Chapter 3
Surface Modification of Dental Implant Improves Implant–Tissue Interface

Takashi Inoue and Kenichi Matsuzaka

Abstract The implant material must have optimum surface compatibility with the host epithelial tissue, connective tissue, and bone tissue. Because, dental implants, which are partially exposed to the oral cavity, must have firm contact with tissues to prevent the bacterial infection. Such materials can be created under well-controlled conditions by modifying the surfaces that contact these tissues. The rough and grooved surfaced implant contributes to a more rapid cell migration and make osseointegration during wound healing. A number of chemical and physical methods for titanium and/or zirconium surface modification have already been established. Recently, plasma treatment can control surface physiochemical properties and affect protein adsorption for bioengineering. Moreover, the "motif-programming" methodology to "biologically" modify titanium and zirconium surfaces has created interfacing artificial proteins that endowed those surfaces with cell-binding activity. These technique should improve firm contact between tissue and dental implant.

Keywords Dental implant • Surface modification • Tissue interface

3.1 Dental Implant–Tissue Interface

Dental implant therapy creates an open wound, and an epithelium-implant interface which is always exposed to the possibility of inflammation is formed [1–3]. Peri-implantitis is a risk factor for a number of age-related diseases, including diabetes, arteriosclerosis, cardiac infarction and NASH (Fig. 3.1). It is important to create a firm implant–tissue interface from such a viewpoint.

Implant–tissue interface formation occurs during the process of wound healing [2, 3]. The oral mucosa is penetrated along the implant surface after the implantation and as a result, peri-implant epithelium is created (Fig. 3.2). Peri-implant epithelium lack the junctional epithelium that are normally formed by

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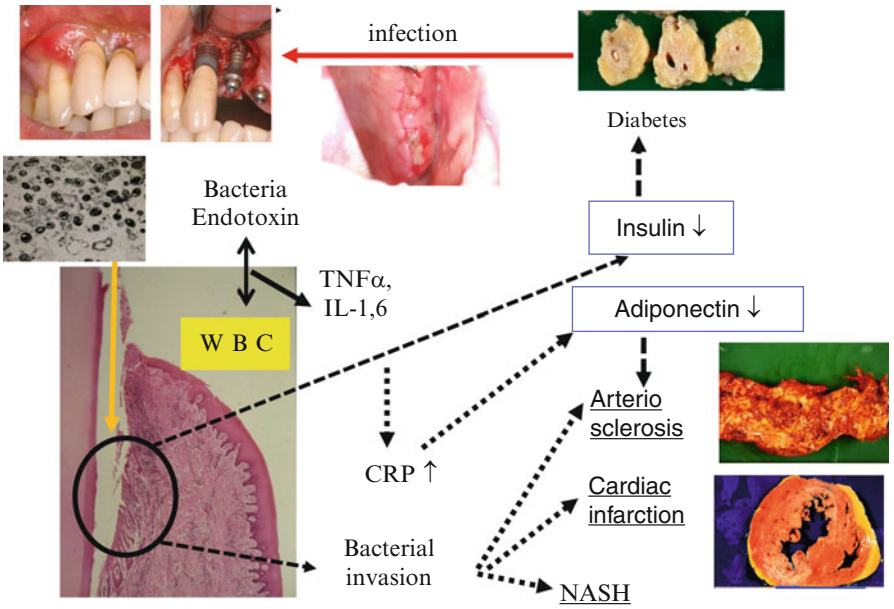
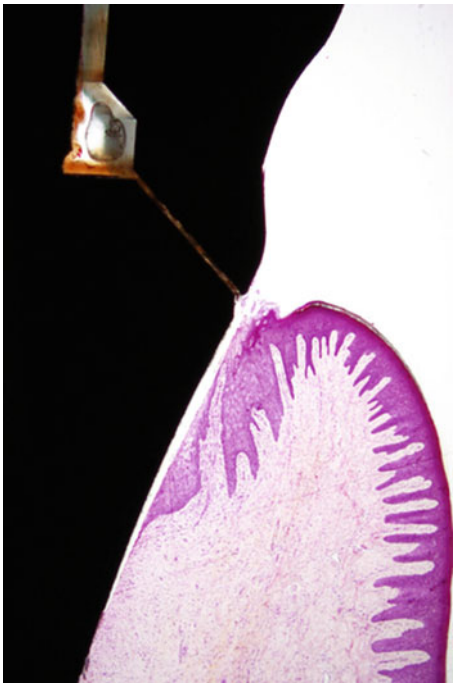


