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IMPACT OF NOVEL DENTAL IMPLANT SURFACE MODIFICATIONS ON OSSEOINTEGRATION AND EARLY HEALING

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Abstract
By adjusting the etching and polishing steps, the surface roughness may be fine-tuned. Using a nanofiber-coated surface is another way to improve osteoconduction. Furthermore, we have shown that covering the titanium surface with nanofibers increases cell adherence. There are a number of methods being researched or suggested to enhance osseointegration by alteration of the implant surface. In addition to increasing and speeding up osseointegration, the use of current surfaces in dental clinical practice may also decrease peri-implant bone loss and enhance the likelihood of re-osseointegration of a damaged surface. In this work, we summarize the literature on the cellular-substrate interaction and in vivo investigations that evaluate the response to these new surfaces. Type I collagen (Koken Co. Tokyo, Japan), polylactic acid (100,000 Da, Aldrich Chemical Company, Inc., St. Louis, U.S.), and 1,1,1,3,3- hexafluoro-2-propanol (HFP, Aldrich Chemical Company, Inc., St. Louis, U.S.) were all employed in the electrospinning process. In addition to increasing and speeding up osseointegration, the use of current surfaces in dental clinical practice may also decrease peri-implant bone loss and enhance the likelihood of re-osseointegration of a damaged surface. An innovative approach to improving osseointegration with the bone regeneration concept is to use functionalized nanofibers as a surface modification for dental implants.

Keywords: Healing, Osseointegration, Dental Implant, clinical, bone

INTRODUCTION

Direct bone contact has allowed osseo integrated implants to serve as both tooth replacements and orthodontic anchors. In 1969, Brnemark et al. first proposed the idea of osseointegration. This created

what the researchers called "a direct functional and structural connection between living bone and the surface of a load carrying implant." Zarb and Albrektsson presented an alternative clinical description of osseointegration as "a process whereby clinically asymptomatic rigid fixation of alloplastic materials is achieved and maintained in bone during functional loading." Titanium dental implants have had their surfaces modified in order to enhance their biological surface qualities, which in turn promotes the osseointegration process. The first generation of implants employed machined surfaces designed using traditional techniques that required many months to accomplish osseointegration. As a result, machined surfaces are no longer often used in therapeutic settings, and instead, topographic and chemical surface alterations are preferred. Blasting with ceramic particles/acid etching, titanium plasma spraying, electrochemical anodization, and calcium phosphate coatings are only some of the ways that have been developed to improve the surface roughness of dental implants. In addition, implant dentistry has seen the emergence of a new area of study focused on the application of bioactive molecules to the surfaces of titanium implants. Bioactive molecules are substances that modify or stimulate the function of living organisms. Bioceramics, ions, and biomolecules are all examples of molecules having the potential to be used for bioactive purposes.

Histologically, osseointegration is characterized as "a direct... contact between living bone and implant". Clinically, the term refers to "the process by which a clinically asymptomatic rigid fixation of an alloplastic material is achieved and maintained in bone during functional loading". Osseointegration is described as "a foreign body reaction where interfacial bone is formed as a defense reaction to shield off the implant from the tissues" by Albrektsson et al. (2017a), who studied the biological response begun at the bone-implant interface after implantation. Either new bone will develop or the area will be encased in connective tissue. In the former case, integration has occurred successfully, but in the second one, it has not and will not. This is why several studies using various methods including light microscopy, electron microscopy, and radiography have focused on the contact between the bone and implant.

LITERATURE REVIEW

Rahimi, et al (2022) The purpose of this review article is to offer a synopsis of the clinical outcomes associated with different surface modifications and therapies for dental implants currently on the market. Understanding the healing process associated with each implant type would help the practicing physician choose the best implant for a given clinical circumstance. PubMed and Scopus databases were searched for relevant articles between January 1990 and July 2021. Dental implants, surface modification, surface treatment, survival rate, and clinical performance were used as keywords in the MeSH database search. This review only discusses research that were published in English-language journal articles. Implant surfaces have been developed in recent years with the goal of facilitating osseointegration at a quicker rate and with better results. To improve clinical performance, several other surface modifications have been created and are being employed, such as the turned (machined), hydroxyapatite-coated surface, titanium plasma-sprayed, grit-blasted, acid-etched, anodization, laser-microtextured, and combinations thereof. Integration into the jawbone is a crucial factor in the long-term success of dental implants. Both the immediate and long-term success of dental implants depend heavily on their geometry and surface texture. Early and rapid loading of dental implants has emerged as a potential alternative to the usual loading strategy as a result of surface alterations that speed up the osseointegration process.

