RSC Advances

ROYAL SOCIETY OF CHEMISTRY

www.rsc.org/advances

RSC Advances

RSCPublishing

REVIEW ARTICLE

Review: Physicochemical modification as a versatile strategy for biocompatibility enhancement of biomaterials

Cite this: DOI: 10.1039/xoxxoooooox

Received ooth January 2015,
Accepted ooth January 2015

DOI: 10.1039/xoxxoooooox

www.rsc.org/

A.A. John[a], A.P. Subramanian[a], M.V. Vellayappan[a], A. Balaji[a], S. K. Jaganathan*[,a], H. Mohandas[c], T. Paramalinggam[b], E. Supriyanto[a], M. Yusof[a]

A biomaterial can be defined as a material intended to interface with biological systems to evaluate, treat, augment or replace any tissue, organ or function of the body. Major problems allied with biomaterials are their properties and the biocompatibility which have to be tackled and resolved before promoting it to the market or implanting it into the biological system. To enhance the biocompatibility of the biomaterials, several surface modification strategies such as physico-chemical, mechanical and biological modifications were explored. In this review, some recent applications of physico-chemical modification technologies such as alteration in the structure of a molecule by chemical modification, surface grafting, abrasive blasting and acid etching, surface coatings, heat and steam treatment for medical materials like polymers, metals, ceramics and nanocomposites were discussed. This article will promote physico-chemical modification as a versatile technology in surface engineering to improve the properties and biocompatibility of medical materials. Further, it will instigate the growth of biomaterial market with various high quality biomaterials.

1. Introduction:

Biomaterial is used to make medical devices that replace a part or a function of the body in a safe, reliable, economic and physiologically acceptable manner [1]. The global biomaterial market is expected to record close to 15% yearly growth for the ten-year period ending 2017 to reach $84 billion [2]. Recently, biomaterials have wide spread application in various biological systems such as skeletal (bone plate, total joint replacements), muscular (sutures, muscle stimulator), nervous (cardiac pacemaker, nerve stimulator), endocrine (microencapsulated pancreatic islet cells), reproductive system (augmentation mammoplasty), dental and maxillofacial applications (cosmetic replacements), drug-delivery system [3, 4]. Biomaterials are majorly classified into the four main categories namely metals, ceramics, polymers and biological substances. The selection of biomaterial depends on the surrounding environment where it will be implanted. The implanted material should not cause any adverse effects like allergy, inflammation and toxicity either immediately after surgery or under post operative conditions [5]. The first requirement of biomaterial is that the material must be biocompatible; it means that the organism should not treat it as a foreign object. Biocompatibility is a fundamental property which decides the excellence of a biomaterial and its application in medical field. The term biocompatibility denotes the ability of a material to perform with an appropriate host response in a specific situation [6]. Biocompatibility has been discussed in lots of works with escalating curiosity in assessing the characteristics of medical materials and devices and also the responses caused by its components. Biocompatibility covers many aspects of the material, including its physical, mechanical and chemical properties, toxic, mutagenic and allergenic effects, so that no noteworthy injuries or harmful effects on the biological function of cells and individuals take place. In order to have better knowledge, readers may refer to the following cited articles [7, 8]. Blood compatibility refers to the events takes place within the biological system when the material surface comes in contact with the blood and its components. Blood compatibility is the outstanding property of the implant material especially for the devices that makes contact with blood [9]. Thus, we can describe blood compatibility as the ability of the material to perform its function in a particular situate without bringing out any blood-related complications. Whenever the blood comes in contact with the implants (biomaterial), it will direct to the complications such as interaction of blood components with surfaces resulting in protein and water adsorption, blood cells interfere with the surface of biomaterial and these events lead to haemostasis and coagulation [10]. Until the biocompatibility of a material is confirmed, it must be

subjected to various studies ranging from in vitro assays to clinical trials, in the areas of pharmaceutics, biology, chemistry and toxicology to validate its use as biomaterial. Secondly, the material should allow the biological system to resume its natural functioning. Thirdly, the material should be mechanically sound; for the replacement of load bearing structures, the material should possess equivalent or greater mechanical stability to ensure high reliability of the graft [11]. It is vital to modify the biomaterial surfaces in order to control the subsequent interaction of implant surface with blood or biological system and it responses for particular applications.

Surface modification approaches namely physico-chemical, mechanical and biological methods are in use. These three categories are further subdivided into sub categories. In our review we mainly focus on physico-chemical modification of biomaterials to improve the blood compatibility of the biomaterial. The most important techniques involved in physicochemical method are modifying the surface by chemical means, surface grafting, abrasive blasting and acid etching, surface coating, high temperature treatments (thermal treatment, vapour and steam treatment), as shown in figure 1.

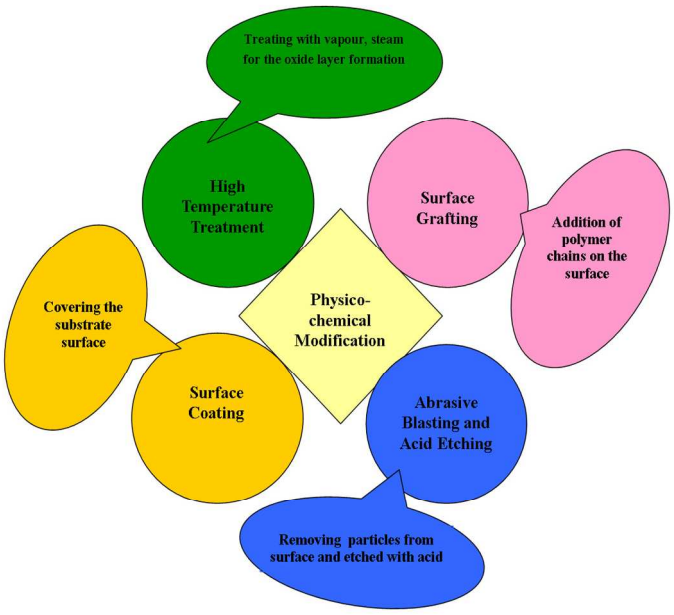

Figure 1: Physico-chemical modification technologies and its strategies

2. Surface treatment:

The surface characteristics of all types of biomaterials act as a key role in determining the biocompatibility of a particular implant. When choosing the biomaterial to communicate with the biological system, the first important criterion to inspect is that of biocompatibility [12]. The biocompatibility and cellular interactions of the biomaterial based on the surface characteristics. The properties such as surface roughness, hardness, temperature, surface chemistry, surface reactivity (inert or active), wettability and surface charge (surface free energy) are the surface characteristics that plays a major function in cell adhesion, cell spreading, cell proliferation and tissue formation [13]. The response of host organism in macroscopic, cellular and protein levels to biomaterials is closely associated with the surface properties of the material. The appropriate physico-chemical properties of a biomaterial required for implantation will depend on the corresponding biomedical application and it can be modified by performing surface treatment techniques. Surface modification is the process of modifying the surface of a material by changing the physical or chemical characteristics different from the ones originally found on the surface of a material which can be carried out in nanoscale or bulk level. The nanoscale surface modification is an important technique in the field of nanotechnology which involves in nanofabrication that can modify both the topography and the chemistry of the surface in nanometric level. In nanoscale modification, the modifications of substrates are carried out in nano domain. For example, in coating technology, the thickness of the films is in the range of nanometers. The nanomolecular layers are less than 10 nm thickness, used to produce nanoscale modifications [14]. After performing the nanoscale modification, the effect of modification was studied by nanometric analysis including scanning tunneling microscopy (STM) and atomic force microscopy (AFM) which shows nanolevel pit formations and average rougness in nanometers. The parameters such as cell adhesion, proliferation involved in interaction mechanism of modified surface with biological substances also controlled in nanoscale [15]. The surface coating with nanoparticles decreases the negative properties of the material and enhances the osteoblast cell adhesion and proliferation when compared with the microparticle coatings. It has been noted that the smooth surface on microlevel surface modification is not necessarily need to be smooth on nanolevel modification [16]. On the other hand, nano engineered surfaces can directly influence the biological properties and will be more useful in the applications of tissue regeneration functionalities than the bulk surface modification [17]. Whereas in bulk modification, a large surface was subjected to modification techniques in order to modify its bulk properties. During bulk surface modification the bulk properties i.e. the properties result from relating to the greater number of atoms present in the sample gets changed. In contrast to nanoscale surface modification, the objective of bulk modification is to change wide range of properties including mechanical, physical and chemical characteristics [18].The interactions between blood and a material surface depend on the blood composition, blood flow and surface characteristics of the implanted material defined by its physico-chemical properties [19]. The modification can be done by different methods with an objective to altering a wide range of surface properties, such as surface roughness, hydrophilicity, surface charge, biocompatibility and reactivity [20-23]. In this review, we will discuss on chemical modification, surface grafting, abrasive blasting and acid etching, surface coating, thermal treatment, vapour and steam treatment, the major techniques of physico-chemical modification to enhance the biocompatibility of the biomaterial in forth coming chapters.

