



# Parallel evolution of polymer chemistry and immunology: Integrating mechanistic biology with materials design

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## ABSTRACT

To develop new therapeutics involves the interaction of multiple disciplines to yield safe, functional devices and formulations. Regardless of drug function and potency, administration with controlled timing, dosing, and targeting is required to properly treat or regulate health and disease. Delivery approaches can be optimized through advances in materials science, clinical testing, and basic biology and immunology. Presently, laboratories focused on developing these technologies are composed of, or collaborate with, chemists, biologists, materials scientists, engineers, and physicians to understand the way our body interacts with drug delivery devices, and how to synthesize new, rationally designed materials to improve targeted and controlled drug delivery. In this review, we discuss both device-based and micro/nanoparticle-based materials in the clinic, our biologic understanding of how our immune system interacts with these materials, how this diverse set of immune cells has become a target and variable in drug delivery design, and new directions in polymer chemistry to address these interactions and further our advances in medical therapeutics.

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## 1. Introduction

The development of novel drug delivery technologies has benefited greatly from the contributions of scientists from multiple backgrounds. Such interdisciplinary collaboration has led to significant breakthroughs in biomedicine and is key to addressing the challenges of the future. Here, we examine contributions of studies in clinical medicine, basic biology and immunology, and polymer chemistry. Through the eyes of chemists, clinicians and biologists, we focus on the parallel evolution of polymer design and mechanistic immunology that has been integral to furthering advances in drug delivery technology. There is evidence of the treatment of disease or pain with pharmaceuticals since the paleolithic age by shamans using medicinal plants. More details of the use of drugs in written history are present throughout ancient Egyptian texts and illustrations, including the use of opium and cannabis for pain relief [1]. In the 19th century, chemists began to isolate and synthesize organic molecules such as quinine, paracetamol, benzocaine, and methamphetamine [2]. Moving into the 20th century, the synthesis of drugs increased exponentially yielding numerous pharmaceuticals that are still in use today. In the 20th and 21st century, new classes of drugs utilizing a design created by our own bodies, such as monoclonal antibodies, began to be generated for the treatment of various conditions, ranging from cancers to hypercholesterolemia.

Regardless of the potency or efficacy of the drug, if it is not delivered to the patient in a controlled manner with precise dosing, location, and clearance, a game-changing drug in vitro could yield a clinical failure. Through the use of controlled degradation and release kinetics afforded by polymeric systems, it became possible to engineer drug delivery technologies to modulate dosing, location, and clearance of therapeutics [3,4]. FDA-approved delivery systems range from polymer-based materials such as the Gliadel wafer (used to deliver carmustine chemotherapy locally to recurrent gliomas), to implantable microelectronics, and to nanoparticle systems such as Doxil (PEGylated liposomal doxorubicin for treatment of various cancers) [5,6].

In addition to therapeutics used in the clinic, there are numerous new materials being developed by engineers and biologists in the laboratory. Through integration of mechanistic biologic knowledge, we are able to identify targets for new therapeutics and understand the interaction of the patient's immune system with implanted materials [7]. As the immune system defends a patient's body from foreign insult, including material implantation, immune cells have become a target and a variable for consideration in materials design. In negative responses to implanted materials, immune cells can produce nitric oxide (NO) radicals capable of expediting material degradation and release of any encapsulated drug cargo [8]. Furthermore, they can create a fibrotic encapsulation of a drug delivery device through deposition of a thick, collagenous matrix around the material in a process known as the foreign body response. Besides their effect on drug delivery devices, the immune system is being further appreciated for its role in multiple pathologies ranging from obvious disorders (autoimmunity) to a variety of seemingly unrelated diseases (Alzheimer's, cardiac disease) [9–12]. Leveraging advances in polymer chemistry, these basic immunologic discoveries have been transformed into improved and targeted therapeutics.

New polymers are able to overcome biologic responses to these materials and afford more bioavailable drugs to promote delivery of therapeutics and translation to the clinic. Chemotherapy-loaded microparticles have been modified into non-spherical shapes to analyze the role of geometry in drug delivery to tumors with PEG-PEMA (poly(ethylene glycol), 2-(pentamethyleneimino) ethyl methacrylate) micelles or filomicelles [13], reversible addition–fragmentation chain transfer (RAFT) polymerization has been used to synthesize polymeric immune targeting moieties to antigens and innate immune ligands [14], and silyl-ether based ring-opening metathesis polymerization (ROMP)-polymers have been utilized to promote degradation and

prevent toxic accumulation of brush-arm star polymers (BASPs) in the liver and spleen [15].

