Study on the effect of titanium-based surface coatings on stainless steel 316L for hip prosthesis

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Abstract. Bone implant materials are selected considering their mechanical properties, their adaptability to the human body and their ability to inhibit inflammation or the manifestation of harmful effects beyond standard tolerance limits in the human body. This study aims at investigating and comparing the mechanical and tribological properties of the commercial Stainless Steel 316L implant, which has a multilayered coating of Titanium Aluminium Nitride and a distinct single layer coating of Titanium Nitride by Physical Vapour Deposition technique. Different experimental characterization techniques that involve electron microscopy and X-ray diffraction helped in appropriate characterization of materials. A pH resistance test was conducted to study the effects of corrosion on the material before and after coating. A cytotoxicity test ensured the biocompatibility factor of the material. The result of this study helps one to infer that the Stainless Steel 316L coated with Titanium Nitride yields relatively better results than Titanium Aluminium Nitride.

1. Introduction

The hip implants are medical implants that are placed inside the human body to regain the mobility of the hip and to also overcome the pain associated with hip diseases and injuries. Medical implants are classified into three – metals, ceramics and polymers. The implants made of metal are usually coated with other materials in order to improve their characteristics. Suitable coatings can be provided on the base metal to modify the surface properties such as resistance to corrosion and wear. The 316L grade of Stainless Steel is the most common grade employed for surgical procedures in the human body as it is suitable for the growth of tissues without any negative impact. Investigations are also done on the prospect to apply hard coatings that are thin, on metallic heads so that the reduction of wear and any release of metal ions can be minimized [1]. The various advancements in the development of implants that promote the bone ingrowth and the importance of certain coatings that provide adequate biocompatibility were also taken not account for existing studies [2]. Researches conducted on various coating materials like Copper, Titanium, Nickel, Iron and Zinc indicated that all materials except Nickel showed uniform coating [3]. Studies are also done to check toxicity levels of biomaterials to check for toxic reactions of the material with the body [4].

2. Materials and methods
The specimens were first machined into different dimensions and coated with Titanium Nitride and Titanium Aluminium Nitride, using the Physical Vapour Deposition technique. Further tests were conducted experimentally to study the characteristic changes brought on to the samples.

2.1 Machining of the SS 316L specimens
Specimens fabricated to suit each test, is from Stainless Steel of grade 316L. The behaviour of the material, when provided with the two different titanium coatings is analysed for the tests conducted. A sheet of Stainless Steel of grade 316L was procured and cut into different dimensions:

- 20 X 20 sq.mm of thickness 1.2mm (20 in number).
- 30 X 30 sq.mm of thickness 1.2mm (20 in number).
- 70 X 10 sq.mm of thickness 1.2mm (20 in number).

The cutting operation is performed using a Laser Cutting machine as it is highly accurate, yields excellent cut quality, offers very small cut width, and also a very small heat affect zone. Rods of the SS 316L are fabricated and machined for tensile test using a lathe machine. TiAlN and TiN coatings were also provided to the specimens which are suitable for the tests conducted. The material is also machined into cylindrical pins of diameter 9mm with 2.5mm length. Etching is done using emry sheets to maintain a minimum surface roughness which can be compared after the test. The grades of the emry sheets used were P240, P280, P320, P360, P400, P600, P800, P1000 and P1200. Once this is done, coatings of TiAlN and TiN were provided.

![Figure 1. Tensile test specimen.](image1)

![Figure 2. Wear test specimen.](image2)

The coatings of Titanium Aluminium Nitride and Titanium Nitride were imparted to the base metal specimens of different dimensions using the Physical Vapour Deposition technique. This technique produces a thin film of the biomaterials on the base metal for which various testing is done to check its compatibility as an implant. Phase change occurs twice; first from condensed form to a vapour form, and then back to its initial condensed form. The process takes place in a vacuum environment for proper deposition of the components to occur on the base material on an atomic level [5].
Figure 3. Process flow of the study.

