

Review Article

A Review on Biomedical Applications of Titanium Dioxide

Pradeep C. Dathan¹, Deepak Nallaswamy¹, S. Rajeshkumar², Suja Joseph¹,
Shahin Ismail¹, Lakshmi Ajithan¹, Leon Jose³¹Department of Prosthodontics, ²Nanobiomedicine Lab, Department of Pharmacology, Saveetha Dental College and Hospitals, SIMATS, Saveetha University, Thandalam, Chennai 602105, India³Department of Mechanical Engineering, Indian Institute of Technology Madras, Chennai 600036, IndiaReceived: 4 February 2023
Accepted: 25 March 2023
Published online: 30 June 2023**Keywords:** titanium, titanium dioxide, titania, nanoparticles, nanotubes, cancer therapy, photodynamic

Titanium is a wonder metal with broad range of application in all aspects of life. Applications include aeronautical, industrial, military, automobiles agriculture and in the field of medicine. Titanium dioxide is the most common compound of titanium it occurs commonly in three crystalline forms namely anatase, rutile and brookite. The nano particles of titanium dioxide have low toxicity, versatile fabrication adaptability which forms the basis for its broad range of application. This review article discusses applications of titanium dioxide nanoparticles in cancer therapy, targeted drug delivery, photodynamic therapy, antibacterial effect, role in bone formation and general applications.

© (2023) Society for Biomaterials & Artificial Organs #20066823

Introduction

The wonder metal titanium was discovered in 1791 by William Gregor in Great Britain. Martin Heinrich Klaproth named it Titanium, after the 'titans' of Greek mythology. The element has an atomic number of 22 and is represented by the symbol Ti in the periodic table. The metal exists in nature as an oxide form and, on reduction, a bright metal with silver color is formed. It has got superior strength, low density and is resistant to corrosion in chlorine, aqua regia and salt water. Kroll and Hunter process is used to extract the metal from its mineral ores [1]. The alloys of Titanium can be formed by alloying titanium with various other metals like molybdenum, iron, vanadium, and aluminum to get light weight alloys with high strength. Its applications are vast, including aeronautical, industrial, military, automobiles and agriculture. In the field of medicine its application includes medical equipment, orthopedic implants, dental implants etc. The alloys can also be used for general application, including jewelry, equipment in sports and building materials. The melting point of titanium is reported as 1668°C, because of this it is commonly used as a refractory metal. The thermal and electric conductivity is low. It is para magnetic and cooling below its critical temperature of 00.49K converts the metal into a superconductor [2-5].

The most common compound of titanium is titanium dioxide (TiO₂) other names include titania or titanium(IV) oxide. It is a naturally occurring compound, formed when oxygen in air reacts with titanium. Titanium tetrachloride (TiCl₄) and titanium trichloride (TiCl₃) are the other compounds [6-8]. The commonly occurring three crystalline forms of titanium dioxide are anatase, rutile and brookite. Anatase as well as brookite are produced in large scale and are important white pigments. Anatase (80%) and Brookite (20%) form the mixed polymers of titanium dioxide and is used extensively for biomedical purposes. American food and drug administration has approved the utilization of titanium dioxide in pharmaceutical products as it is non-toxic and cost effective [9-12].

Nano particles of titanium dioxide have low toxicity, versatile fabrication adaptability and a smaller size which makes it a compound with a broad range of application. They have excellent cytocompatibility and enhances the proliferation, differentiation and spreading of osteoblast cells [12,13]. Titanium dioxide nanoparticles like titania nano tubes (NTs) are stable against disintegration when used as surface coating on implants. They also act as a delivery system that controls the drug release in implants [14]. TiO₂NTs has a porous structure which promotes bone regeneration and repair [15-19]. There is ample evidence in the literature which proves that titanium dioxide nano particles are inert and compatible when used in human body. This review

* Corresponding author
E-mail address: dathanphd@gmail.com (Pradeep C. Dathan)

discusses the future challenges and biomedical applications of titanium dioxide.

Structure and Properties

The dioxide forms of titanium have an octahedral geometry with six oxide anions are bonded to it. These oxide anions remain attached to three titanium centers. One of the dioxide rutile form has got a tetragonal crystal structure, whereas anatase and brookite exhibit orthorhombic structure. The oxygen substructures in rutile shows close packing with slight distortions. Distorted hexagonal close packing of oxide anion is found in anatase, whereas in brookite the crystal structure is cubic close packing and double hexagonal close packing.

Applications of Titanium Dioxide

Titanium in cancer therapy

Chemotherapeutic agents can cause cytotoxicity on normal cells. To overcome this currently nano technology is applied in the diagnosis and treatment of cancer [16-20]. In nano technology nano structures that deliver and release drugs are developed. The therapeutic effect of the drug is increased and at the same time the adverse effect of the chemotherapeutic agents is reduced by aiming only the diseased cells by these nano structures which acts as vehicle [21]. Since the drugs are delivered to the target and drug release is controlled, the healthy normal cells remain unaffected, this reduces the side effects. TiO_2 nano structures has the ability to enhance the therapeutic effect of conventional chemotherapeutic agents by delivering it to specific sites and by controlling the drug release. This is achieved mainly because of its high biocompatibility, non-toxicity and harmonious drug releasing ability.

