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Review Article

Review on material parameters to enhance bone cell function *in vitro* and *in vivo*

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Bone plays critical roles in support, protection, movement, and metabolism. Although bone has an innate capacity for regeneration, this capacity is limited, and many bone injuries and diseases require intervention. Biomaterials are a critical component of many treatments to restore bone function and include non-resorbable implants to augment bone and resorbable materials to guide regeneration. Biomaterials can vary considerably in their biocompatibility and bioactivity, which are functions of specific material parameters. The success of biomaterials in bone augmentation and regeneration is based on their effects on the function of bone cells. Such functions include adhesion, migration, inflammation, proliferation, communication, differentiation, resorption, and vascularization. This review will focus on how different material parameters can enhance bone cell function both *in vitro* and *in vivo*.

Introduction

Bone is a rigid tissue which provides structural support, facilitates locomotion, serves as a reservoir for mineral storage, and protects internal organs and soft tissue. Bone is also an endocrine organ, which serves as a niche for bone marrow and a source of stem cells. This complex tissue consists of an organic component, an inorganic, mineral component and water. The organic component is primarily type I collagen, but also consists of protein polysaccharides, glycosaminoglycans (GAGs) and other non collagenous proteins. Together, the organic constituents are responsible for the toughness of bone [1] which is important to prevent propagation of microfractures. The organic material serves as a template for the nucleation and formation of the inorganic material [2] into both needle and platelet shaped crystals [3] of non-stoichiometric hydroxyapatite (HA), which contains carbonate substitutions. The mineral components give bone its hardness, rigidity and strength [4]. Multiple factors affect bone strength, including the composition, of both the organic and inorganic phases, their arrangement and microarchitecture, and cell presence [5]. Bone fulfils its functions by adapting to its environment across dimensional scale, such as modeling to alter the morphology of thick walled long bones to support mechanical demands, and chemical components, such as the mineral/collagen ratio, which affects physical properties.

Many materials have been used either as permanent prosthetic replacements for bone, or as transient scaffolds to promote bone regeneration. Bone-based biomaterials fall into three broad categories: metals, ceramics, and polymers. Metals used for bone anchored prostheses such as total hip replacements, are primarily cobalt-chromium, and titanium-based alloys [6]. Titanium is notable for its high biocompatibility and ability to form close to direct contact with bone (an interfacial zone of ~100 μm) which is called osseointegration. The ability to achieve osseointegration is commonly exploited in anchoring dental implants to alveolar bone [7]. While metals have favorable mechanical properties for prosthetics, such as high strength and fatigue resistance, titanium is subject to high wear and all metals release metal ions (corrosion), which is more pronounced when concomitant with wear [6,8]. Bioceramics used in bone reconstruction and regeneration includes non-resorbable,

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'bioinert' ceramics that elicit a minimal biological response and are held in bone by mechanical means (press fit, rough surfaces, grouting agents, or tissue ingrowth), surface active ceramics that form chemical bonds with bone, and resorbable ceramics that are gradually replaced by host bone [9]. 'Inert' ceramics are typically used in orthopedic prostheses, particularly the acetabular bearing surface of hip prostheses, as well as endosseous dental implants, and include alumina, zirconia, and silicon nitride [6]. Although ceramics have a low friction coefficient and wear rate, their brittle nature and low fatigue resistance are potential limitations to use in load-bearing applications [8]. Resorbable ceramics includes calcium phosphate ceramics, such as apatites. The solubility and resorbility of apatites vary based on their chemistry and crystallinity. Highly crystalline, stoichiometric HA has poor solubility while biomimetic carbonated apatites similar to native bone mineral are more easily resorbed [10]. Like ceramics, polymers can be divided into resorbable and non-resorbable categories. Non-resorbable polymers include poly-methyl methacrylate which is used as bone cement and grouting agent for anchoring prostheses into bone, and ultra high molecular mass polyethylene which is used as a bearing surface in hip prostheses [6]. Biodegradable polymers such as poly-lactic acid and poly-caprolactone are used as porous scaffolds for bone tissue engineering. While these polymers' tunable degradation rates and ease of functionalization are advantageous for directing cell adhesion and differentiation, their low strength and stiffness limits their use in load-bearing applications [11]. The goal of this paper is to review how material parameters affect functions of bone cells.

The success of implanted biomaterials is based on their effects on the myriad of different bone cells. Primarily four cell types reside in bone: osteoblasts, osteocytes, bone lining cells, and osteoclasts, which work together to grow, remodel and maintain bone volume and function. Osteoblasts are formed from stem cells recruited by cytokines from the bone marrow to the bone surface at sites of growth and remodeling. They are tightly connected to neighboring cells, via gap junctions and adherens junctions. Osteoblasts are active bone cells with a myriad of activities, including secretion and assembly of the organic matrix, and directing mineralization of the matrix by secreting promoters such as alkaline phosphate (ALP). In addition to mineralization, osteoblasts secrete paracrine factors, such as bone morphogenenetic proteins (BMPs), which regulate osteoblast and chondrocyte differentiation [12]. Once the organic matrix is mineralized, most osteoblasts undergo apoptosis, but some become entrapped within the new bone and become osteocytes. Osteocytes are differentiated and encased osteoblasts which live in well ordered lacunae, and communicate via dendritic processes which form complex networks within the canaliculi [13]. Osteocytes are mechanoreceptive cells, can sense mechanical strain in bone, and respond to the physical stimuli by paracrine [14], gap junction communication and focal adhesions. Osteocyte death is caused by natural physiological processes, such as aging and menopause, but also due to microcracks, as they form due to failure of plastic deformation and cause mechanical damage to the osteocytes [15]. Osteoblasts which remain on the bone surface become bone lining cells and become inactive [16]. These cells act as a barrier between bone marrow and bone and digest collagen protruding from Howship's lacunae [17].

Mesenchymal stem cells (MSCs) are a heterogenous population of stem/progenitor cells capable of differentiating into bone, cartilage, and adipose. Originally identified in bone marrow as bone marrow stromal cells (BMSCs), these cells can also be found in periosteum, the perivascular niche, and other tissues; although the extent to which these populations converge/diverge from each other as well as their classification as MSCs is still under debate [18–20]. Additionally, Hematopoietic stem cells (HSCs) regulate osteoblast lineage and support bone marrow vasculature, while circulating cells, which circulate throughout the body and arrive to bone via the vasculature, can help regulate mesenchymal and osteoblastic cells in coordinating hematopoiesis [21]. The main function of osteoclasts is bone resorption, which is vital for bone maintenance, the remodeling stage of bone healing and tooth eruption. These cells are enzymatically active and produce tartrate-resistant acid phosphatase (TRAP), calcitonin receptor (CTR) and matrix metalloproteinases (MMPs), which degrade the matrix. To resorb local areas of bone, osteoclasts form a resorption lacuna within which they release protons to acidify and dissolve mineral in a sealed area and proteases to degrade the bone matrix [22]. Osteoclasts can be activated and recruited to resorb multiple times, before undergoing apoptosis [23].

Bone marrow is soft tissue within the medullary cavities and is an important tissue not only for the skeletal system, as it is a source of MSCs [24], but also for the cardiovascular and immune systems. Hematopoietic stem cells (HSCs), which are located in the bone marrow, are precursors for erythrocytes and immune cells including lymphocytes, eosinophils, neutrophils, T and B cells, as well as osteoblastic cells [25]. Balance between the action of all these cell types is necessary to maintain bone volume and structure, endocrine activity and overall function, and prevent bone diseases such as osteoporosis and osteosclerosis.

