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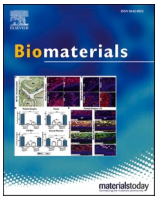

Aseptic and septic prosthetic joint loosening: Impact of biomaterial wear on immune cell function, inflammation, and infection

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ABSTRACT

The success of total joint replacements has led to consistent growth in the use of arthroplasty in progressively younger patients. However, more than 10 percent of patients require revision surgeries due to implant failure caused by osteolytic loosening. These failures are classified as either aseptic or septic and are associated with the presence of particulate wear debris generated by mechanical action between implant components. Aseptic loosening results from chronic inflammation caused by activation of resident immune cells in contact with implant wear debris. In contrast, septic loosening is defined by the presence of chronic infection at the implant site. However, recent findings suggest that subclinical biofilms may be overlooked when evaluating the cause of implant failure, leading to a misdiagnosis of aseptic loosening. Many of the inflammatory pathways contributing to periprosthetic joint infections are also involved in bone remodeling and resorption. In particular, wear debris is increasingly implicated in the inhibition of the innate and adaptive immune response to resolve an infection or prevent hematogenous spread. This review examines the interconnectivity of wear particle- and infection-associated mechanisms of implant loosening, as well as biomaterials-based strategies to combat infection-related osteolysis.

1. Introduction

Total joint arthroplasty is among the most widely used surgical interventions in orthopedics. It is projected that by 2030 there will be 635,000 total hip replacements and 1.28 million total knee replacements annually in the US, with more than 10% requiring revision surgeries due to loosening [1]. Over time, mechanical forces and biological interactions at the joint generate micro- and nano-scale debris, which can initiate localized inflammatory responses mediated by macrophages and foreign-body giant cells, resulting in bone resorption (osteolysis) at the bone-implant interface. Loosening of implants by osteoclastic resorption is a major cause of invasive revision surgeries, and a better understanding of this process is needed to effectively evaluate arthroplasty products, particularly when manufacturers intend to pursue pre-market claims regarding the advantages of specific technologies. Adjustments to the composition of the implants like crosslinking polyethylene, vitamin E enrichment or replacement with novel polymers aim to improve patient outcomes by reducing wear. However, no device on the market is free of wear debris, and the challenges associated with biological responses to wear debris are ongoing.

In general, total joint arthroplasty is a successful technique in replacing the form and function of hip, knee, shoulder, TMJ, and fingers of patients suffering from degenerative diseases such as osteoarthritis. Positive clinical outcomes have expanded the scope and indications for joint reconstructive surgeries, and the typical patient age for hip and knee replacements is now estimated to be less than 65 years [2]. The dramatically increasing number of younger patients receiving total joint arthroplasty [3] is resulting in higher failure rates in these younger and more active populations [4–8]. The most common complication for the procedure is chronic inflammation eventually resulting in bone resorption. Long-term implant survival and considerations of hypersensitivity and infection risk must be adapted for a changing patient population.

The complex interplay between innate immune response, adaptive immune response, biofilms, and particulates is evolving but currently under met in most efforts to understand osteolysis. Importantly,

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Abbreviations

PE	Polyethylene
UHMWPE	Ultra-high molecular weight polyethylene
PMMA	Polymethyl methacrylate
MoM	Metal-on-metal
MoP	Metal-on-polymer
CoC	Ceramic-on-ceramic
ALVAL	Aseptic lymphocyte-dominated vasculitis-associated lesion
EPS	Extracellular polymeric substance
PRR	Pattern recognition receptors
PAMP	Pathogen-associated molecular patterns
LPS	Lipopolysaccharide
LTA	Lipoteichoic acid
ROS	Reactive oxygen species
RANK	Receptor activator of nuclear factor κ-B
RANKL	Receptor activator of nuclear factor κ-B ligand
OPG	Osteoprotegerin
NET	Neutrophil extracellular trap
HBP	Heparin binding protein
PMN	Polymorphonuclear neutrophils
MHC	Major histocompatibility complex
NKT	Natural killer T cell
MRSA	Methicillin-resistant *Staphylococcus aureus*
SCV	Small colony variant
VE-UHMWPE	Vitamin E-enhanced ultra-high molecular weight polyethylene

histological analysis of revision surgeries reveals macrophages, foreign-body giant cells, T cells and B cells even in tissues that were not presumed infected [9]. Wear-associated inflammation in aseptic and septic conditions has been shown recently to be interconnected [10–12]. In particular, modulation of the innate and adaptive immune response by wear debris can contribute to septic loosening through inhibition of the normal response to microbial infection putting patients at greater risk of failure. This review summarizes the generation and consequences of biomaterial wear debris, and highlights the interaction between particulate wear and biofilms as a contributor to both aseptic and septic osteolytic loosening. It also introduces biomaterial strategies for reducing infection risk, as a means to prevent implant failure.