Targeted/smart delivery of drugs

The therapeutic effect of anti-neoplastic drug can be enhanced by surface modified nano particles, these nano particles deliver the chemotherapeutic agent to the affected cell and reduces the toxicity of drug [16,18,20]. Studies have highlighted that TiO_2 nanostructures based chemotherapeutic agents increased the therapeutic effect of antineoplastic drugs. The application of titanium dioxide whiskers (TiO_2 Ws) for drug delivery in cancer treatment was investigated by Li et al and found that the intracellular potency of Daunorubicin (DNR) can be increased by TiO_2 Ws and it also enhances the anti-tumor efficacy of DNR in hepatocarcinoma

cells (SMMC- 7721). This is an indication for the use of TiO_2 Ws as drug delivery vehicle for DNR into the specific cells. They concluded that TiO_2 Ws based drug delivery is a favorable approach in the treatment of cancer [22]. According to Akram et al combination of Doxorubicin and titanium dioxide nano particles (TiO_2 NPs) had synergistic effect in breast cancer cell lines [23].

Venkatasubbu et al. reported the anti-neoplastic effect of paclitaxel attached to modified hydroxyl apatite and TiO_2 NPs. Biochemical analysis showed superior anti-neoplastic activity of surface modified paclitaxel attached to hydroxyl apatite and TiO_2 NPs when compared to paclitaxel [24]. The nano structure mediated smart drug delivery increased the therapeutic effect of anti-neoplastic drugs, minimizes the toxicity and enhances the biological availability and time dependent delivery of drugs.

Photodynamic therapy in cancer treatment

TiO_2 NPs is widely used in photodynamic therapy (PDT). In PDT a photosensitizing agent is administered for localizing the tumor and is activated using a light of specific wavelength [25]. This therapeutic technique has an illumination source a photosensitizer (PS) and an oxygen molecule. A non-toxic photosensitizer is introduced into the living tissues and are activated by photons from the illumination source. This transfers energy into the oxygen molecule and produces singlet oxygen ($^1\text{O}_2$) and reactive oxygen species (ROS) [26, 27]. PDT is considered as a viable alternative non-invasive treatment for cancer [28-30]. This treatment modality is successful in superficial areas and natural cavities where invasive surgery is of great risk [31-33]. Its efficacy in deep seated tumors remains questionable, as light cannot penetrate into deeper tissues and generation of singlet oxygen and other reactive oxygen species with curative quantity becomes difficult. PDT is proven to be more efficient against skin and oral infections [34,35].

In the early stages, TiO_2 NPs where activated with ultraviolet light of less than 385nm wavelength. It can create light induced holes and electrons, which can react with hydroxyl ions of water (OH^-) to form powerful oxidative radicals (OH^\cdot , HO_2^\cdot). These oxidative radicles can destroy bacteria, fungi and tumor cells. High photocatalytic efficiency, low toxicity and excellent photo stability of TiO_2 makes its application in cancer treatment a success. Fujishima et al first reported the photo destructive activity of TiO_2 to malignant cells [36]. Surface of TiO_2 electrode was cultured with Hela cells and UV irradiation was used to anodically polarize the

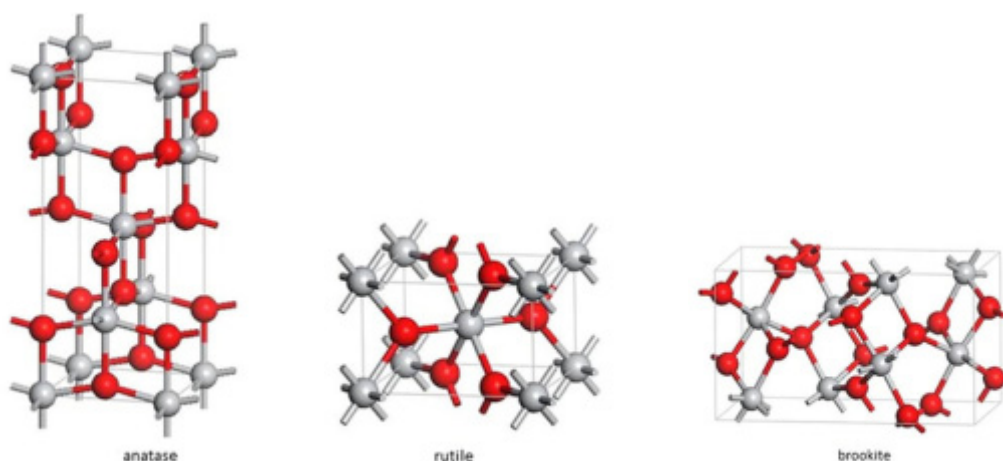


Figure 1: Crystal forms of anatase rutile and brookite

Table 1: Properties of titanium

Chemical formula	TiO ₂
Appearance	White solid
Odour	Odourless
Molecular mass	79.866 g/mol
Boiling point	2972°C
Solubility in water	Insoluble
Magnetic susceptibility (χ)	+5.9 · 10 ⁻⁶ cm ³ /mol
Refractive Index (n_D)	2.488 (anatase)
	2.583 (brookite)
	2.609 (rutile)

electrode. This caused damage of Hela cell membrane. The neoplastic cells were not destroyed when the electrodes were 10 mm away from the surface of cell. A TiO₂ microelectrode which was polarized and illuminated showed selective anti-tumor activity for single T24 cell which are cancerous [37]. Seo et al. used high temperature non hydrolytic method to make water soluble TiO₂NPs. These nanoparticles were short and rod shaped with diameter of 3.5 nm and length of 10.4 nm, which was highly toxic to human melanoma cells (A375) in the presence of UV irradiation [38]. UV light was used to activate TiO₂ but this light source was found to be damaging, therefore visible light was used to activate TiO₂ which produced a positive effect. The optical activation of TiO₂ with visible light was adapted through dye-adsorbed and doping methods. In dye-adsorbed method hypericin B [39], chlorine e [40] and zinc phthalocyanine [41] was used as PDT sensitizers whereas in doping methods metal elements (Pt,Fe) [42-44] and non-metal elements (C,N) [45-47] have been used. Li et al. synthesized nitrogen doped TiO₂NPs (N-TiO₂). Calcination of anatase TiO₂NPs was done under an ammonia atmosphere. The obtained N-TiO₂NPs had a higher absorbance and anti-tumor activity in the visible region than pure TiO₂NPs [48-50].

