, aluminium, nickel and zinc using different microorganisms like *Acidithiobacillus ferrooxidans*, *Acidithiobacillus thiooxidans*, *Aspergillus niger*, etc., to determine the MRR and Ra using various process parameters.

This work seems very promising in terms of the application prospects [17,18]. Because of the lack of previous works about titanium, the obtained results were compared to other metals where bio-machining processes were applied.

Copper, aluminium and nickel were machined using *Acidithiobacillus ferrooxidans* by considering how the cell concentration enhanced the machining rate. The machining relates to the mechanism of sulphur oxidation, and metal dissolution occurs at the cell surface. The specific MRR is measured using the normalization of the surface area, and the removal of nickel occurs more than to copper and aluminium. This high MRR of nickel occurs due to the minimum of microbial stress exposure, and the removal of toxic  $\text{Cu}^{2+}$  needs to be taken in account to obtain a better machining efficiency compared to copper and aluminium when the microbial stress is maximized [19,20]. The bacterial culture supernatant in this study can replace the toxic ferric chloride used for the chemical etching of copper. The biologically produced ferric ions take part in the metal dissolution process, and during this process, the ferric ions are converted into ferrous ions. These ferrous ions can be reused for the growth of bacteria [20]. The material removal clearly indicates that an indirect mechanism exists that is similar to the bio-machining process [19]. Thus, a cleaner machining process can be developed [21]. The specific MRR was observed for copper, nickel and aluminium with *Acidithiobacillus ferrooxidans* through direct or indirect mechanism. A scanning electron microscope (SEM) was used to analyse the surface of a copper workpiece before and after oxidation (before and after the bio-machining process), and during the oxidation of copper, the changes in surface roughness and surface appearance were observed [22].

An indirect leaching mechanism was employed to recover silver from a silver oxide zinc button cell battery using a metal solubilisation technique similar to that of [21], which indicates 98% silver dissolved during the bioleaching process by *Acidithiobacillus ferrooxidans*, which might be implemented to develop a two-stage reactor system [23]. One study investigated the material removal rate and surface appearance of copper workpieces with *Acidithiobacillus ferrooxidans* corresponding to 6, 12 and 18 h of machining time. The material removal rate (MRR) is inversely proportional to the machining time because the decrease in ferrous sulphate and the increase in  $\text{Cu}^{2+}$  and hydrolysis could cause the activity of bacteria to be reduced, thus affecting the MRR [24].

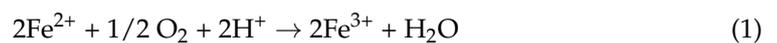
The micro-feature fabrication of copper with *Acidithiobacillus ferrooxidans* is used to produce micro-sized features such as lines, circles, rectangles and combination features. Copper with a size of 3  $\mu\text{m}$  results from the bio-machining process, which is near to the theoretical resolution of bacterial size (1  $\mu\text{m}$ ) [25]. The MRR of enzymatic machining using glucose with copper for 10 h was analysed. The enzymatic MRR is linear with time by adjusting the reaction time [26]. Based on previous studies [15,16], bacterial concentration

is one of the essential parameters during bio-machining to obtain a better removal rate and surface finish for some specified applications [27]. Experimental investigation is not only applicable for exact results, but analytical approaches like Taguchi design, etc., should also be used for identifying better results. The Taguchi design of experiments is used to establish the most influential parameters for a better material removal rate [28]. Successful results of the parameters like temperature, shaking rate and pH value are highly dependent on the material used such as copper, aluminium, zinc, nickel, titanium, etc. [29]. Previous studies were well supported by considering the parameters in order to avoid the progressive decrease in MRR with respect to time [30]. One previous study aimed to address the MRR by using aluminium with *Acidithiobacillus ferrooxidans* and *Aspergillus niger* by considering the cell concentration [31,32]. The results of [32,33] were in agreement with [23] in terms of the metal removal mechanisms due to the metabolic activity of *Acidithiobacillus ferrooxidans*, which is explained by the micro galvanic corrosion through oxidation [32,33].