Meng, Hsiu-Wan & Chien, Esther & Chien, Hua Hong (Ben). (2016) This article aims to provide an up-to-date overview of the latest research on physiologically active dental implant surfaces and their impact on osseointegration. The time span covered by the PubMed search was from January 2006 to January 2016. Only research that examined biomolecular coatings on titanium dental implants in animals or humans after they had been surgically implanted into the bone was included. This review draws from 34 papers that were not previously reviewed. Four types of biomolecular coatings were assessed according to the criteria. Eight articles discuss the possible biomolecules bone

morphogenetic proteins, eight discuss additional growth factors, five discuss peptides, and thirteen discuss the extracellular matrix. The majority of the publications examined a recovery time between one and three months, with a maximum of six months. In addition, with the exception of a single study that looked at implants in both animals and humans, all of the research included the insertion of implants into animals. Biomechanical testing (including removal torque, push-out/pull-out tests, and resonance frequency analysis) and histomorphometric analysis (including percentage of bone-to-implant contact and peri-implant bone density) both point to an improvement in performance after dental implants have been modified with biological molecules. Osseointegration may not always be improved by bioactive surface changes on implant surfaces. However, biomolecular coatings applied to the surface of titanium dental implants seem to encourage peri-implant bone growth, leading to improved osseointegration in the early phases of recovery. Longitudinal clinical trials are required to confirm this finding, however. Clinicians also need to remember that research conducted on animals may not always provide accurate predictions of human clinical outcomes.

Smeets et al (2016) In this research, we'll take a look at how various dental implant surface alterations affect osseointegration. The article talks about both commercially available and experimental surface changes. Provide oral rehabilitation for patients with good bone conditions calling for quick loading procedures or patients with quantitatively or qualitatively damaged bone is the primary problem for modern dental implantologists. Improvements in implant surface design are needed to accommodate these charging circumstances. Researchers have been motivated to develop implant surfaces that mirror the features of natural bone by the discovery of the physiology of bone mending. In order to improve osseointegration in both healthy and damaged bone, this study gives a complete review of surface changes that affect the topography, hydrophilicity, and outer coating of dental implants for the better. Successfully utilized dental implants for many years are the primary topic of this paper's first section, which focuses on sandblasting, acid-etching, and hydrophilic surface textures. The next section discusses cutting-edge methods such as Discrete Crystalline Deposition, laser ablation, and protein, drug, and growth factor surface coatings. Novel dental implant surfaces have been developed, marking a significant technological development. These advances pave the way for rehabilitating patients with high success and consistent survival rates, no matter how dire their circumstances.

Kligman, et al (2021) As the need for oral rehabilitation in patients with healthy and deficient bone has grown, so has the sophistication of implant surface design. For instance, implant surfaces have been changed to bring desirable qualities to a dental implant and so raise the implant success rate and widen their indications in order to combat the most prevalent dental implant-related problems, peri-implantitis and consequent implant loss. Titanium, zirconia, and polyether ether ketone are just a few examples of the wide variety of implant materials that have had their surfaces modified via physical, chemical, and biological methods. By optimizing the implant's surface interface with the surrounding bone, we may speed up the osseointegration process and lower the likelihood of biofilm development caused by bacteria. Critical for clinicians to choose the most suitable materials to improve the success and survival of implantation, this review article aims to comprehensively discuss currently available implant surface modifications commonly used in implantology in terms of their impact on osseointegration and biofilm formation.

Sethi, et al (2023) Long-term success with endosseous dental implants depends on a process called osseointegration. Implant surface parameters, including as roughness, topography, energy, and composition, are the key surface components that determine osseointegration. In order to increase surface area and facilitate the process of osseointegration, implant surface roughness has been improved using both additive and subtractive methods. Anchoring implants securely and over the long term is essential for the long-term success of any orthopedic surgery including an end prosthesis. These alterations provide a more rapid and potent Osseo integration. Hydrophilic features added to roughened surfaces or specific osteogenic peptides coated on surfaces have recently shown increased biocompatibility and have produced speedier Osseo integration than previously modified surfaces.