The anti-neoplastic effect of TiO₂ is effective but tumor cells were not specifically destroyed. Improved TiO₂NPs which identify and bind the receptors of neoplastic cells were developed. This increased the specific destruction of tumor cells and reduced the death of normal cells. Monoclonal antibody proteins (CEA, 83 pre-S1/S2, 84 IL13a2R85 and EGFR86) with high affinity and specificity were immobilized on the surface of TiO₂NPs. The NPs are directed towards the neoplastic cells as the monoclonal antibody proteins are over expressed on their surface.

TiO₂ antibacterial effect

Compared to other antimicrobial agents nanotechnology based materials like metal and metal oxides are effective microbicidal agents with more safety and stability [51-53]. The photocatalytic microbicidal action of TiO₂ was first reported by Matsunaga et al. (1985), when they investigated the antimicrobial effect of TiO₂ against *Lactobacillus acidophilus*, *Saccharomyces cerevisiae* and *E. Coli* [54]. Ahmed et al. found that multi drug resistant *P. aeruginosa* associated infections were effectively treated with TiO₂NPs and antibiotics [55]. Inhibition of bacterial growth by TiO₂NPs was due to its nanometric scale and powerful oxidizing ability.

Antibacterial effect of ZnO₂ and TiO₂NPs against metallo beta lactamase and biofilm producing *Pseudomonas aeruginosa* was reported by Vincent et al. [56]. Antibacterial action against *P.*

aeruginosa was evident when TiO₂NPs was preexposed to UV radiation. ROS which causes cell wall lysis was formed on exposure of sixty minutes [57]. TiO₂ photo catalyst can disinfect a broad spectrum of microorganisms which have a deleterious effect on cellular respiratory activity and can result in cell death [58]. This property is used in tooth paste it also enhances the antimicrobial activity on dental plaque. Rimjim et al. suggested its effectiveness in both aerobic and anaerobic bacteria [59]. The strong oxidizing power of TiO₂ by free radicals like hydroxyl and superoxide anion radicals reduces the growth of *E. coli* and *staphylococcus* [60]. Roy et al. concluded that the antibacterial action of beta lactams, cephalosporins, aminoglycosides, glycopeptides, macrolides, lincosamides and tetracycline was enhanced against methicillin resistant *staphylococcus aureus* [61].

Surface modification of implants with TiO₂NPs provides antibacterial property and enhances osteogenic activity [62,63]. Liu et al. confirmed that implant efficacy improved when TiO₂NPs along with zinc was used in proper concentration this was due to cytocompatibility and antibacterial functions [64]. Ti dental implants when modified with multiple layer of TiO₂ nano network increases the cell adhesion, proliferation and mobility [65]. When heat and plasma treatment was compared it was seen that plasma treatment reduces the adhesion of *prophyromonas gingivalis* without affecting the activity of osteoblast. The heat treated group at 400°C was the most suitable for dental implants because of optimum osteoblast and anti-bacterial activity [66]. Vishnu et al. reported that hydrothermal treatment of etched Ti implants at 225°C for five hours altered the topography and enhanced the antibacterial effect against methicillin resistant *staphylococcus aureus*. These Ti implants improved the calcium deposition by osteoblast and was not cytotoxic against mammalian cells [67].

Titanium dioxide and bone formation

Titanium and its alloys are used as implants in bones as they have superior strength, stability, elastic modulus similar to the elastic modulus of tissues and ability and capability to form a thin stable oxide layer that is resistant to corrosion. TiO₂ nano structures have been successfully used in implants because of its nano topographical characteristics. Low toxicity, flexibility, high corrosion resistance and high tensile strength makes the titanite nanostructures a powerful candidate for implants [68]. Surface properties, micro topography, nanotopography and composition are the key factors for integration of implants with live bone.

Various research has concluded that titanium dioxide surface properties promote attachment of bone forming cells. The bioactivity of crystalline TiO₂ is associated with existence of hydroxyl groups on the surface and negative charges that are induced which in turn draw calcium and then phosphorous ions from the body fluids to the surface of implant. It has also been reported that during change in structure of titanium dioxide layer from anatase to rutile the rate of dissolution of metallic ions are substantially reduced in a stimulated body fluid.

TiO₂ is a bioactive factor which upregulates ALP activity. Haugen demonstrated the ability of porous TiO₂ scaffold to promote new bone formation without causing inflammation and tissue necrosis in a peri implant cortical defect. The bone volume increased significantly when the defects were treated with the TiO₂ scaffolds. Histologically presence of newly formed bone was seen in close proximity with scaffold surface and the cortical section of the defect showed new bone formation. The newly formed bone present in the marrow space put forward the suggestion that the TiO₂ scaffold has the ability to act as a material with osteoconductive properties.

The study concluded by highlighting the ability of TiO₂ scaffold to integrate well with bone and this ability can be utilized to enhance formation of bone and growth of bone adjoining implants as well as bone formation in larger defects [69].

