

Biocompatibility of Titanium Implants in Bone Regeneration Materials

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Abstract: Ti and its alloys are extensively utilized in medical and dental implant devices due to their exceptional corrosion resistance and good hard-tissue compatibility. This study evaluates the biocompatibility of titanium implants in bone regeneration materials. Titanium substrates were prepared, coated with HAp composite via sol-gel method, and analysed. Results showed even ceramic coating, confirmed functional groups, and excellent cell attachment (96.7% viability). Hemocompatibility was confirmed with <3% lysis. Titanium remains a successful implant material due to its biocompatibility and enhanced functionality.

Keywords: Biocompatibility; Titanium implants; osseointegration; HAP.

1. Introduction

The integration of titanium substrates in bone regeneration heralds a groundbreaking chapter in medical science, offering a compelling synthesis of biocompatibility and structural resilience.^[1] This essay delves into the multifaceted aspects of titanium's role in bone regeneration, unraveling its impact on medical procedures and the potential it holds for advancing patient outcomes.^[1,2]

At the core of titanium's efficacy in bone regeneration lies its exceptional biocompatibility. When introduced into the body, titanium elicits minimal immune response, reducing the risk of rejection and adverse reactions.^[1,3] This property is pivotal in the context of orthopedic implants, where foreign materials must seamlessly integrate with the body's natural processes. The ability of titanium to coexist harmoniously with biological systems underscores its suitability for medical applications, particularly in bone regeneration.^[4]

Furthermore, titanium's structural prowess plays a crucial role in providing a stable substrate for bone growth. The mechanical strength of titanium aligns with the demands of load-bearing applications within the skeletal system.^[5] Orthopedic implants made from titanium serve as robust frameworks that endure the mechanical stresses inherent in daily activities, ensuring longevity and functionality.^[6] This durability is a key factor in the success of bone regeneration procedures, as it enables the implant to withstand the dynamic forces exerted on it during the healing process.^[7]

The phenomenon of osseointegration represents a pinnacle achievement facilitated by titanium substrates in bone regeneration. Osseointegration refers to the direct structural and functional connection between living bone and the surface of a load-bearing implant.^[8] Titanium's surface characteristics contribute to this process by promoting the adhesion and proliferation of osteoblasts, the cells responsible for bone formation.^[9] This intimate connection between titanium implants and the surrounding bone tissue enhances stability and functionality, setting the stage for successful bone regeneration.^[10]

The application of titanium substrates extends beyond traditional orthopedic implants to include a spectrum of innovative approaches in bone regeneration. 3D printing technology, for instance, allows for the precise fabrication of titanium scaffolds that mimic the intricate structure of natural bone.^[11] These customized implants not only offer a tailored fit but also provide a conducive environment for cellular activities, accelerating the regenerative process.^[12] The intersection of titanium and advanced manufacturing techniques opens avenues for personalized medicine in orthopedics, marking a paradigm shift in the approach to bone regeneration.^[13]

The field of regenerative medicine continues to evolve, titanium's role in promoting angiogenesis, the formation of new blood vessels, adds another dimension to its significance.^[14,15] By facilitating the development of a vascular network around the implant, titanium substrates enhance nutrient supply to the regenerating tissue.^[14] This vascular support is paramount for the success of bone regeneration, ensuring that the healing process is not only efficient but also sustains the vitality of the newly formed bone.^[16]

As a result, the incorporation of titanium substrates in bone regeneration represents a transformative leap in medical science. Its biocompatibility, structural resilience, and ability to foster osseointegration position titanium as a cornerstone in orthopedic advancements.^[17] From traditional implants to cutting-edge 3D-printed scaffolds, titanium's versatility continues to shape the landscape of bone regeneration, offering hope for improved patient outcomes and the potential for pioneering developments in regenerative medicine.^[18]

2. Materials and methods:

Preparation of Titanium substrate: Titanium substrate was polished using a grid sheet. Further etched with HF acid. After cleaning the Titanium substrate, substrates were coated with Hap composite using sol gel method Titanium substrate were then dried and taken for further studies.