2.2 Mechanical Properties

2.2.1 Tensile test
A tensile test is performed on the specimens in the shape of rods, to check for their tensile strength. A tensile force is imparted on the specimens through a Universal Testing machine and the variation in length is noted. The point of failure of the specimen depicts the ultimate tensile strength and it is checked for the concordance with Hooke’s law. The ultimate Tensile Strength is taken into account for the tensile test performed.

2.2.2 Microvickers Hardness
The microvickers hardness test is done on the specimens cut in the form of sheets, using an optical measurement system. ASTM E-384 specifies a range of light and a diamond indenter is used for creating an indentation, which is in turn converted into a hardness value [6]. The loads applied on the specimens range from a few grams to several kilograms. The specimens must be made smooth before the test for better results as the indentation viewed through the optical system must be clear for measuring the corresponding hardness value.

2.3 pH resistance test
A pH resistance test is also performed in the following procedure:

- Preparation of Phosphate buffer
- pH testing using pH meter
- Immersion of coated material in the buffer

2.3.1 Preparation of Phosphate buffer
A solution of 13.8 grams of (NaH₂PO₄) Sodium dihydrogen Phosphate is dissolved in sufficient amount of water to make a final volume 200ml. Similarly, 14.2 grams of (Na₂HPO₄) disodium phosphate was dissolved in sufficient amount of water to make a final volume of 200ml. These are kept in a separate flask. To make the required pH, the following amount of both the solutions are mixed up.
2.3.2 pH testing using pH meter
The prepared solutions are tested in a pH testing meter. To match the required pH for the test, solutions of one molar of Sodium Hydroxide, NaOH is added if pH needs to be increased and 1 molar of Hydrochloric Acid, HCl is added to decrease the pH. The solutions are again checked for their pH if any additions of NaOH or HCl are provided.

2.3.3 Immersion of coated material in the buffer
three specimens of each TiAlN and TiN specimens in the form of cut sheets is immersed in the pH solution once the required buffer is reached.

Figure 4. Specimens before dipping (left) and after dipping (right).
The specimens are made to react with the solution for twenty four hours and analysis is done through a Scanning Electron Microscope once they are taken out.

2.4 Microstructure analysis for surface parameters
Two tests were conducted to analyse the microstructure for the surface parameters of the specimens.

2.4.1 Scanning Electron Microscope
The Scanning Electron Microscope (SEM) employs high energy focused electron beams to produce the images of the specimen it scans [7]. The SEM equipment has the ability to generate clear in-depth images of microstructure of the specimen. The sample is prepared in such a way that it should have the ability to withstand a vaccum condition and electron beams of very high energy. The sample is placed on a conducting carbon strip which is then supported on a small circular base. The maximum size of the specimen that can be loaded is upto 4 centimeters. The electron beams are pumped and are projected towards the sample through an apperture and high resolution images of the microstructure are generated, which can reveal details lesser than one nanometer. The microstructure is studied once the process the electron beams are made to strike the specimen kept within the chamber.

2.4.2 X-ray Diffraction
The structural information about the crystalline solids can be obtained by the X-ray diffraction method. Diffraction using X-ray beams is done in order to obtain the microstructure of crystalline and amorphous specimens. The specimen to be tested has to be in its powdered state, and a base holder is employed for holding the powdered specimen. Once the X-rays are projected from the transmitter, it passes through scatter slits and then strikes the sample, after which they get diffracted towards the detector plane. The diffraction pattern is thus obtained when the beams reach the detector plane [8]. The count of the elements present can also be identified with respect to the amount of voltage provided in order for the X-rays to strike the powdered specimen.

2.5 Cytotoxicity test
The level of toxicity and the level of adhesion of a material with the cells, are generally checked under the performance in the Cytotoxicity test [9]. The MTT assay is a calorimetric assay used to assess the cell metabolic activity.

Preparation of the MTT solution:
- The MTT is dissolved in a solution of Dulbecco’s Phosphate buffered saline, with a pH at 7.4, to form up to 5mg/ml.
- The MTT solution formed is filter sterilized through a filter of 0.2µm into a container which is sterile and protected from light.
- The MTT solution after filtering is stored either at 4°C for frequent usage, or at 20°C for a long-term storage. The solution has to be protected from light so that it would not form any reaction when exposed to light.