Chung et al. created a uniform non porous structure on titanium implant by coating it with hydroxyl apatite -TiO₂ (HApTiO₂) and immobilize BMP-2 on the surface. The proper porous structure of HApTiO₂ encouraged cell growth and adhesion leading to the time reduction in bone healing at implant bone interface [70]. Mohammedi et al. reported that presence of TiONPs in calcium phosphate cement increases the mechanical strength when used in repair of bone defects. Surface area and porous structure of TiO₂NTs increased due to nanometric thickness, this promoted cell adhesion and improved bone capabilities [72]. TiO₂ NTs diameter influence cell adhesion and osseointegration. Collagen type-I (Col-I) was adsorbed with a higher outcome and faster speed when TiO₂NTs of diameter of 100 nm compared to 30 nm diameter. Hydrogen bond and van der Waals forces that existed between Col-I molecules and TiO₂NTs was the driving force in adsorption mechanism [73]. The length also influenced adhesion, longer the length less was the adhesion. 0.04 μm length of TiO₂NTs have increased adhesion strength compared to NTs with length of 2 μm, this was because of less interfacial stress [74]. Surface topography of dental implants influences osseointegration. Dental implants showed high percentage of osseointegration when the surface of implants were coated with TiO₂ NTs through anodic oxidation and were loaded with BMP₂ [75]. TiO₂ NTs possess good blood compatibility therefore it can be used for surface modification of blood contacting implants, biological molecules like Gly- Arg- Gly- Asp-Ser peptide when immobilized on TiNTs improved osseointegration in dental implants [76]. Jin and his co-workers found that osteoblast adhesion was enhanced when TiO₂ NTs less than 100 nm diameter was used. It also upregulated the alkaline phosphatase enzyme which clearly indicates the bone forming potential and bone tissue interaction of orthopedic implants coated with TiO₂NTs [77]. Huang et al. pointed out that the surface chemistry, structure, wettability and crystalline phase of TiO₂NTs had a positive effect in platelet rich plasma properties. It was found that cell adhesion, migration, proliferation and differentiation improved when properties such as wettability, surface texture and chemistry was altered [78].

Titanium dioxide films and coatings have high biocompatibility and is resistant to corrosion, therefore its use as bone anchored implants are extensively studied. The coatings of TiO₂ on implant surface is accomplished through various methods like laser ablation, dip coating, sol-gel process, heat treatment, electrochemical methods, sputtering, thermal spraying etc. Fabrication of TiO₂ film makes it a bioactive coating and this allows the implant to bond with the surrounding bone by enhancing the growth of calcium phosphate layer on TiO₂ film in presence of body fluids. The nano topography of surface of dental implants enhances blood response, cell adhesion and osseointegration of dental implants and adds to the success of the implants.

Other Applications of Titanium Dioxide

The titanium dioxide has a wide range of applications in pharmaceutical industry. It is used as an active ingredient in various medications. The inherent property of titanium dioxide to absorb UV light and its capacity to scatter light can be utilized to extend the shelf life of pharmaceutical products. TiO₂ protects the active photosensitive ingredient present in the pharmaceutical preparation against UV light and heat degradation and there by enhances the safety and efficacy of drugs [79].

The opacifying and pigmentation ability of titanium dioxide helps to maintain color uniformity over time. As different colors of medication indicate different doses eg: Warfarin, also color indication helps in diagnosis of drug over dosage or intoxication in emergency rooms, therefore color stability of drug plays a significant role, the uniformity and consistency in the maintenance of color ensures pharmaceutical safety [79]. This helps the medical professionals and patients to differentiate medications especially in people with limited eye sight. TiO₂ nano coating on maxillofacial silicone elastomers reduces its color degradation compared with silicone's that are noncoated. Maxillo facial silicones with surface nano coating of ALD TiO₂ was a novel color stable material to be used potentially in extra oral maxillofacial silicone processes [80]. The wound healing ability of titanium dioxide was demonstrated by Sankar et al. TiO₂ nano particles exhibited enhanced wound repair, wound contraction, matured collagen deposition and epithelialization in the wound [81].

A research conducted by Langle et al concluded that TiO₂ can be considered as an appropriate vehicle in the delivery of active substances to treat diabetes mellitus. TiO₂ Stevia Rebaudiana Bertoni (20 and 30 μm) combination has a potent and prolonged anti diabetic activity [82]. A similar conclusion was derived from another research by Samyuktha et al as TiO₂ exhibited alpha amylase inhibition which highlights its anti-diabetic effect [83].

Adverse Effects of Nano TiO₂

Inert nano materials when administered to human body through injections can perform differently. Oral, transdermal/inhalation are the routes of exposure which can cause adverse effects on vital organs including respiratory system [84], gastro intestinal tract, reproductive system, excretory system, circulatory system and nervous system [85]. One of the nano materials commonly studied for pulmonary toxicity is TiO₂ [86]. It is reported that chronic exposure of nano TiO₂ in murine models can cause inflammation, epithelial hyperplasia and pulmonary carcinoma [87]. The size of particles also affects the toxicity; it is reported that large size TiO₂ is comparatively less toxic than smaller sized. It was also seen that in murine models TiO₂ nano particles can cause lesion in brain and fatty degradation of the hippocampus [88]. On the basis of these toxicity of nano structures used in bio medical application needs to be thoroughly evaluated before use. Therefore, careful evaluation of toxicity and clinical outcome of TiO₂ is required before applying in clinical practice.