## 2. Materials and Methods

### 2.1. Bio-machining Mechanism

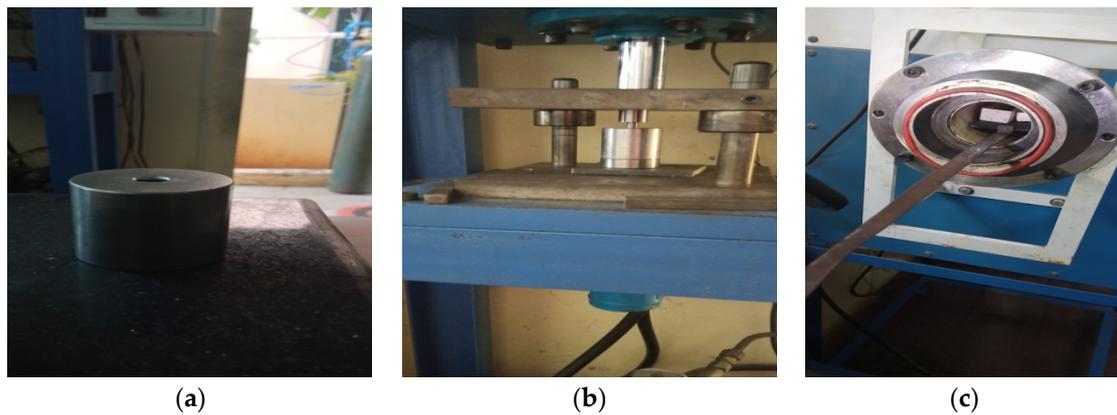
*Acidithiobacillus ferrooxidans* is a rod-shaped bacterium about 1  $\mu\text{m}$  long and 0.5  $\mu\text{m}$  in diameter. *Acidithiobacillus ferrooxidans* metal removal occurs in two stages, namely the direct and indirect mechanisms. In the direct mechanism, the microorganisms and metal are in direct contact through the extracellular polymeric substance, and enzymic activity is responsible for metal oxidation and reduction. In the second stage, the ferrous/ ferric ions dissolve from the surface of the workpiece and there is no direct contact between the metal and workpiece. The indirect mechanism is a cyclic combination of biological and chemical processes, and this mechanism takes two steps. Initially, the reaction starts with ferrous ion  $\text{Fe}^{2+}$  being oxidized into ferric ion  $\text{Fe}^{3+}$  by the microorganisms in order to gain energy. Then, ferric ion acts as an oxidizing agent for metal dissolution, and this reaction yields  $\text{Fe}^{2+}$  from the bacteria. The goal of these two processes is to produce useable energy for the bacteria [21]. Meanwhile,  $\text{H}^+$  ions are consumed continuously, and water is produced. Hence, it can be concluded that the metabolic activity of bacteria can be improved by tuning process conditions such as temperature, shaking rate and  $\text{H}^+$  ions (pH).



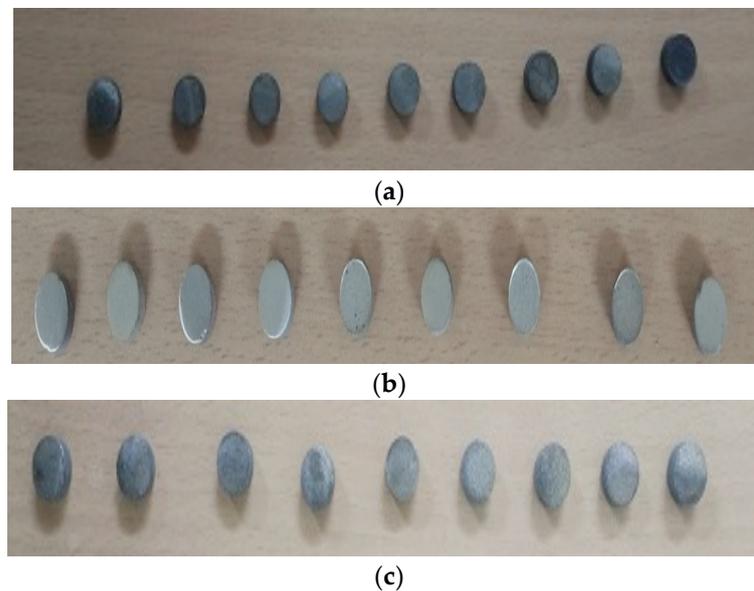
### 2.2. Preparation of Shape Memory Alloys (SMA)

Nickel, titanium and nitinol specimens required for performing bio-machining processes based on the process parameters were fabricated through the powder metallurgy process. The required quantity of nickel, titanium and nitinol powders were purchased from Parshwamani metals (Mumbai, India) with 99.2 %purity. The most suitable specimen size for bio-machining was selected and fixed as 10 mm diameter and 5 mm thickness. To fabricate the expected size, the required quantity of powder particles is measured and poured into an EN32 oil-hardened cylindrical steel die. The compaction pressure range selected based on suitable compaction pressure for the powder metallurgy operation is fixed at 400 MPa [34]. After the compaction process, the green specimen is removed and sintered in a sintering furnace, as shown in Figure 1a–c.

The sintering is carried out in a tubular furnace at 1164 °C for nickel (Ni), 1334 °C for titanium (Ti) and 1040 °C for nitinol (NiTi). After the sintering process, the specimen is removed and cooled. Examples are shown in Figure 2a–c.



**Figure 1.** Process of sample preparation: (a) die and samples: 10 mm × 5 mm; (b) compaction; (c) sintering process.



**Figure 2.** Prepared samples: (a) nickel pellet of 10 mm × 5 mm; (b) titanium pellet of 10 mm × 5 mm; (c) nitinol pellet of 10 mm × 5 mm.

### 2.3. Preparation of *Acidithiobacillus ferrooxidans* Microorganisms

*Acidithiobacillus ferrooxidans* was purchased from SOM Phytopharma Limited (Hyderabad, India). The culture we receive is in liquid form and is packed in plastic cans.

A pure culture of *Acidithiobacillus ferrooxidans* microorganisms is used for this biomachining process. This culture is originally isolated from the medium of 250 mL, which contains various amounts of basal salts like ammonium sulphate—0.875 g, potassium chloride—0.029 g, magnesium sulphate—0.0114 g, calcium nitrate—0.0016 g and dipotassium hydrogen phosphate—0.0145 g and 250 mL distilled water. Then, the medium is sterilized by placing the 250 mL conical flask in an autoclave at 121 °C for 15 min. After sterilization, the culture in this medium was inoculated by pouring in 1 mL of *Acidithiobacillus ferrooxidans* microorganisms using a micropipette. This culture is incubated at 30 °C under stationary conditions for 24 h or kept in a shaker at 140 rpm for 1–2 days until the solution becomes light yellow and semitransparent on the surface of the medium, which will be identified as growth of *Acidithiobacillus ferrooxidans*. All of these operations are conducted under the laminar chamber of a bacterial hood to avoid contamination [15].