Fig. 3.1 Peri-implantitis is a risk factor for a number of diseases

Fig. 3.2 Ground section of the periimplant epithelium



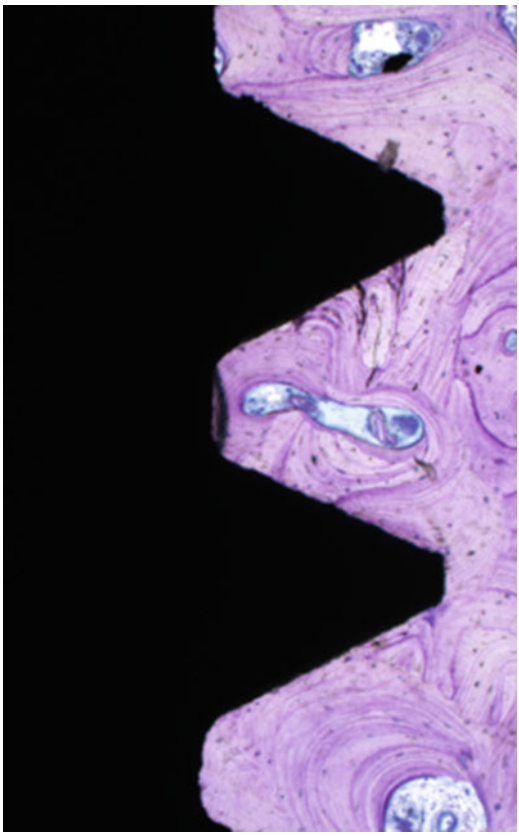


Fig. 3.3 Ground section of osseointegration

hemidesmosomes and the basal lamina, which connect enamel and epithelium of natural tooth [4].

Fibroblast face to the implant surface differentiates into osteoblasts also during the process of wound healing. The osteoblast deposits bone matrix on the implant surface, becomes calcified, and completes osseointegration which is complicatedly associated with the implant materials. It is known that a direct bond between implant and surrounding bone has been demonstrated with implants made of bioactive materials, i.e. bio-glasses and calcium phosphate ceramics [5, 6].

Titanium is known to have a greater ability than other metals to facilitate osseointegration, which is defined as a close contact between bone tissues and implant material such that there is no progressive relative motion of living bone and implant under functional levels and loading for the life of the patient (Fig. 3.3). Even when light microscopy confirms osseointegration of titanium implants, examination by electron microscope reveals that the bone and the implant are not crystallographically continuous (Fig. 3.4). The thin amorphous structures between the bone and the titanium implant, that is, there is no direct contact between titanium and bone was also observed [7].

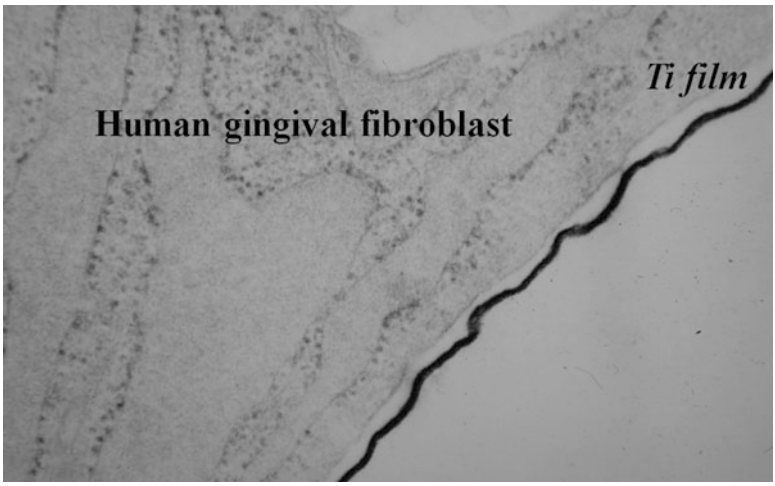


Fig. 3.4 Ultrastructural feature of Human gingival fibroblast titanium film (Ti) interface in vitro