3. Chemical modification:

Chemical modification includes alkali hydrolysis, covalent immobilization and wet chemical methods are only three of the many ways to chemically modify a surface. The surface is prepared with surface activation, where several functionalities are positioned on the material surface to chemically modify the surface and studies have shown that chemical modification enhances the biocompatibility. The wet chemical method is one of the mostly chosen methods where the chemical species are dissolved in an organic solution and reactions take place to reduce the hydrophobic nature. Surface stability is higher in chemical modification than in

physical adsorption. It also offers better biocompatibility towards cell growth and bodily fluid flow. Thus chemical modification is significantly used surface treatment for all types of biomaterials such as polymers, metals, ceramics and nanocomposites.

3.1 Chemical modification of polymers:

Lim *et al*. has established biological progressions including protein adsorption, cell proliferation, and gene expression can be restricted to some extent using chemical methods to modify the surface properties of biocompatible materials leading to controlled surface functionalization of the material [24]. It was stated that poly(lactic-coglycolicacid) can be nanostructured by chemical etching with sodium hydroxide (NaOH), resulting in a material surface features to enhance the activity of various cell types [25, 26]. Balakrishnan *et al.* subjected polyvinyl chloride (PVC) to amination under concentrated solution of ethylenediamine followed by PEG treatment. In that study, platelet studies were performed and conclude that the platelet adhesion is significantly reduced for the modified PVC compared to control PVC. The contact angle measurements depicted an increase in hydrophilicity of modified polymeric surface [27]. Yvette *et al.* tailored the surface of polyurethanes (PUs) by covalent attachment of dipyridamole (trademarked as Persantine), to confirm the inhibition of thrombus formation and adherence of blood platelets upon exposure to human platelet rich plasma (PRP) [28]. The polyethersulfone (PES) membrane was modified into hydrophilic functionality to depress protein adsorption and platelet adhesion. The activated partial thromboplastin time (APTT) for the modified PES membranes was increased, resulted in enhanced blood compatibility [29, 30]. Saravana *et al.* investigated the blood compatibility of metallocene polyethylene (mPE) after treating the polymer surface with hydrochloric acid (HCl). Contact angle analysis of the treated sample indicated an increase in hydrophilicity. FTIR results showed there is no notable changes on functional group, SEM images of modified samples proved that acid tailored surface is engaged with pits formation. Blood coagulation assays like prothrombin time (PT) and activated partial thromboplastin time (APTT) revealed that there is a delay in the clotting on the surface of treated samples. The outcome of haemolytic assay depicted minor damage to red blood cells (RBC) compared to the untreated sample. Platelet adhesion assay showed that the number of platelets adhering on the surface of the treated polymer was considerably less than that on the untreated surface [31]. Surface modification of polyurethane (PU) was performed by blending the sulfonated polyrotaxanes (PRx-SO (3)'s) with a PU solution, followed by solution casting. The incorporation of PRx-SO (3)'s on PU led to the enhancement of hydrophilicity by changing the surface properties of the PU matrix. Thus, surface modification with PRx-SO (3)'s is recommended to be valuable for the fabrication of biocompatible medical devices [32]. Poly(acrylonitrile-co-maleic acid)s (PANCMAs) were tethered with poly(ethylene glycol)s (PEGs). The chemical modification on the membrane surface were characterized by Fourier transform infrared spectroscopy and the hydrophilicity, blood compatibility of the PEG-tethered PANCMA membrane were examined by water contact angle, plasma platelets adhesion and cell adhesion measurements. The results were revealed that the hydrophilicity of the membrane can be improved significantly, and the protein adsorption, platelets adhesion and macrophage attachment on the membrane surface are obviously suppressed [33]. The chemical modification of polymers may produce new polymeric material which cannot be synthesized by polymerization of monomers, considered as an advantage of chemical modification of polymers, even though non-specific interactions also exists [34].

3.2 Chemical modification of metals:

Hansson *et al.* treated the titanium material surface with polyethylene glycol (PEG) and proved that after PEG treatment the material has outstanding blood compatibility by providing resistance to the adhesion of small bio-molecules like fibrinogen and cells such as platelets and leukocytes [35]. The surface chemical composition was also changed by means of alkali modification to improve the biocompatibility as well as bioactivity of titanium (Ti) implant. The chemical modification of Ti implant with hydrofluoric (HF) acid results in the reduction of hydrocarbon content which leads to increase in surface energy and potential of bio-acceptability of Ti-implant [36]. NaOH treatment of titanium implant results in the formation of sodium titanate on the treated surface and the histological assessment, scanning electron microscopy (SEM) observation shows new bone was formed on the surface of alkali-modified implants and the bone grew more rapidly than the unmodified implants [37, 38]. The cytotoxicity evaluation of phytic acid treated WE43 magnesium alloy showed that the biocompatibility of the phytic acid treated WE43 Mg alloy is much better than the blank WE43 magnesium alloy. From the haemolysis test results, we infer that the modified samples with more Phy-Mg complex will have a better biocompatibility [39]. In another study, the pure titanium (Ti) and titanium alloy (Ti-6Al-4V) specimens were implanted into the mice with and without any surface treatment. After 3 months of implantation, the biocompatibilities of unmodified and modified implants were examined by *in vitro* and *in vivo* experiments. The outcome of these experiments point out that the commercial pure Ti and Ti-6Al-4V alloy specimens treated with alkali (KOH) have a better biocompatibility than commercial pure Ti and Ti-6Al-4V alloy specimens without the alkali treatment [40]. Parsapour *et al.* treated the stainless steel with HNO_3 followed by H_2SO_4 and ended the surface treatment with Nb coating to form a passive layer on the surface. The end product of acid treated, Nb coated stainless steel shows improved biocompatibility [41]. The chemical modification of metals produces desired topographical properties with enhanced biocompatibility which directly based on the chemical reagents used for the surface modification for particular application.

3.3 Chemical modification of ceramics:

The surface chemical modification of ceramics improves the biocompatibility and the influence of chemical treatment on cellular behavior was studied. The *in vitro* study inspects the effect of surface chemistry modification of bio-ceramics on human bone-derived cells (HBDCs) and concluded that the surface chemistry affects the cell adhesion [42] and a negative potential was effective in increasing the adhesiveness with increasing wettability, even though living cells have negative charges [43]. Al_2O_3 bio-ceramic was implanted with NH_2 + ions and it was found that the quantity of amidogen radicals implanted on the ceramic surface was proportional to the dosage of NH_2 + ions used during the ion implantation process. In addition, when implantation power of 100 keV was used, highest amount of NH_2 radicals would be implanted on the Al_2O_3 ceramic surface. The results of biocompatibility test shows that the ceramic surfaces implanted with NH_2 + ions have better biocompatibility compared to the unimplanted Al_2O_3 bio-ceramic surface [44]. Calcium silicate (CS), a biodegradable ceramic was chemically modified by partially replacing the calcium sites by strontium. The SEM images of modified ceramic surface indicate improved bioactivity as well the biocompatibility of the ceramic material [45]. The surface of yttria-tetragonal zirconia polycrystal (Y-TZP) was modified by hydrothermal treatment. The topographies

of modified Y-TZP specimens were analyzed by contact angle assay, XRD, FTIR, AFM, and FE-SEM. Then, the RGD-peptide was immobilized on the surface of the Y-TZP by chemical treatment and the resultant surface was analyzed by SEM, FTIR. The results indicate that the cell activity and biocompatibility were better for RGD-peptide immobilized Y-TZP than that on the unmodified Y-TZP [46]. From the reported literatures, the bioactivity of chemically modified ceramics was significantly increased and the selection of chemical reaction for particular application remains a challenge.

3.4 Chemical modification of nanocomposites:

In surface modification of nanocomposite scaffolds, gelatin was initially entrapped onto the surface and heparin was subsequently immobilized on entrapped gelatin. The surface-modification improved the wettability of scaffolds [47]. Nano-hydroxyapatite/poly (e-caprolactone) (PCL) particles were modified with silane coupling agent KH-792 shows positive effect on biocompatibility than the control group [48]. PVA (polyvinyl alcohol)/starch composite were subjected to surface treatment to enhance the biocompatibility. The modified surfaces were studied by FTIR and contact angle measurements. The results of this study conclude that the surface characteristics based on the type and number of incorporated nanoparticles as well as on the treatment applied. [49]. Nano-hydroxyapatite (nHA) was wrapped using polypropylene glycol (PPG) and these nHA particles were successfully introduced on the polyurethane surface. The coagulation assays were performed displays delay in clotting time and MTT assay confirmed the biocompatibility of the modified nano-hydroxyapatite (nHA) composite [50]. Adhikari et al. studied the polymer-matrix nanocomposites based on poly(lactic-co-glycolic) acid (PLGA) and graphene platelets (GNPs) and their biocompatibility was examined and suggested that PLGA/GNP nanocomposites showed better biocompatibility for cell growth with/without graphenes functionalization compared to pure PLGA [51]. The nanomaterial was surface modified with polydopamine (PDA) in a controlled manner compared to the water-phase polymerization. A PDA-shelled nanocomposite depicts reduced toxicity and enhanced biocompatibility [52]. Fe_3O_4 nanoparticles was modified by thiodiglycolic acid (TDGA) and it is used in the preparation of magnetite nanoparticles with improved mechanical properties and the study with fibroblast cell interaction showed that the modified surface have good biocompatibility [53]. The surface of nanocapsules was modified with polyethyleoxide (PEO) and succinic anhydride and the biomedical tests such as haemolysis, thromboelastography (TEG) were conducted over surface modified nanocapsules. The outcome of these experiments depicts that the PEO surface modification greatly reduced the damaging interactions of nanocapsules with red blood cells (RBCs) and platelets [54]. The surface of the micron-sized hydroxyapatite (HA) particles was modified by in situ polymerization of styrene (St), then compounded with high impact polystyrene (HIPS). The surface treated HA particles displayed an improved biocompatibility [55]. The chemical modification of nanocomposites adds an advantage of agglomeration reduction and the effects of modification based on the chemicals used [56].