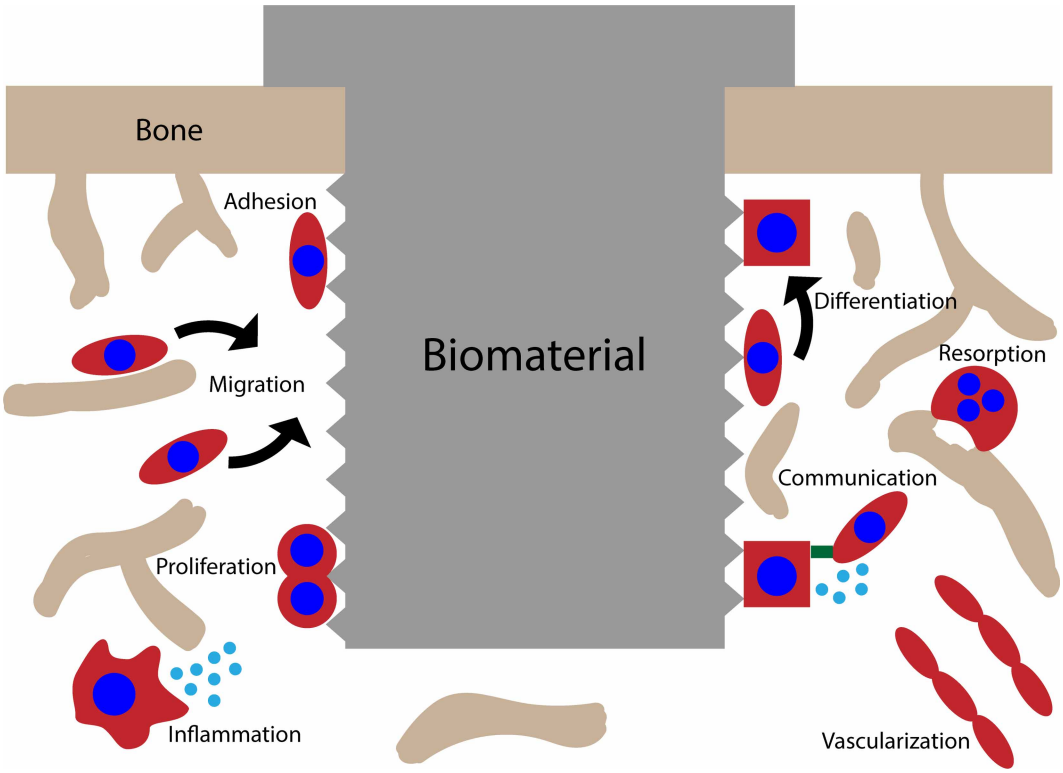

Figure 1. Visual representation of different cell functions affected by biomaterials in bone.
Upon implantation of a biomaterial in bone, an inflammatory response is initiated that stimulates the migration and subsequent adhesion of host cells to the material surface. Osteoprogenitors will then proliferate and later differentiate into osteoblasts to secrete new bone. Eventually new vasculature will form and osteoclasts begin to remodel/resorb the surrounding bone. Cellular communication will occur throughout this process contributing to multiple cell functions.

This paper will review how different material parameters such as stiffness, roughness, surface chemistry, topography, porosity, and protein adsorption affect bone cell functions including inflammation, adhesion, migration, proliferation, communication, differentiation, vascularization, and resorption. The paper will address each of the cell functions individually and present the material parameters most investigated for its effects on the cell function. See Figure 1 for a visual depiction of the cell functions discussed.

Inflammation

Inflammation is critical for proper healing of bone injuries and the implantation of foreign materials typically initiates an inflammatory response. Materials are classified by their biocompatibility into biotolerant, biocompatible, and bioactive categories. Biotolerant materials have low toxicity, but often initiate formation of a fibrous capsule or foreign body giant cell reaction [26]. While biocompatible materials were initially believed to trigger minimal or no inflammatory response, inflammation is critical for proper integration of implants.

Upon exposure to blood *in vivo*, plasma proteins and platelets are quickly absorbed onto the biomaterial surface and activated. Activated platelets initiate the clotting cascade, fibrin forms a provisional matrix, and thrombosis is achieved. Chemokines such as PDGF recruit leukocytes such as neutrophils (initially) and monocytes/macrophages (later). Mast cells also release histamine to mediate the immune response. After the acute inflammation phase, the chronic inflammation phase is characterized by the presence of lymphocytes and plasma cells, but this typically resolves quickly for biocompatible materials. Macrophages remodel the provisional matrix and fibroblasts and endothelial cells are also recruited to form granulation tissue around the implant [26].

In osseointegration, both M1 (pro-inflammatory) and M2 (anti-inflammatory) macrophages are recruited to the peri-implant tissue [27]. Polarization of macrophages towards the M2 phenotype is critical for implant

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integration and maintenance [28,29]. M2 macrophages secrete BMP-2 to recruit osteoblasts to form new bone around the implant surface [30].

Surface modifications can modulate the inflammatory response of biomaterials. Coating titanium implants with bioglass increases macrophage adhesion while reducing secretion of pro-inflammatory cytokines [31]. Other surface treatments can modulate the inflammatory response. For example, surface hydrophobicity down-regulates pro-inflammatory cytokines such as TNFa and IL1a [32]. Although many materials provoke only minimal inflammation in bulk form, biocompatible materials can become inflammatory when in micro/nano particle form. Such particles can be released as prosthetic debris into the surrounding tissue during wear, contributing to inflammation, osteolysis, and implant failure [33]. Macrophages will attempt to phagocytize these particles and secrete pro-inflammatory cytokines including TNF-a, IL-1a, IL-1B, IL-6, IL-8, IL-11, IL-15, TGF-a, GM-CSF, M-CSF, and PDGF. Osteoclast formation is subsequently stimulated through the RANKL pathway, contributing to peri-implant bone resorption and osteolysis [34].

Adhesion

One of the first steps for biomaterial-cell interactions is cellular adhesion. Cellular adhesion is mediated by integrins, heterodimeric membrane proteins that facilitate attachment of cells to the extracellular matrix as well as biomaterials. It is important to recognize that cells do not attach to 'naked' materials. Material surfaces are conditioned by the surrounding fluid/serum. Upon immersion in biological fluids, the material surface is quickly saturated with proteins. Surface energy influences protein adsorption [35], but the relationship is not simple, as other material parameters including hydrophilicity, stiffness, charge and topography also direct protein adsorption. Serum proteins such as vitronectin and fibronectin are important for osteoblast adhesion to materials [36]. These proteins contain adhesive peptide sequences such as RGD that form attachments with integrins [37]. Fibronectin supports the survival of attached osteoblasts [38] and vitronectin is critical for cell attachment and spreading *in* [39]. Osteopontin is another matrix protein present at the bone-biomaterial interface. Osteopontin quickly accumulates at the tissue-implant interface to form a cement line. This osteopontin rich layer helps support cell adhesion, regulates mineralization, and may play a vital role in anchoring the implant to the surrounding tissue. Materials found to accumulate an osteopontin coating include HA, titanium, and cell culture dishes [40].

Physical properties of the bulk material and surface also influence cellular adhesion. For example, fibroblasts spread more, form stress fibers, and increase integrin expression when on stiffer substrates [41]. Surface roughness and topography can have effects on the magnitude of cell adhesion, which is often exploited when designing dental/orthopedic implants to enhance osseointegration. For example, sand blasting, acid etching, and anodization enhances cell adhesion and osseointegration of titanium implants [42], and greater numbers of osteoblasts attached to grooved titanium than rough titanium because of patterning and alignment [43]. The influence of topography on cell adhesion extends to the nanoscale [44] and nanoporous titanium promotes maturation of focal adhesion and filopodia in osteogenic cells compared with polished titanium [45]. Cell adhesion, however, can be reduced on rough titanium surfaces in some situations. Human osteoblast-like MG63 cells have lower attachment to grit-blasted titanium (roughness 2.0–3.3 μm) versus machined titanium (0.2 μm roughness) [46]. The seemingly contradictory information regarding the effect of roughness on cell attachment illustrates the complex relationship between roughness and surface topography. Definitions of 'rough' surfaces vary from study to study and many studies do not characterize surface topography, or do so only with qualitative techniques [47].