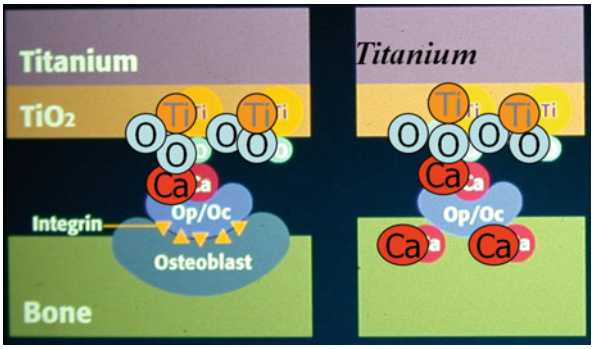


Fig. 3.5 Hypothetical osseointegration on the titanium surface. *Ti* Titanium, *O* Oxygen, *Op* Osteopontin, *OC* Osteocalcin, *Ca* Calcium [2]

Generation of the titanium oxide film on the surface of titanium is one reason for this ability and its high level of corrosion resistance. In addition, the degree of the deposition of calcium phosphates in body fluid is greater on titanium than on other metals. Presently, adsorption of osteogenic proteins such as osteocalcin (Oc) and osteopontin (Op) to the titanium surface is a main function of the osseointegration of titanium [2, 3]. There are two mechanisms involved in the adsorption of osteogenic proteins. Titanium oxide has a similar number of isoelectric points (pI) at approximately pH = 5 as those of pH = 4.7–4.9 on osteogenic proteins. Accordingly, at around pH 7, both titanium oxide and osteogenic proteins are negatively charged. The calcium ion-mediated mechanism caused by the positively charged calcium ions, Ca^{2+}. The hydration effect of terminal OH radicals which are positively charged, is also considered as playing a role in protein adsorption (Fig. 3.5) [8].

In view of the direct bone-implant contact, it plays important roles of surface geometry and surface chemistry of the implant material, and cell behavior surrounding implant.

3.2 Effect of the Surface Geometry

Brunette et al. [9] discuss that the grooved surface changes the cytoplasm by offering a microenvironment and prepares a good condition for generating the calcifying tissue, and that cells which are arranged along the groove help the osteogenic cell differentiate to osteoblast (Fig. 3.6). Further, vinculins, which is one of the proteins of attaching to the substrate, are also oriented to the microgroove (Fig. 3.7). To support cellular attachment, spreading and growth, and improve cellular function, a lot of reports have been published about roughened implant surfaces (Fig. 3.8) [10–13], as well as on controlled microtopography [9, 13–19]. We also explored how the fibroblast originated from human gingiva reacts against the titanium discs of various surface geometries. In the phase-contrast microscopic view of titanium disc with mechanically polished grooves on the surface, it is observed that cells are surrounding the disc. In the scanning electron microscopic view, cells are arranged along the groove (Contact guidance (Fig. 3.9) [10]. On the other hand, in the phase-contrast microscopic view of titanium disc with rough surface, cells are arranged vertically to disc. In the scanning electron microscopic view, cells are arranged in free directions to disc, and cell bridge is geometryed (Two center effect: Fig. 3.10) [10].

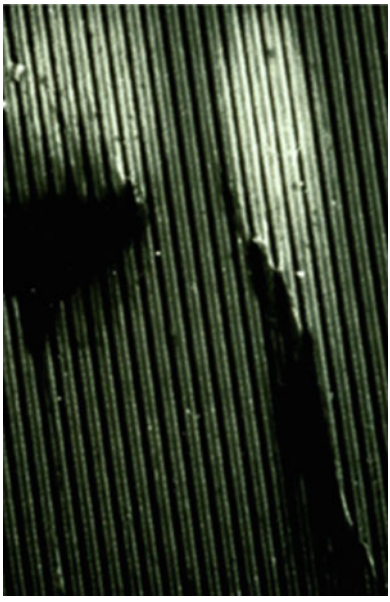


Fig. 3.6 Scanning electron microscopic findings of fibroblast on microgrooved surface