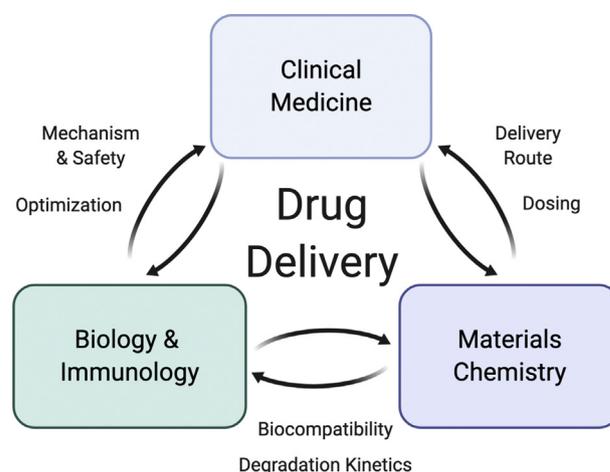
Further work in the fields of materials science, basic immunology, and clinical medicine, will continue to push forward development of new drug delivery technologies that help address unmet needs in human health and disease (Fig. 1). In this review, we discuss clinical advances and limitations, understanding and modulating the body's response to materials and approaches to engineering the immune system, and finally new approaches in materials chemistry and material development, that interact to push forward the interdisciplinary field of drug delivery.

## 2. Clinical translational of drug delivery technologies

The goal of all drug delivery technologies is to positively impact patient care. It is through the efforts of multiple scientific and engineering disciplines that this work is achieved, and numerous clinically used and paradigm-shifting technologies have resulted from bridging these different fields [16]. Clinically successful materials share this developmental passage through multiple scientific disciplines, and we are still learning from current outcomes of drug delivery technologies to further develop and identify needs for improvement in materials design.

### 2.1. Keys to clinical translation of technologies

The most basic tenet of creating and translating new drug delivery technologies is addressing a clinical need using novel scientific and engineering methods. Through work with physicians on the frontlines of medicine, these unmet clinical needs can be identified and addressed. Integral pieces involving the translation of new technologies are having a team of hardworking successful individuals, the proper investment to undertake the work, and timing [17]. Thomas Friedman said that “vision without resources is only a hallucination”. Investment from grants, angel investments, venture capital, and other opportunities will provide the resources, and at times additional wisdom, to assist in the transition. Proper timing for translating a project is essential, as launching a company too early may be an issue in finding investors and may influence the lifespan of patents. Investment of time and resources into the development of drug delivery technologies has resulted in the translation of



**Fig. 1.** Drug Delivery Requires Contributions of Clinical, Biological, and Chemical Studies to Support Innovation. Interactions of clinical practice with biology and immunology through the studies of safety, drug mechanism, and patient tolerance of implanted materials yield critical directions for novel material design. Materials chemistry and immunology interact to understand biocompatibility and effect of host response on degradation kinetics of polymers. Ultimately materials chemistry and clinical need interact to control drug dosing and delivery.

several therapeutics to address previously unmet needs in human health and disease (Fig. 2).

There are many examples of such technologies that have been used in the clinic and continue to be used in patients. Of these technologies, there are a number of different approaches to improving drug delivery, including nanomaterials, solid polymeric depots, and remotely triggered microchips.

## 2.2. Nanomaterials

One approach to drug delivery is packaging drug payloads in engineered nanoparticles. There are a multitude of nanomaterials that have reached clinical use and clinical trials [18,19]. The most common clinical indications for these materials are cancer and inflammatory diseases [20,21]. Here, we will focus on liposomal doxorubicin, Particle Replication in Nonwetting Templates (PRINT) formulation LIQ861, and lipid nanoparticles (LNP), as these nanomaterials have generated significant interest based upon their unique technology.