Surface chemistry of materials also directs cell adhesion. Cells typically have greater adhesion on hydrophilic surfaces [39]. Contamination of titanium surfaces with hydrocarbons has a negative effect on adhesion, whereas UV photofunctionalization enhances osteoblast adhesion by catalyzing the oxidation of hydrocarbon contaminants, increasing hydrophilicity and surface energy [48]. There is much interest in engineering material surfaces with specific ligands to influence cell attachment. One of the most common ligands is the RGD peptide sequence, which is found in extracellular matrix proteins such as collagen, fibronectin, and vitronectin. RGD can bind to integrins such as α5β1 and αVβ3 [37]. Cyclic RGD stimulates osteoblast adhesion [49] and RGD helps osteoblasts attach to chitosan [50]. Since RGD binds integrins, it can have conflicting interactions with serum proteins, resulting in negative effects on cell adhesion. For example, RGD coating can enhance MSC attachment to HA depending on density, but can also interfere due to competing interactions with serum proteins [51,52]. *In vivo* studies on the effect of RGD coatings on osseointegration have had conflicting results,

with RGD coating on hydroxyapatite inhibiting osseointegration [53] but stimulating bone formation when conjugated to titanium [54]. Other peptides have been used as well, including the P-15 collagen peptide (GTPGPQGIAGQRGW) that increases cell attachment to bovine anorganic bone mineral [55]. Dual-peptides containing material-binding domains and cell-binding domains have proven useful for creating adhesive interfaces between cells and materials. The Glu7-Pro-Arg-Gly-Asp-Thr peptide containing the mineral binding polyglutamate sequence as well as RGD facilitates attachment of osteoblasts to HA [56]. Phage display is a powerful tool that facilitates the discovery of novel sequences with specificity to certain cells or materials. This technique resulted in the discovery of the MSC-binding DPIYALSWSGMA (DPI) sequence and biomimetic mineral binding VTKHLNQISQSY (VTK) sequence. Combining these sequences into dual-peptide DPI-VTK increases the magnitude and specificity of MSC attachment to mineralized biomaterials [57]. *In vivo*, DPI-VTK functionalized scaffolds seeded with human MSCs resulted in greater volume of regenerated bone and vasculature [58].

Migration

Like adhesion, integrins play a critical role in cell migration. Cell migration requires that adhesions are dismantled on the back end of the cell while new ones are created on the leading edge. In order for this to occur, adhesion must be strong enough to facilitate attachment while weak enough to allow detachment. Therefore, cell migration is greatest at intermediate ligand concentration/attachment strength [59]. Cells migrate at different rates on different materials. For example, bone marrow stromal cells have reduced motility on mineralized vs non-mineralized PLGA [60]. For directional migration to occur, cells must be able to sense a gradient. This can occur through two main modes, haptotaxis with surface-bound gradients of ligands, or chemotaxis with soluble gradients. Haptotaxis can be influenced by the hydrophilicity of a substrate, with bone cells migrating towards hydrophilic substrates due to increased vitronectin adsorption (Dalton 1998). Functionalization of scaffolds with ligands can increase migration *in vivo*. For example, α2β1 integrin specific peptides coupled to a hydrogel enhances migration of osteoprogenitors [61]. Growth factors adsorbed or encapsulated within materials can be released in a controlled manner, creating a soluble gradient to induce chemotaxis of osteogenic cells. For example, SDF-1 induces migration of MSCs when released from scaffolds [62], and calcium, released by certain ceramic materials, induces MSC migration [63] by increasing osteopontin expression [18].

Proliferation

Cell proliferation is necessary to replace cells lost from apoptosis during both bone homeostasis and regeneration. Of particular importance during bone healing is the proliferation of MSCs, which will subsequently differentiate into osteoblasts to form new bone [64]. Different material parameters can stimulate or inhibit this process. Chemical cues, such as extracellular calcium released from calcium containing bioceramics can stimulate proliferation of MSCs [18]. Integrins such as α5β1 regulate cell proliferation in osteoblasts. Blocking the α5 subunit in osteoblasts resulted in reduced proliferation [65]. In contrast, silencing integrin α2β1 in osteoblast-like cells increased proliferation. Proliferation and differentiation are typically mutually exclusive processes, as evidenced by the pro-differentiation and anti-proliferation effects of α2β1 [66]. Micropatterning of surfaces can stimulate cell proliferation by directing mechanical strain along the axis of cell elongation [67]. Microroughness, in contrast, can reduce proliferation and induce differentiation of MSCs, and this general trend holds for many cell types [36,66]. Controlling protein adsorption can impact cell proliferation, with fibronectin stimulating proliferation [68]. Functionalizing surfaces with adhesive ligands such as RGD, rather than the whole protein, can also increase proliferation [50].

Communication

The complex and dynamic functions of bone tissue require cell coordination on a spatial and temporal level, which is regulated by cell communication. Gap junction communication between osteocytes coordinate mechanotransduction, mineral deposition and paracrine communication via BMPs, VEGF and RANKL. Paracrine communication via BMP-2 and osteocalcin coordinate osteogenic differentiation of stem cells [69], osteoclastogenesis, healing and other cell activities for bone. The most important function of gap junction communication in bone is mechanotransduction, allowing other functions of bone, such as remodeling, to occur. Mechanical properties of the material, as well as the forces around the material affect gap junction communication. Permeability of bone, which allows for mechanotransduction, is a function of bone porosity and viscosity

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of the fluids within the pores. Mechanotransduction is mediated through gap junction communication and occurs by shear flow of the fluid in the pores caused by mechanical forces. Fluid shear directly triggers perturbation of α5β1 integrins [70], but also cadherins and caveolae [71] to activate the ERK1/2 pathway and open gap junction hemichannels [72]. The reaction to mechanical stimulus is more complex than the proposed Mechanostat Theory, which indicated that at high mechanical stimuli bone mass is increased, at lower stimuli is decreased and an intermediate stimulus is maintained. Within the range of physiological strains, there is overlap in resorption and formation rates, which indicates that load is not the only factor controlling bone remodeling.

Although mediated through gap junction communication, mechanotransduction, through gap junction communication, can promote paracrine communication as a result. Mechanotransduction allows cells to create a biochemical signal within the cell from a mechanical signal from the surrounding matrix. The direct mechanical material-cell interaction prompts osteocytes to produce paracrine signals such as BMPs, Wnts, PGE2 and NO, which influence cell behavior such as recruitment, differentiation and activity of osteoblasts and osteoclasts [73–76]. The effect of gap junction communication on paracrine communication and other cell functions, such as differentiation, can be manipulated by micropatterning surfaces. Engineering patterns allows for contact between multiple cells and encourages gap junction communication increased differentiation [77]. Many studies do not delve beyond observing the effects certain biomaterials make on cell behavior. Understanding the underlying mechanisms may lead to better understanding the role of gap junction communication in cell behavior.

Cell communication is also modulated by surface chemistry. For example, cell attachment to calcium silicate induces cross-talk between osteocytes and endothelial cells, inducing paracrine communication of VEGF [78]. Col-I attachment can modulate $[Ca^{2+}]$- and cAMP-signaling pathways in osteoblasts [79]. Material characteristics, such as aligned pattern and surface chemistry, can be used in tandem to affect cell behavior. For example, Xu et al. [80] used aligned ECM and bioglass to increase Cx43 expression in MSCs, which has an important role in MSC differentiation. Biomaterials containing silica have been used for bone regeneration as the presence of silica increases Cx43 mediated gap junction communication [81,82], proliferation and differentiation in MSCs [83], while decreasing osteoclastogenesis [84]. Both paracrine and gap junction communication can modulate other cell functions, thus it is important to assess the effect of the material on cell communication.

Differentiation

Mesenchymal stem cells can be sourced from multiple locations, including the bone marrow, and are multipotent stem cells, which differentiate into osteoblasts, adipose cells, cartilage, neural or muscle cells [85].

MSC differentiation can be directed by physical cues such as material stiffness and surface geometry, as MSCs are mechanotransductive and convert mechanical signals from the substrate surface into biochemical signals. Mechanical parameters such as fluid shear stress and substrate strain also provide cues for MSC differentiation, also through means of mechanotransduction [86]. The mechanical to chemical signal transduction can be observed by focal adhesion formation and the YAP gene program [87]. More specifically, α2β1 integrin binding [66] promotes osteogenic differentiation.