New insights into the properties, responses, and behavior of materials might be gained through the study of surface engineering, which in turn could lead to the development of innovative materials, modification techniques, and the design of bioimplants.

RESEARCH METHODOLOGY

Implant surface and osseointegration

The steps involved in osseointegration have been studied in animals and are briefly summarized here. Ions (e.g., Ca) and plasma proteins (e.g., albumin, globulin, and fibrin) bind to the implant surface immediately after surgical trauma (0-4 hours) on the implant side of the incision (Figure 1A). Because this protein coat is what allows inflammatory and tissue-forming cells to adhere to the implant surface, it is a crucial step in the osseointegration process. Titanium's biocompatibility and osseointegration are predicated on the formation of an oxide layer upon contact with blood. Nanoscale examination of Osseo integrated implants revealed a slow incorporation of Ca and P ions into the Ti oxide layer. Bone ingrowth into the nanostructured features of the implant surface is supported by the fact that the bone-implant interface is not a crisp line of transition but rather a 3-D gradient of Ca, P, and O decreasing from the bone to the implant side where Ti ions rise. Jarmar discussed the possible impacts of surface roughness on osseointegration, as well as the development and characteristics of titanium oxide on the nanoscale. Platelet adherence, microparticle creation, and P-selectin expression may all be influenced by the implant's surface microtexture, which in turn improves the device's osteoconductivity.

Figure 1 (A) Clot formation in the blood. Inflammation is triggered by the release of pro-inflammatory, angiogenic, and chemotactic chemicals (pGF, fh, fbg) from platelets (P) and red blood cells (RBC) trapped in the fibrin network. Drilling produces bone debris, some of which may be seen (DEBR), as well as neutrophils (N), and bacteria (B). Inflamed state; case B. Together with neutrophils, macrophages emit growth factors (mGF) that trigger the ensuing proliferative (angiogenesis and the production of osteoid provisional matrix [OM]) by digesting bacteria (B) and bone debris (DEBR). Osteoclasts (Oc) are cells that resorb bone from the surface. The development of granulation tissue (C). Endothelial cells in the healthy bone infiltrate the wound site and begin forming new blood vessels. The OM bridges the defect and builds around the new blood vessels. Osteoprogenitor cells get activated and express components for the tissue mineralization conducted by osteoblasts (Ob) when bone drilling exposes bone morphogenetic proteins (BMP), growth factors, and differentiation factors contained in the bone matrix. New immature bone (IB) forms on the implant and pristine bone surface (D). The OM, lined by the Ob, serves as the ossification center and undergoes progressive remodeling in lamellar bone. Bone resorption-inducing proteins (RANKL) tend to cluster in construction zones. (E) Lamellar bone growth and Oc and Ob-mediated IB remodeling.