One of the most well-established nanomaterials is liposomal doxorubicin (Doxil). As with most drugs, cancer cells have drug-resistance mechanisms including: 1) drug efflux pumps, 2) high levels of glutathione and other scavengers that can remove free radicals, 3) decreased topoisomerase II expression or mutations in the enzyme activity preventing effectiveness of topoisomerase II inhibitor, and 4) defects in cell cycle checkpoints or apoptotic signaling pathways which result in enhanced cell viability [22]. To circumvent drug efflux pumps as a mechanism of resistance, encapsulation of doxorubicin in polyethylene glycol (PEG)-ylated liposomes has resulted in the development of Doxil [23]. This novel drug delivery approach could bypass efflux pumps, because liposomal doxorubicin enters the cells through endocytosis of the particle, rather than diffusion through the bilayer. Also, the targeted delivery leads to increased drug concentration within the cell, which could potentially saturate the drug efflux pumps [24]. Liposomal doxorubicin received approval by the FDA for the treatment of Kaposi's sarcoma in 1995. Initial studies in Acquired Immunodeficiency Syndrome (AIDS)-related Kaposi's sarcoma patients demonstrated partial remission in approximately 70% of patients and stable disease in the rest. Overall, it was well-tolerated with the most common adverse event being myelosuppression [25].

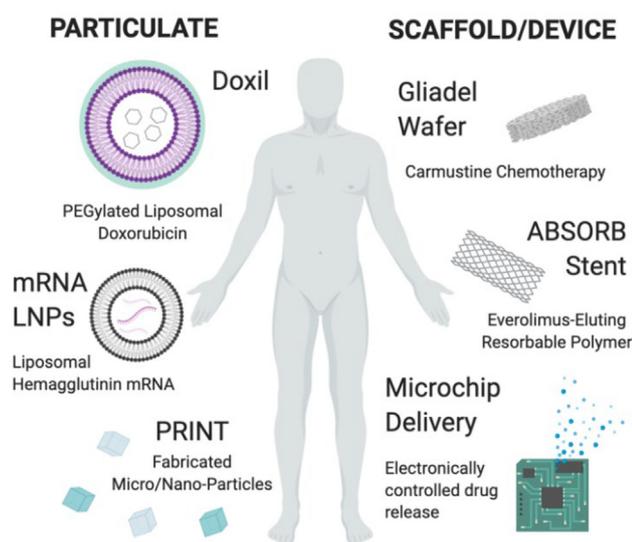
In contrast to some small molecule drugs, nucleic acid therapeutics incur a major delivery problem due to the mRNA and siRNA lability, cellular uptake of nucleic acids, and targeting. There are a number of LNP-based systems that have achieved success in progressing from pre-clinical studies to clinical studies due to the ability of the LNP in protecting nucleic acids and increasing cellular uptake. A major example of this is the use of LNPs for the delivery of messenger RNA (mRNA) for management of infectious diseases. Through the combined efforts of research teams in industry and academia, mRNA vaccines using novel LNP systems were created. These LNPs enable expression of full-length, membrane-bound form of the hemagglutinin (HA) glycoprotein from the H10N8 and H7N9 influenza strains. In two randomized, placebo-controlled, double-blind, phase 1 clinical trials, patients were administered two-dose vaccination three weeks apart at varying dose levels. With the primary endpoints being safety and tolerability, they demonstrated very few adverse events with the most common being local injection site pain (10%) at the highest dose for the avian influenza strain, H7N9. For the secondary endpoints of immunogenicity, they found significant hemagglutination inhibition (HAI) seroconversion rates of 78.3% for H10N8 and 96.3% for H7N9 in patients at the highest doses. H10N8 HAI antibody titers ( $\geq 1:40$ ) in 100% of patients and H7N9 HAI antibody titers ( $\geq 1:20$ ) in 89.7% of patients at their highest respective doses. Persistence of detectable HAI titers for 6 months after vaccination suggests development of a memory B-cell response [26]. Furthermore, there are other clinical examples of nucleic acid therapeutics, including an siRNP-loaded LNP formulation for the treatment of peripheral nerve disease (polyneuropathy) caused by hereditary transthyretin-mediated amyloidosis [27,28].

The particle replication in nonwetting templates (PRINT) technology was one of the first top down fabrication methods for generating nano- and micro-particles. This technology enabled the fabrication of particles of any size, shape, composition, and surface chemistry [29]. PRINT technology has been used to generate novel formulations for influenza vaccines and treatment of pulmonary artery hypertension (PAH). In an open-label phase 3 trial, known as INSPIRE (Investigation of the Safety and Pharmacology of Dry Powder Inhalation of Treprostinil), they evaluated the safety and tolerability of LIQ861 in PAH patients. They found similar adverse events profiles compared to nebulized treprostinil inhalation solution, including cough, headache, and throat irritation (Tyvaso). Furthermore, using a patient oriented measure of the adverse effects of heart failure, they found improvement in both the physical and emotional well-being compared to Tyvaso [30].