### 3. Results

#### Bio-Machining Process

The surface of the work samples was polished with different grits of silicon carbide (SiC) paper. Before bio-machining, the work samples were cleaned using ethanol, then dried and weighed. The culture was taken and the workpiece was placed in *Acidithiobacillus ferrooxidans* culture in a laminar chamber, a closed chamber to avoid contamination.

All the prepared conical flasks were sealed with cotton and placed in a shaker, and different parameters were set: shaking speed 140 rpm, temperature 20 °C, 25 °C, 30 °C, pH value 1.8, 2.0, 2.5 and presence of ferrous 25%, 50% and 75%. Every 24 h, the samples were taken out from the flasks and dried at room temperature. The weight of the samples after machining was measured and is reported in Tables 1–3. The weight of the samples before and after the experiment was measured using an analytical digital weighing balance with an accuracy of 0.0001 g. The following mathematical expression is used to find the MRR (3).

$$MRR = (\text{Initial weight of the work sample } (m_i) - \text{Final weight of the sample } (m_f)), \quad (3)$$

By using these different ranges of parameters, higher and better material removal rates were identified, and they are reported in Tables 1–3. A Zeiss TSK—Surfcom touch 50A contact type roughness tester was used to measure the surface roughness (Ra) value with the evaluation length of 5 mm [16]. Three trials were performed, and the average values are reported in Tables 1–3.

**Table 1.** MRR and Ra of nickel with *Acidithiobacillus ferrooxidans*.

No.	Shaking Rate (RPM)	Temperature (°C)	pH	Ferric Content (%)	Initial Weight (g)	MRR (g)			Ra (µm)
						24 h	48 h	72 h	
1	140	20 °C	1.8	25	2.5449	0.0321	0.0459	0.0802	1.84
2	140		2.0	50	2.2994	0.0192	0.0264	0.1226	1.02
3	140		2.5	75	2.3091	0.0087	0.0093	0.1079	1.23
4	140	25 °C	1.8	75	2.0411	0.0073	0.0154	0.0486	1.52
5	140		2.0	25	2.507	0.0325	0.0397	0.0864	1.04
6	140		2.5	50	2.3348	0.0061	0.0174	0.0315	0.55
7	140	30 °C	1.8	50	2.3829	0.0044	0.0166	0.1217	<b>0.33</b>
8	<b>140</b>		2.0	<b>75</b>	<b>2.5835</b>	<b>0.0092</b>	<b>0.0396</b>	<b>0.2342</b>	0.41
9	140		2.5	25	2.3883	0.0019	0.0228	0.2179	0.73

**Table 2.** MRR and Ra of titanium with *Acidithiobacillus ferrooxidans*.

No.	Shaking Rate (RPM)	Temperature (°C)	pH	Ferric Content (%)	Initial Weight (g)	MRR (g)			Ra (µm)
						24 h	48 h	72 h	
1	140	20 °C	1.8	25	1.243	0.2479	0.2678	0.4109	2.31
2	140		2.0	50	0.944	0.342	0.1591	0.428	2.58
3	140		2.5	75	1.164	0.2211	0.2221	0.3186	2.32
4	140	25 °C	1.8	75	1.0242	0.0086	0.0171	0.0386	1.54
5	140		2.0	25	1.0349	0.0056	0.0247	0.3286	1.80
6	140		2.5	50	1.0311	0.0325	0.2485	0.3791	1.19
7	140	30 °C	1.8	50	1.0165	0.0168	0.0172	0.2782	1.99
8	<b>140</b>		2.0	<b>75</b>	1.0621	0.0765	0.3297	0.4091	<b>1.55</b>
9	140		2.5	25	<b>1.3356</b>	<b>0.2304</b>	<b>0.3394</b>	<b>0.4594</b>	3.08