1.1. Implant materials and wear debris

Total joint arthroplasty devices use low-friction designs pioneered by Charnley in the 1960s for hip replacements [13]. These devices are typically comprised of a metal femoral shaft fitted with a ball made of metal (e.g. Co–Cr, Co–Ti, Ti6Al4V, SS) or ceramic (alumina or zirconia) [14], which articulates against an acetabular cup made of metal (Co–Cr), ceramic (alumina, zirconia), or polymer (ultra-high molecular weight polyethylene [UHMWPE]) implanted in the pelvic bone or femur using bone cement (polymethyl methacrylate [PMMA]). A popular early hip replacement model consisted of an UHMWPE cup and Co–Cr head, but particles generated by chronic wear of the UHMWPE has motivated the use of alternative materials and designs.

Approximately 35% of total hip arthroplasties performed in the US in 2005 used metal-on-metal (MoM) configurations [15]. MoM implants and less invasive resurfacing techniques were considered promising alternatives for active, younger patients seeking to resume high-impact activity [16]. MoM devices are mechanically strong with hard surfaces and the larger metallic heads provide great stability and range of motion [17,18] while generating less volumetric wear compared to their metal-on-polymer (MoP) counterparts. Co–Cr alloy has a linear wear rate of 0.1 μm per year (10^6 cycles) and 316 L stainless steel and Ti–6Al–4V releases 0.2 μm and 1 μm of wear per year (10^6 cycles), respectively [19]. While the mechanical properties of MoM joint replacements are ideal for short term activity, they have a significantly higher rate of osteolysis when compared to MoP or ceramic-on-ceramic (CoC) [20]. The wear debris from MoM are smaller, more numerous [21], and readily phagocytosed by host immune cells [22]. Metal ions leeched from metal wear cause metal hypersensitivities [23] and ion concentration in blood serum continues to increase up to 10 years after surgery [24]. Additional risks from metal wear including pseudotumor formation [25] and aseptic lymphocyte-dominated vasculitis-associated lesion (ALVAL) [26] contributed to a decline in popularity. In light of these complications, no MoM hip replacements have been approved by the FDA for use in the US since 2016.

For decades, conventional UHMWPE has been the predominant material of choice in total joint arthroplasty devices because of its biocompatibility, low cost, low coefficient of friction and high compressive and impact strength. However, UHMWPE generates relatively large amounts of volumetric wear when interfacing with the hard metallic head of implants. Carbon crosslinking methods such as gamma ray irradiation, chemical induction, and silane induction were implemented to increase resistance to wear [27] and has become the new standard. Irradiated polyethylene is subjected to thermal stabilization and vitamin E enrichment to increase oxidation resistance and remove free radicals that could cause surface delamination [28,29]. Pseudo capsular histological samples of revision surgeries show highly cross-linked UHMWPE wear is 26% smaller in diameter and results in a >90% reduction of wear volume compared to conventional polyethylene [30]. The size of UHMWPE particles can vary depending on the wear mechanism, ranging from large particles from adhesive wear to smaller particles from fragmentation of larger particles or exfoliation from the surface [19]. Wear from acetabular hip devices over an average 12.8-year lifespan generates an average of 785 mm^3 total volumetric wear (59.6 mm^3/year), including an average of $5.68 \pm 2.16 \times 10^{12}$ particles ($4.31 \pm 1.52 \times 10^{11}$/year), or approximately 1.3×10^{10} wear particles per mg of PE [31].

Numerous causes for aseptic loosening have been proposed, which can be broadly categorized into mechanical and/or biological responses that ultimately lead to fibrous soft tissue formation and osteolysis. Various histological studies of aseptic loosening sites have indicated the presence of wear particles from implant components, consisting of metal and polymer particles with sizes ranging from submicron to several hundred microns [19,30,32,33]. Corresponding immunohistochemical studies have shown the presence of macrophages and inflammatory mediators at the site [33]. The phagocytic uptake of wear particles by resident macrophages and the subsequent release of cytokines result in chronic bone resorption or osteolysis around the implant by decreasing local bone formation and increasing osteoclastic activity. By definition, aseptic loosening occurs without the presence of clinical or microbiological evidence of infection. However, numerous *in vitro* and *in vivo* studies support the concept that bacteria can have a role in aseptic loosening [34–36], and therefore, "aseptic loosening" may be a misnomer in numerous cases. Though wear particle generation from total joint arthroplasty devices is considered inevitable, the degree of particle generation is affected by surgical technique contributing to poor alignment, inadequate fixation, and mechanical instability reducing the lifetime of the device [37], and the effects of these particles may be accelerated by infection.