4. Surface grafting:

Surface grafting refers to the addition of polymer chains to the surface to change the surface properties. The thin film on material surface can be formed through spin casting, precipitation, langmuir-blodget technique, polymer adsorption and chemical grafting. Among these techniques, chemical grafting gains more advantage over the other methods because of ease and controllable addition of number of polymer chains on the same material surface with high surface density, precise localization and long stability of grafted layers. Surface grafting offers existing materials with new functionalities such as hydrophilicity, adhesion, biocompatibility and anti-fogging [57].

4.1 Surface grafting of polymers:

The addition of sulfur based (SB) functional groups direct to a decrease of hydrophobicity and roughness of the surface. Alves *et al.* grafted the polyurethane film with sulfonic group and the results of surface characterization tests and blood compatibility studies indicates an enhancement of the modified polyurethane biological performance with increased blood compatibility [58]. Feng *et al.* tailored polycarbonateurethane (PCU) surface with Poly(ethylene glycol) monoacrylates (PEGMAs) with a molecular weight between 400 and 1,000 g $mol^{(-1)}$ to improve the hydrophilicity and haemocompatibility of the surface of polycarbonateurethane (PCU). The surface-grafted PCU films were characterized by fourier transformation infrared spectroscopy, X-ray photoelectron spectroscopy, contact angle, SEM, atomic force microscopy measurements and the blood compatibility of the surface was evaluated by platelet adhesion tests. The results showed that the hydrophilicity of the modified film had been improved significantly by grafting PEGMAs, and platelets adhesion onto the film surface was noticeably reduced. In addition, the molecular weight of PEGMAs had a great influence on the hydrophilicity and haemocompatibility of the PCU films after surface modification and increased with increasing molecular weight of PEGMAs [59]. Three zwitterionic polymers were grafted from silicone rubber (SR) membrane. Observing the experimental results, all the zwitterionic polymer modified surfaces have better resistance to protein adsorption and have excellent resistance to platelet adhesion, showing significantly improved blood compatibility [60]. Poly(vinyl alcohol) (PVA) was added on chitosan (CS) membrane surface and the biocompatibility was evaluated by FTIR, XRD and SEM examinations. The results suggest that adding PVA into CS membrane could greatly improve CS membrane's flexibility and wettability [61]. Acetylated 1-thio-β-D-glucopyranose and 1-thio-β-D-galactopyranose was grafted onto a homopolymer of pentafluorostyrene (PFS) and onto a block copolymer of styrene and PFS. Finally the results depicts that the grafted PFS are biocompatible for 3T3 fibroblast and MC3T3-E1 preosteoblast cell lines [62]. PEG cellulose is obtained by grafting PEG chains onto the cellulosic polymer. The results shows that the modified cellulose indicates an useful approach to improve biocompatibility of the dialysis membrane for hemodialysis[63]. Sulfonated poly(ethylene oxide) (PEO) grafted polyurethane (PU) (PU-PEO-SO(3)) was examined using scanning electron microscopy, platelet adhesion and thrombus formation appeared to be appreciably lesser formed on the PU-PEO-SO(3) coated implants compared with control PUs. The effectiveness of PU-PEO-SO(3)-coated implants in terms of blood compatibility, bio-stability and calcification resistance may provide them as a promising biomedical material in the application for blood/tissue contacting implants and artificial organs [64]. Surface grafting of polymers gains an advantage that the addition of number of polymer chains on the polymer surface can be carried out easily but the surface modification occurs through reversible physical adsorption which is a drawback of grafting technique [65].

4.2 Surface grafting of metals:

The surface of stainless steel was modified by carbohydrate polymer grafting followed by acid-treatment. The surface investigation confirmed that the surface was changed from hydrophobic to hydrophilic and from rough to smooth. The biological experiments revealed that the surface-modified stainless steel not only inhibited non-specific fibrinogen adsorption but also repelled most of proteins from human blood. The treated stainless steel surfaces have improved biocompatibility when compared to bare stainless steel-based medical device [66]. Kyomoto *et al*. grafted a 2-methacryloyloxyethyl phosphorylcholine (MPC) polymer onto the surface of a cobalt-chromium-molybdenum (Co-Cr-Mo) alloy to develop a highly biocompatible hip joint for total hip arthroplasty and it was confirmed that the grafted metal surface have good biocompatibility than the raw cobalt alloy surface [67]. The alkaline phosphatase (ALP) enzyme was grafted to titanium and its alloy surface and it is proved that the grafted metal surface shows improved bioactivity as well as the biocompatibility [68]. 1H,1H,2H,2H-perfluorodecyl acrylate was added to the surface of diamond-like carbon (DLC) deposited titanium metal and the resulted surface was analyzed using X-ray photoelectron spectroscopy (XPS), contact angle measurement (CA), and 3D surface profiler. All results suggest that the biocompatibility and functional properties of the modified Ti_6Al_4V substrates were improved [69]. The addition of polymeric chains over the metal surface was little complicated and the grafting was not durable but it helps to attain the desired properties such as corrosion and abrasion resistance [67].

4.3 Surface grafting of nanocomposites:

The surface of $BaTiO_3$ nanoparticles was grafted with 1-tetradecylphosphonic acid (TDPA) to functionalize the surface of $BaTiO_3$ nanoparticles. The acid grafted nanoparticle surface was analyzed by FTIR, XPS and XRD. These results illustrated that the modified surface have improved flexibility and biocompatibility [70]. Cheng *et al.* modified the surface of nano-hydroxyapatite fibrous scaffold with polyethylene glycol (PEG) to enhance the hydrophilicity of n-HA particles. The results of his study proved improved wettability of modified surface [71]. The poly(ε-caprolactone) PCL-grafted HAp in nanocomposites provided more favorable environments for protein adsorption with better biocompatibility compared to unmodified HAp. Nanocomposites containing PCL-grafted nanophase HAp showed positive effects on fibroblast cell adhesion [72]. The surface of BG nanoparticles was grafted with L-lactide to yield poly(L-lactide)(PLLA) grafted gel particle(PLLA-g-BG). The nanocomposite with 20% PLLA-g-BG exhibited superior surface properties, including roughness and enhanced cell adhesion. The results depict that the application of PLLA-g-BG with a certain blend ratio can improve the bioactivity and biocompatibility [73]. Iron/carbon nanoparticles (Fe@CNPs) are nanomaterials that are grafted with polymer and their relation between biocompatibility and surface chemistry was investigated. The outcome of the investigation proved that the surface chemistry has major effect on the biocompatibility of the grafted Fe@CNPs [74]. The surface grafting of nanocomposites avoids the aggregation of nanoparticles and used to form a stable suspension in organic solvents but there is possibilities of negative effects on surface chemistry.