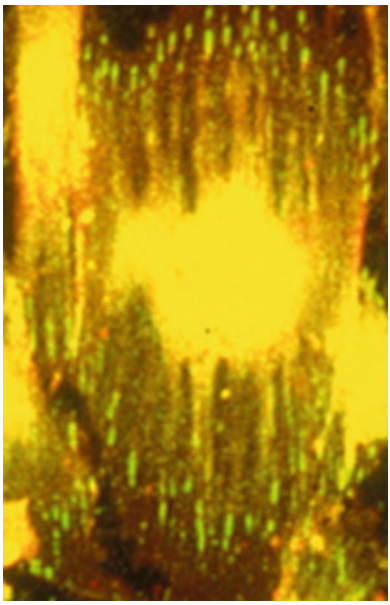


Fig. 3.7 Confocal scanning microscopic findings of osteoblast using vinculin on microgrooved surface

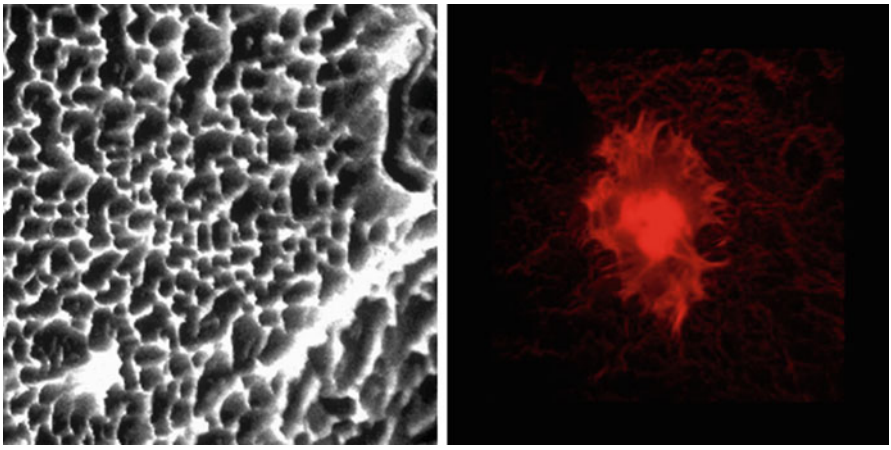


Fig. 3.8 Scanning electron microscopic findings of rough surface implant (SLA: *Left*) and Immune-fluorescence microscopic findings of cells on the SLA

3.3 Control of Surface Chemistry

Surface chemistry involves the adsorption of proteins and cells on biomaterials. This adsorption reflects the affinity between two substances, and the strength of adsorption follows the order: chemical adsorption including covalent bonds and ionic bonds > electrostatic force found in electrokinetic potential or zeta

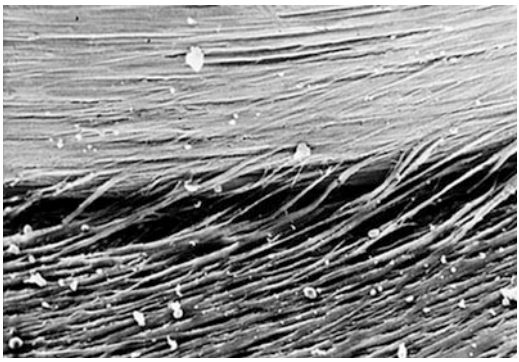


Fig. 3.9 Numerous cellular bridges that extend from the multilayer to the oriented cell sheet and orientated parallet to the grooves (contact guidance)