1.2. Biofilm formation on implants

Septic loosening from biofilm-related infections is another major

cause of implant failure, and may be more prevalent than previously thought. Recent findings suggest that many revision surgeries attributed to aseptic loosening are actually septic in nature with the presence of subclinical biofilms that are overlooked in standard examinations [35,36,38]. Advancements in microbial detection and analysis such as metagenomic NGS [39] and combining 16 S rRNA PCR with reverse line blot hybridization [35] are more accurate and sensitive compared to targeted PCR and can consistently identify a broad range of microorganisms that compose biofilms. These biofilms are multicellular colonies of bacteria, fungi, and protozoa attached to a surface that are typically resistant to the host immune system and antibiotics. Biofilms may be attributed to contamination during surgery, but circulating bacteria in the bloodstream, known as bacteremia, can cause hematogenous periprosthetic joint infections from unrelated infection sites after surgery [40]. It is estimated that 60–70% of hospital-acquired infections are associated with medical device bacterial biofilms [41]. These infections eventually cause septic loosening in 25% of total knee arthroplasty and 15% of total hip arthroplasty revision surgeries [42]. Large-scale evaluations of microbial contributions to implant complications are limited, but one such study from the UK in 2010 found 44% of knee and hip periprosthetic infections were caused by *Staphylococcus aureus,* making it the most endemic microbe. An additional 31% of infections involved coagulase-negative staphylococci such as *Staphylococcus epidermidis* [43]. The coagulase-positive characteristic of *S. aureus* makes it particularly virulent as it can generate a staphylothrombin complex that promotes blood clot formation from platelet aggregation through fibrin conversion of fibrinogen [44]. Geographic studies of drug resistance and biofilm composition suggest variable biofilm development and evolution in different regions [45]. For example, a study from Spain observed an increasing trend of multi-drug resistant periprosthetic joint infections over the course of 10 years attributed to a 2.9% increase in multi-drug resistant gram-negative bacilli [46].

Biofilm formation begins when planktonic (free floating) bacteria attach to a surface and become sessile, as shown schematically in Fig. 1. In the case of septic loosening, the surface they attach to is the fibrous capsule that forms around the implant due to the host response [47]. Serum proteins adhere to the surface of the implant immediately after surgery, initiating a series of events that results in the formation of a fibrous capsule [41]. The capsule surface provides an attractive substrate of collagen, laminin, fibronectin, elastin, and fibrinogen for attachment of bacteria via adhesin proteins on their surface [48]. After adherence to a surface, bacteria begin to secrete extracellular polymeric substances (EPS) that transform a disconnected population of bacteria into a networked and fortified biofilm. The EPS is influenced by the local mechanobiology and is composed of polysaccharides, extracellular DNA (eDNA), and proteins [49]. The EPS encapsulates the bacterial population to make a complex interconnected community that functions as a single multicellular system. As the population of bacteria changes, they demonstrate quorum sensing to regulate gene expression, which allows the biofilm to respond to environmental changes as a unit [50]. The maturing biofilm is characterized by growth in cell number, increased intercellular communication, and coalescing of smaller colonies [51]. As the biofilm reaches a critical size and population, the bacterial colony exhausts its resources and begins the process of dispersal. Individual bacteria or pieces of the biofilm are released from the surface to attach elsewhere and propagate the infection. This release is prompted by the degradation of the ECM, surfactant-induced decrease of surface tension, or cell death [52–55]. These infections can go undetected when evaluating patient biopsies, making microbiological identification inconsistent.

1.3. Macrophage and neutrophil response to biofilm and wear

The macrophage-mediated innate immune response is the first line of defense against pathogens and acts in a non-specific manner to resolve most bacterial infections, as shown schematically in Fig. 2. Bacteria present specific molecular structures referred to as pathogen-associated molecular patterns (PAMPs), and which include lipopolysaccharide (LPS) for Gram-negative bacteria [56] and lipoteichoic acid (LTA) for Gram-positive bacteria [57]. The initial response to infection occurs when anti-inflammatory, tissue-resident M2 macrophages are activated through pattern recognition receptors (PRRs) that recognize the PAMPs of bacteria, leading to the initiation of M1 polarization through TNFα [58,59]. Surrounding monocytes, fibroblasts, and endothelial cells produce MCP-1/CCL2 to recruit more monocytes to the infected tissue [60–62]. In addition to differentiating into macrophages and dendritic cells, monocytes help to combat pathogens via phagocytosis and reactive oxygen species (ROS) generation [63].

Macrophage response to wear particles also induces activation of the canonical pro-inflammatory M1 phenotype. Sustained M1 activation and stimulation of the inflammatory cascade leads to subsequent osteoclast activation and osteolysis. Biochemical indicators found in explanted tissues include the inflammatory cytokines TNFα [64], IL-1 [65], and IL-6 [66], especially in conjunction with pathologic