Surface preparation: The surface morphology and cross section of the coatings were observed by scanning electron microscopy (SEM, Quanta FEG 450). The basic composition of the coatings was analyzed by Fourier transform infrared (FTIR) spectroscopy.

Hemocompatibility assay:

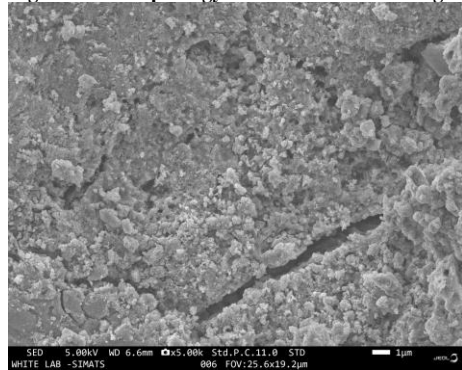
An assay for hemocompatibility assessed the interaction of erythrocytes with MTCN. This study used the same procedures as earlier papers. In order to isolate erythrocytes (RBCs) by separating plasma, whole human blood treated with EDTA was ethically taken from patients in vacutainers and centrifuged at 1500 × g for five minutes. After three rounds of washing in phosphate-buffered saline (pH 7.4), the concentration of these RBCs was diluted to 10%. PBS was used to blend samples of 200 μL erythrocyte suspension (12.5, 25, 50, 100, and 200 μg/mL) to a final volume of 1 mL. After an hour of incubation at 37°C, the mixture was centrifuged for five minutes. Cells treated with PBS served as negative controls, whereas cells treated with deionized water served as positive controls.

Biocompatibility and Osseointegration: Hydroxyapatite, a biocompatible material, ensures good adhesion of the Ti-64 coating to the surrounding tissue. In addition, the structure of hydroxyapatite with natural bone mineral promotes osseointegration (the direct connection of the implant to the surrounding bone tissue), which contributes to the overall stability of the implant.

Clinical Applications: The application of Ti-64 coated implants in clinical settings has high potential. These coatings are able to improve patient outcomes by reducing the risk of infection, reducing the risk of complications and contributing to the long-term success of treatment.

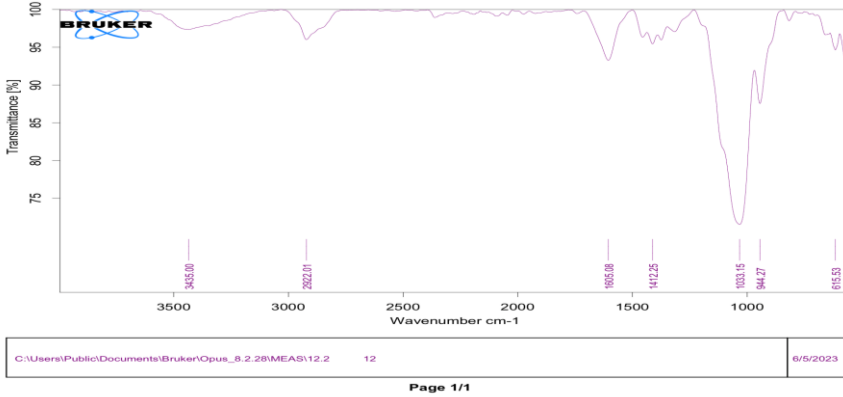
3. Results:

Fig-1 SEM morphology shows that even coating of ceramic (CaP) CPTi grade 2 and uniform morphology.



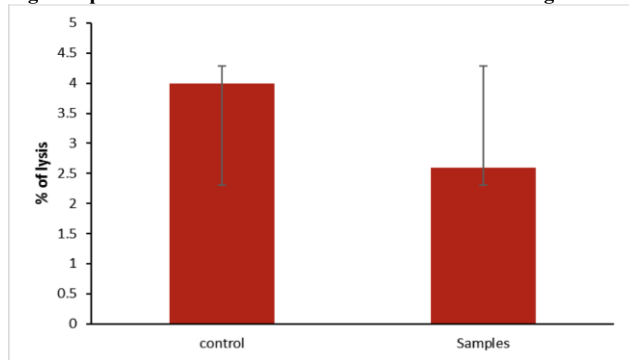
SEM micrograph shows the surface morphology of calcium phosphate coating which exists on commercially pure titanium material at grade 2. The image reveals an even and uniform coating with fine homogeneous surface morphology. The scale bar serves its purpose to show the level of magnification.