**Table 3.** MRR and Ra of nitinol with *Acidithiobacillus ferrooxidans*.

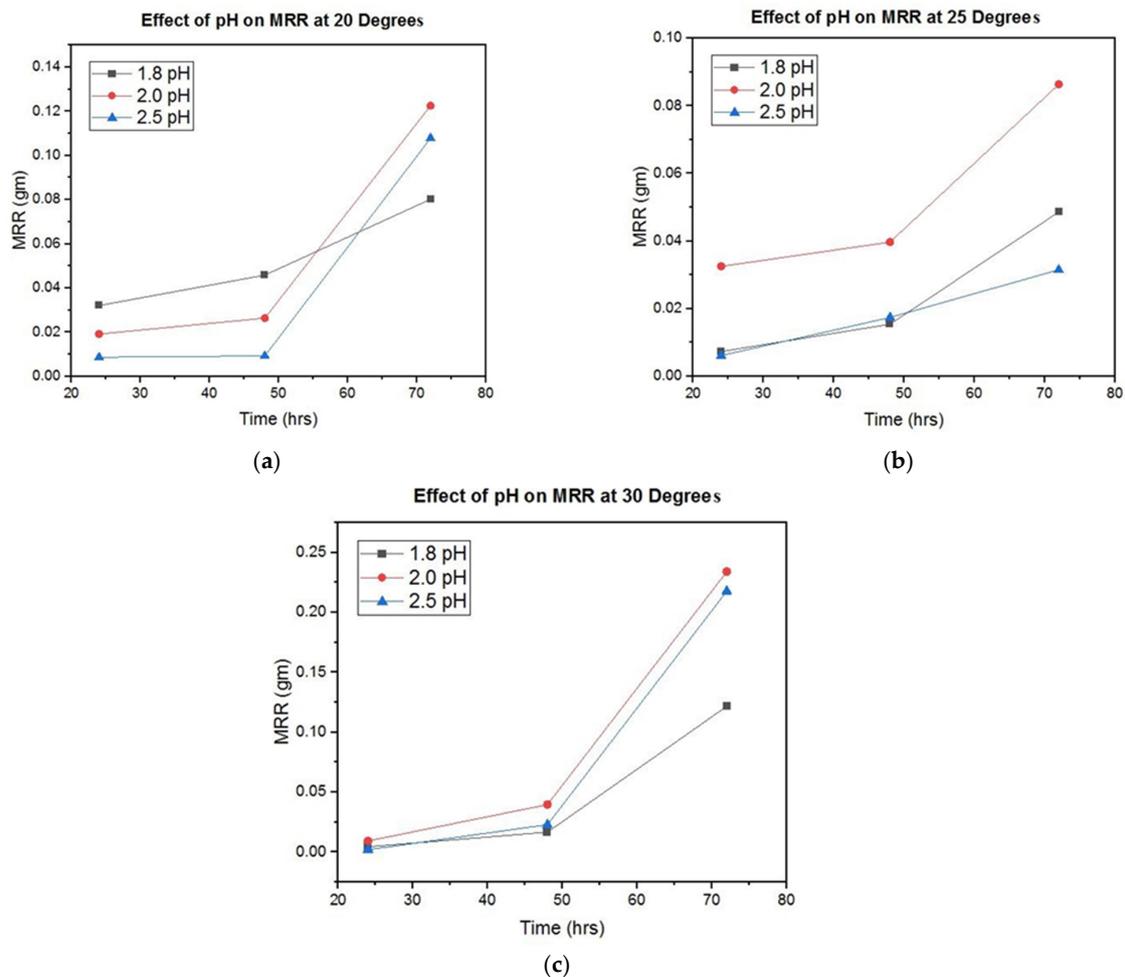
No.	Shaking Rate (RPM)	Temperature (°C)	pH	Ferric Content (%)	Initial Weight (g)	MRR (g)			Ra (µm)
						24 h	48 h	72 h	
1	140	20 °C	1.8	25	2.3321	0.0587	0.1287	0.2547	1.92
2	140		2.0	50	2.1547	0.1874	0.2354	0.3587	1.09
3	140		2.5	75	2.3587	0.1654	0.1952	0.2842	1.32
4	140	25 °C	1.8	75	2.1254	0.1985	0.2365	0.2987	1.67
5	140		2.0	25	2.6324	0.2417	0.2986	0.3254	1.09
6	140		2.5	50	2.3875	0.2586	0.3541	0.3826	1.57
7	140	30 °C	1.8	50	<b>2.4578</b>	<b>0.3247</b>	<b>0.4812</b>	<b>0.5854</b>	<b>0.98</b>
8	140		2.0	75	2.1879	0.3954	0.4892	0.5214	1.54
9	140		2.5	25	2.5205	0.4125	0.5112	0.4991	1.72

With the help of the observed results, the effect of process parameters on MRR and Ra is explained.

**4. Results and Discussion**

**4.1. Effect of pH on MRR—Nickel**

In Figure 3, the parameters obtained for nickel are presented.



**Figure 3.** Effect of pH on MRR—nickel: (a) machining time vs. MRR at 20 °C; (b) machining time vs. MRR at 25 °C; (c) machining time vs. MRR at 30 °C.

In Figure 3a–c, an increase in MRR corresponding to the machining time (h) with respect to pH values and ferric concentration can be observed, and the maximum metal removal rate of 0.2342 g occurs at a temperature of 30 °C, a pH value of 2.0 and a ferric content of 75% for 72 h of bio-machining.

4.2. Effect of pH on MRR—Titanium

In Figure 4, the parameters obtained for titanium are presented.

In Figure 4, at a pH value of 1.8 and 2.5, there is an increase in MRR. At a pH of 2.0, the MRR decreases from 0.342 g to 0.1591 g from 24 h to 48 h, and the maximum metal removal of 0.428 g was reached for 72 h of bio-machining. The decrease in MRR can be eliminated by adding H<sup>+</sup> ions in the form of H<sub>2</sub>SO<sub>4</sub> to compensate for the H<sup>+</sup> ions consumed during bio-machining.

Figure 4b,c shows an increase in MRR corresponding to machining time (h) with respect to pH values and ferric concentration, and the maximum metal removal rate of 0.4594 g occurs at a temperature of 30 °C, pH value of 2.5 and ferric content of 25% for 72 h of bio-machining.