## 2.3. Solid polymeric depots

Beyond nanoparticle-based approaches, large macro-devices have been engineered for the controlled release of drugs from a local depot. Solid polymer drug delivery depots hold substantial clinical promise for their ability to deliver large doses of drugs over prolonged periods of time. These typically require surgical or interventional implantation of the solid depot in a location of interest. Two primary solid polymer depots include the carmustine (Gliadel) wafer for gliomas and an everolimus-eluting coronary artery bioresorbable stent. These systems have very different indications and primary functions, but both were developed from multi-disciplinary teams to address major needs within healthcare.

Carmustine wafers were created to address the major need for better treatments for glioblastoma multiforme (GBM). The prognosis for patients with GBM is poor. These tumors are managed by surgery, radiation, and chemotherapy and traditionally respond well to therapy, at least initially [31]. Unfortunately, tumors typically recur either at the primary tumor site or distantly. A major challenge for drug treatment of these tumors involve overcoming the blood brain barrier (BBB). Through the combined efforts of neurosurgeons, engineers, and chemists, the Gliadel wafers were generated for implantation at the time during surgical resection of the tumor. These wafers are composed of a poly



**Fig. 2.** Examples of clinically applied drug delivery systems. Particulate and device-based systems are currently being used or tested in humans. These support the delivery of small molecule drugs (chemotherapeutics, immunosuppressants), mRNA & siRNA, and peptides. Their mechanism of action is either by internalization of delivery vehicles (lipid nanoparticles), local release at the site of action (chemotherapy), or local release for systemic modulation (microchip peptide delivery).

(carboxyphenoxypropane/sebacic acid) [p(CPP:SA)] matrix combined with carmustine and can deliver the drug over a 3–4-week period. The polymer matrix degrades over 6–8 weeks and its degradation products are ultimately metabolized and renally excreted. There have been multiple randomized clinical studies involving the surgery, radiation, and Gliadel wafers showing improved survival compared to those that did not receive the Gliadel wafers with few side effects in patients with newly diagnosed and recurrent malignant gliomas [32–35]. Larger trials with trimodality therapy, including surgery, radiation, and chemotherapy, plus Gliadel wafers could be used to understand optimal therapies within the surgical and oncology community [36].

Another prime example of clinically used solid polymeric drug delivery devices are drug-eluting cardiac stents. Drugs, including sirolimus, everolimus, paclitaxel, and others, have been incorporated onto bare metal stents to improve the rates of restenosis in coronary arteries that have undergone percutaneous intervention [37]. Sirolimus was one of the initial drugs tested for release from a polymeric coating on a metal stent. In pre-clinical studies, a mixture of poly-*n*-butyl methacrylate and polyethylene-vinyl acetate (PBMA-PVA) copolymers and sirolimus was applied to the surface of a stainless-steel, balloon-expandable stent to achieve a thickness of 5  $\mu\text{m}$  followed by a coating of the PBMA-PVA copolymers [38]. This stent was designed to release approximately 80% of the sirolimus over 1 month after implantation. In a randomized, double-blind trial comparing the sirolimus-eluting stent to a bare metal stent, it was found that the sirolimus-eluting stents significantly reduced the rates of restenosis compared to bare metal stents at 6 months [39]. There has been rapid clinical adoption of these stents within the field of interventional cardiology after FDA approval in the early 2000s [40,41]. Newer cardiac stents are undergoing evaluation to further improve long-term restenosis (ref).

#### 2.4. Wirelessly controlled microchips

A novel implantable technology reported by Santini et al. showcased the use of an implantable multi-well microchip capable of being externally controlled to release drug at user- or provider-defined times [42]. Potential drug classes delivered using this system include peptides and hormones given the drug potency and need for unique dosing schedules. The main challenges for human translation of this system included hermetic sealing of the microchips' drug reservoirs, release mechanism, and aseptic filling of the drug in each well of the microchip [43].