5. Abrasive blasting and acid etching:

Abrasive blasting is the process of forcibly propelling a stream of abrasive material against a surface under high pressure to smooth a rough surface, roughen a smooth surface, shape a surface, or remove surface contaminants or some other substances from the material surface [75]. Grit blasting involves projection of ceramic particles such as alumina, titanium oxide and calcium phosphate particles through a nozzle at high velocity by means of compressed air. Depending on the size of the ceramic particles, different surface roughness can be produced on medical implants. The blasting material should be chemically stable, biocompatible and should not produce negative effects on the material surface under treatment [76, 77]. The clinical benefits in haemodialysis therapy is the removal of substances such as beta2-microglobulin (beta2-m) have been reported by several authors: elimination of large-molecular weight "uremic toxins" is now generally acknowledged as being advantageous to the overall quality of life of patients by improving the membrane compatibility with human blood [78]. Kim projected the ceramic particles towards the surface of titanium implants at high velocity to obtain high surface roughness. Then the blasted surface of implants was modified by micro-arc oxidation treatment. A porous TiO2 layer was formed on the surface that can attribute to the excellent biocompatibility [79]. The stable oxide layer over the metal surface plays a pivotal role in biocompatibility and so an oxide layer was formed on the surface of stainless steel through grit-blasting followed by micro-arc oxidation. The modified metal surface shows enhanced biocompatibility compared to the control group and the modified stainless steel implant is suitable for cementless arthroplasty because of its outstanding biocompatibility due to oxide layer formation [80]. Lampin *et al.* sand blasted the poly (methyl methacrylate) (PMMA) with alumina particles and the treated surface was characterized in comparison with untreated samples. The results depicts that the sand blasted PMMA has increased surface roughness as well as the hydrophilicity of the polymer surface [81]. Hossein *et al.* sand blasted the titanium ($Ti_{13}Zr_{13}Nb$) surface with alumina particles followed by H_2SO_4 etching at 25º C for 20 seconds. The SLA treated surface was characterized with the aid of field emission scanning electron microscope (FESEM) and the chemical composition is measured through energy dispersive x-ray (EDX). The results of FESEM and EDX depicted that SLA treated surface have better compatibility [82]. Li *et al.* established a comparison study on various surface treatments of titanium implant such as sand blasting, acid etching and finally ended with UV radiation. As a whole, UV irradiation was recognized as a trustworthy method for surface cleaning without change of topography and roughness and lead to greater biocompatibility of sandblasted and acid-etched titanium surface [83]. The titanium metal implant was treated with hydrofluoric acid solution (HF) and then the study of modified surface displayed an increased roughness, lower cytotoxicity level and better biocompatibility than the untreated implant surface [84]. The abrasive blasting technology involves in the removal of contaminants or other substances by forcibly propelling of abrasive material that may affects the mechanical properties of the material under modification but it was the best choice for surface cleaning of all type of biomaterials[75, 82].

6. Surface Coating:

Coating is an effective method of surface modification to improve the biocompatibility of medical implants [85]. Various methods have been developed to coat medical materials such as, plasma spraying [86, 87], sputter deposition [88, 89], sol-gel coating [90], electrophoretic deposition [91, 92] or biomimetic precipitation [93] and dipping method [94], the advantages and disadvantages of these coating methods are given in table 1.

Table 1: List of merits and demerits of surface coating technologies

Coating Technique	Thickness of coating layer	Merits	Demerits
Plasma Spraying	50-250µm	Can coat complex materials	Needs extremely high temperature, de-bonding of coated layer.
Sputter Deposition	0.02-1µm	Uniform coating thickness	Expensive Time consuming Cannot coat complex surfaces
Sol-gel Coating	<1µm	Can coat complex shapes, Low processing temperatures	Requires controlled atmosphere processing
Electrophoretic Deposition	0.1-2.0mm	Uniform coating thickness Rapid deposition rates Can coat complex materials	Difficult to produce crack-free coatings, Requires high sintering temperatures
Biomimetic deposition	<30 µm	Coating of complex geometries, Co-deposition of bio-molecules	Time consuming, Requires controlled pH
Dipping Method	0.05-0.5mm	Inexpensive Coatings applied quickly Can coat complex substrates	Requires high sintering temperatures, Thermal expansion mismatch

Organophosphonic acids and organophosphonates are initially used for metal and metal oxide coatings for surface modification and modification of metal nanoparticles because of its inherent biocompatibility [95]. Mostly medical implants are coated (by plasma spraying or other methodologies) with layers of hydroxyapatite (HA), because it is rapidly integrated into the human body than the other coating materials [96], calcium phosphate to improve biocompatibility [97] or mixtures of the two[98]. In Bicon Implant System (Boston, USA), Star lock implants (Park Dental Research Corp, USA), Osstem (South Korea) surfaces are HA coated [99]. Since the application of plasma-sprayed hydroxyapatite coatings onto metallic bone implants in the 1980s, the concept of implant coatings has shifted from passive protecting thin films to active and instructive immobilized layers. Currently, a plethora of coating techniques is being investigated to actively coordinate a desired biological response at the interface between artificial implants and the surrounding living tissue [100]. Butruk *et al*. modified the polyurethane (PU) surface with soybean-derived phosphatidylcholine (PC) by one-step dip coating technique. To estimate blood compatibility of the resulted material, modified samples were contacted with human blood. The PC-treated surfaces were thoroughly analyzed and tested for fibrinogen resistance, the ability to oppose platelet adhesion, haemolysis ratio and plasma re-calcification time. Outcome of this analysis demonstrated a significant reduction in fibrinogen deposition to PC-modified materials as compared to non-modified PU. The proportion of non-aggregated platelets remaining in blood samples contacted with PC-coated materials exceeded 70%. The same parameter measured for control PU was significantly lower and is about 28% [93]. The medical polymer polyurethane was coated with polyaniline (PANi) and polyaniline-silver nanoparticle composite (PANi-AgNp) and the coated surface characteristics were investigated. Contact angle measurement indicates hydrophilic surfaces that are compatible to cells when compared unmodified surfaces. These modifications make the surface more biocompatible than the original PU. The coating of polymers is easy to implement but it decreases the resistance to heat and the coating layer is not durable [101]. HA coated bone implants have improved biological fixation, shows better fixation after 4 weeks of implantation. It can be concluded that HA coating was an effective method for improving bone formation for orthopaedic implants with enhanced biocompatibility [102]. A titanium oxide ceramic coating of 2000 Å to 2500 Å thicknesses on the titanium implant surface was subjected to heat treatment to enhance the ceramic coating adherence with the metal surface. The resulted coated metal surface shows higher surface hardness and it was suitable for orthopaedic and dental implants [103]. The ceramic coating increases the strength of chemical bonding that holds the atoms and molecules together that further improves the hardness of the material [104]. The gelatin nano gold (GnG) composite is used for surface coating of titanium in addition to insure biocompatibility. The surface characterization tests were performed to evaluate the haemocompatibility of the modified surface and the results depicts that the GnG coated surface have better compatibility than the pure titanium [105]. Kim *et al.* coated the magnesium (Mg) surface with hydroxyapatite (HA) in an aqueous solution containing calcium and phosphate sources to improve its in vitro and in vivo bio-corrosion resistance, biocompatibility and bone response. The preliminary in vivo experiments also showed that the bio-corrosion of the Mg implant was significantly retarded by HA coating, which resulted in good mechanical stability and improved biocompatibility [106]. A surface coating of poly(1,3-trimethylene carbonate) (PTMC) on magnesium (Mg) alloy was investigated. The haemocompatibility and histocompatibility of coated surface were examined and compared with control sample. The results revealed that PTMC-coated surface led to less haemolysis than on the controls [107]. The nanocomposites of fibronectin (FN) and gold nanoparticles AuNPs (FN-Au) were surface modified and analyzed by the atomic force microscopy (AFM), UV-Vis spectrophotometry (UV-Vis), and fourier transform infrared spectroscopy (FTIR). The biocompatibility of the nanocomposites was assessed by the response of monocytes and platelets to the material surface in vitro. These results recommend that the FN-Au nanocomposite thin film coating may serve as a potential and uncomplicated solution for the surface modification of blood-contacting medical devices [108]. For

all types of raw materials, the advantages of coating technique include deposition of holes present in the surface and low processing temperature but the selection of coating material still remains a challenge [95, 104].

7. Thermal Treatment:

Heat treating is a group of industrial and metal working processes used to alter the physical and chemical properties of a material. Kawase *et al.* made a comparison study on control platelet-rich fibrin (PRF) with the heat-compressed PRF. The heat treated sample appeared plasmin-resistant and remained stable for longer than 10 days. Moreover, in animal implantation studies, the heat-compressed PRF was experimented at least for 3 weeks after implantation *in vivo* whereas the control PRF was completely degraded within 2 weeks. Therefore, these findings suggest that the heat-compression technique decrease the rate of biodegradation of the PRF membrane without sacrificing its biocompatibility [109]. Titanium was modified by means of hydrothermal treatment with a maximum pressure of 6.3 MPa at 280º C in Calcium oxide (CaO) solution or water to improve bioactivity and biocompatibility. As a result, calcium titanate was formed on the titanium surface and has improved bioactivity and biocompatibility [110]. Titanium was also treated with NaOH solution at 60º C for 24 hours followed by heat treatment upto 600º C for 1 hr. The results infer that alkali solution-heat treated surface have improved biocompatibility [111]. Titanium implant was sintered with tricalcium phosphate (TCP) by spark plasma at 1200º C, the final TCP-Ti composite shows improved cell viability and proliferation. *In vivo* study confirmed that within 3 months of implantation in an animal body, 70% TCP-Ti had an excellent bone-implant interface compared with a pure Ti metal implant [112]. In addition to bioactivity, orthopaedic implants require porosity for tissue regeneration, heating at high temperatures (500-1000ºC) resulted in porosity and directing to positive consequent modifications in the mechanical properties and biocompatibility, biodegradability and bioactivity of the material surface [113, 114]. The effect of heat treatment on the alloys were studied and documented that the microstructure of an alloy was changed due to the thermal effect; the end results could be useful in further understanding the relationship between the biocompatibility, wear and corrosion resistance of the alloy, so as to allow the development of a promising biomedical material [115]. Bimbo *et al*. demonstrated that thermally hydrocarbonized porous silicon (THCPSi) nanoparticles did not induce any significant toxicity, oxidative stress, or inflammatory response in Caco-2 and RAW 264.7 macrophage cells. On the whole, these silicon-based nanosystems exhibit outstanding *in vivo* stability, biocompatibility, low cytotoxicity and non-immunogenic profiles, ideal for oral drug delivery purposes [116]. Cui *et al*. applied hot water and heat treatments to transform the titania layers from an amorphous structure into a crystalline structure with enhanced compatibility. The loads of Ti-OH groups formed by hot water treatment could contribute to apatite formation on the surface of titanium metals, and subsequent heat treatment would enhance the bond strength between the apatite layers and the titanium substrates. Thus, bioactive titanium metals could be prepared via hot water followed by heat treatment that would be suitable for applications under load-bearing conditions [117]. The high operating temperature of thermal modification is a major drawback. The polymeric and ceramic materials failed to withstand this high level temperature and so thermal treatment is mostly preferred for metallic biomaterials, possessing high melting point that lead to optimistic changes in mechanical and physico-chemical properties of metals.