Conclusion

Titanium dioxide has a wide range of application in all aspects of daily life. As titanium dioxide has the ability to absorb ultra violet light it protects the skin from ultraviolet radiation. This property extends its use in sunscreen lotions, cosmetic products and skin care products. Photo catalytic activity of titanium dioxide results in thin coatings which exhibits self-cleansing and disinfection properties on exposure to ultra violet radiations. Alloys of titanium are characterized by being light weight, very high tensile strength even at high temperatures, high corrosion resistance and ability to withstand high temperature makes the wonder metal titanium as an ideal metal to be used in air crafts, pipes for power plants, Armour plating, naval ships, space crafts and missiles. Unique properties such as non-toxicity, bio compatibility and affordability made TiO₂ nano structure and their composites to gain attention and use in biomedical field. Researches demonstrate that TiO₂ nano structures are safer than any other nano material for bio medical application. Chemo therapeutic agents for clinical applications when modified by TiO₂ NPs improved the bioavailability and the release

time of chemotherapeutic agents. The uniqueness of TiO₂ nano materials is non toxicity and nano topographical characteristics. These characteristics made the material available for the use in orthopedic and dental implants. There are numerous studies in literature that give evidence for the potential of titanium and its oxides to be used as bone, dental and drug releasing implants. Considering all the above mentioned aspects it can be concluded that titanium, titanium dioxide and nano materials based on titanium dioxide can be judiciously and effectively used as valuable material for dental implants and other biomedical applications.