The mechanical property of surface stiffness particularly, affects MSC cell fate. MSCs on softer substrates, with elasticity mimicking brain elasticity ($E \sim 0.1–1$ kPa) have neuron-like morphology past 7 days, while stiffer matrices (25–40 kPa) results in polygonal MSC morphology, resembling osteoblasts [88–90]. Micropatterning regulates cell shape, which determines fate and commitment of MSCs. [91,92] Convex geometries, such as pentagons, lead to an adipocyte lineage, while concave geometries, such as star shapes, lead to an osteogenic lineage [93]. Smaller micropatterns (1024 μm^2 islands) lead MSCs into chondrocyte lineage, while large micropatterns (10 000 μm^2 islands) drive MSCs into myocytes [94].

Vascularization

Vascularization is necessary for bone survival, but also healing and endocrine signaling. Angiogenesis, formation of blood vessels from existing vessels, is tightly bound with osteogenesis and bone remodeling, and the collagen that is deposited by osteoblasts onto the surface of the new blood vessels is also a template for mineral deposition [95]. During bone development and healing, the processes of angiogenesis and osteogenesis are coupled, and together are regulated by vascular endothelial growth factor (VEGF) and BMP-2. Type H vessels,

which are in the vicinity of bone growth plates direct bone growth by gap junction communication as well as paracrine communication, stimulating progenitor cells to proliferate and differentiate [95].

Multiple material parameters affect quality and quantity of newly formed vessels. For bone regeneration, both pore size and interconnection are important parameters. Interconnected pores between 100–150 μm are beneficial for vessel development [96]. β-TCP scaffolds made with larger pores implanted without cells lead to the formation of larger blood vessels, but scaffolds with larger interconnections result in both larger vessels and more vessels, with effect plateauing at pore size of 400 μm [86].

Increased roughness and surface energy of both metal [97,98] and polymeric surfaces [99,100] improves cell adhesion by increasing adhesion density and cell aspect ratio, even on a nanometer scale. To undergo angiogenesis, endothelial cells need to be adherent and motile, therefore increasing endothelial cell adhesion density is critical to induce angiogenesis and maintain the neovasculature. Surface stiffness also affects endothelial cell morphology, which affects angiogenic potential. Endothelial cells develop a spread morphology and have more actin fibers when seeded on stiffer surfaces. ($E > 2$ kPa) [41,101].

Resorption

Bone remodeling is a dynamic process of replacing old bone matrix with new matrix and maintaining bone volume. Bone resorption is an essential process for bone to adapt to physiological changes throughout life [102]. Resorption is a complex process including migration of osteoclasts to a focal site, followed by attachment and polarization, then dissolution of hydroxyapatite (HA) and degradation of the organic matrix. Once these steps are complete, the osteoclasts undergo apoptosis.

Resorption by osteoclasts can be modulated by altering surface parameters such as material surface chemistry and surface roughness. Resorption of calcium-phosphate ceramics is dependent on Ca/P ratio and crystallinity. For example, osteoclasts resorb more biphasic calcium phosphate when HA/β-tri calcium phosphate (β-TCP) ratio is 25/75 versus 75/25. [8] Strontium substituted calcium phosphate inhibits osteoclast resorption by delaying osteoclast differentiation [103,104]. Carbonated apatite has increased resorption [105]. The nature of organic materials also impacts resorption, as fibrinogen modified chitosan has greater osteoclastic resorption than on unmodified chitosan [106]. Besides surface chemistry, surface roughness affects osteoclast resorption. When on rough biomimetic hydroxyapatite, osteoclasts have reduced resorption [107], but on rough titanium, there is no change in MMP expression or morphology [28,108]. This is due to surface wettability and surface energy [109,110].

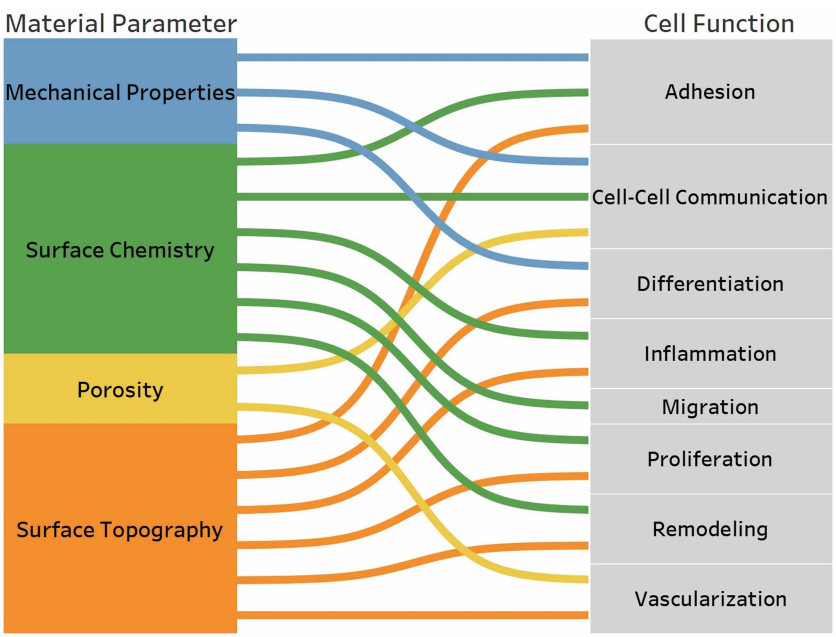

Figure 2. Relationship between most influential material parameters and impacted cell functions.

The diagram highlights the connections between cell functions and material parameters discussed in the review paper.

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Conclusion

Cell functions can be stimulated or inhibited by choice of material, which guides surface chemistry, protein adsorption and material stiffness, and material surface and bulk modification, such as surface roughness, topography, porosity and strain. Additionally, modifications of many of these material properties can promote one cell function over another. Many cell functions can be modulated through modifying integrin binding and cell adhesion, as osteocytes and MSCs are mechanotransductive cells and sense surface stiffness, roughness and shear forces through focal adhesions which have cascade effects throughout the cell and tissue. An overview of the relationships between material parameters and cell functions can be seen in Figure 2.

Better understanding of the effect of how physical parameters drive bone cell function has allowed for improved therapy and implant design. As bone is a dynamic organ, which is interconnected with the immune system and vascular system, future material designs need to better account for not only osteogenic potential, but also vascular and immune potential. Additionally, most of the studies cited assessed the effect of one parameter on one cell action, which although informative, may be giving a limited image of the potential of manipulating a particular variable on effective tissue engineering strategies. The future of material design would encompass various cell functions and modulating material parameters to ensure not only one, but multiple cell functions are promoted in order to achieve optimal effects.

Perspectives

- Highlight the importance of the field: Both the choice of material and its design specifications have a significant effect on many cell functions, and it is vital to material success to understand their effect on cell functions.
- A summary of the current thinking: Material parameters have a multitude of effects on cells and can be used to promote cell functions. Many cell functions are modulated through integrin binding of cells to material, thus modifications of cell attachment are important to consider in material design.
- A comment on future directions: Current research focuses on modification of one or two properties. Understanding complexities of bone function, such as healing, differentiation and remodeling, enables materials to be designed that direct multiple cell functions.

Competing Interests

The authors declare that there are no competing interests associated with the manuscript.

Author Contributions

E.M. and M.M. equally contributed to planning and writing the manuscript, with oversight, guidance, editing and funding from D.K.

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Abbreviations

ALP, alkaline phosphate; BMPs, bone morphogenenetic proteins; HA, hydroxyapatite; HSCs, hematopoietic stem cells; MSCs, mesenchymal stem cells; TCP, tri calcium phosphate; VEGF, vascular endothelial growth factor.