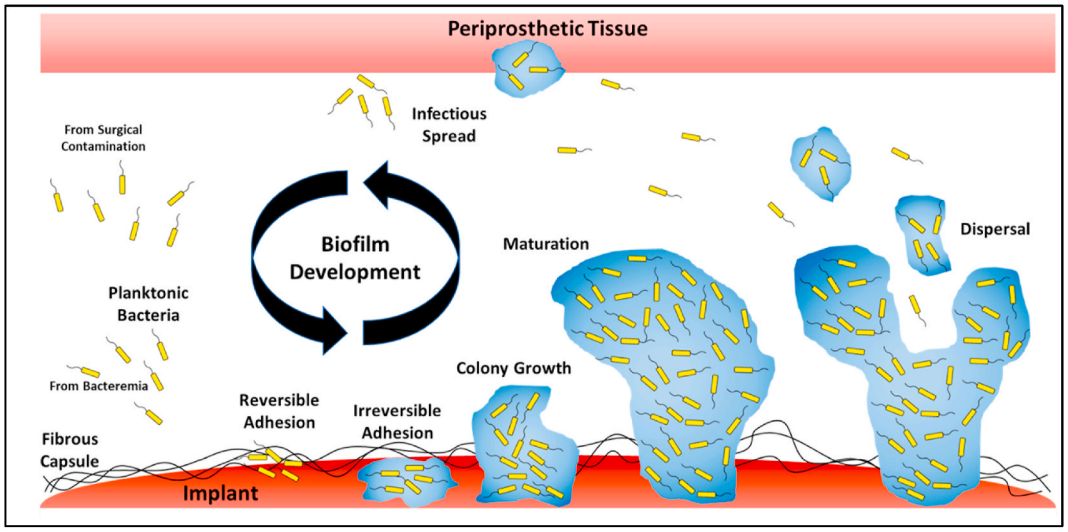

Fig. 1. Biofilm development in total joint arthroplasty. Planktonic bacteria from surgical contamination or bacteremia attach to the fibrous capsule around the implant. Bacteria become sessile as they secrete protective EPS to form a larger biofilm. The bacteria of the biofilm grow rapidly to form a tower-like structure before dispersing individual bacteria and masses of the biofilm. The dispersal infects cells and embeds into the surrounding periprosthetic tissue to propagate the infection.