References

1. Lide, D. R., ed., CRC Handbook of Chemistry and Physics (86th ed.). Boca Raton (FL): CRC Press. (2005).
2. Titanium, Columbia Encyclopedia (6th ed.). New York: Columbia University Press. 2000–2006 (2000).
3. Stwertka, Albert, Titanium, Guide to the Elements (Revised ed.). Oxford University Press. pp. 81–82 (1998).
4. Baan R, Straif K, Grosse Y, Secretan B, El Ghissassi F, Coglianò V. Carcinogenicity of carbon black, titanium dioxide, and talc. *Lancet Oncol.*, 7(4), 295 (2006).
5. Bavykin DV, Friedrich JM, Walsh FC. Protonated titanates and TiO₂ nanostructured materials: synthesis, properties, and applications. *Adv Mater.*, 18(21), 2807–2824 (2006).
6. Encyclopaedia Britannica. 2006. Retrieved 19 January 2022
7. Steele, M.C.; Hein, R.A., Superconductivity of Titanium. *Phys. Rev.* 92(2), 243-247 (1953).
8. Thiemann, M. et al., Complete electrostatics of a BCS superconductor with eV energy scales: Microwave spectroscopy on titanium at mK temperatures, *Phys. Rev. B.* 97(21), 214516 (2016).
9. McCullagh C, Robertson JM, Bahnemann DW, Robertson PK. The application of TiO₂ photocatalysis for disinfection of water contaminated with pathogenic micro-organisms: a review. *Res Chem Intermed.*, 33(3–5), 359–375 (2007).
10. Bonetta S, Bonetta S, Motta F, Strini A, Carraro E. Photocatalytic bacterial inactivation by tio2-coated surfaces. *AMB Express.*, 3(1), 59 (2013).
11. El, Goresy; Chen, M; Dubrovinsky, L; Gillet, P; Graup, G, An ultradense polymorph of rutile with seven-coordinated titanium from the Ries crater. *Science.* 293(5534), 146770 (2001).
12. Gao A, Hang R, Huang X, et al. The effects of titania nanotubes with embedded silver oxide nanoparticles on bacteria and osteoblasts. *Biomaterials.*, 35(13), 4223–4235 (2014).
13. Hang R, Gao A, Huang X, et al. Antibacterial activity and cytocompatibility of cu–ti–o nanotubes. *J Biomed Mater Res A.*, 102(6), 1850-1858 (2014).
14. Xu Z, Jiang X. Rapid fabrication of tio2 coatings with nanoporous composite structure and evaluation of application in artificial implants. *Surf Coat Technol.*, 381, 125094 (2020).
15. Weetall HH. Biosensor technology what? Where? When? And why? *Biosens Bioelectron.*, 11(1–2), i–iv (1996).
16. Razavi H, Janfaza S. Medical nanobiosensors: a tutorial review. *Nanomed J.*, 2(2), 74–87 (2015).
17. Janfaza S, Nojavani MB, Nikkhal M, Alizadeh T, Esfandiari A, Ganjali MR. A selective chemiresistive sensor for the cancer-related volatile organic compound hexanal by using molecularly imprinted polymers and multiwalled carbon nanotubes. *Mikrochim Acta.*, 186(3), 137 (2019).
18. Hashemzadeh H, Allahverdi A, Peter E, N-M H. Comparison between three-dimensional spheroid and two-dimensional monolayer in a549 lung cancer and pc9 normal cell lines under treatment of silver nanoparticles. *Modares J Biotechnol.*, 10(4), 573–580 (2019).
19. Esfandiyari J, Shojaedin-Givi B, Mozafari-Nia M, Hashemzadeh H, Naderi-Manesh H. Diatom biosilica shell manipulation with gold, spion nanoparticles and trastuzumab with aims of diagnostics of her2 cells. *Modares J Biotechnol.*, 10(4), 581–588 (2019).
20. Zabarjadi A, Hashemzadeh H, Taravat E. Telomere, chromosome end, and telomerase enzyme as a cancer biomarker. *Genet Third Millennium.*, 11(1), 3018–3027 (2013).
21. Hashemzadeh H, Javadi H, Darvishi MH. Study of structural stability and formation mechanisms in dspc and dpsm liposomes: a coarse-grained molecular dynamics simulation. *Sci Rep.*, 10(1), 1837 (2020).
22. Li Q, Wang X, Lu X, et al. The incorporation of daunorubicin in cancer cells through the use of titaniumdioxide whiskers. *Biomaterials.*, 30(27), 4708–4715 (2009).
23. Akram MW, Raziq F, Fakhar-e-Alam M, et al. Tailoring of au-TiO₂ nanoparticles conjugated with doxorubicin for their synergistic response and photodynamic therapy applications. *J Photochem Photobiol a Chem.*, 384, 112040 (2019).
24. Venkatasubbu GD, Ramasamy S, Reddy GP, Kumar J. in vivo and in vivo anticancer activity of surface modified paclitaxel attached hydroxyapatite and titanium dioxide nanoparticles. *Biomed Microdevices.*, 15(4), 711–726 (2013).