Initial studies in humans were focused on the delivery of human parathyroid hormone fragment (hPTH) indicated for treatment of osteoporosis. The incidence of osteoporotic fractures is extraordinarily high up to 9 million patients per year, as estimated by the World Health Organization (WHO) [44]. At the time of the study, these fractures were estimated to cost up to \$20 billion, and resulted in a significant reduction in quality of life [45]. The clinical trial for the study involved implantation of the device in subcutaneous tissue of 8 female osteoporotic patients between 65 and 70 years of age. The surgical wound was required to heal for 8 weeks in order to enable stable capsule formation. The primary endpoints assessed were the pharmacokinetic (PK) parameters and safety measures of fibrosis, with secondary endpoints being bioactivity of the drug and reproducibility of the drug release. The device was compared to teriparatide injections ( $2 \times 20 \mu\text{g}$ ). In general, the implanted device was well-tolerated by patients and did not require opioid pain medications. It was found that there was comparable PK between the microchip and the FORSTEO injections, with pulsatile response from the hPTH. As the device is expected for prolonged implantation ( $\geq 1$  year), future devices are anticipated to contain tens to hundreds of wells for delivery. In addition, safety mechanisms are expected to be designed to reduce any potential for device failure. There are potentially significant opportunities for use of this device, particularly in situations where frequent injections are required [43].

#### 2.5. Challenges of clinical translation

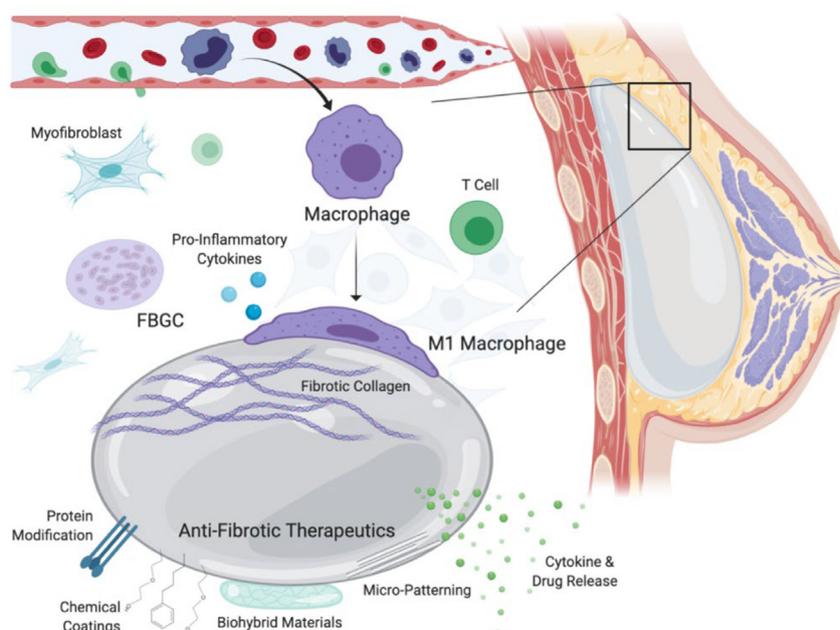
One goal of every drug delivery technology is clinical translation, but major hurdles remain before the technology is translated to the clinic. Each technology may have different requirements based upon the device function, size, length of implantation, and potential toxicities or side effects. Major considerations for translation include intellectual property, manufacturing, overall cost-effectiveness in comparison to current therapies, and of course biocompatibility and safety [20,46]. Beyond biocompatibility, understanding how a patient's body interacts with technologies used in drug delivery will allow for a more thorough mechanistic understanding of these devices *in vivo*, to create targeted, rationally designed therapeutics that function as intended and have long-lasting therapeutic activity.

### 3. The immune system as a target and variable in material design for drug delivery

Advances in bioengineering have come with a wealth of different materials and devices that are able to regain function of missing or non-functional tissues. Pacemakers, glucose monitors, knee replacements, breast implants, all help replace missing form or function. However, when these materials are implanted, our immune system responds by first attempting to phagocytose the material (successfully in the case of nano- and some micro-particles) and if unable to, they then try to degrade the invading material, proceeding to envelop it in a dense fibrotic capsule [47]. This process is known as the foreign body response and can greatly interfere with the function of drug delivery devices [48]. As such, the immune system has not only become a target of drug delivery for direct applications (i.e. cancer immunotherapy, novel vaccine design) but also is an important consideration for all of bioengineering as it can greatly help or hinder the function of medical device implants (Fig. 3).