References

1 Jepsen, K.J., Goldstein, S.A., Kuhn, J.L., Schaffler, M.B. and Bonadio, J. (1996) Type-I collagen mutation compromises the post-yield behavior of Mov13 long bone. *J. Orthop. Res.* **14**, 493–499 https://doi.org/10.1002/jor.1100140320

2 Rubin, M.A., Jasiuk, I., Taylor, J., Rubin, J., Ganey, T. and Apkarian, R.P. (2003) TEM analysis of the nanostructure of normal and osteoporotic human trabecular bone. *Bone* **33**, 270–282 https://doi.org/10.1016/S8756-3282(03)00194-7

3 Reznikov, N., Bilton, M., Lari, L., Stevens, M.M. and Kröger, R. (2018) Fractal-like hierarchical organization of bone begins at the nanoscale. *Science* **360**, eaao2189 https://doi.org/10.1126/science.aao2189
4 Jäger, I. and Fratzl, P. (2000) Mineralized collagen fibrils: a mechanical model with a staggered arrangement of mineral particles. *Biophys. J.* **79**, 1737–1746 https://doi.org/10.1016/S0006-3495(00)76426-5
5 Fonseca, H., Moreira-Gonçalves, D., Coriolano, H.J.A. and Duarte, J.A. (2014) Bone quality: the determinants of bone strength and fragility. *Sport Med.* **44**, 37–53 https://doi.org/10.1007/s40279-013-0100-7
6 Hu, C.Y. and Yoon, T.R. (2018) Recent updates for biomaterials used in total hip arthroplasty. *Biomater. Res.* **22**, 1–12 https://doi.org/10.1186/s40824-017-0112-8
7 Bosshardt, D.D., Chappuis, V. and Buser, D. (2017) Osseointegration of titanium, titanium alloy and zirconia dental implants: current knowledge and open questions. *Periodontol 2000* **73**, 22–40 https://doi.org/10.1111/prd.12179
8 Steffi, C., Shi, Z., Kong, C.H. and Wang, W. (2018) Modulation of osteoclast interactions with orthopaedic biomaterials. *J. Funct. Biomater.* **9**, 18 https://doi.org/10.3390/jfb9010018
9 Kohn, D.H. and Ducheyne, P. (1992) Materials for Bone, Joint and Cartilage Replacement. In *Medical and Dental Materials* (Williams, D.F., ed.), pp. 29–109, VCH
10 Ramaswamy, J., Ramaraju, H. and Kohn, D.H. (2011) Bone-like mineral and organically modified bone-like mineral coatings. *Biol. Biomed. Coatings Handb. Process Charact.* 1–27 https://doi.org/10.1201/b10871
11 Gentile, P., Chiono, V., Carmagnola, I. and Hatton, P.V. (2014) An overview of poly (lactic-co-glycolic) acid (PLGA)-based biomaterials for bone tissue engineering. *Int. J. Mol. Sci.* **15**, 3640–3659 https://doi.org/10.3390/ijms15033640
12 Urist, M. and Strates, B. (1971) Bone morphogenic protein. *J. Dent. Res.* **50**, 1392–1406 https://doi.org/10.1177/00220345710500060601
13 Aarden, E.M., Nijweide, P.J. and Burger, E.H. (1994) Function of osteocytes in bone. *J. Cell Biochem.* **55**, 287–299 https://doi.org/10.1002/jcb.240550304
14 Lean, J.M., Mackay, A.G., Chow, J.W. and Chambers, T.J. (1995) Osteocytic expression of mRNA for c-fos and IGF-I: an immediate early gene response to an osteogenic stimulus. *Am. J. Physiol. Endocriniol. Metab.* **270**, E937–E945 https://doi.org/10.1152/ajpendo.1996.270.6.E937
15 Noble, B.S. and Reeve, J. (2000) Osteocyte function, osteocyte death and bone fracture resistance. *Mol. Cell Endocrinol.* **159**, 7–13 https://doi.org/10.1016/S0303-7207(99)00174-4
16 Miller, S., de Saint-Georges, L. and Bowman B, W.J. (1989) Bone lining cells: structure and function. *Scanning Microsc.* **3**, 953–960 PMID:2694361
17 Everts, V., Delaissié, J.M., Korper, W., Jansen, D.C., Tigchelaar-Gutter, W., Saftig, P. et al. (2002) The bone lining cell: its role in cleaning howship's lacunae and initiating bone formation. *J. Bone Miner. Res.* **17**, 77–90 https://doi.org/10.1359/jbmr.2002.17.1.77
18 Lee, M.N., Hwang, H.S., Oh, S.H., Roshanzadeh, A., Kim, J.W., Song, J.H. et al. (2018) Elevated extracellular calcium ions promote proliferation and migration of mesenchymal stem cells via increasing osteopontin expression. *Exp. Mol. Med.* **50**, 1–16 https://doi.org/10.1038/s12276-018-0170-6
19 Debnath, S., Yallowitz, A.R., McCormick, J., Lalani, S., Zhang, T., Xu, R. et al. (2018) Discovery of a periosteal stem cell mediating intramembranous bone formation. *Nature* **562**, 133–139 https://doi.org/10.1038/s41586-018-0554-8
20 Kfoury, Y. and Scadden, D.T. (2015) Mesenchymal cell contributions to the stem cell niche. *Cell Stem Cell* **16**, 239–253 https://doi.org/10.1016/j.stem.2015.02.019
21 Bunney, P.E., Zink, A.N., Holm, A.A., Billington, C.J. and Kotz, C.M. (2017) Orexin activation counteracts decreases in nonexercise activity thermogenesis (NEAT) caused by high-fat diet. *Physiol Behav.* **176**, 139–148 https://doi.org/10.1016/j.physbeh.2017.03.040
22 Parvizi, J. and Kim, G. (2010) *High Yield Orthopaedics*, Elsevier
23 Kanehisa, J. and Heersche, J.N.M. (1988) Osteoclastic bone resorption: in vitro analysis of the rate of resorption and migration of individual osteoclasts. *Bone* **9**, 73–79 https://doi.org/10.1016/8756-3282(88)90106-8
24 Wexler, S.A., Donaldson, C., Denning-Kendall, P., Rice, C., Bradley, B. and Hows, J.M. (2003) Adult bone marrow is a rich source of human mesenchymal 'stem' cells but umbilical cord and mobilized adult blood are not. *Br. J. Haematol.* **121**, 368–374 https://doi.org/10.1046/j.1365-2141.2003.04284.x
25 Zhang, J., Niu, C., Ye, L., Huang, H., He, X., Tong, W. et al. (2003) Identification of the haematopoietic stem cell niche and control of the niche size. *Nature* **425**, 836–841 https://doi.org/10.1038/nature02041
26 Anderson, J.M., Rodriguez, A. and Chang, D.T. (2008) Foreign body reaction to biomaterials. *Semin. Immunol.* **20**, 86–100 https://doi.org/10.1016/j.smim.2007.11.004
27 Trindade, R., Albrektsson, T., Galli, S., Prgomet, Z., Tengvall, P. and Wennerberg, A. (2018) Osseointegration and foreign body reaction: titanium implants activate the immune system and suppress bone resorption during the first 4 weeks after implantation. *Clin. Implant. Dent. Relat. Res.* **20**, 82–91 https://doi.org/10.1111/cid.12578
28 Insua, A., Monje, A., Wang, H.L. and Miron, R.J. (2017) Basis of bone metabolism around dental implants during osseointegration and peri-implant bone loss. *J. Biomed. Mater. Res. A.* **105**, 2075–2089 https://doi.org/10.1002/jbm.a.36060
29 Sridharan, R., Cameron, A.R., Kelly, D.J., Kearney, C.J. and O'Brien, F.J. (2015) Biomaterial based modulation of macrophage polarization: a review and suggested design principles. *Mater Today* **18**, 313–325 https://doi.org/10.1016/j.mattod.2015.01.019
30 Jennissen, H.P. (2016) A macrophage model of osseointegration. *Curr. Dir. Biomed. Eng.* **2**, 53–56 https://doi.org/10.1515/cdbme-2016-0015
31 Scislowska-Czarnecka, A., Menaszek, E., Szaraniec, B. and Kolaczkowska, E. (2012) Ceramic modifications of porous titanium: effects on macrophage activation. *Tissue Cell* **44**, 391–400 https://doi.org/10.1016/j.tice.2012.08.002
32 Hamlet, S., Alfarsi, M., George, R. and Ivanovski, S. (2012) The effect of hydrophilic titanium surface modification on macrophage inflammatory cytokine gene expression. *Clin. Oral Implants Res.* **23**, 584–590 https://doi.org/10.1111/j.1600-0501.2011.02325.x
33 Bitar, D. and Parvizi, J. (2015) Biological response to prosthetic debris. *World J Orthop.* **6**, 172–189 https://doi.org/10.5312/wjo.v6.i2.172
34 Kzhyshkowska, J., Gudima, A., Riabov, V., Dollinger, C., Lavalle, P. and Vrana, N.E. (2015) Macrophage responses to implants: prospects for personalized medicine. *J. Leukoc. Biol.* **98**, 953–962 https://doi.org/10.1189/jlb.5VMR0415-166R
35 Boyan, B.D., Hummert, T.W., Dean, D.D. and Schwartz, Z. (1996) Role of material surfaces in regulating bone and cartilage cell response. *Biomaterials* **17**, 137–146 https://doi.org/10.1016/0142-9612(96)85758-9