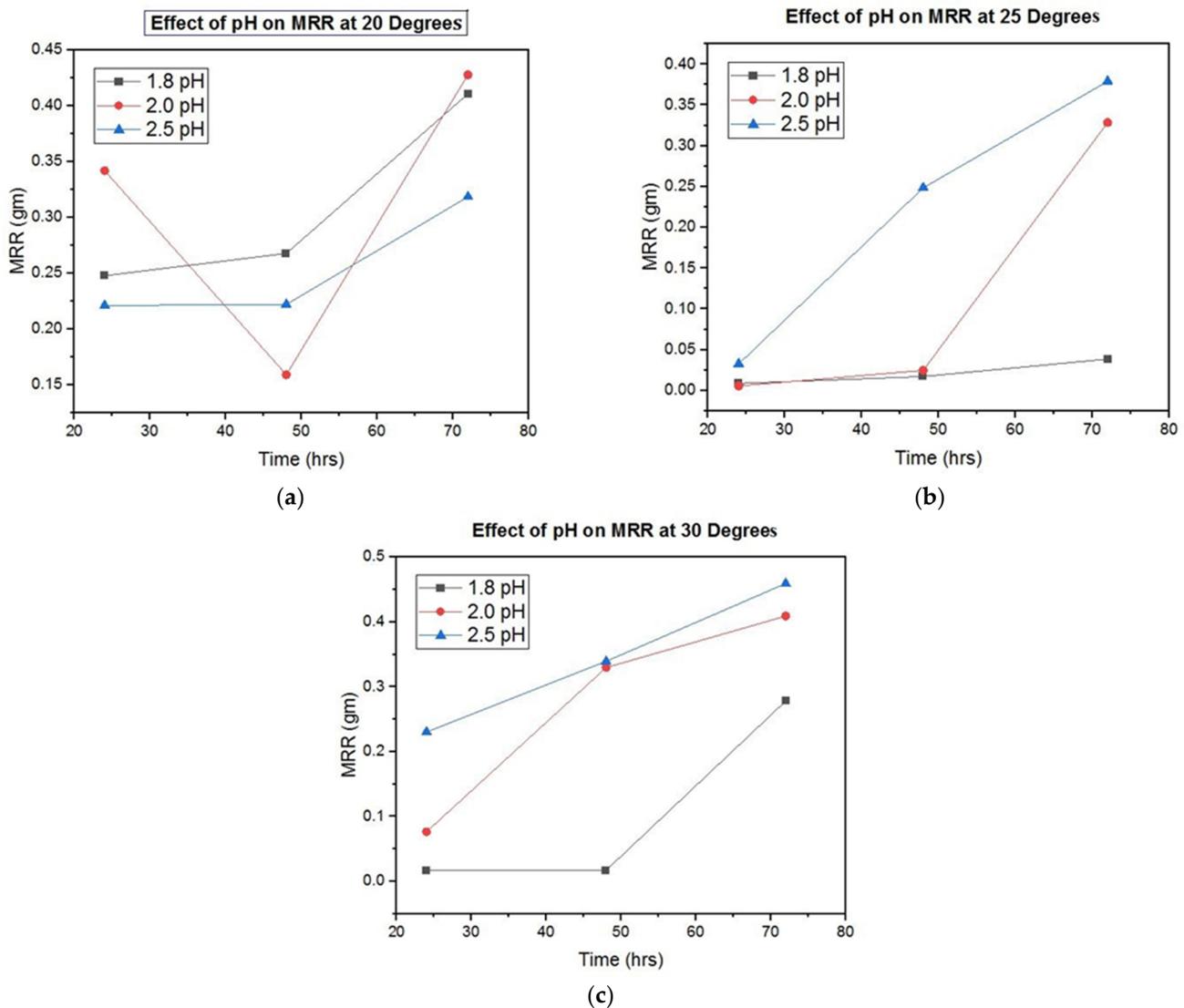


Figure 4. Effect of pH on MRR—titanium: (a) machining time vs. MRR at 20 °C; (b) machining time vs. MRR at 25 °C; (c) machining time vs. MRR at 30 °C.

4.3. Effect of pH on MRR—Nitinol

In Figure 5, the parameters obtained for nitinol are presented.

Figure 5a–c shows that an increase in MRR can be observed corresponding to machining time (h) with respect to pH values and ferric concentration, and the maximum metal removal rate of 0.5854 g occurs at a temperature of 30 °C, pH value of 1.8 and ferric content of 50% for 72 h of bio-machining.

4.4. Effect of pH on Surface Roughness

In Figure 6, the surface roughness results obtained for nickel, titanium and nitinol are presented.