#### 3.1. The immune system as an adversary

Early in the design of bioengineered materials, the immune system was seen as an adversary. Immune cells require a diabetic patient to move the needle of their insulin pump every three days, they can contribute to the degradation of bone and inflammation resulting in a need for revision surgeries of knee or hip replacements [57], and inflammation around hernia meshes can cause further tissue damage [51]. In the context of drug delivery macro-devices, they can encapsulate and prevent the release of the loaded drug cargo. There have been multiple studies throughout the years focusing on the mechanisms of the foreign body response. Since the 1980's, multiple laboratories have shown that macrophages are critical mediators of this response [58]. Macrophages can secrete oxide radicals, such as nitric oxide, that are meant to degrade the implanted material [59]. Furthermore, these cells can secrete cytokines and chemokines that alter the response of surrounding cells, such as T cells, or stromal cells, such as fibroblasts [60]. These cells can fuse to form multinucleate giant cells (foreign body giant cells and Langerhans giant cells), which are present in other forms of persistent inflammation, such as tuberculosis and xanthoma [61]. This process follows frustrated phagocytosis – in which a macrophage is unable to phagocytose the foreign body – and relies on the cytokine interleukin-4 (IL-4), which has been implicated in various cellular fusion processes including fusion of foreign body giant cells [61–63]. Reports have noted that this fused-cell response is more pathologic than a response that only induces recruitment of mononuclear myeloid cells, possibly due to higher levels of compounds that promote the degradation of the polymer [64]. These pro-inflammatory macrophages and foreign body giant cells (FBGCs) express high levels of M1 macrophage-associated marker CD86, along with MHCII, suggesting communication with the adaptive immune system and are correlated with hypervascularization of the implant and surrounded area [47,65]. Ultimately, if the material is



**Fig. 3.** Approaches to modulating immune responses to implanted materials. Tactics to modulate immune cell behavior have involved protein coatings and surface modifications [49], small molecule chemical coatings [50], hybrid materials with synthetic and biologic polymers [51,52], micropatterning and mechanical cues [53,54], as well as protein and drug release from implants [55,56].

not degraded, these responses lead to the deposition of a dense matrix, composed mainly of fibrillar collagen in a process known as encapsulation. This dense encapsulation can interfere with the intended action of a medical device or, alternatively, result in cell death inside any cell-laden scaffolds or devices.

The foreign body response can have pathologic effects on multiple different types of medical device implants. Broadly these can be viewed as non-integrating and integrating materials. For non-integrating materials, such as cosmetic implants, drug delivery devices, pacemakers, needles of glucose monitors, and similar, the desired interaction with the immune system is either ignorance or tolerance. This means, that either the immune system does not recognize the device at all (ignorance) or recognizes the device and receives signals to be tolerated to the material (tolerance). For integrating materials, such as regenerative medicine scaffolds or cell-based therapeutics that require vascularization for oxygenation, the immune system must assist in scaffold integration to prevent scarring, encapsulation, or excessive damaging inflammation.

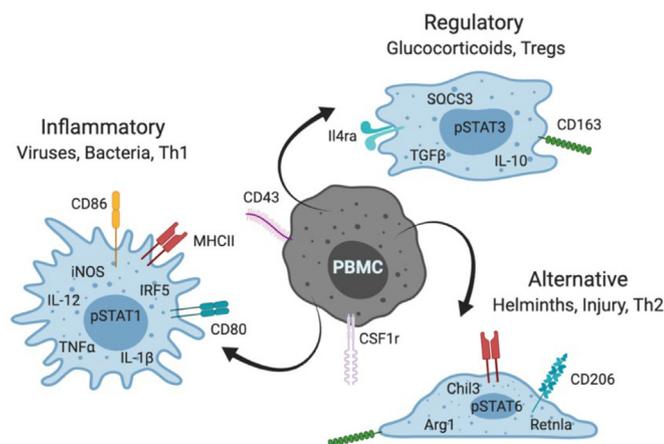
Pathologic responses to medical device implants have resulted in the blocking of translation or requirement of subsequent procedures to mitigate the fibrosis that has occurred due to the foreign body response. Since the initial applications of polymers, including polyvinyl alcohol (PVA) sponges and silicone implants, for plastic and reconstructive surgery in the 1950's, encapsulation and inflammation has been observed around the implants [66]. More recently, approaches for implantation of devices that must interact with the surrounding environment have greatly been hindered by fibrotic encapsulation. Cell-based drug delivery devices for the treatment of type-1 diabetes through implantation of insulin-secreting pancreatic islet cells inside biomaterials, such as alginate, led to a prevention of direct killing of donor cells by cytotoxic T lymphocytes (CD8+ T cells) and natural killer cells, but presented a new issue of fibrotic encapsulation of the device and inhibition of proper nutrient and waste exchange [67–70]. This ultimately leads to poor islet survival and insufficient secretion of insulin [68]. Inhibition of fibrosis by these materials and others used in various applications of bioengineering can potentially be achieved through cell-killing protein coatings such as Fas ligand and anti-Fas antibodies, novel chemical coatings, anti-inflammatory drug release from scaffolds, hybrid materials such as