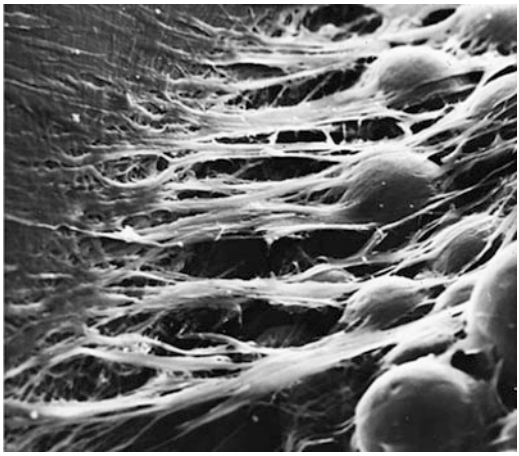


Fig. 3.10 The migration of fibroblasts onto rough surface and numerous cellular bridges oriented at right angle to the rim of the disc (Two center effect)

potential > hydrogen bonds involved in hydrophilic groups such as –OH, –COOH, and $–NH_2$ > hydrophobic interaction (i.e., adsorption of hydrophobic substances in water) > van der Waals forces. Adsorption characteristics are primarily evaluated by hydrophobicity (wettability), which can be determined by measuring the surface energy (contact angle), and electrokinetic potential (zeta potential, isoelectric point), which reflects surface electric charges and these affect creation of firm integration between implant and cells.

3.4 Protein Application

As for the surface chemistry, methods of modifying the titanium surface using adhesive proteins such as osteonectin, fibronectin or laminin-5 compatible with the soft tissue/implant interface have been proposed. For the implant surface in contact with subepithelial connective tissues, tresyl chloride treatment is used to

adhere the selected proteins such as fibronectin to the amino residues [20]. Thus the gingival epithelium attached to dental implants through the formation of hemidesmosomes using laminin-5 [21]. However, a stable coating and prevention of protein denaturation at the time of implantation are necessary using motif-programming or plasma treatment.

3.5 Application of Motif-Programming

Motif-programming is a method for creating artificial proteins by combining functional peptide motifs in a combinatorial manner. This method is particularly well suited for developing liaison molecules that interface between cells and inorganic materials. Here we describe creation of artificial proteins through the programming of two motiefs, a natural cell attachment motif (RGD) and an artificial Ti-binding motif (Fig. 3.11). Although the interaction with Ti was not covalent, the proteins recapitulated several functions of fibronectin, and thus, could serve as an artificial ECM on Ti materials. Because the motif-programming system could be easily extended to create artificial proteins having other biological functions and material specificities, it should be highly useful for application to dental implant and tissue

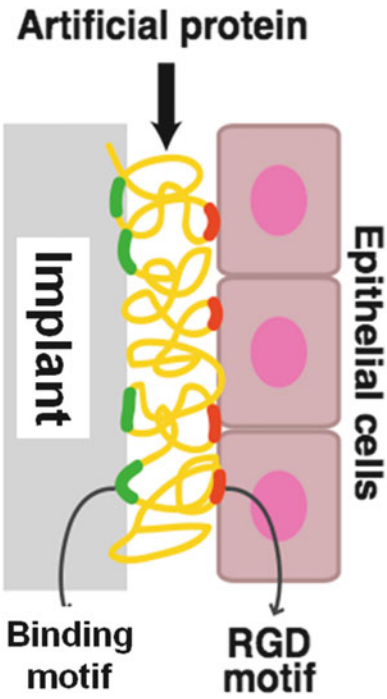


Fig. 3.11 Schematic representation of an artificial protein intermediating between the surface of titanium and cells. Artificial proteins are shown as yellow containing Ti binding motif (TBP: *green*) and a cell binding motif (RGD: *red*) [22]

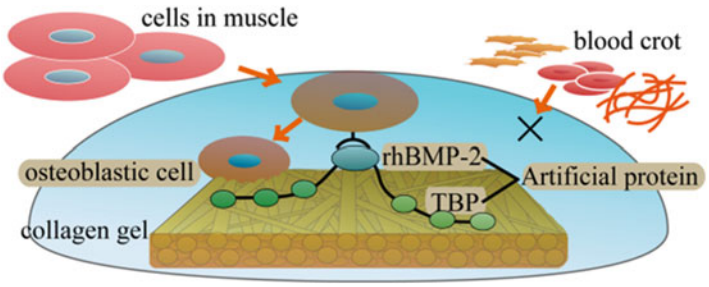


Fig. 3.12 Schematic image of the AFT between BMP-2 and TBP [24]