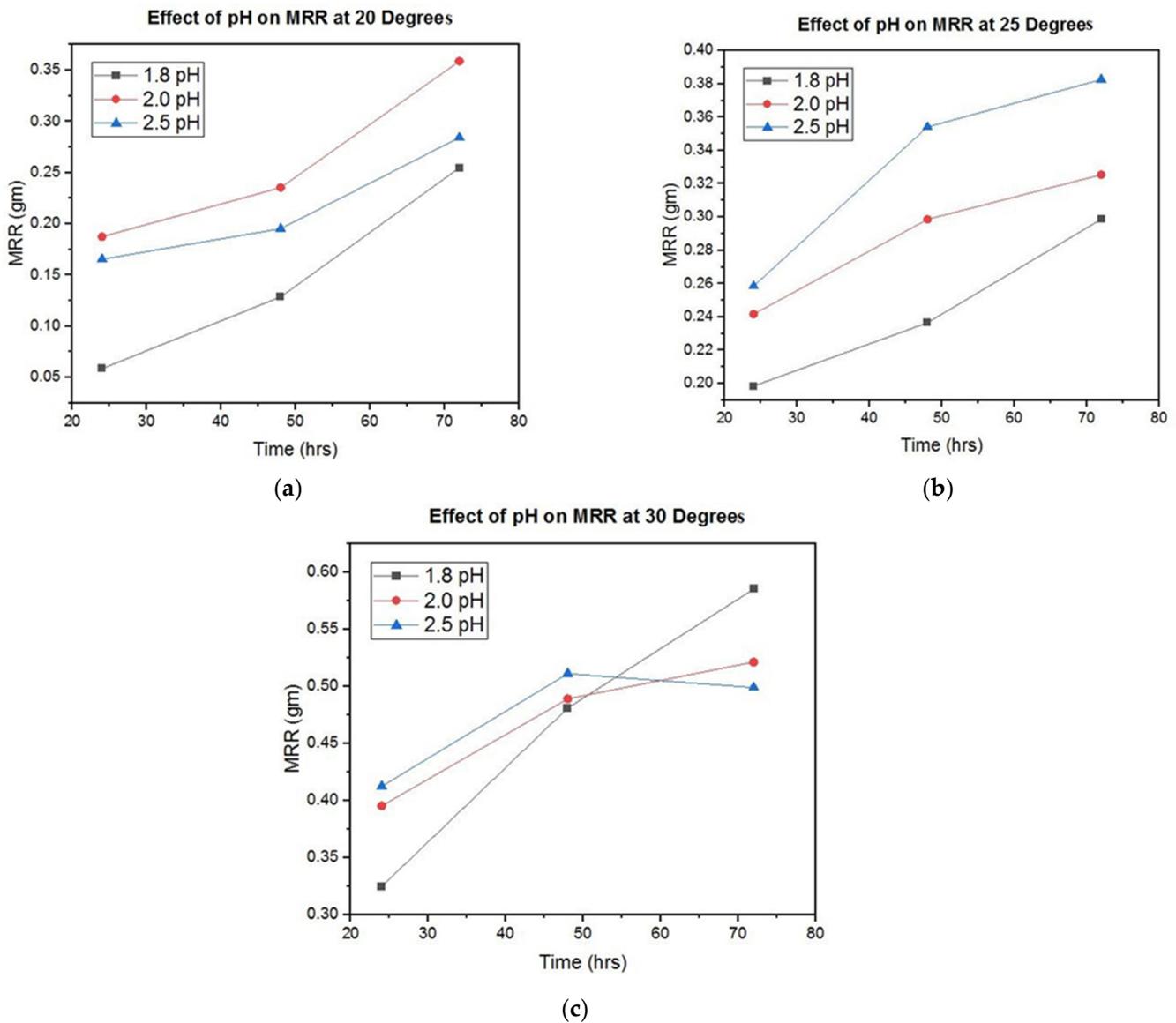


Figure 5. Effect of pH on MRR—nitinol: (a) machining time vs. MRR at 20 °C; (b) machining time vs. MRR at 25 °C; (c) machining time vs. MRR at 30 °C.