8. Vapour and Steam treatment:

Water vapour is little water droplets that exist in the air, while steam is water heated to the point that it turns into gas. In simplified science, both are referred to as the gaseous state of water. Steam is believed to be basically water vapour at a higher temperature. A vapour is a matter in the gas state at a temperature below to its critical point and it is used in the field of biomaterial science to improve the blood compatibility of the biomaterials [118]. Jensen *et al.* etched polydimethylsiloxane (PDMS) for 8-min in water vapour at a pressure of 50 m Torr and power of 400 W that resulted in unwavering long-term wettability and excellent *in vitro* cell compatibility. Finally it was concluded that water vapour plasma may be used to improve biointegration of PDMS implants and thereby evade clinical problems related with the formation of a dead space [119]. Silicon carbide (SiC), a chemical vapour deposition coating for cardiovascular implants resulted in decline in platelet adhesion and also less inflammatory reactions. Diamond like carbon has comparable advantages as SiC and also it offers higher hardness, lower frictional coefficient, chemical inertness, bio-stability, and also good blood compatibility making it as graceful alternative for the application on vascular stents [120]. Wang *et al*. used human hepatoma cells (BEL-7402) as model cells to examine cell adhesion, spreading and proliferation of cells on zein films before and after surface treatment with water vapour and he concluded hydrophilicity, cell adhesion were significantly improved after the treatment on zein films [121]. Non-woven polyethylene terephthalate (NW-PET) was subjected to surface modification under water vapour to enhance the compatibility and the outcome of this study illustrated that the water vapour treated NW-PET have improved platelet compatibility [122]. The hydroxyapatite (HA) coatings were kept in water vapour at 125º C, with a pressure of 0.15 MPa for 6 hr to modify amorphous phase in the coating into crystalline HA and improved the stability of the coating [123]. Lee *et al.* established that the water vapour treatment is an easy and valuable technique to fabricate hydroxyl groups on the polymer surfaces, which possibly have a positive effect on cell adherence with increased wettability [124].

Steam is the technical term for the gaseous phase of water, which is formed when water boils, frequently used surface treatment for biomaterials to seal the interconnected pores and to enhance the superficial properties of the material. The treatment with steam forms an oxide layer on the material surface that fills the pores on the surface thereby increasing the density and also has effect on the hardness of the material [125]. Steam treating is the controlled oxidation of metals to produce a thin layer of oxide on the surface of a component. This process can be used to provide a component with improved corrosion resistance, better wear resistance, increased surface hardness, wettability [126]. The ferrous components were subjected to steam treatment to improve the mechanical properties through the formation of oxide layer that gives better biocompatibility and the reduced heat required for steam production and its environmental benefits makes steam treatment technology as a growing application in surface modification [127]. The high biocompatibility of Ti and alloys of Ti is due to the formation of oxide layer during the process of implant preparation and it can be modified by steam sterilization that results in increased oxide layer thickness with respect to the unsterilized samples [128]. Rai *et al*. sterilized the poly(glycerol sebacate) (PGS) with steam and concluded that the sterilized samples maintain their mechanical properties, compatibility and the treated PGS was used for wider applications in medical devices [129]. Chen *et al.* performed hydrophilization treatment on graphene surface using steam to

reduce the interfacial impedance during cardiac and neural recording through converting it into hydrophilic nature that resulted in enhanced biocompatibility [130]. The mechanical properties of chitosan/hydroxyapatite (HAp) nanocomposites were improved by heat treatment with saturated steam which led to the hydrogen bond formation between chitosan molecules. The treated nanocomposites were implanted into the rats and after 3 weeks of post implantation it was found that the cells were seen around the composite accompanied with surface roughness showed enhanced biocompatibility [131]. The limitation of vapour and steam treatment includes that the material under the surface modification should tolerate the heat of vapour and steam. It is ease, cost effective and there is no need of further sterilization of modified surface are the merits of vapour and steam treatment [124].

9. Conclusion:

The surface properties and biocompatibility of the biomaterials were improved by various surface modification techniques namely physico-chemical, mechanical and biological methods. Among these techniques, physico-chemical method, a versatile strategy is widely used in recent days which include both physical and chemical means, frequently the covalent addition of some reagents and high temperature treatment. The merits and demerits of each physico-chemical modification technology for each raw material are listed in the table 2. From the results and remarks of various works summed up, we can conclude that most of the research has been aimed on chemical modifications to improve the biocompatibility by altering the surface characteristics of the medical material.

Mostly all modalities under physico-chemical modification were performed on metals and its alloys due to the high mechanical strength of metallic bonding that can withstand high temperature without losing its shape. The surface treatment of polymers can be mainly achieved by surface grafting and coating mechanisms. However further experimentation of polymers using other modalities may further promote it as a promising biomaterial for various biomedical applications. The recent generation of biomaterials such as ceramics and nanocomposites are not much subjected to surface modifications because of their tailor made characteristics for particular biomedical applications. Despite of its pre-defined characteristics, there are some deficits with biocompatibility, so ceramics and nanocomposites were still expected to be investigated in depth to develop insights about the prospect of physico-chemical treatments in those materials.

The physico-chemical modification induced changes in the physical properties of all biomaterials like modification in their surface roughness and wettability. These physical changes due to physico-chemical modification are graphically represented in the figure 2. These physical variations leads to the improved biocompatibility decreased platelet adhesion, enhanced protein adsorption and reduced red blood cell damage as shown in the figure 3. Biomedical materials subjected to physico-chemical modification makes them more attractive choice for diverse applications like cardiovascular, tissue regeneration and orthopaedic applications since the surfaces can be optimized for the particular applications. Hence proper exploitation of this strategy will quench the thirst of long time unmet demands of biocompatibility.

Table 2: List of merits and demerits of surface modification technologies for each raw material.

Modification Technology	Type of Material	Merits	Demerits
Chemical modification	Polymer	• Produces new polymeric material which cannot synthesized by polymerization reaction	• Non specific interaction
	Metal	• Desired topographical properties	• Effects based on chemical reagents or molecules used for modification
	Ceramics	• Increased bioactivity	• Selection of chemical reagent remains challenge
	Nanocomposites	• Reduction in agglomeration	• Effects based on chemical reagents or molecules used for modification
Surface Grafting	Polymer	• Controllable addition of polymeric chains	• Physical adsorption taking place is reversible
	Metal	• Better corrosion resistance • Improved abrasion resistance	• Non specific localization of grafted molecules • Complicated process
	Nanocomposites	• Aggregation of nanoparticles are avoided • Forms a stable suspension in organic solvents	• Possibilities of negative effects on surface chemistry.
Abrasive blasting and acid etching	Polymer, Metal , Ceramics and Nanocomposites	• Removes contaminants • Best choice of surface cleaning	• Forcibly propelling of abrasive material may damage the surface • Mechanical properties may affected
Surface Coating	Polymer	• Deposition of holes • Easy to implement	• Decreased heat resistance
	Metal	• Deposition of holes • Better corrosion resistance	• Coating layer is not durable
	Ceramics	• Increases the strength of chemical bonding • Increased hardness	• Selection of coating material is complicated
	Nanocomposites	• Enhanced interaction of coated material with the surface	• Non stable coating layer
Thermal treatment	Polymer, Metal , Ceramics and Nanocomposites	• Optimistic changes in mechanical properties • Improved physico-chemical properties	• Very high operating temperature • Only suitable for the materials with high melting point
Vapour and steam treatment	Polymer, Metal , Ceramics and Nanocomposites	• Cost effective • No need of further sterilization	• Production of high temperature • Produce adverse thermal effects