Preparation of the Solubilization solution:
- At 2% (volume/volume) of glacial Acetic acid, prepare a 40% (volume/volume) Dimethyl Formamide.
- To it, add 16% (weight/volume) of Sodium Dodecyl Sulphate and let it dissolve.
- The pH is adjusted to 4.7.
- The solution prepared is stored at room temperature to avoid any precipitation.

The MTT assay protocol:
- The cells are prepared to test the compounds in ninety-six well plates with a volume of 100 microliters for each well.
- Incubation is done for the desired period of exposure.
- MTT solution of 10 microliters is added to each well to reach a concentration of 0.45mg/ml for the solutions.
- Incubation is again done for a period of 4 hours at 37°C.
- Solubilization solution of 100 microliters is added to each well so that the crystals of Formazan dissolve.
- The solutions are properly mixed to ensure solubilization is done properly.
- The absorbance is recorded at 570nm for the solutions in the wells.

The cytotoxicity test was performed with the help of the above mentioned MTT kit and the test was performed for 24 hours. The test was performed by keeping SS 316L as a control limit. The SS 316L coated with TiN and TiAlN were placed in the system for the specified time.

3. Results and Discussion
The samples were characterized using various surface analysis techniques and cytotoxicity tests for biocompatibility. The results indicated that there is a uniform coating. The TiN and TiAlN coatings on stainless steel resulted in excellent adhesion and hydrophilicity. Also, the corrosion for the coated material was determined by conducting the pH resistance test. Biocompatibility of the coated material has been ensured by conducting the cytotoxicity test. From all the above stated experimental investigation, SS 316L coated with TiN yields better results in comparison with TiAlN.

3.1 Wear Test
The wear test has been done by using the wear and the friction monitor setup under dry condition at different loads. The values were tabulated.

| Thickness (microns) | Load (N) | Speed (rpm) | Time (s) | Before weight (gm) | After weight (gm) | Friction coefficient (µ) | RB (µm) | RA (µm) |
|---------------------|---------|-------------|----------|-------------------|------------------|------------------------|--------|--------|

Table 1. Friction monitor observation.
|                |                |                |                |                |                |                |
|----------------|----------------|----------------|----------------|----------------|----------------|----------------|
| Uncoated SS 316L |                |                |                |                |                |                |
| 0              | 5              | 100            | 60             | 12.532         | 12.524         | 0.029          | 0.43           | 0.544          |
| 0              | 10             | 125            | 75             | 12.889         | 12.797         | 0.214          | 0.27           | 0.373          |
| 0              | 15             | 150            | 90             | 12.751         | 12.672         | 0.246          | 0.31           | 0.572          |
| 0              | 20             | 175            | 105            | 12.957         | 12.801         | 0.386          | 0.27           | 0.561          |
| TiN coated SS 316L |                |                |                |                |                |                |
| 2              | 5              | 100            | 60             | 12.973         | 12.951         | 0.052          | 0.54           | 0.564          |
| 2              | 10             | 125            | 75             | 12.392         | 12.369         | 0.149          | 0.56           | 0.587          |
| 2              | 15             | 150            | 90             | 12.113         | 12.035         | 0.237          | 0.40           | 0.443          |
| 2              | 20             | 175            | 105            | 11.523         | 11.509         | 0.255          | 0.45           | 0.575          |
| TiAlN coated SS 316L |                |                |                |                |                |                |
| 2              | 5              | 100            | 60             | 12.091         | 12.089         | 0.085          | 0.70           | 0.712          |
| 2              | 10             | 125            | 75             | 12.371         | 12.369         | 0.101          | 0.49           | 0.521          |
| 2              | 15             | 150            | 90             | 12.421         | 12.419         | 0.198          | 0.91           | 1.145          |
| 2              | 20             | 175            | 105            | 12.468         | 12.341         | 0.317          | 0.50           | 0.833          |

Plot for the various values of coefficient of friction for different materials is also done.