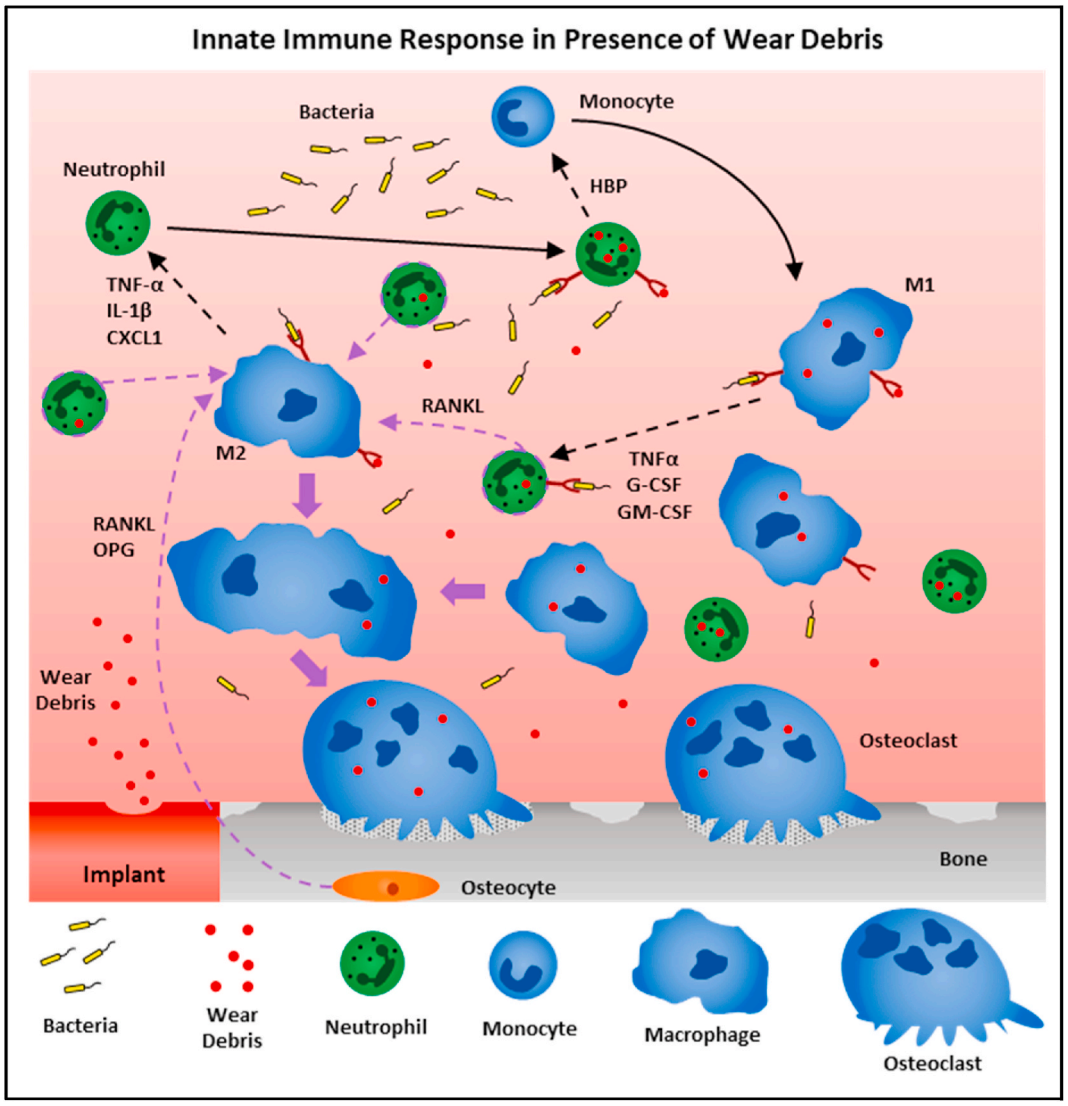

Fig. 2. The innate immune response to infection and wear debris. Tissue-resident macrophages encounter pathogens, polarize from the M2 to the M1 state, and recruit neutrophils to the site. Neutrophils recruit more monocytes and employ granules to destroy bacteria. Wear debris similarly causes M1 polarization but particulate phagocytosis by either macrophages or neutrophils reduces successive phagocytosis of pathogens. Increased TNFα and RANKL production overpowers OPG signaling from osteocytes to induce osteoclast formation by fusion of macrophages. The inability to clear infection through macrophages and neutrophils perpetuates the inflammatory state with no resolution causing continued osteoclastogenesis. Purple arrows represent osteoclastogenesis pathway. Dotted lines indicate cell signaling relationships. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

osteolysis [67]. In addition, explanted inflamed tissue contains colony stimulating factors (CSFs) [68,69], prostaglandin E_2 (PGE2) [65,66,70], as well as proteolytic enzymes (collagenase and gelatinase) [65]. Macrophages can fuse together to form foreign body giant cells during fibrous capsule formation at the surface of the implant, or to phagocytose wear particles too large for isolated macrophages [71]. Exposure to small wear particles also initiates differentiation of macrophages to osteoclasts by fusion as a result of an increase in TNFα [29], resulting in bone resorption in the proximity of the interface. Normally osteoclastogenesis is promoted by an increase in receptor activator of nuclear factor κ-B (NF- κB) ligand (RANKL) produced by inflammatory cells relative to osteoprotegerin (OPG) produced by osteoblasts. RANKL and OPG compete to bind to receptor activator of nuclear factor κ-B (RANK) on the surface of macrophages and monocytes. During infection, the RANKL from inflammatory cells increases to shift the balance to osteoclastogenesis. There is evidence that wear particles can reduce the effectiveness of the inflammatory response, since polyethylene and ceramic particles have been shown to induce apoptosis in macrophages [72]. Macrophages that have interfaced with polyethylene particles exhibit an inability to clear bacterial populations [10] leaving the host at risk of chronic or persistent infection. The chronic inflammation and immunosuppression induced in macrophages by wear debris can perpetuate osteolysis adversely affecting patient outcomes.

The most abundant leukocytes in circulation, neutrophils are the primary actors of the innate immune response to bacteria. Macrophage secretion of the chemokines TNFα, IL-1β, and CXCL1 signals neutrophil recruitment [73–75]. Polymorphonuclear neutrophils (PMN) are equipped with antimicrobial granules of proteases and the capacity to produce toxic oxygen metabolites, known as a respiratory burst [76]. Neutrophils also recognize PAMPs through PRRs and thereby can phagocytose bacteria. The earliest neutrophils to make contact with bacteria secrete a variety of chemoattractive signals that elicit a neutrophil swarm to surround the infection and insulate the surrounding tissue from harm [77]. The assembly of neutrophil extracellular traps (NETs) by release of chromatin and granules into the extracellular space is required to combat formation of larger clusters of bacteria in biofilms [78,79]. Among the proteases composing neutrophil antimicrobial granules, azurocidin (also called heparin-binding protein, HBP) can

recruit circulating monocytes and trigger M1 polarization [80–82]. Additional release of annexin A1 by neutrophils promotes phagocytic activity of macrophages [83]. M1 macrophages then phagocytose bacteria through PAMP recognition, however macrophages lack the antimicrobial granules that neutrophils use to deconstruct bacteria. As bacteria are cleared, neutrophils undergo apoptosis and degranulation, releasing their antimicrobial granules to the extracellular space. M1 macrophages can enhance their antimicrobial capacity through uptake of these granules, or trigger neutrophil apoptosis through membrane-bound TNFα and phagocytose them directly [84,85]. Neutrophils are recruited in an active infection by M1 macrophage and the release of TNFα, GM-CSF, and G-CSF that maintain neutrophil survival and migration [86]. At the resolution of a successful innate immune response, macrophages phagocytose the neutrophils and revert to the anti-inflammatory M2 phenotype important for tissue repair [87].