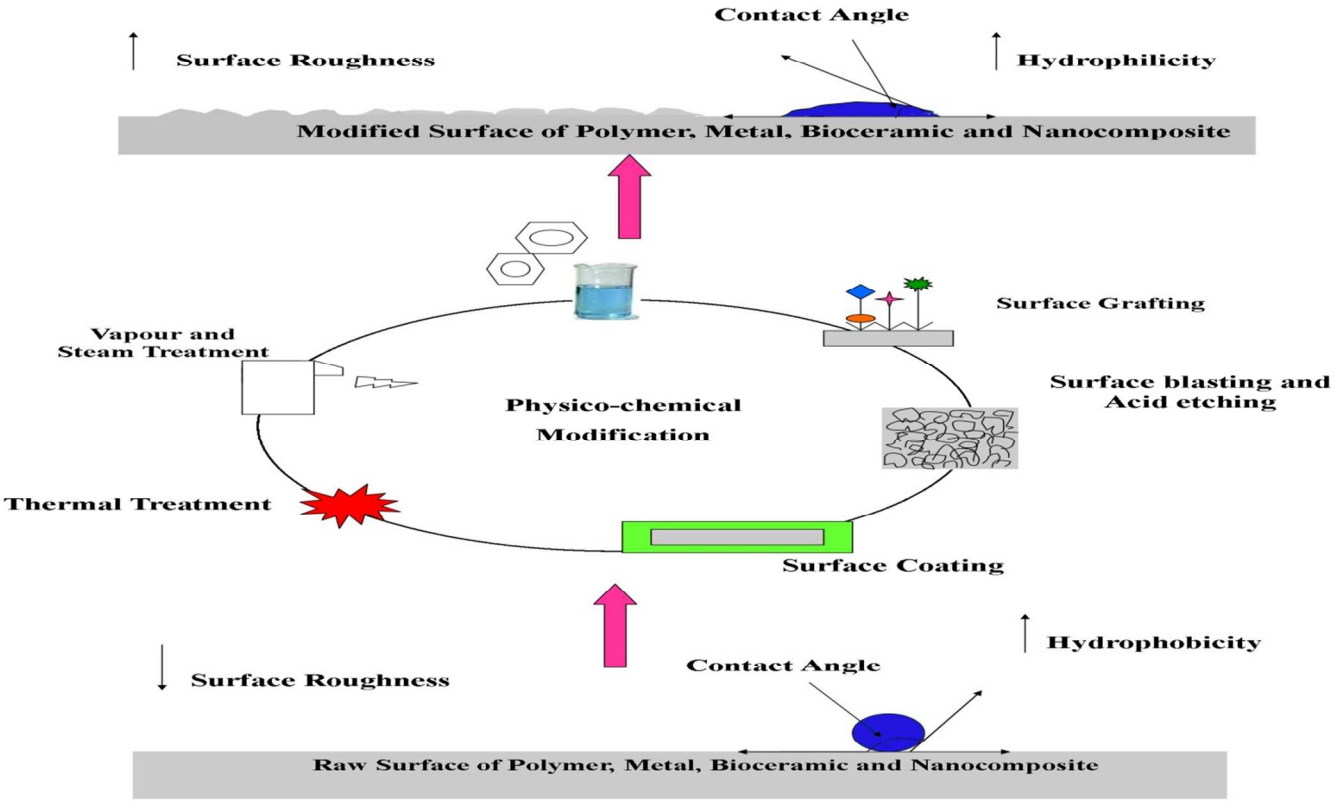

Figure 2: Physical changes of the biomaterial surface due to physico-chemical modification

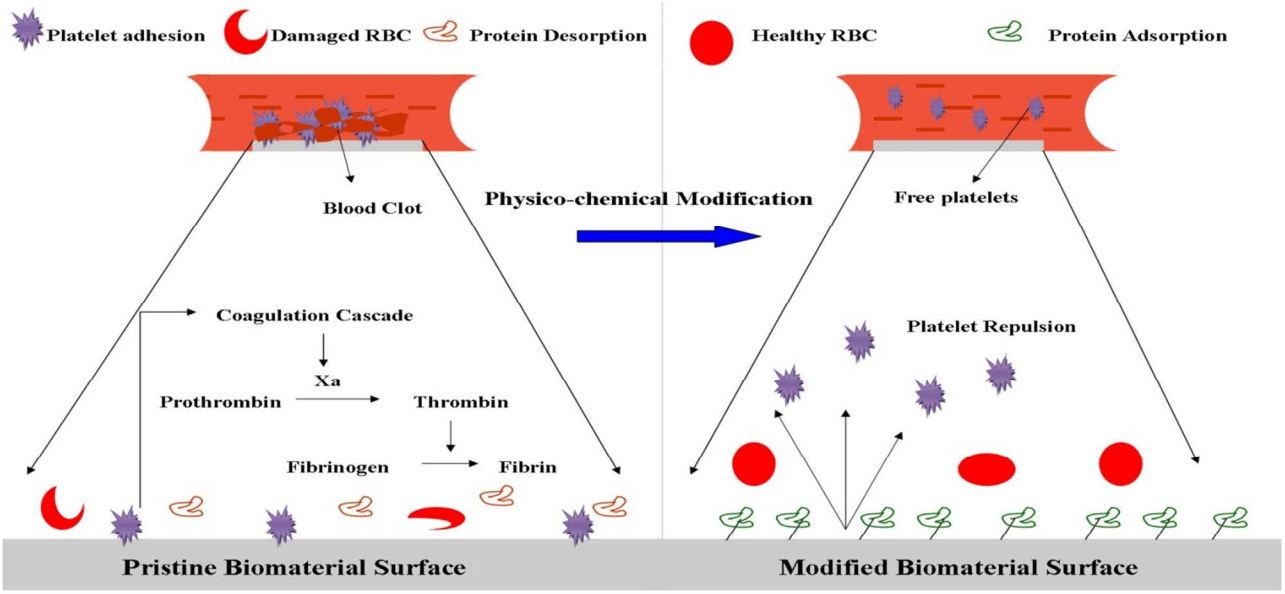

Figure 3: Enhanced biocompatibility changes induced by the physico-chemical modification

ACKNOWLEDGEMENTS

This work was supported partly by the Ministry of Higher Education Malaysia with the Grant Vot No: R.J130000.7809.4F444 and also acknowledges the support of UPMU, UTM.

Notes and references

[a]IJN-UTM Cardiovascular Engineering Centre, Faculty of Biosciences and Medical Engineering, Universiti Teknologi Malaysia, Johor Bahru 81310, Malaysia.
[b]Department of Chemistry, Faculty of science, Universiti Teknologi Malaysia, Johor Bahru 81310, Malaysia.
[c]Department of Biomedical Engineering, University of Texas Arlington, Texas,United States- TX 76019.

[*]Corresponding author
Dr Saravana Kumar Jaganathan, IJN-UTM Cardiovascular Engineering Centre, Faculty of Biosciences and Medical Engineering, University Teknologi Malaysia, Johor Bahru 81310, Malaysia Email: jaganathaniitkgp@gmail.com
Tel : 00607-5558548:Fax: 07-5558553