![Graph showing coefficient of friction](image)

**Figure 5.** Coefficient of friction plot.

It has been inferred that Titanium Nitride coated with SS 316L has less coefficient of friction when compared with Titanium Aluminium Nitride coated with SS 316L. In addition, the Titanium coated steels have less coefficient of friction when compared with uncoated SS 316L.

### 3.2 Material Characterization

The results of the SEM tests are represented below. The material microstructure of the specimens is observed and carefully taken into account.
Figure 6. SEM images of (a) SS 316L (b) TiN coated SS 316L and (c) TiAlN coated SS 316L.

Figure 6 (b) and (c) clearly show formation of titanium coatings on the base metal SS 316L. Thus, it indicates that the coating is uniform and is properly coated by using Physical Vapour Deposition. The microstructure is analysed for all the three specimens of TiAlN.

Figure 7. SEM images of microstructures of (a) TiAlN coated SS 316L at a pH 6 (b) TiAlN coated SS 316L at a pH 6.5 (c) TiAlN coated SS 316L at a pH 7.

From figure 7, it was observed that the microstructure is similar to that of figure 7 (c) which was taken before the test. Thus, the TiAlN coated SS 316L did not undergo corrosion and coating remains the same after the test and it did not peel off. The microstructure is similarly analysed for the three specimens of TiN coated SS 316L for which pH resistance test was conducted.

Figure 8. SEM images of (a) TiN coated SS 316L at a pH 6 (b) TiN coated SS 316L at a pH 6.5 and (c) TiN coated SS 316L at pH 7.

The results obtained from Energy Dispersive Spectroscopy were also taken into account.
From figure 9, the presence of Titanium and Nitride has been verified clearly for the plot obtained for TiN and the corresponding weight percentage of Ti was 5.49% and nitride was 11.69%. Similarly, the presence of Titanium, Aluminium and Nitride has been verified clearly for the plot obtained for TiAlN. The weight percentage of Al was 17.4 Titanium was 56.84% and nitride was 12.83%.

The results obtained from X-ray Diffraction showed the number of various elements present within the samples, with which the element with the highest concentration was recognized.

From the result of XRD for the uncoated specimen of Stainless Steel 316L, it was noticed that there was a peak concentration of Iron, then Chromium, and then Manganese. When Titanium coatings were provided, the concentration peak changed for the elements, with Titanium becoming the element with most concentration level in Titanium Nitride coated specimen and second highest concentration in Titanium Aluminium Nitride coated specimen.
The XRD result for TiAlN indicated that Aluminium has the most concentration and the result for TiN indicated that Titanium has the most concentration. This showed that the coatings provided on the base metal were uniform.

3.3 Mechanical Properties

The results of Micro Vickers hardness test and tensile test were specifically noted individually for analysing the nature of the specimens under an external force applied on them. For Micro Vickers hardness, the test was conducted on three different points on the specimen and the mean value has been taken as the hardness value for the test with a constant load of 100gm.

| Material           | Hardness (HV) |
|--------------------|---------------|
| SS 316L            | 57.95         |
| TiN coated SS 316L | 70.48         |
| TiAlN coated SS 316L | 66.7         |

It was observed that Titanium Nitride coated SS 316L has higher hardness when compared with the uncoated SS 316L and Titanium Aluminium nitride coated SS 316L. The results of tensile test were also accounted to check the ultimate tensile strength of the coated specimens and the uncoated specimen.
Table 3. Ultimate tensile strength values by tensile test.

| Material                  | UTS (MPa) |
|---------------------------|-----------|
| SS 316L                   | 960.655   |
| TiN coated SS 316L        | 968       |
| TiAlN coated SS 316L      | 964       |

Figure 13. Comparison of Tensile Strength.

It was observed that there is an increase in the ultimate tensile strength of Titanium coated SS 316L alloys. In addition, the UTS of the Titanium Nitride coated SS 316L was higher than the Titanium Aluminium Nitride coated and uncoated SS 316L.

