CHAPTER 2

METHODS AND MATERIALS

This chapter deals with the description of various materials and methods involved to achieve the objectives of the present investigation. The schematic representation of the methodology and the various techniques adopted in this study is given in Figure 2.1

2.1 MATERIALS

Pure titanium (Kobe steel co., Kobe, Japan), Ti-6Al-4V and Ti-6Al-7Nb alloys were used as the substrate for present investigation. The elemental compositions of the titanium and its alloys were given in the Table 2.1.

2.2 SPECIMEN PREPARATION

Titanium and its alloys were polished using silicon carbide (SiC) water proof papers up to 2500 grit. Final polishing was done using alumina paste in order to produce scratch-free and mirror-finish surface. Then the polished specimens were washed with detergent solution, degreased with acetone and thoroughly washed with distilled water. This was followed by ultrasonic cleaning in acetone for 10 minutes and then dried.
Figure 2.1 Schematic representation of the work plan for the present investigation
### Table 2.1 Chemical compositions of titanium and its alloys (wt. %)

<table>
<thead>
<tr>
<th>Metal</th>
<th>Al</th>
<th>V</th>
<th>Nb</th>
<th>O</th>
<th>C</th>
<th>N</th>
<th>H</th>
<th>Ni</th>
<th>Fe</th>
<th>Ta</th>
<th>S</th>
<th>Si</th>
<th>Ti</th>
</tr>
</thead>
<tbody>
<tr>
<td>Titanium</td>
<td>0.005</td>
<td>&lt;0.030</td>
<td>-</td>
<td>0.140</td>
<td>0.002</td>
<td>0.010</td>
<td>0.001</td>
<td>0.008</td>
<td>0.024</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>Bal.</td>
</tr>
<tr>
<td>Ti-6Al-4V</td>
<td>6.750</td>
<td>4.500</td>
<td>-</td>
<td>0.200</td>
<td>0.080</td>
<td>0.050</td>
<td>0.015</td>
<td>-</td>
<td>0.400</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>Bal.</td>
</tr>
<tr>
<td>Ti-6Al-7Nb</td>
<td>5.850</td>
<td>0.010</td>
<td>6.950</td>
<td>0.170</td>
<td>0.001</td>
<td>0.010</td>
<td>0.003</td>
<td>0.010</td>
<td>0.200</td>
<td>0.050</td>
<td>0.008</td>
<td>0.630</td>
<td>Bal.</td>
</tr>
</tbody>
</table>

Bal. = Balance
2.3 PROCEDURE FOR PRETREATMENT

The specimens were chemically washed at 60°C for 2 minutes in a 1:3:6 mixture of 55 mass % HF aqueous solution, 60 mass % HNO₃ aqueous solution and distilled water. This was done to remove the naturally formed oxide layer. Then it was ultrasonically cleaned for 15 minutes and this step was repeated thrice in distilled water and then dried.

2.4 PROCEDURE FOR HYDROGEN PEROXIDE TREATMENT

The H₂O₂ solution of different concentrations viz., 5 wt.%, 15 wt.% and 25 wt.% were prepared using distilled water. The chemical treatment on the surface of pretreated specimen was carried out in solution containing 5 wt.% of H₂O₂ and 0.1 M HCl, with each sample being immersed in 40 ml of H₂O₂ solution and kept at 80°C in an oven for 1 h. The specimens were then heat treated at 400°C for 1 hr in air atmosphere. Finally, it was cleaned ultrasonically with distilled water for 2 minutes to remove the loosely attached particles. Similar experimental procedure was adopted for other concentrations of H₂O₂.

2.5 PREPARATION OF SIMULATED BODY FLUID SOLUTION

Simulated body fluid (SBF) solution being a metastable solution containing calcium and phosphate ions already saturated with respect to the apatite, any irregularity in the preparation lead to precipitation of Ca²⁺ and PO₄³⁻ ions. Hence, it is required to take utmost care in the preparation of SBF solution. The following procedure was adopted for the preparation of SBF solution. The chemical composition of SBF solution is given in Table 2.2.
Table 2.2 Reagents used in the preparation of SBF solution

<table>
<thead>
<tr>
<th>Order</th>
<th>Reagents</th>
<th>Grams in 1000 mL</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>NaCl</td>
<td>8.035</td>
</tr>
<tr>
<td>2</td>
<td>NaHCO₃</td>
<td>0.355</td>
</tr>
<tr>
<td>3</td>
<td>KCl</td>
<td>0.225</td>
</tr>
<tr>
<td>4</td>
<td>K₂HPO₄·3H₂O</td>
<td>0.231</td>
</tr>
<tr>
<td>5</td>
<td>MgCl₂·6H₂O</td>
<td>0.311</td>
</tr>
<tr>
<td>6</td>
<td>1 Kmol/cm³ HCl</td>
<td>40 cm³</td>
</tr>
<tr>
<td>7</td>
<td>CaCl₂</td>
<td>0.292</td>
</tr>
<tr>
<td>8</td>
<td>Na₂SO₄</td>
<td>0.072</td>
</tr>
<tr>
<td>9</td>
<td>((HOCH₂)₃CNH₂)</td>
<td>6.118</td>
</tr>
<tr>
<td>10</td>
<td>1 Kmol / cm³ HCl</td>
<td>Appropriate amount for adjusting the pH</td>
</tr>
</tbody>
</table>

All the apparatus used in preparing the SBF solution were cleaned with dilute HCl solution, rinsed with ultra-pure water and again immersed in dilute HCl over night. The next day the apparatus was washed several times with ion-exchanged water and the mouths were covered with a wrapping film and were placed in a drier below 50°C.