1. D.Bronzino, The Biomedical Engineering Handbook, 2nd edition,1 , 2000.
2. http://reportlinker.com/ci02234/Biomaterial.html
3. P.Parida, A.Behera, S.Chandra Mishra, *International Journal of Advances in Applied Sciences*, 2012, 125-129.
4. B.Singh, L.Pal, *J Mech Behav Biomed Mater*, 2012, 9-21.
5. G.Manivasagam, D.Dhinasekaran, A. Rajamanickam, *Recent Patents on Corrosion Science,* 2010, 40-54.
6. D.Williams, Medical device technology 14 (8): 10–3, 2003.
7. J.Lemmons, J.Natiella, *Dent Clin North Am.* 1986,30,3–23.
8. G.Schmalz, *J Dent Res*, 2002, 81,660–663.
9. S.K.Jaganathan, E.Supriyanto, S. Murugesan, A.Balaji, M.K.Asokan, *BioMed Research International*. 2014.
10. M.Shuchman, *New England Journal of Medicine.* 2006, 355, 1949–1952.
11. DF.Williams,*Biomaterials,*2008,29,2941–2953.
12. E.Gentleman, M.D.ball, M.M.stevens. *Biomaterials*. Vol. 2.
13. S.K.Jaganathan, A.Balaji, M.V. Vellayappan, A.P.Subramanian, A.A. John, M.K.Asokan, E.Supriyanto, *J Mater Sci,* 2014.
14. L. Richard McCreery and Adam Johan Bergren, 2012.
15. V Safonov, A Zykova, J Smolik, R Rogovska, N Donkov and V Georgieva, *Journal of Physics:ConferenceSeries,* 2010.
16. Iva Milinkovi, Rebeka Rudolf, Karlo T. Rai, Zoran Aleksi, Vojkan Lazi, Aleksandar Todorovi, Dragoslav Stamenkovi, *Materials and technology*, 2012, 251–256.
17. Fabio Variola, John B. Brunski, Giovanna Orsini, Paulo Tambasco de Oliveira, Rima Wazenm and Antonio Nanci, *Nanoscale,* 2011, 3, 335–353.
18. Farhang Abbasi, Hamid Mirzadeh and Ali-Asgar Katbab, *Polymer International*, 51:882–888, 2002.
19. KNJ.Stevens, Maastricht, 2011.
20. AP.Alekhin, GM.Boleiko, SA.Gudkova, AM.Markeev, AA.Sigarev, VF.Toknova, AG.Kirilenko, RV.Lapshin, EN.Kozlov, DV.Tetyukhin, *Nanotechnologies in Russia*,2010,5,696–708.
21. S.Bertazzo, WF.Zambuzzi, da Silva, CV.Ferreira, CA.Bertran, *Clinical Oral Implants Research,* 2009, 20, 288-293.
22. Gabo, Kuang-Yen Chen, GT. Carroll, B. L.Feringa, *A Eur J.* 2013, 19 (32),10690–10697.
23. A.S.Hoffman, *Chin Jour Polym Sci,* 1995, 3,13.
24. JY.Lim, HJ.Donahue, *Tissue Engineering,* 2007, 13(8),1879-91.
25. A.Thapa, DC.Miller, TJ.Webster, KM.Haberstroh, *Biomaterials,* 2003, 24(17),2915-26.
26. DC.Miller, A.Thapa, KM.Haberstroh, TJ.Webster, *Biomaterials,* 2004, 25(1),53-61.
27. B.Balakrishnan, DS.Kumar, Y.Yoshida, A. Jayakrishnan, 2005, 26(17),3495-502.
28. BJ.Yvette, Aldenhoff, L.H.Koole, *Eur Cells Mater*, 2003,5,61-67.
29. T.Xiang, WW.Yue, R.Wang, S.Liang, SD.Sun, CS.Zhao,*Colloids Surf B Biointerfaces,* 2013, 110,15-21.
30. S.Nie, J.Xue, Y.Lu, Y.Liu, D.Wang,S.Sun, F.Ran, C.Zhao, *Colloids Surf B Biointerfaces,* 2012, 116-25.
31. SK.Jaganathan, H.Mohandas, G. Sivakumar, P.Kasi, T.Sudheer, S.Avineri Veetil, S.Murugesan, E.Supriyanto, *BioMed Res Int*,2014.
32. HD.Park, WK.Lee, T.Ooya, KD.Park, YH.Kim, N.Yui, *J Biomed Mater Res A*. 2003, 66(3),596-604.
33. ZK.Xu, FQ.Nie, C.Qu, LS.Wan, J. Wu, K. Yao, *Biomaterials,* 2005, 26(6),589-98.
34. Ahmed Akelah, *Technology and Engineering*, 2013.
35. KM.Hansson, S.Tosatti, J.Isaksson et al. *Biomaterials,*2005,26:861–872.
36. M.Danila Korotin, S.Bartkowski, EZ. Kurmaev, M.Meumann, B.Eugeniya Yakushina, RZ.Valiev, O.S.Cholakh, *J Biomater Nanobiotech,* 2012, 3,87-91.
37. W.Xue, X.Liu, X.Zhen, C.Ding, 2005, 26(16),3029-37.
38. X.Zhao, X.Liu , C.Ding , PK Chu, *Surf Coat Tech*. 2006, 5487 - 5492.
39. CH Yea, YF Zhenga, SQ Wang, TF Xia, YD Li, *Appl Surf Sci*, 2012, 258, 3420– 3427.
40. MH.Lee, D.Yoon, D.Won, T.Bae, F. Watari, *Met Mater Int*. 2003, 9, 35-42.
41. A.Parsapour, S. Nouri Khorasani, M. Hossein Fathi, *J Mater Sci Technol*, 2012, 28(2), 125-131.
42. H.Zreiqat, P.Evans, CR.Howlett, *J Biomed Res*, 1999, 44, 389-396.
43. K.Nishizawa, M.Toriyama, T.Suzuki, Y.Kawamoto, Y.Yokogawa, H.Nagae, *J Ferm Bioeng*, 1993, 75, 435-437.
44. Q.Zhao, GJ.Zhai, Ng DHL, XZ.Zhang, ZQ.Chen, *Biomaterials,* 1999, 20, 595-599.
45. W.Zhang, W.Liu, W.Gu, L.Chen, Y. Shen, *Adv Mater Res*, 2012, 391-392, 195-199.

46. Sk.Hsua, H.Hsua, WF.Hob, CH. Yaoc, P.Changd, SC.Wu, The 41st International Conference on Metallurgical Coatings and Thin Films, 2014, 572,91–98.
47. B.Duan, M.Wang, *J R Soc Interface*, 2010, 7, S615–S629.
48. C. Deng , X. Xiao , N. Yao , XB. Yang, J. Weng, *Int J Polym Mater Po*. 2011, 60,12,969-978.
49. MC. Pascu, MC. Popescu, C.Vasile, *J. Phys. D: Appl Phys,* 2008, 41, 12.
50. M.Selvakumar, SK.Jaganathan, GB. Nando, S.Chattopadhyay, *J Biomed Nanotech*, 2015, 11, 291-305.
51. AR.Adhikari, I.Rusakova, A.Haleh, J.Luisi, NI.Panova, Laezza F, WK.Chu, *J Appl Phy*, 2014, 115.
52. Q.Yue, M.Wang, Z.Sun, C.Wang, Y. Deng, D.Zhao, *J Mat Chem B,* 2013,1, 6085-6093.
53. A.Mohammadi, M.Barikani, M.Barmar, 2013, 48,7493-7502.
54. AV.Jovanovic, JA.Flint, M.Varshney, TE.Morey, DM.Dennis, RS.Duran, *Biomacromolecules,* 2013, 7(3):945-9.
55. XH.Gong, CY.Tang, HC.Hu, XP.Zhou XP, XL.Xie, *J Mater Sci Mater Med*, 2004, 15(10),1141-6.
56. LS. Wang and RY. Hong, *Advances in Nanocomposites*, 289-322.
57. S.Minko,*Polymer surfaces and interfaces* 2008, 215-234.
58. P.Alves, S.Pinto, P.Ferreira, JP.Kaiser, A.Bruinink, HC.de Sousa, MH, *J Mater Sci Mater Med*,2014,2017-26.
59. Y.Feng, H. Zhao, M.Behl, A.Lendlein , J.Guo, D.Yang, *J Mater Sci Mater Med* 2013,24(1),61-70.
60. P.Liu, Q.Chen, B.Yuan, M.Chen, S.Wu, S.Lin, J.Shen, *Mater Sci Eng C Mater Biol Appl,* 2013,33(7),3865-74.
61. P. Zhuanga, Y.Lia, L.Fanb, J.Linb, Q. Hu, *Int. J. Biol. Macromol,* 2012,50,658–663.
62. K.Babiuch, CR.Becer, M.Gottschaldt, JT.Delaney, J.Weisser, B.Beer, R.Wyrwa, M.Schnabelrauch, US.Schubert, *Macromol Biosci,* 2011, 11, 535-548.
63. Sirolli V, S. Di Stante, S.Stuard, L.Di Liberato, L.Amoroso, P.Cappelli, M.Bonomini, *Int J Artif Organs*, 2000, 23(6),356-64.
64. L.Yang, JC.Davis, R.Vankayala, KC.Hwang, J.Zhao, B.Yan, *Biomaterials.* 2010, 31,5083-5090.
65. Mitsuhiro Ebara et al., smart biomaterials, 2014.
66. C.Kanga, Y.Lee, *J Ind Eng Chem*, 2012,18,1670–1675.
67. M.Kyomoto, T.Moro, Y.Iwasaki, F.Miyaji, H.Kawaguchi, Y.Takatori, K.Nakamura, K.Ishihara, *J Biomed Mater Res A,* 2009, 91, 730-741.
68. S.Ferraris, S.Spriano, CL.Bianchi, C.Cassinelli, E.Vernè, *J Mater Sci - Mater Med,* 2011,22,1835-1842.
69. HD.Park,WK. Lee, T.Ooya, KD.Park, YH.Kim, N.Yui, *J Biomed Mater Res A*, 2003, 66(3),596-604.
70. HJ.Ye, WZ.Shao, L.Zhen, *Colloids Surf A,*2013,427, 19-25.
71. Z.Cheng, G.Pang, H.Wang, J.Li, X.Zhao, *Adv Mat Res*, 2012,1095-1099.
72. HJ.Lee, SE.Kim, HW.Choi, CW.Kim, KJ.Kim, SC.Lee, *Eur Polym* J, 2007, 43, 1602-1608.
73. SJ.Dong, T.Yu, JC.Wei, XB.Jing, YM. Zhou, PB.Zhang, XS.Chen, *Chem J Chinese,* 2009,30,1018-1023.
74. Y.Liu, X.Fu, Y.Bu, J.Zhang, J.Zhang, Wu.Longmin, *Surf Coat Tech*, 2012, 208, 51–56.
75. B.Pattanaik, S.Pawar, S.Pattanaik, 2012,23 (3),398-406.
76. A.Wennerberg, T.Albrektsson, B.Andersson, *Int J Oral Maxillofac Implants*, 1996, 11,38-45.
77. A.Citeau, J.Guicheux, C.Vinatier, P.Layrolle, TP.Nguyen, P.Pilet, et al. *Biomaterials*, 2005, 26,157-65.
78. SK.Bowry, *Int J Artif Organs*, 2002, 25(5),447-60.
79. YW.Kim, *Mater Manuf Processes,* 2010, 25, 307-310.
80. YM.Lim, SY.Kwon, DH. Sun, YS.Kim, *Clin Orthop Relat R,* 2011, 469,330-338.
81. M.Lampin, R.Warocquier-Clérout, C.Legris, M.Degrange, MF.Sigot-Luizard, *J Biomed Mater Res*, 1997, 36, 99-108.
82. HM.Khanlou, *AJBAS,* 2012, 6(6), 125-131.
83. S.Li, J.Ni, X.Liu, X.Zhang, S.Yin, M. Rong, Z.Guo, L.Zhou, *J Biomed Mater Res Part B,*2012,100B,1587–1598.
84. SF.Lamolle, M.Monjo, M.Rubert, HJ.Haugen, SP.Lyngstadaas, JE.Ellingsen, *Biomaterials.*2009,(5),736-42.
85. T.Shirzadian, S.Bagheri, H.SaeidiBorojeni, P.Ghaffari, F.Foroughi, M.Mahboubi, *Congress of Iranian Neurosurgeons,* 4, 2012.
86. Z.Strnad, J.Strnad, C.Povýsil, K.Urban, *Int J Oral Maxillofac Implants*. 2000,15,483-90.
87. M.Gottlander, CB.Johansson, T.Albrektsson, *Clin Oral Implants Res*. 1997, 8,345-55.
88. M.Yoshinari, Y.Ohtsuka, T.Dérand, *Biomaterials,* 1994,15,529-35.
89. Y.Yang, KH.Kim, JL.Ong, *Biomaterials,* 2005; 26:327-37.
90. HQ.Nguyen, DA.Deporter, RM.Pilliar, N.Valiquette, R.Yakubovich, *Biomaterials*, 2004, 25,865-76.
91. D.Lakstein, W.Kopelovitch, Z.Barkay, M.Bahaa, D.Hendel, N.Eliaz, *Acta Biomater,* 2009, 5, 2258-69.
92. J.Wang, P.Layrolle, M.Stigter, K.de Groot, *Biomaterials*, 2004,25,583-92.
93. F.Barrère, P.Layrolle, CA.Van Blitterswijk, K.De Groot, *J Mater Sci Mater Med,* 2001, 12,529-34.
94. B.Butruk-Raszeja, M.Trzaskowski, T.Ciach, *J Biomater Appl,* 2014.
95. C.Queffélec, M.Petit, P.Janvier, D.Andrew Knight, B.Bujoli, *Chem Rev,* 2012,112, 3777−3807.
96. M.Niinomi, M.Nakai, J.Hieda, K.Cho, T.Kasuga, T.Hattori, T. Goto, T.Hanawa, *Int J Surf Sci Eng,*2012, 8,138-152.
97. K.De Groot, *J Biomed Mater Res,* 1989,23,1367-71.
98. HW.Kim, G.Georgiou, JC.Knowles, YH. Koh,HE.Kim,*Biomaterials*,2004,25(18),4203-13.
99. EJ Lee, SH.Lee, HW.Kim, YM.Kong, HE.Kim, *Biomaterials*, 2005, 26,3843-51.
100. R.Bosco, J.Beucken, S.Leeuwenburgh, J. Jansen, *Coatings,* 2012,2, 95-119.
101. PK.Prabhakar, S.Raj, PR.Anuradha, SN.Sawant, M.Doble, *Colloid Surface B,* 2011,86,146-153.
102. A.Moroni, VL.Caja, EL.Egger, L.Trinchese,EY. Chao, *Biomaterials,* 1994, 15(11),926-30.
103. G.Szabó, L.Kovács, K.Vargha, J.Barabás, NZ. Émeth, *J Long Term Eff Med Implants*, 1999, 9,247-259.
104. Iva Milinkovi, Rebeka Rudolf, Karlo T. Rai, Zoran Aleks, Vojkan Lazi, Aleksandar Todorovi, Dragoslav