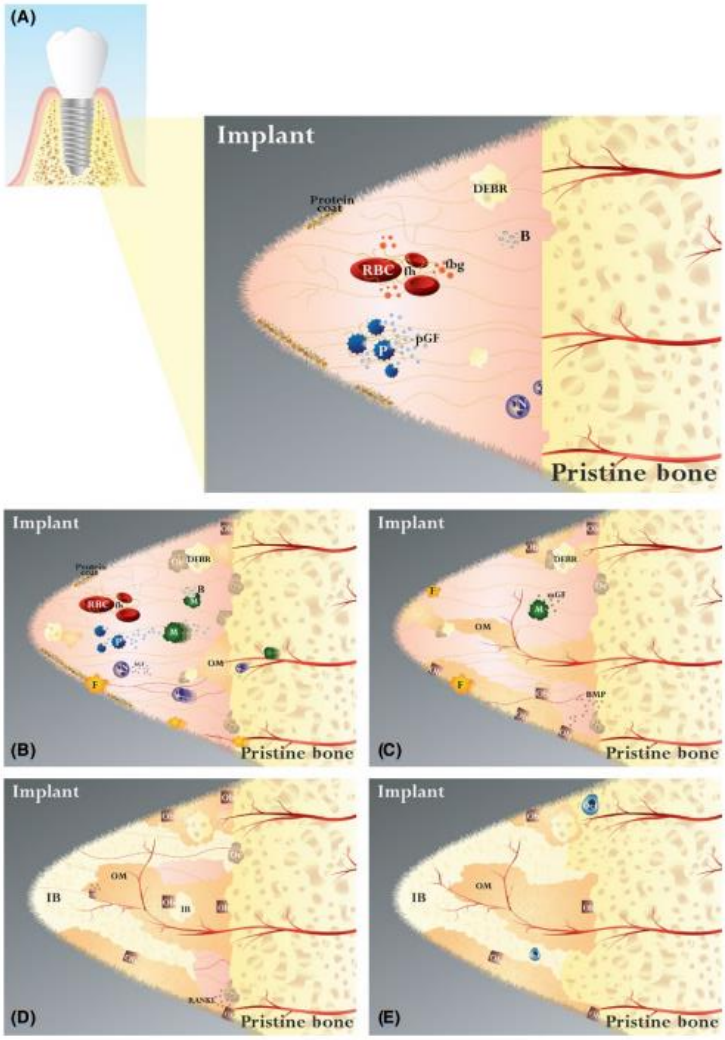

Figure 1: Interaction of the implant with the bone just after surgery (A), a few days later (B), a week later (C), two weeks later (D), and four weeks later (E).

In a few days (three to four), following surgery Figure 1B shows that the spongy area of virgin bone is where angiogenesis and blood clot organization are most active in granulation tissue, while the coagulum with a large number of erythrocytes is still present on the implant side of the incision. A thin coating of osteoid matrix is placed directly onto the metal surface lining the implant about 1 week following surgery (Figure 1C). Human implant surface cells were shown to upregulate genes involved in the development of extracellular matrix and collagen fibril organization. Immature bone apposition along the pristine bone and the implant surface increases from the places where old bone contacts the implant in succeeding healing stages (after 2 weeks) (Figure 1D). As shown in Figure 1E, the first evidence of bone remodeling conducted by osteoclasts arise in primary osteons, a provisional matrix lined with osteoblasts that functions as a center of ossification and is eventually reformed into lamellar bone. Table 1 displays the most important implant surface-related parameters that may influence osseointegration at various stages of the healing process.

Table 1: Surface topography's effect on osseointegration throughout the healing process

Healing phases	Influence of the surface on osseointegration
Early healing phase: immediately after surgery	Affinity, adsorption, and adhesion of plasma proteins and coagulum to the implant surface, and the titanium oxide formation seemed to be influenced by the microtopography and physical-chemical properties (hydrophilicity or hydrophobicity) of the surface, as the latter affect blood clot adhesion and stabilization, and titanium oxide formation. Higher blood clot affinity and earlier and greater deposition of the osteoid matrix were found on chemically-modified (chemically active) SLA (same sandblasting and etching procedure as SLA, but rinsed under N_2 protection and directly stored in an isotonic NaCl solution, again protected by N_2 filling) than on untreated SLA surface.
Few days after surgery (3-4 days)	A greater number of cells (also fibroblast-like cells) and improved bone metabolism were found on rough (SLA and oxidized) implant surfaces than on machined surfaces. Higher levels of genes for proteins inducing bone formation (osteoprotegerin, osteocalcin, and alkaline phosphatase) and bone resorption (cathepsin K, RANKL) were found expressed by cells on oxidized implant surfaces than on machined surfaces. Machined implants induced higher expression of pro-inflammatory cytokines (tumor necrosis factor at day 1 and interleukin 1β at days 1 and 6).
1 week	Cuboidal osteoblast-like cells producing the osteoid matrix with a higher amount of newly-formed bone have been observed on SLA surfaces. Fibroblast-like cells on machined implants.
2-4 weeks	A higher amount of newly-formed bone was found on the SLA active surface than on the SLA surface. Studies in humans reported that bone implant contact progressively augmented from approximately 12.2%-14.8% after 2 weeks on SLA and SLA-active surfaces, respectively, to 32.4% (SLA) and 48.3% (SLA active) at 4 weeks, and 62% at 6 weeks for both surfaces.

RANKL, receptor activator of nuclear factor κ-B ligand; SLA, sandblasted and acid-etched implants.