zwitterionic hydrogels, physical patterning of the surfaces of materials, and novel polymers [49–52,55,71]; however, there are concerns of immune activation not only by the material but also by the compounds secreted by the cells themselves. Recently, scientists have shown an activation of the adaptive immune system by other proteins secreted by donor islets and that an inflammatory reaction can be induced to the medical device without direct contact with the material [72]. Other drug delivery macro-devices, such as those intended for timed release of therapeutics like hormonal birth control or antibiotics, have also been effected by the foreign body response [73].

### 3.2. Engineering biology through modulating macrophage phenotype

As macrophages have been implicated in both the foreign body response and tissue remodeling, these cells have been targeted for a multitude of therapeutics ranging from cancer immunotherapy, to tissue regeneration, to infectious disease [54,74–82]. In the context of implanted devices (such as those intended for long-term drug delivery) the general focus has been on maintaining the M1–M2 macrophage polarization balance (Fig. 4). M1 macrophages as mentioned previously, are inflammatory macrophages that are found during the foreign body response and naturally are important in defense against viruses and bacteria. M2 macrophages, or alternatively activated macrophages do not have the same canonical inflammatory profile, and are found during helminth infection, allergy and asthma, as well as wound healing [83–85]. A major M2-associated cytokine, IL-4 (secreted by Th2 T cells), is implicated both in positive aspects of tissue remodeling [86,87], and in stages of the foreign body response – specifically fusion of macrophages to form FBGCs [62], displaying the heterogeneity of macrophage and immune responses and a need to focus on the function of those cells over the phenotypic categorization. It is important to note, that although the terminology “M1” and “M2” remain for nomenclature of polarization states, their phenotype is not as binary and categorical as this naming convention may suggest [60].

Generally, in tissue engineering there have been strides to either engineer materials to promote an M2 phenotype, or engineer macrophages themselves to adopt a more M2-like phenotype, avoiding the expression of pro-inflammatory proteins such as IL-12, Interferon



**Fig. 4.** Immune cell polarization and activation. Immune cells can activate towards a spectrum of different effector functions dependent upon the challenge that they are faced with. These polarization states are often categorized including Type-1 responses (M1/Th1) the classical inflammatory state that fights viral infection, Type-2 responses (M2/Th2) associated with response to multicellular pathogens and wound healing, and regulatory responses that can inhibit (regulate) other immune polarization states to prevent over-inflammation and tissue damage. Pathologies associated with over-activation of any state includes damaging inflammation (M1/Th1), excessive scarring (M2/Th2), and tumor immune escape (regulatory).

gamma (IFN $\gamma$ ), and Tumor Necrosis Factor alpha (TNF $\alpha$ ) [88,89]. In the context of regenerative medicine, collagen-based decellularized scaffolds promote an CD163+/CD206+ M2-like macrophage phenotype; however, if these scaffolds are cross-linked, they lose their ability to integrate into surrounding tissue and result in high levels of CD86+ M1 macrophages [90,91]. Other studies modulate macrophage behavior via micropatterning to elongate the cells, promoting a cell shape that is similar to the spindle-like M2 macrophage, thereby resulting in a polarization towards an M2-like state [53]. Alterations in mechanical properties of materials (e.g. stiffness) also have an effect on macrophage polarization [65,78,92–94].