36 Wilson, C.J., Clegg, R.E., Leavesley, D.I. and Pearcy, M.J. (2005) Mediation of biomaterial-cell interactions by adsorbed proteins: a review. *Tissue Eng.* **11**, 1–18 https://doi.org/10.1089/ten.2005.11.1
37 Ruoslahti, E. (2003) The RGD story: a personal account. *Matrix Biol.* **22**, 1–7 https://doi.org/10.1016/S0945-053X(03)00083-0
38 Globus, R.K., Doty, S.B., Lull, J.C., Holmuhamedov, E., Humphries, M.J. and Damsky, C.H. (1998) Fibronectin is a survival factor for differentiated osteoblasts. *J. Cell Sci.* **111**, 1385–1393 PMID:9570756
39 Anselme, K. (2000) Osteoblast adhesion on biomaterials. *Biomaterials* **21**, 667–681 https://doi.org/10.1016/S0142-9612(99)00242-2
40 McKee, M.D. and Nanci, A. (1996) Osteopontin at mineralized tissue interfaces in bone, teeth, and osseointegrated implants: ultrastructural distribution and implications for mineralized tissue formation, turnover, and repair. *Microsc. Res. Tech.* **33**, 141–164 https://doi.org/10.1002/(SICI)1097-0029(19960201)33:2<141::AID-JEMT5>3.0.CO;2-W
41 Yeung, T., Georges, P.C., Flanagan, L.A., Marg, B., Ortiz, M., Funaki, M. et al. (2005) Effects of substrate stiffness on cell morphology, cytoskeletal structure, and adhesion. *Cell Motil. Cytoskeleton* **60**, 24–34 https://doi.org/10.1002/cm.20041
42 Kim, M.H., Park, K., Choi, K.H., Kim, S.H., Kim, S.E., Jeong, C.M. et al. (2015) Cell adhesion and in vivo osseointegration of sandblasted/acid etched/anodized dental implants. *Int. J. Mol. Sci.* **16**, 10324–10336 https://doi.org/10.3390/ijms160510324
43 Jayaraman, M., Meyer, U., Bühner, M., Joos, U. and Wiesmann, H.P. (2004) Influence of titanium surfaces on attachment of osteoblast-like cells in vitro. *Biomaterials* **25**, 625–631 https://doi.org/10.1016/S0142-9612(03)00571-4
44 Mendonça, G., Mendonça, D.B.S., Aragão, F.J.L. and Cooper, L.F. (2008) Advancing dental implant surface technology: from micron- to nanotopography. *Biomaterials* **29**, 3822–3835 https://doi.org/10.1016/j.biomaterials.2008.05.012
45 Guadarrama Bello, D., Fouillen, A., Badia, A. and Nanci, A. (2017) A nanoporous titanium surface promotes the maturation of focal adhesions and formation of filopodia with distinctive nanoscale protrusions by osteogenic cells. *Acta Biomater.* **60**, 339–349 https://doi.org/10.1016/j.actbio.2017.07.022
46 Schwartz, Z., Raz, P., Zhao, G., Barak, Y., Tauber, M., Yao, H. et al. (2008) Effect of micrometer-scale roughness of the surface of Ti6Al4V pedicle screws in vitro and in vivo. *J. Bone Joint Surg. Am.* **90**, 2485–2498 https://doi.org/10.2106/JBJS.G.00499
47 Wennerberg, A. and Albrektsson, T. (2009) Effects of titanium surface topography on bone integration: a systematic review. *Clin. Oral Implants Res.* **20**, 172–184 https://doi.org/10.1111/j.1600-0501.2009.01775.x
48 Yamada, M., Miyauchi, T., Yamamoto, A., Iwasa, F., Takeuchi, M., Anpo, M. et al. (2010) Enhancement of adhesion strength and cellular stiffness of osteoblasts on mirror-polished titanium surface by UV-photofunctionalization. *Acta Biomater.* **6**, 4578–4588 https://doi.org/10.1016/j.actbio.2010.07.010
49 Kantlehner, M., Schaffner, P., Finsinger, D., Meyer, J., Jonczyk, A., Diefenbach, B. et al. (2000) Surface coating with cyclic RGD peptides stimulates osteoblast adhesion and proliferation as well as bone formation. *Chembiochem* **1**, 107–114 https://doi.org/10.1002/1439-7633(20000818)1:2<107::AID-CBIC107>3.0.CO;2-4
50 Tsai, W.B., Chen, Y.R., Liu, H.L. and Lai, J.Y. (2011) Fabrication of UV-crosslinked chitosan scaffolds with conjugation of RGD peptides for bone tissue engineering. *Carbohydr. Polym.* **85**, 129–137 https://doi.org/10.1016/j.carbpol.2011.02.003
51 Sawyer, A.A., Hennessy, K.M. and Bellis, S.L. (2005) Regulation of mesenchymal stem cell attachment and spreading on hydroxyapatite by RGD peptides and adsorbed serum proteins. *Biomaterials*. **26**, 1467–1475 https://doi.org/10.1016/j.biomaterials.2004.05.008
52 Hennessy, K.M., Pollot, B.E., Clem, W.C., Phipps, M.C., Sawyer, A.A., Culpepper, B.K. et al. (2009) The effect of collagen I mimetic peptides on mesenchymal stem cell adhesion and differentiation, and on bone formation at hydroxyapatite surfaces. *Biomaterials* **30**, 1898–1909 https://doi.org/10.1016/j.biomaterials.2008.12.053
53 Hennessy, K.M., Clem, W.C., Phipps, M.C., Sawyer, A.A., Shaikh, F.M. and Bellis, S.L. (2008) The effect of RGD peptides on osseointegration of hydroxyapatite biomaterials. *Biomaterials* **29**, 3075–3083 https://doi.org/10.1016/j.biomaterials.2008.04.014
54 Ferris, D. (1999) RGD-coated titanium implants stimulate increased bone formation in vivo. *Biomaterials* **20**, 2323–2331 https://doi.org/10.1016/S0142-9612(99)00161-1
55 Qian, J.J. and Bhatnagar, R.S. (1996) Enhanced cell attachment to anorganic bone mineral in the presence of a synthetic peptide related to collagen. *J. Biomed. Mater. Res.* **31**, 545–554 https://doi.org/10.1002/(SICI)1097-4636(199608)31:4<545::AID-JBM15>3.0.CO;2-F
56 Fujisawa, R., Mizuno, M., Nodasaka, Y. and Yoshinori, K. (1997) Attachment of osteoblastic cells to hydroxyapatite crystals by a synthetic peptide (Glu7-Pro-Arg-Gly-Asp-Thr) containing two functional sequences of bone sialoprotein. *Matrix Biol.* **16**, 21–28 https://doi.org/10.1016/S0945-053X(97)90113-X
57 Ramaraju, H., Miller, S.J. and Kohn, D.H. (2017) Dual-functioning peptides discovered by phage display increase the magnitude and specificity of BMSC attachment to mineralized biomaterials. *Biomaterials* **134**, 1–12 https://doi.org/10.1016/j.biomaterials.2017.04.034
58 Ramaraju, H. and Kohn, D.H. (2019) Cell and material-specific phage display peptides increase iPS-MSC mediated bone and vasculature formation in vivo. *Adv. Healthc. Mater.* **8**, 1801356 https://doi.org/10.1002/adhm.201801356
59 DiMilla, P.A., Stone, J.A., Quinn, J.A., Albelda, S.M. and Lauffenburger, D.A. (1993) Maximal migration of human smooth muscle cells on fibronectin and type IV collagen occurs at an intermediate attachment strength. *J. Cell Biol.* **122**, 729–737 https://doi.org/10.1083/jcb.122.3.729
60 Leonova E, V., Pennington, K.E., Krebsbach, P.H. and Kohn, D.H. (2006) Substrate mineralization stimulates focal adhesion contact redistribution and cell motility of bone marrow stromal cells. *J. Biomed. Mater. Res. A* **79**, 263–270 https://doi.org/10.1002/jbm.a.30786
61 Shekaran, A., García, J.R., Clark, A.Y., Kavanaugh, T.E., Lin, A.S., Guldberg, R.E. et al. (2014) Bone regeneration using an alpha 2 beta 1 integrin-specific hydrogel as a BMP-2 delivery vehicle. *Biomaterials* **35**, 5453–5461 https://doi.org/10.1016/j.biomaterials.2014.03.055
62 Chen, G. and Lv, Y. (2017) Matrix elasticity-modified scaffold loaded with SDF-1α improves the in situ regeneration of segmental bone defect in rabbit radius. *Sci. Rep.* **7**, 1–12 https://doi.org/10.1038/s41598-017-01938-3
63 Aquino-martínez, R., Angelo, A.P. and Pujol, F.V. (2017) Calcium-containing scaffolds induce bone regeneration by regulating mesenchymal stem cell differentiation and migration. *Stem Cell Res. Ther.* **8**, 265 https://doi.org/10.1186/s13287-017-0713-0
64 Li, G., White, G., Connolly, C. and Marsh, D. (2002) Cell proliferation and apoptosis during fracture healing. *J. Bone Miner. Res.* **17**, 791–799 https://doi.org/10.1359/jbmr.2002.17.5.791
65 Keselowsky, B.G., Wang, L., Schwartz, Z., Garcia, A.J. and Boyan, B.D. (2007) Integrin alpha(5) controls osteoblastic proliferation and differentiation responses to titanium substrates presenting different roughness characteristics in a roughness independent manner. *J. Biomed. Mater. Res. A* **80**, 700–710 https://doi.org/10.1002/jbm.a.30898
66 Lai, M., Hermann, C.D., Cheng, A., Olivares-Navarrete, R., Gittens, R.A., Bird, M.M. et al. (2015) Role of α2β1 integrins in mediating cell shape on microtextured titanium surfaces. *J. Biomed. Mater. Res. A* **103**, 564–573 https://doi.org/10.1002/jbm.a.35185