Advanced electrospinning for biomimetic surface modification of dental implant materials

- **Mechanical Polishing/ etching:**

Mechanical polishing using 320 grit and 400 grit SiC papers yielded a mirror finish on discs of pure Titanium (15mm diameter) and Titanium alloy (Ti- 6Al- 4V) (25mm diameter) acquired from the Northwest Institute for Non - Ferrous Metal Research (Xian, Shanxi, P.R. China). Alumina (1M) cloth was then used to further polish the discs. The discs were then ultrasonically cleaned with ethanol for 15 minutes. This guarantees the elimination of any contaminants introduced by the mechanical polishing process. After the mechanical process was complete, the surface was etched chemically for 1 minute using 4% HNO3 in ethanol. The discs were left out in the open air to dry.

Poly(lactic-co-glycolic acid) (PLGA) and PLGA/Collagen nanofibers were electrospun on the Ti discs

Type I collagen (Koken Co. Tokyo, Japan), polylactic acid (100,000 Da, Aldrich Chemical Company, Inc., St. Louis, U.S.), and 1,1,1,3,3- hexafluoro-2-propanol (HFP, Aldrich Chemical Company, Inc., St. Louis, U.S.) were all employed in the electrospinning process. Pellets of poly(lactic acid) (PLGA) (75:25) were dissolved in 15% HFP by weight. Table 2 displays the results of optimizing the electrospinning settings, the w/v ratio of PLGA in HFP, and the fiber deposition duration until uniform nanofibers were formed without bead formation. The same method (Table 3) was used for electrospinning a combination of PLGA and Collagen (50:50 w/w). In order to get the fibers from the spinneret (27G1/2 needle, Becton Dickinson, BD, N.J, U.S.) onto the collector plate, over which the Ti disc was placed, a high voltage electric field was applied (DC high voltage power supply from Gamma High Voltage Research, Florida, U.S.). In Figure 2 we can see the experimental setup. To make consistent and homogeneous nanofibers, the spinneret's tip was flattened by grinding. A syringe pump (KD Scientific Inc., Maryland, United States) was used to apply a consistent feed rate of 1 mL/h.

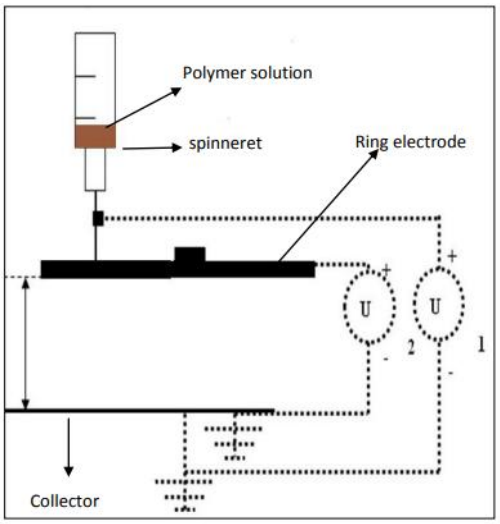

Figure 2: Electrospinning set up

DATA ANALYSIS

New surface alterations improve dental implant osseointegration

Recent biomedical and engineering studies have aimed to better create, maintain, and reestablish osseointegration by creating biomaterials and surfaces that can regulate tissue repair. According to the literature, osseointegration occurs when mineralized matrix is deposited by osteoblasts onto the implant surface. The titanium oxide layer that grows on the titanium surface facilitates this process. Several methods are being studied or newly presented in clinical practice to enhance osseointegration through this process. Most surface treatments aim to improve osteoblast differentiation, adhesion, and osteogenic activity by forming a thick layer of titanium oxide, changing the surface chemical composition by incorporating bioactive molecules and drugs, or modifying the surface topography.

Roughness of a surface

Research into the osseointegration of titanium implants has shown that the surface roughness has an effect on the behavior of osteogenic and inflammatory cells. Preclinical research evaluating implants with a similar nano-topography was driven by promising results from cell culture experiments performed on TiO2 nanotubes made by anodic oxidation. According to von Wilmowsky's findings in a pig calvaria model, titanium implants coated with TiO2 nanotubes with a diameter of 30 nm seemed to increase osteoblast function at an early stage of healing; however, apparently no significant increase in bone mineralization process and in the osteoblast proliferation rate is produced compared to untreated implants. Further studies on rabbits found that implants with nanostructured surfaces (TiO2 nanotubes with mean pore diameters of 108 and 37 nm) had greater osseointegration strength (measured by torque removal and pullout test) and new bone formation than implants with machined or microrough surfaces (blasted). Also, Lim et al. found that after 4 weeks of healing, there was considerably more bone to implant contact on the total surface (cortical and medullary region) of anodized implants than machined implants. Anodized implants were still better after 12 weeks of recovery, but only in the medullary region. Researchers have studied the effects of various nanotube diameters on bone tissue by inserting four titanium cylinder implants with machined or anodized surfaces into the skull of a miniature pig. The latter included a wide range of nanotube sizes, from 30 to 100 nm. When compared to machined implants, anodized implants showed greater osteogenesis gene expression, bone-implant contact, and bone deposition. Stem cell development into the osteogenic lineage seemed to be hastened by exposure to bigger diameter (70 and 100 nm) nanotubes. According to the findings, nanotubes with a diameter of 70 nm are the most effective in boosting implants' osteo conductivity and osseointegration.