Stamenkovi, *Materials and technology*, 2012, 3, 251–256.
105. YH. Lee, G.Bhattaraia, S.Aryal *et al. Appl Surf Sci,* 2010, 256, 5882–5887.
106. SM.Kim, JH.Jo, SM.Lee, MH.Kang, HE.Kim, Y.Estrin, JH.Lee, JW.Lee, YH.Koh, *J Biomed Mater Res A.* 2014,102(2),429-41.
107. J.Wanga, Y.Hea, MF.Maitz *et al*. *Acta Biomater,* 2013, 9, 8678–8689.
108. HS.Hung, CM.Tang, CH.Lin, SZ.Lin, MY.Chu et al. *PLoS ONE*, 8(6), 2013.
109. T.Kawase, M.Kamiya, M.Kobayashi, T.Tanaka, K.Okuda, LF. Wolff, H.Yoshie, *J Biomed Mater Res B Appl Biomater*. 2014.
110. R.Sultana, M.Kon, LM.Hirakata, E.Fujihara, K.Asaoka, T.Ichi kawa, *Dent Mater J*, 2006,25(3),470-9.
111. K.Feng, E.Wu, Y.Pan, K.Ou, *Mat Trans*. 2007, 48, 2978-2985.
112. D.Mondal, L.Nguyen, IH.Oh, BT.Lee,*J Biomed Mater Res A,* 2013, 101(5),1489-501.
113. P.Majumdar, SB.Singh, S.Dhara, M.Chakraborty, *J Mech Behav Biomed Mater,* 2012, 10,1-12.
114. Y.Zhou, H.Li, K.Lin, W.Zhai, W.Gu, J.Chang, *J Mater Sci Mater Med*, 2012, 23(9),2101-8.
115. CZ.Wu, SC.Chen, YH.Shih, JM.Hung, CC.Lin, LH. Lin, KL.Ou,*J Mech Behav Biomed Mater,* 2011, 4(7),1548-53.
116. LM.Bimbo, M.Sarparanta, HA.Santos, AJ.Airaksinen, E.Mäkilä, T.Laaksonen, L.Peltonen, VP.Lehto, J.Hirvonen, J.Salonen, *ACS Nano,* 2010, 4(6),3023-32.
117. X.Cui, HM.Kim, M.Kawashita, L.Wang, T.Xiong, T.Kokubo, T.Nakamura, *J Mater Sci Mater Med,* 2008, 19(4),1767-73.
118. MJ.Jackson, GM.Robinson, N.Ali, Y.Kousar, S.Mei, J.Gracio, H.Taylor, W.Ahmed, *J Med Eng Technol,* 2006, 30(5),323-9.
119. C. Jensen, L. Gurevich, A. Patriciu, JJ. Struijk, V.Zachar, CP. Pennisi, *J Biomed Mar Res A,* 2012, 100 A.
120. M. Fedel, A. Motta, D. Maniglio, C. Migliaresi, *J Biom Mat Res B,* 2009, 90,338–349.
121. HJ.Wang, JX.Fu, JY.Wang, *Colloids Surf B Biointerfaces,* 2009, 69(1),109-15.
122. EH.Kostelijk, AJ.Klomp, GH.Engbers, CW.Gouwerok, AJ.Verhoeven, WG.Van Aken, J.Feijen, D.de Korte, *Transfus Med,* 2001, 11(3),199-205.
123. Y.Cao, J.Weng, J.Chen, J.Feng, Z.Yang, X.Zhang ,*Biomaterials*, 1996, 17(4),419-24.
124. JH.Lee, JW.Park, HB.Lee, *Biomaterials,* 1991, 12(5),443-8.
125. P. Nader, H.Zahra, N.Mohammad. Comat.2012.
126. Y.Cao, F.Chan, YH.Chui, H.Xiao, *Bio Resources,* 2012, 7, 4109-4121.
127. SL.Feldbauer, Steam Treating; Enhancing the surface properties of metal components**.**
128. NL.Hernández de Gatica, GL.Jones, JA.Gardella Jr, *Appl Surf Sci*, 1993, 68,107-121.
129. R.Rai, M.Tallawi, JA.Roether, R.Detsch, N.Barbani, E.Rosellini, J.Kaschta, DW.Schubert, AR.Boccaccini , *Mater Lett,* 2013, 105,32-35.
130. CH.Chen, CT.Lin, WL.Hsu, YC.Chang, SR.Yeh, LJ.Li, DJ.Yao, *Nanomedicine*, 2013, 9(5),600-4.
131. H.Kashiwazaki, Y.Kishiya, A.Matsuda, K.Yamaguchi, T.Iizuka, J.Tanaka, N.Inoue, *Bio-Med Mater Eng,* 2009, 19,133-140.