67 Théry, M. (2010) Micropatterning as a tool to decipher cell morphogenesis and functions. *J. Cell Sci.* **123**, 4201–4213 https://doi.org/10.1242/jcs.075150
68 Dolatshahi-Pirouz, A., Jensen, T., Kraft, D.C., Foss, M., Kingshott, P., Hansen, J.L. et al. (2010) Fibronectin adsorption, cell adhesion, and proliferation on nanostructured tantalum surfaces. *ACS Nano* **4**, 2874–2882 https://doi.org/10.1021/nn9017872
69 Li, F., Whyte, N. and Niyibizi, C. (2012) Differentiating multipotent mesenchymal stromal cells generate factors that exert paracrine activities on exogenous MSCs: implications for paracrine activities in bone regeneration. *Biochem. Biophys. Res. Commun.* **426**, 475–479 https://doi.org/10.1016/j.bbrc.2012.08.095
70 Batra, N., Burra, S., Siller-Jackson, A.J., Gu, S., Xia, X., Weber, G.F. et al. (2012) Mechanical stress-activated integrin α5β1 induces opening of connexin 43 hemichannels. *Proc. Natl Acad. Sci. U.S.A.* **109**, 3359–3364 https://doi.org/10.1073/pnas.1115967109
71 Nelson, W.J. and Nusse, R. (2004) Convergence of Wnt, β-Catenin, and cadherin pathways. *Science* **303**, 1483–1487 https://doi.org/10.1126/science.1094291
72 Burra, S., Nicolella, D.P., Francis, W.L., Freitas, C.J., Mueschke, N.J., Poole, K. et al. (2010) Dendritic processes of osteocytes are mechanotransducers that induce the opening of hemichannels. *Proc. Natl Acad. Sci. U.S.A.* **107**, 13648–13653 https://doi.org/10.1073/pnas.1009382107
73 Robling, A.G., Castillo, A.B. and Turner, C.H. (2006) Biomechanical and molecular regulation of bone remodeling. *Annu. Rev. Biomed. Eng.* **8**, 455–498 https://doi.org/10.1146/annurev.bioeng.8.061505.095721
74 Tan, S.D., de Vries, T.J., Kuijpers-Jagtman, A.M., Semeins, C.M., Everts, V. and Klein-Nulend, J. (2007) Osteocytes subjected to fluid flow inhibit osteoclast formation and bone resorption. *Bone* **41**, 745–751 https://doi.org/10.1016/j.bone.2007.07.019
75 You, L., Temiyasathit, S., Lee, P., Kim, C.H., Tummala, P., Yao, W. et al. (2008) Osteocytes as mechanosensors in the inhibition of bone resorption due to mechanical loading. *Bone* **42**, 172–179 https://doi.org/10.1016/j.bone.2007.09.047
76 Santos, A., Bakker, A.D. and Klein-Nulend, J. (2009) The role of osteocytes in bone mechanotransduction. *Osteoporos. Int.* **20**, 1027–1031 https://doi.org/10.1007/s00198-009-0858-5
77 Tang, J., Peng, R. and Ding, J. (2010) The regulation of stem cell differentiation by cell-cell contact on micropatterned material surfaces. *Biomaterials* **31**, 2470–2476 https://doi.org/10.1016/j.biomaterials.2009.12.006
78 Li, H., Xue, K., Kong, N., Liu, K. and Chang, J. (2014) Silicate bioceramics enhanced vascularization and osteogenesis through stimulating interactions between endothelia cells and bone marrow stromal cells. *Biomaterials* **35**, 3803–3818 https://doi.org/10.1016/j.biomaterials.2014.01.039
79 Green, J., Schotland, S., Stauber, D.J., Kleeman, C.R. and Clemens, T.L. (1995) Cell-matrix interaction in bone: type I collagen modulates signal transduction in osteoblast-like cells. *Am. J. Physiol. Cell Physiol.* **268**, C1090–C1103 https://doi.org/10.1152/ajpcell.1995.268.5.C1090
80 Xu, Y., Wu, Z., Dong, X. and Li, H. (2017) Combined biomaterial signals stimulate communications between bone marrow stromal cell and endothelial cell. *RSC Adv.* **7**, 5306–5314 https://doi.org/10.1039/C6RA28101J
81 Uribe, P., Johansson, A., Jugdaohsingh, R., Powell, J.J., Magnusson, C., Davila, M. et al. (2020) Soluble silica stimulates osteogenic differentiation and gap junction communication in human dental follicle cells. *Sci. Rep.* **10**, 1–11 https://doi.org/10.1038/s41598-020-66939-1
82 Popara, J., Accomasso, L., Vitale, E., Gallina, C., Roggio, D., Iannuzzi, A. et al. (2018) Silica nanoparticles actively engage with mesenchymal stem cells in improving acute functional cardiac integration. *Nanomedicine* **13**, 1121–1138 https://doi.org/10.2217/nnm-2017-0309
83 Costa-Rodrigues, J., Reis, S., Castro, A. and Fernandes, M.H. (2016) Bone anabolic effects of soluble si: in vitro studies with human mesenchymal stem cells and cd14+ osteoclast precursors. *Stem Cells Int.* **2016**, 5653275 https://doi.org/10.1155/2016/5653275
84 Mladenović, Ž, Johansson, A., Willman, B., Shahabi, K., Björn, E. and Ransjö, M. (2014) Soluble silica inhibits osteoclast formation and bone resorption in vitro. *Acta Biomater.* **10**, 406–418 https://doi.org/10.1016/j.actbio.2013.08.039
85 Moroni, L. and Fornasari, P.M. (2013) Human mesenchymal stem cells: a bank perspective on the isolation, characterization and potential of alternative sources for the regeneration of musculoskeletal tissues. *J. Cell Physiol.* **228**, 680–687 https://doi.org/10.1002/jcp.24223
86 Bai, F., Wang, Z., Lu, J., Liu, J., Chen, G., Lv, R. et al. (2010) The correlation between the internal structure and vascularization of controllable porous bioceramic materials in vivo: a quantitative study. *Tissue Eng. Part A* **16**, 3791–3803 https://doi.org/10.1089/ten.tea.2010.0148
87 Wang, N., Tytell, J.D. and Ingber, D.E. (2009) Mechanotransduction at a distance: mechanically coupling the extracellular matrix with the nucleus. *Nat. Rev. Mol. Cell Biol.* **10**, 75–82 https://doi.org/10.1038/nrm2594
88 García, A.J. and Reyes, C.D. (2005) Critical reviews in oral biology & medicine bio-adhesive surfaces to promote osteoblast differentiation and bone formation. *J. Dent. Res.*, 407–413 https://doi.org/10.1177/154405910508400502
89 Xi, J., Kong, L., Gao, Y., Gong, Y., Zhao, N. and Zhang, X. (2005) Properties of poly(3-hydroxybutyrate-co-3-hydroxyhexanoate) films modified with polyvinylpyrrolidone and behavior of MC3T3-E1 osteoblasts cultured on the blended films. *J. Biomater. Sci. Polym. Ed.* **16**, 1395–1408 https://doi.org/10.1163/156856205774472344
90 Engler, A.J., Sen, S., Sweeney, H.L. and Discher, D.E. (2006) Matrix elasticity directs stem cell lineage specification. *Cell* **126**, 677–689 https://doi.org/10.1016/j.cell.2006.06.044
91 McBeath, R., Pirone, D.M., Nelson, C.M., Bhadriraju, K. and Chen, C.S. (2004) Cell shape, cytoskeletal tension, and rhoA regulate stem cell lineage commitment. *Dev. Cell* **6**, 483–495 https://doi.org/10.1016/S1534-5807(04)00075-9
92 Wang, X., Nakamoto, T., Dulińska-Molak, I., Kawazoe, N. and Chen, G. (2016) Regulating the stemness of mesenchymal stem cells by tuning micropattern features. *J. Mater. Chem. B* **4**, 37–45 https://doi.org/10.1039/C5TB02215K
93 Kilian, K.A., Bugarija, B., Lahn, B.T. and Mrksich, M. (2010) Geometric cues for directing the differentiation of mesenchymal stem cells. *Proc. Natl Acad. Sci. U.S.A.* **107**, 4872–4877 https://doi.org/10.1073/pnas.0903269107
94 Gao, L., McBeath, R. and Chen, C.S. (2010) Stem cell shape regulates a chondrogenic versus myogenic fate through rac1 and N-cadherin. *Stem Cells* **28**, 564–572 https://doi.org/10.1002/stem.308
95 Shoham, A.B., Rot, C., Stern, T., Krief, S., Akiva, A., Dadosh, T. et al. (2016) Deposition of collagen type I onto skeletal endothelium reveals a new role for blood vessels in regulating bone morphology. *Development* **143**, 3933–3943 https://doi.org/10.1242/dev.139253
96 Klenke, F.M., Liu, Y., Yuan, H., Hunziker, E.B., Siebenrock, K.A. and Hofstetter, W. (2008) Impact of pore size on the vascularization and osseointegration of ceramic bone substitutes in vivo. *J. Biomed. Mater. Res. A* **85**, 777–786 https://doi.org/10.1002/jbm.a.31559
97 Raimondo, T., Puckett, S. and Webster, T.J. (2010) Greater osteoblast and endothelial cell adhesion on nanostructured polyethylene and titanium. *Int. J. Nanomed.* **5**, 647–652 https://doi.org/10.2147/IJN.S13047