Further, in a 1000 ml beaker, 750 ml of water was taken and the temperature was maintained at 36.5°C with continuous stirring over a clean bench. Reagents were added to the solution in the same order as given in the Table 2.2. All the chemicals were weighed in a weighing bottle and were added after the complete dissolution of the previous chemical.
The pH meter was calibrated before the preparation of each fresh solution. After the addition of (CH$_2$OH)$_3$CNH$_2$, the temperature of the solution was accurately maintained at 36.5°C and the pH was adjusted to 7.4 using HCl solution. After the adjustment of the pH, the solution was transferred in to a volumetric flask of 1000 ml. The beaker was rinsed several times with ultra-pure water and the solution was added in the flask. The volume of the solution was made up only after the temperature of the solution dropped to 20°C. The flask was shaken well after the make-up. The as prepared solution was stored in polyethylene bottle maintained at 3-5°C in a refrigerator.

Stability of the solution was examined before using. This was done by keeping 50 ml of the solution in an incubator at a temperature of 36.5°C for 2-3 days. Lack of any precipitation was an indication of a stable solution. The solution was discarded and a fresh solution was prepared in case of any suspensions.

### 2.6 SURFACE CHARACTERISATION

#### 2.6.1 Raman Spectroscopy

The crystallographic structures of the titanium oxide layer after the surface modification using H$_2$O$_2$ was analysed by Raman spectroscopy, in the range 3200 to 200 cm$^{-1}$ using 3000 HR System equipped with 514.5 nm argon-ion laser.

#### 2.6.2 Fourier Transform Infra Red Spectroscopic Studies

The functional groups of the apatite formed on the surface were characterized using FT-IR Perkin-Elmer, USA. The apatite particles were carefully scratched from the surface of specimen and the resulting powder from the surface of titanium substrate was mixed with KBr and pressed to
form a pellet. This pellet was analysed using FT-IR in the range of 400 – 4000 cm\(^{-1}\) wave number with a resolution of 4 cm\(^{-1}\).

2.6.3 Scanning Electron Microscopy / Energy Dispersive X-ray Analysis

Surface morphology and elemental composition of the H\(_2\)O\(_2\) treated followed by heat treated specimens before and after immersion in SBF solution were analysed using Scanning Electron Microscope (SEM) coupled with Energy Dispersive X-ray (EDAX). The instrument used was an (Hitachi Model-S 3400).

2.7 ELECTROCHEMICAL STUDIES

2.7.1 Cell Assembly

Figure 2.2 shows the electrochemical set up conventionally used for polarisation and electrochemical impedance studies. The cell set up was made of a 500 ml capacity flask equipped with a platinum mesh electrode as the counter electrode, saturated calomel electrode as the reference electrode and hydrogen peroxide treated titanium and its alloys as the working electrode with the size of 1 cm\(^2\) were exposed in an electrolyte solution.

2.7.2 Open Circuit Potential Measurement

The change in potential with respect to time was monitored for a period of time. The potential of the working electrode with respect to reference electrode after stabilization period was termed as open circuit potential (OCP) or corrosion potential (E\(_{corr}\)).
2.7.3 Potentiodynamic Polarisation Studies

The potentiodynamic polarisation studies were carried out for untreated and H₂O₂ treated titanium and its alloys immersed in SBF solution. All the potential measurements were carried out at a scan rate of 0.167 mVs⁻¹ in an aerated medium. Potentiostat (model PGSTAT 12 with FRA, Autolab, The Netherlands B.V.) controlled by a personnel computer with dedicated software viz., General Purpose Electrochemical System (GPES version 6.0) was used for conducting the polarisation experiments. In order to obtain reliable results, polarization experiments were triplicated in SBF solution.

2.7.4 Electrochemical Impedance Spectroscopic Studies

Electrochemical Impedance Spectroscopic (EIS) studies were carried out at open circuit potential using an electrochemical system
Frequency Response Analyzer (FRA), which included a potentiostat model PGSTAT 12. The H$_2$O$_2$ treated specimens with an exposed surface area of 1 cm$^2$ were used as the working electrode. The impedance spectra were acquired in the frequency range of $10^4$ Hz to $10^{-2}$ Hz with a 10 mV amplitude sine wave generated by a frequency response analyzer.

Impedance spectra were represented in Bode phase angle and resistance plots. The impedance spectra were modeled by fitting these data with an equivalent circuit. The software {EQUIVCRT} model was used in this study for EIS data modeling (Boukamp 1986). The impedance spectra were fitted using a non-linear least square (NLLS) fitting procedure for H$_2$O$_2$ treated titanium and its alloys in SBF solution.