Surface characterization analysis

Using an electrostatically driven jet of polymer solution, the simple and flexible method of electrospinning may create polymer nanofibers with diameters ranging from a few nanometers to several micrometers. The typical electrospinning apparatus is seen in Figure 2. A polymer solution droplet is kept at the end of a capillary tube, and an electrical potential is supplied between the droplet and a grounded target. As the droplet's surface tension is overcome by the external electric field, a charged jet of polymer solution is released. The electric field determines the path taken by the charged jet. Electrospinning naturally facilitates the precise placement of polymer fibers because to this characteristic. Nanofibers were positioned and aligned on a tapered and grounded wheel-like bobbin using an electrostatic field-assisted assembly approach detailed in the paper. Our results indicate that nanofibers were selectively deposited onto discs made of commercially pure Ti (cpTi) and Ti alloy (Ti6Al4V) during the deposition process. Hohman, Shin, Rutlege, and Brenner also revealed findings from their research on the stability of electrically induced fluid jets and instabilities during electrospinning. It was shown that the electrical instability grows with the intensity of the field. As can be seen in Figure 3, these findings agreed with those of our investigation employing a cutting-edge electrospinning method. The left-hand diagram has 18kV at the needle and 10kV at the ring, whereas the right-hand one has 14kV at the ring and 18kV at the needle. Inconsistencies in the electric field may be seen beginning around 14kV.

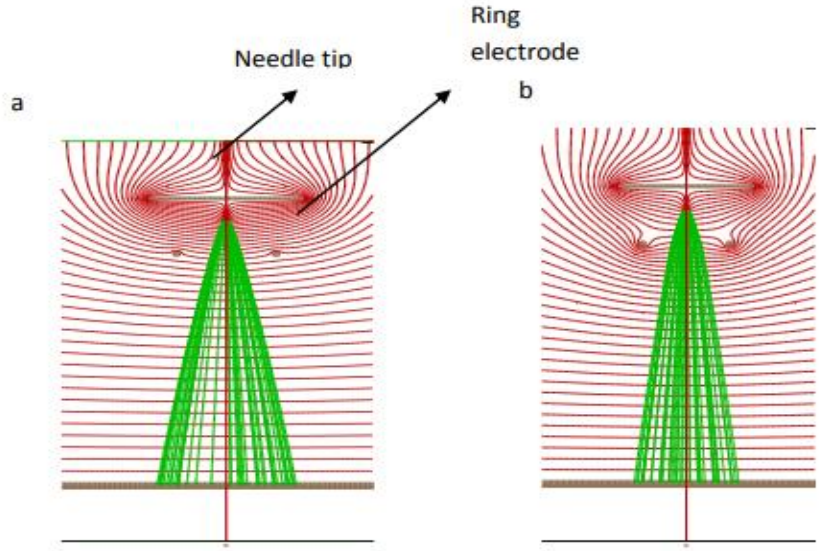

Figure 3: Form of an electric field the needle is charged to 18kV, while the ring electrode is charged to 10kV. b) 18 kilovolts (kV) at the needle's tip and 14 kV (kV) at the ring electrode