The generation of wear particulate can disrupt the normal functions of neutrophils upsetting the innate immune response and contributing to chronic infection and inflammation. The number of PMNs is a key criterion in diagnosing periprosthetic joint infection requiring revision surgery as their persistence is indicative of chronic infection [88]. In response to wear debris, it is established that PMNs can phagocytose polyethylene, Co–Cr, and Ti particles and degranulate [89]. In particular, the introduction of UHMWPE wear debris to neutrophils has been shown to inhibit their ability to phagocytose bacteria, though they have increased metabolic activity indicative of increased ROS production [12]. Small particle phagocytosis also sequesters neutrophil elastase, a key factor in the formation of NETs to combat biofilm [90]. Particle-compromised neutrophils may not be able to effectively clear bacteria, leading to further recruitment of monocytes/M1 macrophages, which in turn could promote the survival and recruitment of more neutrophils. Neutrophils have also been shown to express RANKL both at the membrane surface and intracellularly to facilitate osteoclastogenesis [91]. Macrophages that have phagocytized wear particles have increased inflammatory marker expression and can still form functional osteoclasts that contribute to implant loosening [92]. This feedback loop (Fig. 2) exacerbates the inflammatory conditions needed to disrupt the RANKL/OPG balance maintained by osteocytes and cause increased osteoclastogenesis associated with implant failure [93].

1.4. Dendritic cell and T cell responses to biofilm and wear

Dendritic cell and T lymphocyte interactions with wear debris are not fully understood, but the available information suggests they may have important implications in implant loosening. Dendritic cells are professional antigen presenting cells that act as a bridge between the innate and adaptive immune response, as shown in Fig. 3. Tissue-resident and monocyte-derived dendritic cells possess PRRs that identify PAMPs of different bacteria types, activating the dendritic cell and inducing a pathogen-specific change in mRNA expression and release of chemokines [94]. Dendritic cells internalize bacterial antigens by phagocytosis, endocytosis, and micropinocytosis [95], and are then transported from the infection site by afferent lymphatic vessels to

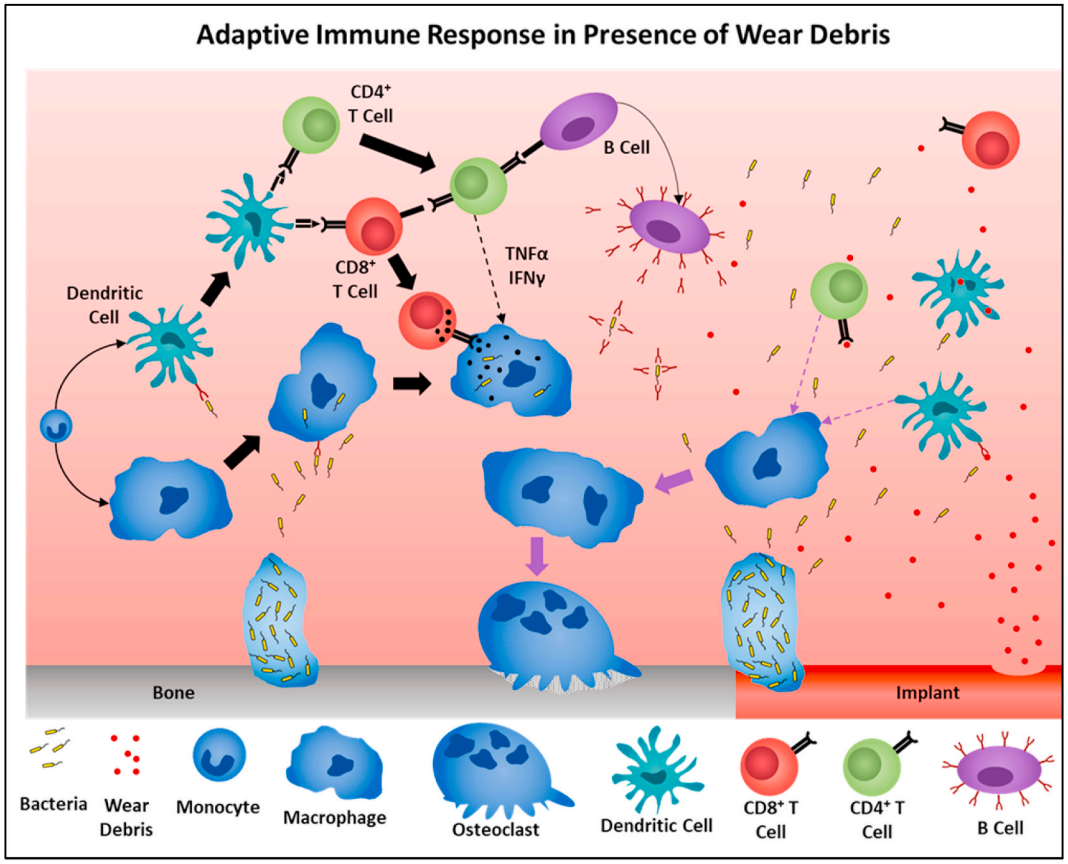

Fig. 3. The adaptive immune response to infection and wear debris. Dendritic cells identify pathogens and present corresponding MHC class I or MHC class II to $CD8^+$ and CD4+T cells respectively. $CD8^+$ T cells directly destroy pathogen-containing cells, e.g., macrophages. CD4+T cells affect the inflammatory polarization of macrophages and activate $CD8^+$ T cells and B cells that release antimicrobial antibodies to attack the infection. Contact with wear debris causes dendritic cells to promote osteoclast formation from macrophages. Phagocytosis of wear debris prevents antigen presentation from dendritic cells and causes cell death. Reduced dendritic cell function similarly decreases activation of T cells. Particles directly contacting CD4+T cells can induce inflammatory secretome release significant for osteoclast formation. Immune disruption allows for infection spread and osteolytic resorption. Purple arrows represent osteoclastogenesis pathway. Dotted lines indicate cell signaling relationships. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

secondary lymphoid organs including draining lymph nodes and the spleen. Here dendritic cells present antigens in major histocompatibility complex (MHC) class I and MHC class II to activate $CD8^+$ cytotoxic T cells and $CD4^+$ helper T cells, respectively.