Biochemical Society Transactions (2020) **48** 2039–2050
https://doi.org/10.1042/BST20200210

98 Khang, D., Lu, J., Yao, C., Haberstroh, K.M. and Webster, T.J. (2008) The role of nanometer and sub-micron surface features on vascular and bone cell adhesion on titanium. *Biomaterials* **29**, 970–983 https://doi.org/10.1016/j.biomaterials.2007.11.009

99 Chung, T.W., Liu, D.Z., Wang, S.Y. and Wang, S.S. (2003) Enhancement of the growth of human endothelial cells by surface roughness at nanometer scale. *Biomaterials* **24**, 4655–4661 https://doi.org/10.1016/S0142-9612(03)00361-2

100 Miller, D.C., Thapa, A., Haberstroh, K.M. and Webster, T.J. (2004) Endothelial and vascular smooth muscle cell function on poly(lactic-co-glycolic acid) with nano-structured surface features. *Biomaterials* **25**, 53–61 https://doi.org/10.1016/S0142-9612(03)00471-X

101 Byfield, F.J., Reen, R.K., Shentu, T.P., Levitan, I. and Gooch, K.J. (2009) Endothelial actin and cell stiffness is modulated by substrate stiffness in 2D and 3D. *J. Biomech.* **42**, 1114–1119 https://doi.org/10.1016/j.jbiomech.2009.02.012

102 Proff, P. and Römer, P. (2009) The molecular mechanism behind bone remodelling: a review. *Clin. Oral Investig.* **13**, 355–362 https://doi.org/10.1007/s00784-009-0268-2

103 Schumacher, M., Wagner, A.S., Kokesch-Himmelreich, J., Bernhardt, A., Rohnke, M., Wenisch, S. et al. (2016) Strontium substitution in apatitic CaP cements effectively attenuates osteoclastic resorption but does not inhibit osteoclastogenesis. *Acta Biomater.* **37**, 184–194 https://doi.org/10.1016/j.actbio.2016.04.016

104 Bonnelye, E., Chabadel, A., Saltel, F. and Jurdic, P. (2008) Dual effect of strontium ranelate: stimulation of osteoblast differentiation and inhibition of osteoclast formation and resorption in vitro. *Bone* **42**, 129–138 https://doi.org/10.1016/j.bone.2007.08.043

105 Spence, G., Patel, N., Brooks, R., Bonfield, W. and Rushton, N. (2010) Osteoclastogenesis on hydroxyapatite ceramics: the effect of carbonate substitution. *J. Biomed. Mater. Res. A* **92**, 1292–1300 https://doi.org/10.1002/jbm.a.32373

106 Torres, A.L., Santos, S.G., Oliveira, M.I. and Barbosa, M.A. (2013) Fibrinogen promotes resorption of chitosan by human osteoclasts. *Acta Biomater.* **9**, 6553–6562 https://doi.org/10.1016/j.actbio.2013.01.015

107 Costa, D.O., Prowse, P.D.H., Chrones, T., Sims, S.M., Hamilton, D.W., Rizkalla, A.S. et al. (2013) The differential regulation of osteoblast and osteoclast activity bysurface topography of hydroxyapatite coatings. *Biomaterials* **34**, 7215–7226 https://doi.org/10.1016/j.biomaterials.2013.06.014

108 Brinkmann, J., Hefti, T., Schlottig, F., Spencer, N.D. and Hall, H. (2012) Response of osteoclasts to titanium surfaces with increasing surface roughness: an in vitro study. *Biointerphases* **7**, 1–9 https://doi.org/10.1007/s13758-012-0034-x

109 Sartoretto, S.C., Alves, A.T.N.N., Resende, R.F.B., Calasans-Maia, J., Granjeiro, J.M. and Calasans-Maia, M.D. (2015) Early osseointegration driven by the surface chemistry and wettability of dental implants. *J. Appl. Oral Sci.* **23**, 272–278 https://doi.org/10.1590/1678-775720140483

110 Breding, K., Jimbo, R., Hayashi, M., Xue, Y., Mustafa, K. and Andersson, M. (2014) The effect of hydroxyapatite nanocrystals on osseointegration of titanium implants: an in vivo rabbit study. *Int. J. Dent.* **2014**, 171305 https://doi.org/10.1155/2014/171305