Fig-2 Functional group analysis confirms that formation of carboxyl, hydroxyl, phosphate presented in bio ceramic coating.



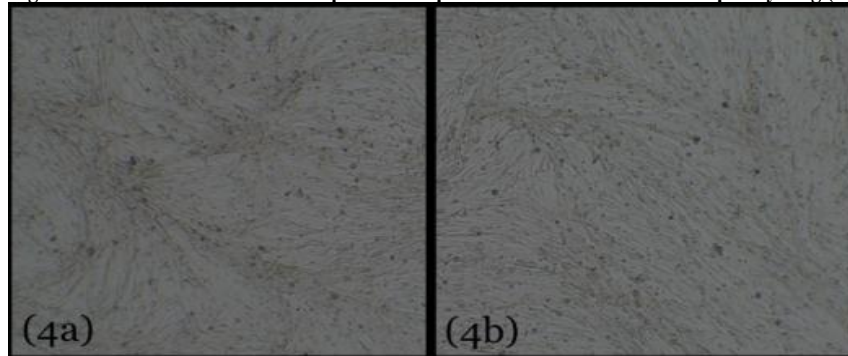
The FTIR analysis confirms the chemical integrity and bioactivity of the ceramic coating. The spectrum shows a strong absorption peak at 1033.15 cm⁻¹, which acts as the main "fingerprint" for phosphate PO₄³⁻ groups and includes a secondary peak at 944.27 cm⁻¹. The combination of phosphate vibrations with the wide hydroxyl (-OH) band that appears around 3435 cm⁻¹ indicates that the coating demonstrates bioactive properties of Hydroxyapatite (HAp) material. The broad hydroxyl peak demonstrates high surface energy which helps proteins to adsorb and cells to attach after the implant enters clinical use. The C-H stretching peaks 2922 cm⁻¹ show low intensity which proves that the coating consists mostly of inorganic material with only slight organic contamination thus confirming high purity of the synthesized bio-ceramic layer.

Fig-3 As per the ASTM Standard it is found that the coating shows less than 3% lysis and hence it is found to be hemocompatible.



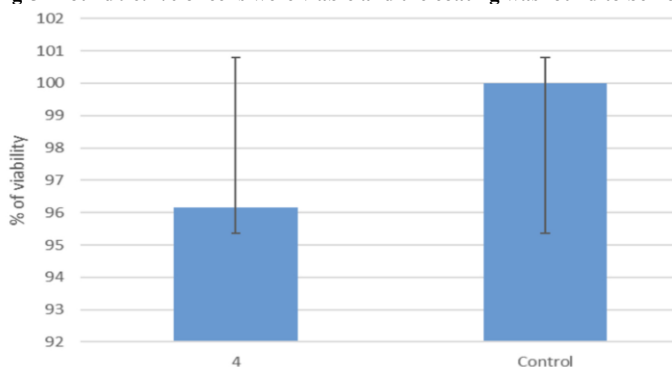
The synthesized coating shows a hemolytic rate of 3% which falls below the 5% threshold used to assess hemocompatibility of materials. The coating demonstrates biocompatibility through its ability to maintain red blood cells because it only produces minimal cell lysis when tested against the control group. The findings demonstrate that the coating does not create harmful effects on blood components thus making it suitable for extended use in medical settings.

Fig-4 Better cell attachment and improved cell proliferation is found microscopically. Fig (4a) is a control, Fig (4b) is a sample.



The micrographs display cell growth patterns which occur on the control surface and the experimental bio-ceramic coating. The sample exhibits higher cell density together with distinct cell shapes which indicate better biocompatibility.

Fig-5 Around 96.7% of cells were viable and the coating was found to be non toxic.



The experimental sample reached a cell viability rate of 96.7% when compared to its control group. The bio-ceramic coating demonstrates non-toxic properties which promote cellular health according to the high percentage of viable cells. The statistical analysis results show no significant difference in cell viability between the two groups which supports the safety profile of calcium phosphate-coated Ti for use in clinical settings.