T lymphocytes are the major actors in the adaptive immune response and upon activation circulate to the site of infection. $CD4^+$ T cells are responsible for two main functions: 1) release of inflammatory cytokines and 2) activation of $CD8^+$ T cells and B cells. The inflammatory secretome of activated $CD4^+$ T cells includes interferon gamma (IFNγ) and TNFα, which promote M1 polarization and activity of macrophages [96,97]. Conversely, monocytes/macrophages have the capacity to polarize $CD4^+$ T cells into an inflammatory phenotype through direct cell-cell interactions, continuing the cycle until the infection is resolved [98]. $CD8^+$ T cells employ perforin and granzyme granule release to directly destroy cells that contain bacteria such as phagocytes or infected cells holding them in vacuoles [99]. T cell granule destruction is more target-specific than the granule release by neutrophils because it is released along an immune synapse between antigen presenting cells and the T lymphocyte. After contact with an antigen, B cells are activated to produce antimicrobial antibodies and $CD4^+$ T cells amplify their production [100]. The adaptive immune response to bacteria is required to clear bacterial infection when the innate immune response fails to do so.

Contact with nanoscale UHMWPE debris activates dendritic cells secretion of the inflammatory cytokines IL-6 and IL-1β [101]. Dendritic cells exposed to UHMWPE also increase IFN-γ production by natural killer T (NKT) cells, which polarizes macrophages to an M1 phenotype and increases macrophage production of TNFα [102]. Subsequently, dendritic cells amplify osteoclastogenesis in macrophages responding to wear particles, thereby aggravating aseptic loosening [103]. Dendritic cells are capable of phagocytizing wear particles and secreting IL-1β, but the resulting damage to endosomes can cause release of cathepsins into the cytosol that may lead to cell death [104]. Damage to endosomes responsible for loading and transferring antigens could also disrupt antigen presentation by dendritic cells to T lymphocytes in the secondary lymphoid organs. Histological analysis of the spleen, lymph nodes, and liver of patients has revealed the presence of metallic and polyethylene particles with higher incidence in patients who suffered implant failure [105]. However, polyethylene is difficult to detect at lower concentrations and long term effects in these tissues are unknown. In cancer, dysfunction of antigen presentation by dendritic cells is associated with T cell proliferation and contributes to tumor survival [106]. Activation of T lymphocytes in the presence of biomaterial wear debris typically occurs through antigen-presenting cells, but studies suggest that metal wear debris can act as an antigen and directly influence T cells. CoCr nanoparticles, for example, reduce the proliferation of $CD8^+$ T cells in patients with metal-on-metal implants [107]. Analysis of the effect of metal particles on human blood-isolated $CD4^+$ T cells revealed activation and secretion of both inflammatory and anti-inflammatory cytokines, depending on T helper cell subtype [108–110]. Taken together, these findings suggest that disruption of the cells in adaptive immune response by wear particles may inhibit the resolution of bacterial infections and increase incidence of septic loosening.

1.5. Biomaterials-based strategies to counteract biofilm formation

The development of biomaterials that can prevent or counteract biofilm formation offers an opportunity to prevent septic loosening of implants. Although wear can be reduced through material modifications [29,30,111], the generation of wear debris is essentially unavoidable. The antioxidant vitamin E was initially incorporated into UHMWPE to improve the integrity of the implant by improving oxidation resistance [112]. Studies characterizing wear particulates and the resulting inflammatory response showed peripheral blood mononuclear cells produce significantly fewer of the osteolytic cytokines TNFα, IL-1β, IL-6, and IL-8 in response to vitamin E-enhanced UHMWPE (VE-UHMWPE), compared to virgin UHMWPE [29]. Vitamin E also provides protection against biofilm formation when added to media *in vitro* and VE-UHMWPE has had similar results in bacterial culture [113,114]. While vitamin E is effective at preventing biofilm formation of *S. aureus* and *E. coli* [115], it has shown less definitive effects in preventing MRSA infection [116]. Compared to virgin PE, macrophages incubated with VE-UHMWPE exhibited greater survival, reduced TNFα production, and greater pathogen response to *S. aureus*. *In vivo* results in a mouse calvarial model corroborated the enhanced clearance of *S. aureus* and reduced osteolysis when VE-UHMWPE was implanted, compared to virgin UHMWPE [10]. Although promising, the mechanisms by which vitamin E reduces osteolysis and produces antimicrobial effects are still unclear.