25. T. J. Dougherty, C. J. Gomer, B. W. Henderson, G. Jori, D. Kessel, M. Korbek, J. Moan and Q. Peng, Photodynamic therapy: *Natl Cancer Inst.*, 90(12), 889-905 (1998).
26. T.A. Heinrich, A.C. Tedesco, J.M. Fukuto and R.S. da Silva, *Dalton Trans.*, 43, 4021–4025 (2014).
27. M.A. Biel, in *Biofilm-based Healthcare-associated Infections*, Springer, 119–136 (2015).
28. M. Triesscheijn, P. Baas, J.H. Schellens and F.A. Stewart, *Photodynamic therapy in oncology, Oncologist*, 11(9), 1034-44 (2006).
29. R. R. Allison and C. H. Sibata, photodynamic therapy photosensitizers: a clinical review, *7(2)*, 61-75 (2010).
30. A. Fujishima, J. Ohtsuki, T. Yamashita and S. Hayakawa, *Photomed. Photobiol.*, 8, 45 (1986).
31. R.J. Skyrme, A.J. French, S.N. Datta, R. Allman, M.D. Mason and P.N. Matthews, *BJU Int.*, 95, 1206–1210 (2005).
32. L. E. Rhodes, M. de Rie, Y. Enström, R. Groves, T. Morken, V. Goulden, G. A. Wong, J.-J. Grob, S. Varma and P. Wolf, *Arch. Dermatol.*, 140, 17–23 (2004).
33. H.I. Hung, J.M. Schwartz, E.N. Maldonado, J. J. Lemasters and A.L. Nieminen, *J. Biol. Chem.*, 288, 677–686 (2013).
34. D. Ferguson, A. Perry, M.E. Wood, P.G. Winyard and M. Whiteman, *Free Radical Biol. Med.*, 76, S135 (2014).
35. R.G. Lopes, M.E.S.O. Santi, B.E. Franco, A.M. Deana, R.A. Prates, C.M. França, K.P.S. Fernandes, R.A.M. Ferrari and S.K. Bussadori, *Lasers Med. Sci.*, 5, 146–152 (2014).
36. R. Cai, K. Hashimoto, Y. Kubota and A. Fujishima, Induction of cytotoxicity by photoexcited TiO₂ particles, *Chem. Lett.*, 52(8), 2346-8 (1992).
37. Sakai, H., Baba, R., Hashimoto, K., Fujishima, A. and Heller, A., Local Detection of Photoelectrochemically Produced H₂O₂ with a Wired Horseradish-Peroxidase Microsensor. *The Journal of Physical Chemistry*, 99, 11896-11900 (1995).
38. J.W. Seo, H. Chung, M.Y. Kim, J. Lee, I.H. Choi and J. Cheon, *Small*, 3, 850 (2007).
39. S. Xu, J. Shen, S. Chen, M. Zhang and T. Shen, *J. Photochem. Photobiol.*, B, 67, 64 (2002).
40. Y. Tokuoka, M. Yamada, N. Kawashima and T. Miyasaka, *Chem. Lett.*, 35, 496 (2006).
41. T. Lopez, E. Ortiz, M. Alvarez, J. Navarrete, J. A. Odriozola, F. Martinez-Ortega, E. A. Pa'ez-Mozo, P. Escobar, K. A. Espinoza and I. A. Rivero, *Nanomed.: Nanotechnol., Biol. Med.*, 6, 777 (2010).
42. A Jan, Wolnicka-Gubisz, Urbanska, G. Stochel, W. Macyk, Photodynamic activity of platinum(IV) chloride surface-modified TiO₂ irradiated with visible light. *Photochem. Photobiol.*, B, 92, 54 (2008).
43. R Damoiseaux 1, S George, M Li, S Pokhrel, Z Ji, B France, T Xia, E Suarez, R Rallo, L Mädlar, Y Cohen, E M V Hoek, A Nel: No time to lose-high throughput screening to assess nanomaterial safety, 3(4), 1345-60 (2011).
44. S. George, S. Pokhrel, Z. Ji, B. L. Henderson, T. Xia, L. Li, J. I. Zink, A. E. Nel and L. Mädlar, *J. Am. Chem. Soc.*, 133, 11270 (2011).
45. Z. Hu, Y. Huang, S. Sun, W. Guan, Y. Yao, P. Tang and C. Li, *Carbon*, 50, 994 (2012).
46. Asahi, R., Morikawa, T., Ohwaki, T., Aoki, K. and Taga, Y. (2001) Visible-Light Photocatalysis in Nitrogen-Doped Titanium Oxides. *Science*, 293, 269-271.79 J. Zhong, F. Chen and J. L. Zhang, *J. Phys. Chem. C*, 114, 933 (2010).
47. Z. Li, L. Mi, P. N. Wang and J. Y. Chen, *Nanoscale Res. Lett.*, 2011, 6, 356.
48. Y. Cong, J. Zhang, F. Chen, M. Anpo, and D. He, "Preparation, Photocatalytic Activity, and Mechanism of Nano-TiO₂ Co-Doped with Nitrogen and Iron (III)," *J. Phys. Chem. C*, 111, 10618-10623 (2007).
49. M. Abdulla-Al-Mamun, Y. Kusumoto and M. S. Islam, Synergistic cell-killing by photocatalytic and plasmonic photothermal effects of Ag@TiO₂ core-shell composite nanoclusters against human epithelial carcinoma (HeLa) cells. *Chem.*, 22, 5460 (2012).
50. J. Xu, Y. Sun, J. Huang, C. Chen, G. Liu, Y. Jiang, Y. Zhao and Z. Jiang, *Bioelectrochemistry*, 71, 217 (2007).
51. McCullagh C, Robertson JM, Bahnemann DW, Robertson PK. The application of TiO₂ photocatalysis for disinfection of water contaminated with pathogenic micro-organisms: a review. *Res Chem Intermed.*, 33(3–5), 359–375 (2007).