3.4 pH resistance test

The pH resistance was done for two different Titanium coatings at three different buffer solutions of pH 6, 6.5 and 7. The varying buffer solution values for Titanium Nitride and Titanium Aluminium Nitride coated SS 316L were plotted with respect to the control values.
Figure 14. pH compatibility test for TiAlN coated SS 316L at pH 6.0-7.0.

The graph was plotted for the corresponding variation in wavelength in nanometres to the change in optical density. It was noticed that the optical density decreased with the increase in wavelength and after a certain limit, the optical density remained constant with the increase in wavelength. This behaviour was much similar in the varied pH concentrations of the TiAlN coated SS 316L specimen. It was observed that there was very less variation in the characteristics of pH of Titanium Aluminium Nitride coated over SS 316L with respect to the control. A similar plot was done for Titanium Nitride coated over SS 316L for the pH behaviour.

Figure 15. pH compatibility test for TiN coated SS 316L at pH 6.0-7.0.

It has been observed that there is no significant variation with respect to the control for the two coated titanium specimens. Thus, it can be said that the titanium coated materials are resistant to corrosion at given a pH of 6, 6.5 and 7.

3.5 Cytotoxicity
The cytotoxicity test was done on both Titanium coated SS 316L materials by introducing the cervical cancer cells on top of the coated surfaces for a span of 24 hours.

Figure 16. (a) Living Hela cells on surface of TiN coated SS 316L (b) living cells on the surface of TiAlN coated SS 316L
The plot shows the percentage of cells alive in reference with SS 316L which was proven to be biocompatible.

![Cytotoxicity](image)

**Figure 17.** Percentage of living cells.

It has been observed that the Titanium Aluminium Nitride coated SS 316L and Titanium Nitride coated SS 316L shows no cytotoxicity. However, there is a value change and the TiN coated SS 316L is slightly cytotoxic than TiAlN coated SS 316L. Since there is no significant variation in the values, it can be said that both the materials are biocompatible.

4. Conclusion
The aim of the study was to compare and investigate the mechanical and tribological properties of the commercial Stainless Steel 316L implant with a multilayer coating of Titanium Nitride and Titanium Aluminium Nitride, so as to check for the suitability of these materials for the application of bioimplants. From the results, it can be observed that the Titanium Nitride coated with SS 316L alloy would be a better replacement for SS 316L as it enhanced the mechanical, tribological and microbial properties. It can be concluded that further studies can be carried out on the above study to enhance the use of the material as prosthesis. Cell culture tests can be done to check how the materials behave when it is implanted in the human system. The same methodology can be adopted for the implant materials coated with nano particles to investigate the behaviour of the implants over existing implant material that is being used.

5. References

[1] J.A Ortega-Saenz 2013 Bio tribological study of multilayer coated metal-on-metal hip prostheses in a hip joint simulator, *Mexico: Universidad Autonoma de Nuevo Leon, FIME*. 234-242

[2] Bill G.X Zhang 2014 Bioactive coating for Orthopaedic Implants- recent trends in development of Implant coatings, *Australia: University of Melbourne*.11878-11921

[3] Liang Hao 2013 Influence of Metal Properties on the formation and evolution of metal coating during mechanical coating, *ASM International: Chibo University*. 1-33

[4] H.J Johnson 1985 Biocompatibility test procedures for materials evaluation in vitro II. Objective methods of toxicity assessment, *Illinois: Laboratories*. 19. 489-508

[5] Tadeusz Burakowski and Tadeusz Wierzchon 1999 Surface Engineering of Metals: Principles, Equipment, Technologies, *New York: CRC press*. 586-634

[6] Peggs, G.N and Leigh, I.C 1983 Recommended Procedure for Micro-indentation Vickers Hardness Test, *Great Britain: National Physical Laboratory*. 37-48

[7] Piyush Charan, E.R. 2015 *Scanning Electron Microscope*.1-5

[8] Bertram Eugene Warren 1990 *X-ray Diffraction*. 151-157
[9] L. Reclaru 2014 New generation super alloy candidates for medical applications: Corrosion behaviour, cation release and biological evaluation, *Germany: Johannes Gutenberg University Mainz*. 411-420