An alternate strategy to prevent septic loosening is antibiotic loading of biomaterials. Antibiotic loaded cement spacers are already implemented in revision surgeries for septic loosening, resulting in success rates similar to aseptic revision surgeries [117]. Alternatively, the use of antibiotic-loaded bone grafts in revision surgeries has shown promising results in reducing infection [118], though excessive loading can reduce the mechanical integrity [119]. Despite the success of antibiotics, bacteria have many mechanisms of resistance. Genetic transfer from one organism to another, can result in multi-drug resistance. Genetic transfer of an alternate penicillin binding protein (PBP2a) caused the emergence of methicillin-resistant *S. aureus* (MRSA) in the 1960s, which resulted in new understanding regarding overuse of antibiotics and resulting drug resistance [120]. The widespread use of antibiotics like penicillin has been linked to β-lactam resistance by MRSA, revealing that drug resistance was developed against an entire class of antibiotics more than a decade prior to the introduction of methicillin [121,122]. Small colony variants (SCV) are slow-growing subpopulations of bacteria under metabolic deficiency that provide another form of antibiotic resistance by alterations in cellular respiration that reduce potential for drug uptake. Under normal conditions the faster growing populations of bacteria will outcompete the SCV, but antibiotics eliminate the more proliferative bacteria thereby encouraging SCVs to persist. Furthermore, the stress from antibiotics can induce the SCV phenotype [123]. However, these bacteria do not remain in this nearly dormant state and offspring can mutate into a faster growing population that maintains the drug resistance [124]. Bacteria also employ multidrug efflux pumps to provide added protection by ejecting antibiotics and metal ions from the biofilm. However, these pumps can be inhibited, making them generally vulnerable to antibiotics and, importantly, metal ions [125].

Bactericidal metal ions incorporated into oxidative or ceramic surface coatings represent a developing field of materials research. Naturally antimicrobial silver and copper coating of titanium-aluminum (TiAl) alloys releases ions that inhibit bacterial growth [126,127]. Incorporation of Ag and Cu particles into hydroxyapatite coatings provide antibacterial properties [128–130] and combining growth factors like bone morphogenetic protein-2 (BMP-2) shows promise in osseointegration [131]. It has been shown that Ag particles can combine synergistically with the effect of antibiotics in preventing implant-associated infections, including MRSA [126,132,133]. Despite their potential, metal coatings can have cytotoxic effects on host tissue and can only utilize a finite amount of metal ions initially attached [134].

Whereas antibiotic and metallic coatings can provide short-term protection against infection post-surgery, “smart” material coatings respond to changes in the microenvironment to react specifically when bacteria are present. The stimuli that actuate smart coatings can be categorized as bacterial byproducts or external activation. The biofilm microenvironment can vary in pH, but infection generally fosters hypoxia that creates an acidic microenvironment. The acidity degrades specialized hydrogels [135] or thin layers of hydroxyapatite [136] to release metal ions and/or antibiotics. Similarly, ROS produced by bacteria and local inflammation can degrade polymer coatings containing antimicrobial agents [137]. Immune-modulatory polymers can respond

to infection by releasing adsorbed peptides, nucleic acids, or cytokines to polarize macrophages to either inflammatory or anti-inflammatory phenotypes [138,139]. External control of smart materials is facilitated by UV and electrical stimulation to release antimicrobial factors into the infection site [140,141]. These innovations illustrate the evolution from bioinert materials to materials that exhibit intelligent, bioactive responses.

2. Conclusion

Aseptic and septic loosening of total join replacements have commonly been considered separate processes, but new research shows the impact of wear debris on the formation and survival of biofilm, even at subclinical levels. Routine microbiological diagnostics may misdiagnose septic loosening caused by subclinical biofilms, resulting in ineffective treatment and recurring implant failure. The interaction of implant wear debris with macrophages and neutrophils impairs the ability of the innate immune system to remove bacteria, thereby allowing greater maturation and proliferation of biofilms that are already equipped with immunosuppressive mechanisms. In addition to compromising the innate immune system, wear particles disrupt the dendritic cells and T lymphocytes of the adaptive immune response, which are needed to coordinate a targeted reaction to periprosthetic joint infection. This disruption exacerbates the inflammatory pathways associated with both aseptic and septic loosening, causing chronic inflammation and greater osteolytic resorption. Even in the absence of subclinical biofilms resulting from surgery, the impact of wear debris on the immune response makes periprosthetic tissue more susceptible to infection from bacteremia. Continued biomaterials research on reduction of wear debris is important, but does not address the need to proactively prevent biofilm formation and resulting osteolysis. Modifications of implant biomaterials through the release of antibiotic drugs, antimicrobial ions, and antioxidants are potential strategies to reduce risk of infection. These infection resistance approaches offer great opportunities to enhance implant stability and lifespan. However, further research is needed to understand the long term consequences of wear particle invasion into the periprosthetic tissue and the secondary lymphatic organs.

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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