52. Calamak S, Shahbazi R, Eroglu I, Gultekinoglu M, Ulubayram K. An overview of nanofiber-based antibacterial drug design. *Expert Opin Drug Discov.*, 12(4), 391–406 (2017).
53. Kaviyarasu K, Geetha N, Kanimozhi K, et al. in vivo cytotoxicity effect and antibacterial performance of human lung epithelial cells a549 activity of zinc oxide doped tio2 nanocrystals: investigation of biomedical application by chemical method. *Mater Sci Eng C.*, 74, 325–333 (2017).
54. Matsunaga T, Tomoda R, Nakajima T, Wake H. Photoelectrochemical sterilization of microbial cells by semiconductor powders. *FEMS Microbiol Lett.*, 29(1–2), 211–214 (1985).
55. Ahmed FY, Farghaly Aly U, Abd El-Baky RM, Waly NGFM. Comparative Study of Antibacterial Effects of Titanium Dioxide Nanoparticles Alone and in Combination with Antibiotics on MDR *Pseudomonas aeruginosa* Strains. *Int J Nanomedicine.*, 15, 3393-3404 (2020).
56. Vincent MG, John NP, Narayanan PM, Vani C, Murugan S. In vitro study on the efficacy of zinc oxide and titanium dioxide nanoparticles against metallo beta-lactamase and biofilm producing *Pseudomonas aeruginosa*. *J Appl Pharm Sci.*, 4(7), 41–46 (2014).
57. Arora B, Murar M, Dhumale V. Antimicrobial potential of Tio2 nanoparticles against MDR *Pseudomonas aeruginosa*. *J Exp Nanosci.*, 10(11), 819–827 (2015).
58. Maness P, Smolinski S, Blake DM, Huang Z, Wolfrum EJ, Jacoby WA. Bactericidal activity of photocatalytic TiO₂ reaction: toward an understanding of its killing mechanism. *Appl Environ Microbiol.*, 65(9), 4094–4098 (1995).
59. Rimjhim AT. Analysis of Antimicrobial Activity of Titaniumdioxide Nanoparticles on Aerobic and Anaerobic Dental Isolates.
60. de Dicastillo, C.L. , Correa, M.G. , Martínez, F.B. , Streitt, C., Galotto, M.J. Antimicrobial Effect of Titanium Dioxide Nanoparticles. In: Mare, M. , Lim, S.H.E. , Lai, K. , Cristina, R. , editors. *Antimicrobial Resistance - A One Health Perspective* [Internet]. London: IntechOpen (2020).
61. S. Roy, Aashis; Parveen, Ameena; R. Koppalkar, Anil; Prasad, M. V. N. Ambika, Effect of Nano - Titanium Dioxide with Different Antibiotics against Methicillin-Resistant *Staphylococcus Aureus*. *Journal of Biomaterials and Nanobiotechnology*, 1(1), 37–41 (2010).
62. Azzawi ZG, Hamad TI, Kadhim SA, Naji GA-H. Osseointegration evaluation of laser-deposited titanium dioxide nanoparticles on commercially pure titanium dental implants. *J Mater Sci Mater Med.*, 29(7), 96 (2018).
63. Souza JC, Sordi MB, Kanazawa M, et al. Nano-scale modification of titanium implant surfaces to enhance osseointegration. *ActaBiomater.*, 94, 112–131 (2019).
64. Liu W, Su P, Chen S, et al. Antibacterial and osteogenic stem cell differentiation properties of photoinduced tio2 nanoparticle-decorated tio2 nanotubes. *Nanomedicine.*, 10(5), 713–723 (2015).
65. Choi SH, Jang YS, Jang JH, Bae TS, Lee SJ, Lee MH. Enhanced antibacterial activity of titanium by surface modification with polydopamine and silver for dental implant application. *J Appl BiomaterFunct Mater.*, 17(3), 2280800019847067 (2019).
66. Ji M-K, Oh G, Kim J-W, et al. Effects on antibacterial activity and osteoblast viability of non-thermal atmospheric pressure plasma and heat treatments of tio2 nanotubes. *J Nanosci Nanotechnol.*, 17(4), 2312–2315 (2017).
67. Vishnu J, Manivasagam VK, Gopal V, et al. Hydrothermal treatment of etched titanium: a potential surface nano-modification technique for enhanced biocompatibility. *Nanomedicine.*, 20, 102016 (2019).
68. Williams DF. On the mechanisms of biocompatibility. *Biomaterials.*, 29(20), 2941–2953 (2008).
69. Lars M. Bjursten; Lars Rasmusson; Seunghan Oh; Garrett C. Smith; Karla S. Brammer; Sungho Jin, Titanium dioxide nanotubes enhance bone bonding in vivo. *J Biomed Mater Res.*, 92(3), 1218-24 (2010).
70. Chen H-I, Shu H-Y, Chung C-J, He J-L. Assessment of bone morphogenic protein and hydroxyapatite–titanium dioxide composites for bone implant materials. *Surf Coat Technol.*, 276, 168–174 (2015).
71. Mohammadi M, Hesaraki S, Hafezi-Ardakani M. Investigation of biocompatible nanosized materials for development of strong calcium phosphate bone cement: comparison of nano-titania, nano-silicon carbide and amorphous nano-silica. *Ceram Int.*, 40(6), 8377–8387 (2014).
72. Brammer KS, Frandsen CJ, Jin S. TiO₂ nanotubes for bone regeneration. *Trends Biotechnol.*, 30(6), 315–322 (2012).
73. Yang W, Xi X, Ran Q, Liu P, Hu Y, Cai K. Influence of the titania nanotubes dimensions on adsorption of collagen: an experimental and computational study. *Mater Sci Eng C.*, 34, 410–416 (2014).
74. Yang W, Xi X, Ran Q, Liu P, Hu Y, Cai K. Influence of the titania nanotubes dimensions on adsorption of collagen: an experimental and computational study. *Mater Sci Eng C.*, 34, 410–416 (2014).
75. Hu N, Wu Y, Xie L, et al. Enhanced interfacial adhesion and osseointegration of anodic TiO₂ nanotube arrays on ultra-finegrained titanium and underlying mechanisms. *Acta Biomater.*, 106(1), 360–375 (2020).
76. Kim G-H, Park S-W, Lee K, et al. Evaluation of osteoblast-like cell viability and differentiation on the gly-arg-gly-asp-ser peptide immobilized titanium dioxide nanotube via chemical grafting. *J Nanosci Nanotechnol.*, 16(2), 1396–1399 (2016).
77. K.S. Brammer, S. Oh, C.J. Cobb, L.M. Bjursten, H. van der Heyde and S. Jin, *Acta Biomater.*, 5, 3215–3223 (2009).
78. Huang HH, Chen JY, Lin MC, Wang YT, Lee TL, Chen LK. Blood responses to titanium surface with tio2 nano-mesh structure. *Clin Oral Implants Res.*, 23(3), 379–383 (2012).
79. Das, R., Ambardekar, V., Bandyopadhyay, P.P. Titanium Dioxide and Its Applications in Mechanical, Electrical, Optical, and Biomedical Fields. In: Ali, H. M. , editor. *Titanium Dioxide - Advances and Applications* [Internet]. London: IntechOpen
80. <https://tdma.info/the-essential-role-of-titanium-dioxide-in-pharmaceuticals/>
81. Bishal AK, Wee AG, Barão VAR, Yuan JC, Landers R, Sukotjo C, Takoudis CG. Color stability of maxillofacial prosthetic silicone functionalized with oxide nanocoating. *J Prosthet Dent.*, 121(3), 538-543 (2019).
82. Langle, Ariadna; González-Coronel, Marco Antonio; Carmona-Gutiérrez, Genaro; Moreno-Rodríguez, José Albino; Venegas, Berenice; Muñoz, Guadalupe; Treviño, Samuel; Díaz, Alfonso, Stevia rebaudiana loaded titanium oxide nanomaterials as an antidiabetic agent in rats. *Revista Brasileira de Farmacognosia*, 25(2), 145–151 (2015).
83. Samyuktha PS, Ganapathy DM, Rajeshkumar S. In Vitro Study Of Antidiabetic Effect Of Green Synthesised Titanium Dioxide Nanoparticles. *Natural Volatiles & Essential Oils Journal*, 25, 7260-70 (2021).
84. Sankar, Renu; Dhivya, Ravishankar; Shivashangari, Kanchi Subramanian; Ravikumar, Vilwanathan, Wound healing activity of *Origanum vulgare*engineered titanium dioxide nanoparticles in Wistar Albino rats. *Journal of Materials Science: Materials in Medicine*, 25(7), 1701–1708 (2014).
85. B. L. Baisch, N. M. Corson, P. Wade-Mercer, R. Gelein, A. J. Kennell, G. Oberdorster and A. Elder, *Part. Fibre Toxicol.*, 11, 5 (2014).
86. W. McKinney, M. Jackson, T. Sager, J. Reynolds, B. Chen, A. Afshari, K. Krajnak, S. Waugh, C. Johnson and R. Mercer, *Inhalation Toxicol.*, 24, 447–457 (2012).
87. Nogueira CM, de Azevedo WM, Dagli ML, Toma SH, Leite AZ, Lordello ML, Nishitokukado I, Ortiz-Agostinho CL, Duarte MI, Ferreira MA, Sipahi AM. Titanium dioxide induced inflammation in the small intestine. *World J Gastroenterol.*, 18(34), 4729-35 (2012).
88. G. Oberdörster, E. Oberdörster and J. Oberdörster, *Environ. Health Perspect.*, 113, 823–839 (2005).