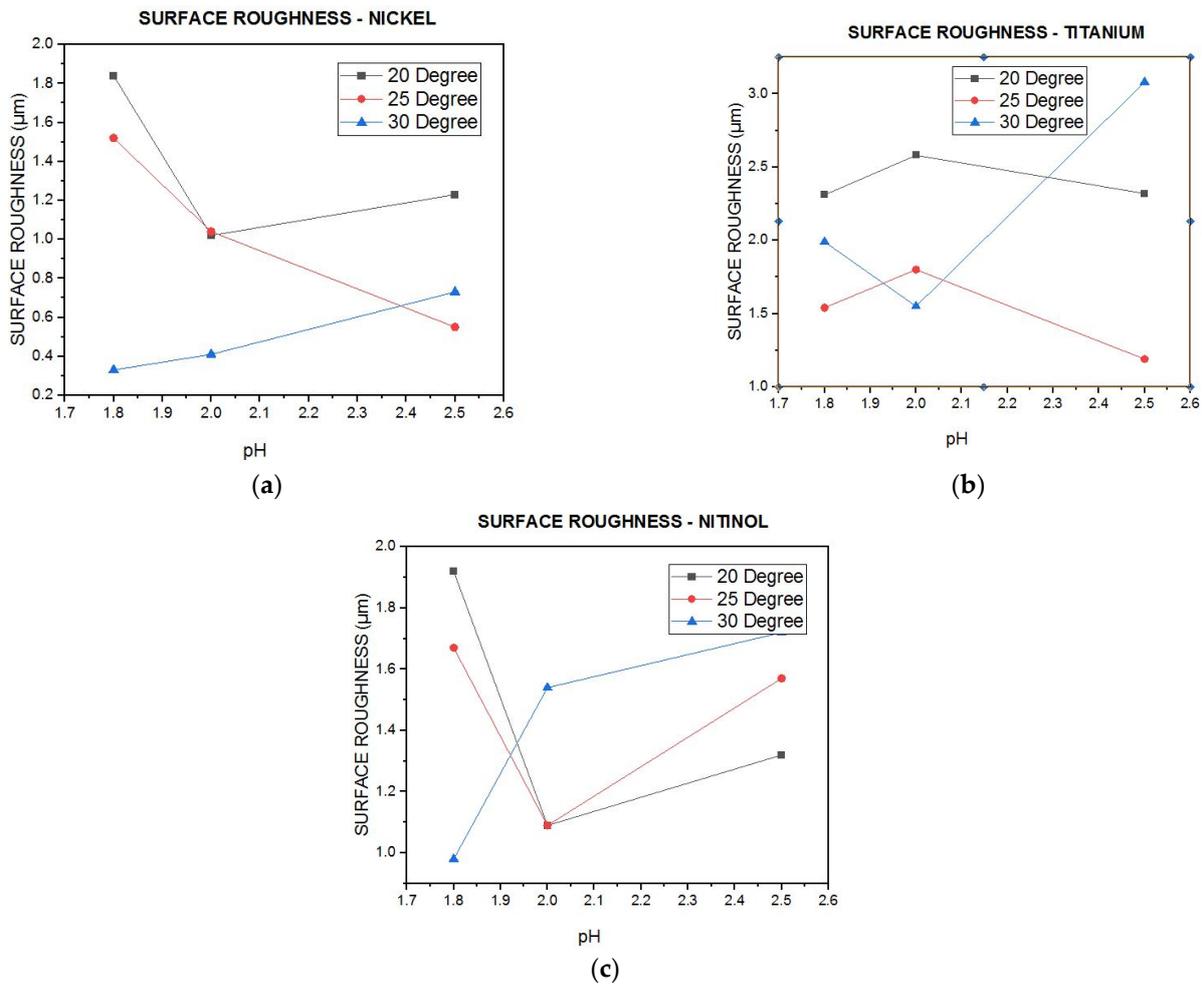


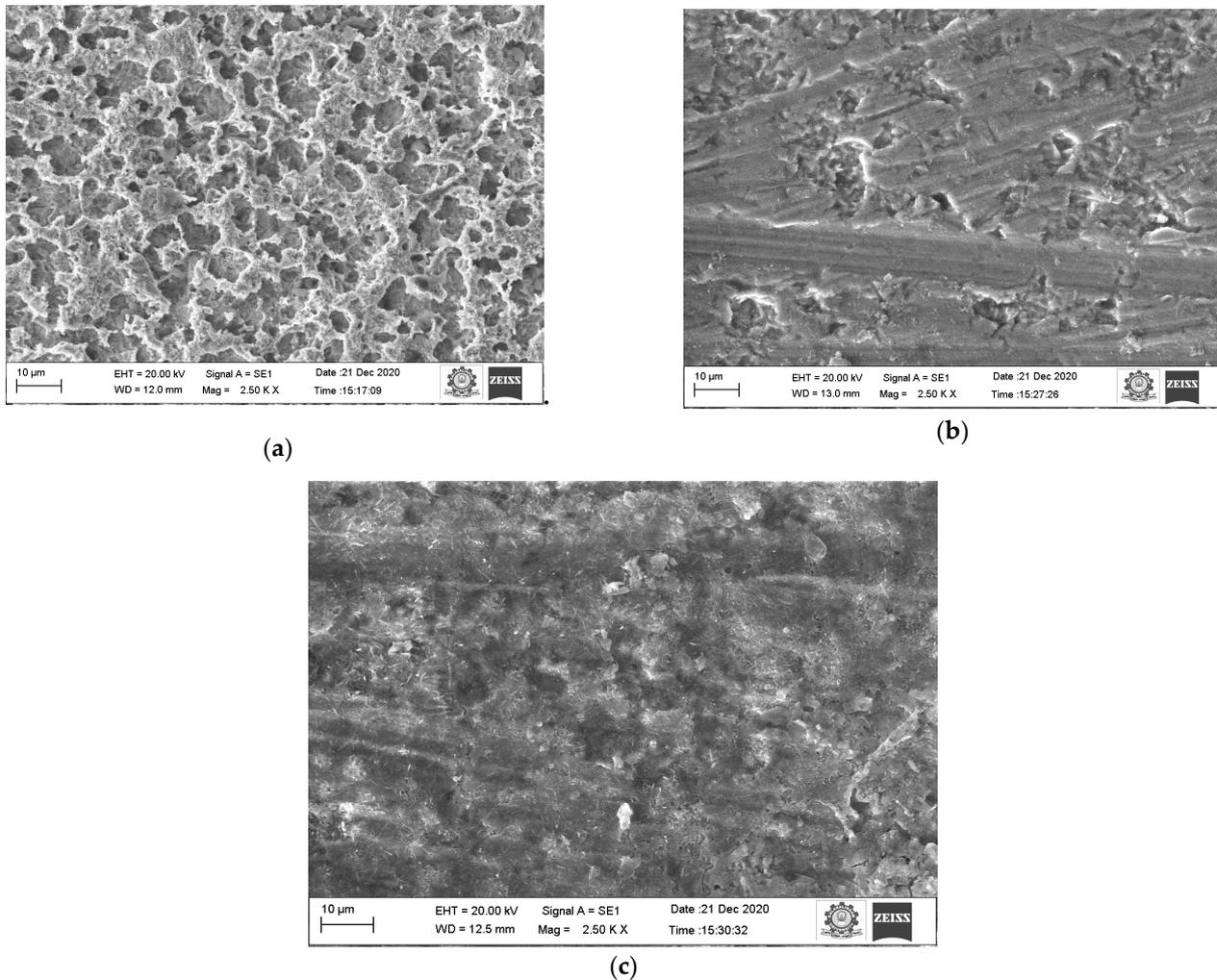
Figure 6. Effect of pH vs. Ra (a) Nickel; (b) titanium; (c) nitinol.