4. Discussion:

Dental implant insertion success is correlated with both their hemocompatibility and biocompatibility. Dental implants can be made to have acceptable levels of hemocompatibility and biocompatibility by modifying their surfaces in different ways.^[3] Even though the implant has been placed in a jawbone that is not sufficient, the surface alteration causes the implant's surface to become rough, which will lower its survival rate.^[10] The dental implant's porosity and roughness will act as a scaffold to support the formation of bone, strengthening its attachment to the jaw. Dental implants with surface modifications have pores that can support the formation of cells and tissue and permit the diffusion of different nutrients. An optimal porosity for cell adhesions is provided by the SLA surface modification.^[19]

Studies on biocompatibility have demonstrated the relationship between, among other things, bone ingrowth and cell proliferation and design flexibility. The findings raise the possibility of using EBM titanium more widely to repair particular bone abnormalities. In one study, the porous Titanium implants' surfaces were modified by coating them using the EBM process, and then the surfaces were coated using a biomimetic technique. For implants with and without biomimetic apatite coating, the biocompatibility both in vitro and in vivo were assessed. Positive results for cell adhesion, proliferation, and morphology were obtained from the in vitro biocompatibility of the EBM porous titanium. Implants with and without biomimetic coating were compared, and the results indicated that porous EBM Ti-64 implants may support cell growth on par with coated implants. After 12 weeks, the in vivo histological study revealed that the rates of ingrowth and bone formation between coated Ti-64 and EBM Ti-64 were similar.^[20] Similarly our study shows better cell attachment and improved cell proliferation is found microscopically.

The Ti implant's surface wettability behavior had an impact on RBC aggregation as well. The surface hydrophilicity of biomedical titanium dental implants rises, facilitating the absorption of different plasma proteins including fibrinogen, which is essential for the implant's recovery and the growth of new bone surrounding it. The FE-SEM micrographs of red blood cell (RBC) morphology following Ti implant, both with and without surface alteration, are shown in a previous work. While the IDCT-modified Ti implants displayed an implant surface rich in RBC aggregation and fibrin filament formation, it is evident that less RBC aggregation was observed on Ti implants without surface modification. Similarly our study shows around 96.7% of cells were viable and the coating was found to be non toxic.^[2] SEM morphology shows that even coating of ceramic (CaP) CPTi grade 2 and uniform morphology.^[8] As a result, the altered titanium dental implants may help with osseointegration, which is necessary for the success of dental implants. Nevertheless, additional research is required for practical implementation, utilizing a more extensive sample size and a diverse duration of follow-up to reinforce the proof of the swift osseointegration brought about by dental implants.

5. Limitations and Future Scope

While the findings are promising, long-term clinical performance assessment is necessary. Future research should focus on optimizing manufacturing protocols and conducting extensive clinical trials to evaluate reinforced ceramics with customized porosity architectures.

6. Conclusion:

Titanium has an outstanding record of being used effectively as an implant material. This success is attributed to titanium's exceptional biocompatibility, which is caused by the stable oxide layer that forms on its surface. This is because of the related major benefits, which include enhanced functionality, lower processing costs, less waste, energy efficiency, and flexible design. To enhance the performance of sintered bioceramics for clinical use, further research is still required, despite the wide range of techniques that have been investigated over the previous few decades. Future material scientists should concentrate on developing novel forming techniques that can produce reinforced ceramics with customized porosity architecture, opening up new avenues for enhanced bone surgery applications.