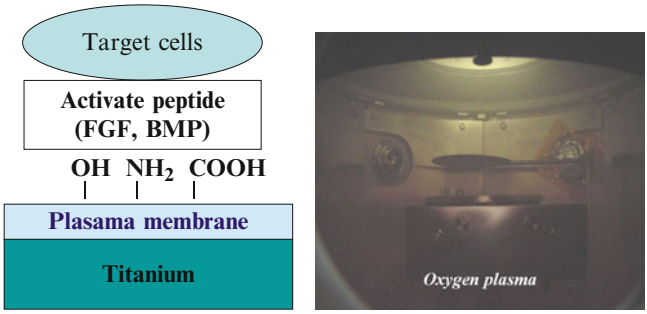


Fig. 3.13 Plasma treatment was performed using the VEP-1000 system (*right*) and schematic image shows the attachment of cells on the plasma treatment titanium surface (*left*)

engineering [22, 23]. Yuasa et al. using an artificial fusion protein between bone morphogenetic protein 2 and titanium-binding peptide and reported that this artificial protein accelerates osteogenesis in the muscle tissue and suggests its possible use in dental implant for better osseointegration (Fig. 3.12) [24].

3.6 Plasma Treatment of Implant Surface

Plasma treatment is a well-established method of surface processing in the microelectronics industry for effective surface modification, exhibiting high surface energies, good wettability and cleaning (Fig. 3.13). This technique can control surface physicochemical properties and affect protein adsorption, and is of particular interest in biomedical engineering. Matsuzaka et al. reported that bone morphogenetic protein-2(BMP-2) and fibronectin could be immobilized using oxygen plasma treatment. Immobilization of GFG-2 on an implant using modified surface topography might allow proliferation of periodontal ligament cells around the implant [25, 26].

3.7 Calcium Phosphate (Ca-P) Coating by Plasma Spraying

Ca-P implants, including hydroxyapatite (HA), are well known for good osteoconductivity (the early stage of osteogenesis) as well as for direct binding to bone tissue in vivo. Alkaline phosphatase expression and parathyroid hormone response were higher in cultures grown in HA than in cultures grown in titanium [27] and the in vitro formation of extracellular matrices was greater on Ca-P coatings than on titanium.

In spite of their rapid and strong bonds to living bone tissues and favorable osteogenic ability, Ca-P ceramics alone cannot be used for implants because of their lack of strength. Accordingly, Ca-P coatings on Ti implants produced by the plasma spraying have frequently been used [28]. These Ca-P coated implants, however, often develop fractures in their coatings as well as at the titanium interface after implantation. The reason for this is thought to originate in the comparatively thick, porous, non-uniform (crystalline surrounded by an amorphous mass), and poorly adherent Ca-P layer produced by plasma spraying. These fragments of a certain size cause phagocytosis by macrophages, leading to inflammation. It is therefore desirable for the materials to be rapidly and completely absorbed in the host tissues and to be entirely replaced with bone tissue. When osteogenesis occurs at the site where old bones are absorbed (remodeling of bones), the Ca-P coatings should be no thicker than necessary.

3.8 Thin Ca-P Coatings

Attempts have recently been made to solve problems, the cold plasma, ion-plating [29] and the ion sputtering [28], which are a kind of physical vapor deposition (PVD), are used to produce implant materials consisting of a thin, homogeneous, and adherent Ca-P coating. Ion beam dynamic mixing (IBDM) was also introduced as a suitable technique for fabricating a thin and adherent ceramic layer [30]. This method is a combination of ion implantation and PVD, and has the advantages of a high deposition rate, producing defect-free transparent thin films, and excellent adhesion compared to conventional thin-film deposition techniques.

3.9 Future of Dental Implant

The next generation should be the surface modification of any of the materials for bio-functionalization of dental implants. Such materials can be created under well-controlled conditions by modifying the surfaces of metals that contact those tissues. “Tissue-compatible implants,” which are compatible with all host tissues, must integrate with bone tissue, easily form hemidesmosomes, and prevent biofilm accumulation.

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