An extra ring electrode was added to concentrate the electrospinning current. The ring electrode was attached to a high voltage electric field source, as indicated in Figure 2. Figure 3 depicts the electric field pattern that was analyzed to determine the optimal voltages across the two-power supply. The electrospinning process was found to be limited when the needle voltage was 18kV and the ring electrode voltage was 14kV. This was due to the fact that, as seen in Figure 3, the electric field began to exhibit anomalies at 14kV. When the ring electrode voltage was lowered to 10kV, however, the electric field was smooth and free of any irregularities that would impede the electrospinning process. The spot where fiber was deposited shrank when the voltage was raised. After electrospinning, the nanofibers were vacuum-dried to get rid of any lingering solvent. When the voltage was increased, the size of the irregularity increased, too, suggesting that the diameter of the deposited fibers shrank. In reality, it is working against the ring's electric field, but it is too weak to completely halt the electrospinning process. As opposed to this, the electric field at the ring for 10kV is quite uniform, with no distinct characteristics that may interfere with the electrospinning process. The reduction in fiber deposition site shown by green lines in Figure 3 is for illustrative purposes only.

Table 2: Electrospinning PLGA nanofibers with optimized parameters by adjusting duration and concentration

Electrospinning PLGA nanofibers at various concentrations (15%, 18% and 20% w/v) and time periods (5 seconds to 2 minutes)			
Time	**Nanofiber diameter ± SD (micro meter)**		
	15%	**18%**	**20%**
2 min	0.68 ± 0.282	1.761 ± 0.371	1.800 ± 0.213
1.5 min	0.774 ± 0.227	1.212 ± 0.39	0.914 ± 0.151
1min	0.543 ± 0.153	1.114 ± 0.357	1.089 ± 0.267
30sec	0.768 ± 0.314	1.326 ± 0.479	1.067 ± 0.194
15 sec	0.957 ± 0.357	1.615 ± 0.472	1.731 ± 0.386
10sec	0.996 ± 0.344	1.721 ± 0.413	1.264 ± 0.269
5sec	0.759 ± 0.415	1.535 ± 0.594	1.381 ± 0.449

Table 3: Electrospinning parameters for PLGA/Collagen nanofibers optimization through time and concentration changes

Electrospinning PLGA/Collagen nanofibers at various concentrations (10% and 15% w/v) and time periods (5 seconds to 2 minutes)		
Time	**Nanofiber diameter ± SD (micro meter)**	
	10%	**15%**
2 min	0.549 ± 0.213	0.827 ± 0.116
1.5 min	0.279 ± 0.085	0.898 ± 0.176
1min	0.368 ± 0.089	0.801 ± 0.147
30sec	0.251 ± 0.093	0.776 ± 0.136
15 sec	0.378 ± 0.068	0.817 ± 0.151
10sec	0.410 ± 0.093	0.828 ± 0.185

As can be seen in Tables 2 and 3, the fiber diameter was optimized by adjusting the deposition duration and concentration factors. SEM micrographs revealed that the quantity of fiber deposited increased with deposition times longer than 15 seconds. Therefore, it is preferable that the cells make contact with both the nanofibers and the nano-topography of the Ti substrate to guarantee osseointegration. The SEM pictures show that the fiber deposition was not uniform for the 10-second and 5-second deposition times. Since then, 15 seconds has been settled upon as the sweet spot for deposition in subsequent research. Tables 2 and 3 show that the diameter of the fibers became larger as the polymer concentration rose. Even though the fiber diameter was reduced for polymer concentrations below 10% and 15% for PLGA/Collagen and PLGA correspondingly, bead formation and nonuniformity of fibers were observed. Therefore, PLGA nanofibers and PLGA/Collagen nanofibers were both spun with a polymer concentration of 15% and a deposition period of 15 seconds for further research.

CONCLUSION

This paper concludes that in order to promote and speed up implant osseointegration, as well as to decrease the incidence of peri-implant bone loss and favor the re-osseointegration of afflicted surfaces, the use of contemporary surfaces should be advocated in dental clinical practice. Through our research, we confirmed that a nanotextured surface on titanium could be produced using just acid and alkali. By adjusting the etching and polishing steps, the surface roughness may be fine-tuned. Furthermore, we have shown that covering the titanium surface with nanofibers increases cell adherence. This is due to the fact that the nanofibers function similarly to the body's natural ECM, enhancing cell adhesion. Our electrospinning setup allowed us to deposit fibers with high accuracy in a short amount of time. Our apparatus is more effective at depositing fibers than the standard method of electrospinning. In addition, we demonstrated that the synergistic impact of collagen and n-HA on cpTi and Ti alloy samples coated with biomineralized PLGA/Collagen nanofibers resulted in the highest adhesion efficiency of the hMSCs compared to the other samples.

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