A fine surface finish was obtained: 0.33  $\mu\text{m}$  occurs at a temperature of 30  $^{\circ}\text{C}$ , pH value of 1.8 and ferric content of 50% for nickel; 1.55  $\mu\text{m}$  occurs at a temperature of 30  $^{\circ}\text{C}$ , pH value of 2.0 and ferric content of 75% for titanium and 0.98  $\mu\text{m}$  occurs at a temperature of 30  $^{\circ}\text{C}$ , pH value of 1.8 and ferric content of 50% for Nitinol for 72 h of bio-machining.

The material removal rate (MRR) for the microbial, chemical and enzymatic methods for 10 h were 1.501 g, 17.625 g and 0.1005 g. The material removal rate of chemical machining is higher than the others. Controlling this is difficult because acid penetration into the grooves created on the workpiece causes unfavourable surface properties [16]. Compared with the above-mentioned methods, the growth and activity of this microorganism lead to a better material removal rate with fine dimensional tolerances, making it suitable for implant materials in medical applications

The surface roughness analysis using glucose oxidase shows that a roughness value of 0.89  $\mu\text{m}$  and in a range greater than 2 is highly rough [16], and this research shows that biological machining leads to a fine roughness value of 0.33  $\mu\text{m}$ , which mean it can be considered for implant materials in medical applications.

Figure 7a–c shows the SEM images of the machined specimens of nickel, titanium and nitinol.



**Figure 7.** SEM Images: (a) Machined specimen of Nickel; (b) Machined specimen of Titanium; (c) Machined specimen of Nitinol.

It is noted from the SEM images that the particles are uniformly distributed, compressed and sintered, and the surface of the specimen is machined with the microorganisms. The material removal rate starts from the weakest section (i.e., grain boundaries) as an initial stage throughout the surface. It is compared with different temperatures, pH values and percentages of ferric content, but keeping the shaking speed constant.

Figure 7a shows a material removal rate of 0.2342 g at a temperature of 30 °C, a pH value of 2.0 and a ferric content of 75% for a time period of 72 h. Figure 7b shows a material removal rate of 0.4594 g at a temperature of 30 °C, pH value of 2.5 and ferric content of 25% for a time period of 72 h. Figure 7c shows a material removal rate of 0.5854 g at a temperature of 30 °C, pH value of 1.8 and ferric content of 50% for a time period of 72 h. Shaking speed is one of the essential parameters for obtaining a fine MRR, which shows that the supply of nutrients spread throughout the medium aids in the direct metal removal mechanism.

## 5. Conclusions

The provided research allows us to show the usefulness of a bio-machining process for nickel, titanium and nitinol with *Acidithiobacillus ferrooxidans* microorganisms. The obtained results confirm that the material removal rate and surface roughness are performance indicators that can be used to check the bio-machining performance for future scope when

considering various parameters like shaking speed, temperature, pH and percentage of ferric content.

- Nickel, titanium and nitinol materials are suitable for medical applications when they are fabricated using the powder metallurgy technique and machined with a bio-machining process to finely finish their microstructure with its optimal compaction pressure and sintering temperature, no heat generation and with fine dimensional accuracy.
- *Acidithiobacillus ferrooxidans*, a novel microorganism that was cultured and grown in basal salts, can be employed to machine nickel, titanium and nitinol materials.
- The material removal rate and surface roughness were investigated to obtain a better material removal rate and surface roughness by considering various parameters like shaking speed, temperature, pH and percentage of ferric content.
- The maximum material removal rate of 0.2342 gm occurs at a temperature of 30 °C, a pH value of 2.0 and a ferric content of 75% for nickel; 0.4594 gm occurs at a temperature of 30 °C, pH value of 2.5 and a ferric content of 25% for titanium and 0.5854 gm occurs at a temperature of 30 °C, a pH value of 1.8 and a ferric content of 50% for nitinol for 72 h of bio-machining.
- The average surface roughness obtained by machining with *Acidithiobacillus ferrooxidans* found by the authors was about 0.89 µm, and a fine surface roughness was obtained: 0.33 µm occurs at a temperature of 30 °C, a pH value of 1.8 and a ferric content of 50% for nickel; 1.55 µm occurs at a temperature of 30 °C, a pH value of 2.0 and a ferric content of 75% for titanium and 0.98 µm occurs at a temperature of 30 °C, a pH value of 1.8 and a ferric content of 50% for nitinol for 72 h of bio-machining.
- Nitinol shows a better material removal rate of 0.5854 gm with the effect of parameters like shaking speed 140 rpm, temperature 30 °C, pH value 1.8 and ferric content 50%, while nickel shows a fine surface roughness of 0.33 µm with the effect of parameters like shaking speed 140 rpm, temperature 30 °C, pH value 1.8 and ferric content of 50%.
- These bio-machining parameters proved that the material removal rate and surface roughness can be improved by optimizing the process parameters, and should be considered for implant materials in biomedical applications. This bio-machining process can replace other conventional and unconventional machining processes.

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