7. References:

- [1] An HW, Lee J, Park JW (2023) Surface characteristics and in vitro biocompatibility of surface-modified titanium foils as a regenerative barrier membrane for guided bone regeneration. *J Biomater Appl* 37:1228–1242.
- [2] Zhu Y, Zhou D, Zan X, Ye Q, Sheng S (2022) Engineering the surfaces of orthopedic implants with osteogenesis and antioxidants to enhance bone formation in vitro and in vivo. *Colloids Surf B Biointerfaces* 212:112319.
- [3] Cao W, Jin J, Wu G, Bravenboer N, Helder MN, Schulten EAJM, et al (2022) Kappa-carrageenan-Functionalization of octacalcium phosphate-coated titanium Discs enhances pre-osteoblast behavior and osteogenic differentiation. *Front Bioeng Biotechnol* 10:1011853.
- [4] Sarraf M, Rezvani Ghomi E, Alipour S, Ramakrishna S, Liana Sukiman N (2022) A state-of-the-art review of the fabrication and characteristics of titanium and its alloys for biomedical applications. *Biodes Manuf* 5:371–395.
- [5] Al-Shalawi FD, Mohamed Ariff AH, Jung DW, Mohd Ariffin MKA, Seng Kim CL, Brabazon D, et al (2023) Biomaterials as Implants in the Orthopedic Field for Regenerative Medicine: Metal versus Synthetic Polymers. *Polymers* 15:2601.
- [6] Wang R, Wang M, Jin R, Wang Y, Yi M, Li Q, et al (2023) High Strength Titanium with Fibrous Grain for Advanced Bone Regeneration. *Adv Sci* 10:e2207698.
- [7] Xu C, Ivanovski S (2025) Clinical translation of personalized bioengineered implant scaffolds. *Nat Rev Bioeng* 3:390–407.
- [8] Hou PJ, Syam S, Lan WC, Ou KL, Huang BH, Chan KC, et al (2020) Development of a Surface-Functionalized Titanium Implant for Promoting Osseointegration: Surface Characteristics, Hemocompatibility, and In Vivo Evaluation. *Appl Sci* 10:8582.
- [9] Ramya, Rajasekar A (2021) Enhanced Antibacterial effect of Titanium Dioxide Nanoparticles mediated Grape Seed Extract on oral pathogens - *Streptococcus mutans* and *Lactobacillus*. *J Evol Med Dent Sci* 10:1656–1661.
- [10] Larsson C, Thomsen P, Aronsson BO, Rodahl M, Lausmaa J, Kasemo B, et al (1996) Bone response to surface-modified titanium implants: studies on the early tissue response to machined and electropolished implants with different oxide thicknesses. *Biomaterials* 17:605–616.
- [11] Sidambe AT (2014) Biocompatibility of Advanced Manufactured Titanium Implants—A Review. *Materials* 7:8168–8188.
- [12] Beloti MM, Rosa AL (2024) Bone Regeneration and Repair Materials. *Materials* 17:1396.
- [13] Brie IC, Soritau O, Dirzu N, Berce C, Vulpoi A, Popa C, et al (2014) Comparative in vitro study regarding the biocompatibility of titanium-base composites infiltrated with hydroxyapatite or silicitanate. *J Biol Eng* 8:14.
- [14] Rajaraman V, Nallaswamy D, Ganapathy D, Rajeshkumar S, Ariga P, Ganesh K (2021) Effect of Hafnium Coating on Osseointegration of Titanium Implants: A Split Mouth Animal Study. *J Nanomater* 2021:7512957.
- [15] Swarna Meenakshi S, Sankari M (2021) Effectiveness of Chitosan Nanohydrogel as a Bone Regenerative Material in Intrabony Defects in Patients With Chronic Periodontitis: A Randomized Clinical Trial. *J Adv Oral Res* 12:222-228.
- [16] Peskova M, Hradilova S, Jiravova J (2020) Comparison of the biocompatibility of titanium and its alloys for dental implants. *Biomed Pap Med Fac Univ Palacky Olomouc Czech Repub* 164:14-23.
- [17] Preethi S, Rajeshkumar S (2021) Antibacterial and Cytotoxic activity of Magnesium oxide nanoparticles mediated by Ginger and Garlic. *Int J Dent Oral Sci* 8:4349-4354.
- [18] Abarna S, Ariga P (2020) Influence of surface treatment on the bond strength of various veneering ceramics to zirconia-A systematic review. *Int J Res Pharm Sci* 11:4793-4799.
- [19] Burdick JA, Mauck RL. *Biomaterials for Tissue Engineering Applications: A Review of the Past and Future Trends*. Springer Science & Business Media; 2010.
- [20] Shokry S (2017) *Wear Behavior of Ti-6Al-4V for Joint Implants Manufactured by Electron Beam Melting*. [Thesis/Publication].