Various nanoparticle-based platforms have been utilized to modulate macrophage behavior to decrease inflammatory M1 macrophage activation [95]. Polyethylenimine (PEI), chitosan, and lipidoid nanoparticles have been used to deliver anti-TNF $\alpha$  small interfering Ribonucleic Acid (siRNA), decreasing in TNF $\alpha$  in vitro and modulating disease phenotype in several animal models, including collagen-induced arthritis and LPS-induced colitis [96–98]. Further studies in the context of cancer-based therapeutics have focused on the other side of this coin: promoting an inflammatory (M1) phenotype to inhibit cancer growth and target malignant cells for destruction by the immune system [74]. M2-like macrophages are associated with a poor prognosis in tumor growth, although ultimately it is the presence of immune cells and inflammation (regardless of polarization state) that help fight tumors, with M1-like macrophages having the strongest tumor-fighting effect [99].

Beyond modulating immune cells with changing gene expression through mRNA or siRNA delivery, there have been efforts to modulate their behavior with direct delivery of native or modified cytokines [100–102]. There are limitations to cytokine delivery, due to their short half-life, instability at room temperature, and cost of production of the cytokines themselves [103]. Approaches to some of these issues include direct PEGylation of the cytokines to lengthen half-life and creation of fully synthetic cytokines [104–108]. These designer cytokines can alter the behavior of both innate immune cells, such as macrophages, as well as adaptive immune cells, such as T cells and B cells.

### 3.3. Adaptive immunity and cell engineering

There are many facets of the immune system that can be modulated via drug delivery. Rapid acting macrophages and dendritic cells of the

innate immune system have been targeted for cancer immunotherapy, and until recently were viewed as transient actors of the immune system, though recent work on trained immunity has revealed a memory-like behavior dependent upon epigenetic modifications that can have long-lasting effects on the patient's immune system [109]. In a more classical view of long-lasting immune memory, adaptive immune cells such as T cells and B cells can have life-long remembrance of previous infections, which is a critical facet of disease prevention. The adaptive immune system has long been a target of immune modulation through vaccination [110–112]. Its exquisite specificity and long-term memory are keys to both therapeutics in cancer treatment and infectious disease, as well as autoimmunity [113–116]. Long-term efforts in engineering the immune system have focused mainly in oncology. Cancer immunotherapy, which activates the immune system to attack cancer as opposed to only using chemotherapeutics to kill actively dividing cells, is a class of therapeutics that involves multiple approaches for targeting cancer cells [117–119]. Checkpoint blockade utilizes antibodies to block the action of inhibitory proteins such as programmed death ligand-1 (PD-L1) and (cytotoxic T lymphocyte associated protein-4) (CTLA-4) which are upregulated by tumors [120–125]. A subset of cancers has identifiable antigens, and those can be targeted by chimeric antigen receptor (CAR) T cells. These T cells have been engineered to specifically target a single antigen that is unique to the malignant cell. The first FDA-approved CAR-T therapeutic was indicated for use in B cell acute lymphoblastic leukemia (B-ALL). T cells have been isolated from the patient, then engineered to react specifically against CD19, a protein expressed on the surface of mature B cells [126]. As patients can survive without B cells, CD19 is a viable target for both B-ALL and large B cell lymphoma. Beyond T cells, natural killer (NK) cells have also been engineered to express specific receptors to target cancer cells. CS1-targeted CAR-NK cells were able to prolong survival of mice injected with human IM9 multiple myeloma (MM) cells and suppress tumor growth [127]. CAR NK-92, a clonally expanded NK cell line is efficacious in glioblastoma (mouse, EGFR & EGFRVIII) [128], breast cancer brain metastases (mouse, EGFR) [129], B cell leukemia (mouse, CD19) [130] and refractory acute myeloid leukemia (human safety trial, CD33) [129]. Beyond desired inflammatory effects, researchers have also utilized CAR cells for anti-inflammatory regulatory purposes targeting inflammatory disorders. This includes tolerance to self-antigens, myelin in experimental autoimmune encephalitis (EAE), and exogenous proteins, factor VIII in FVIII-treated hemophiliacs [131,132].

T cells have also been targeted for anti-inflammatory purposes in the context of autoimmune-like conditions. Nanoparticle-based therapeutics have evolved to deliver both tolerizing signals such as rapamycin along with the antigen of interest (i.e. myelin basic protein in mouse EAE models or collagen in LPS-induced arthritis) to educate host immune cells towards a more tolerogenic phenotype [133–136]. Other approaches have involved delivering tolerizing cytokines naturally produced by the body to promote regulatory responses which have been reported to be critical beyond autoimmunity in both medical device acceptance and wound healing [137]. Delivery of IL-10 (normally secreted by regulatory T cells) has been used in animal models to prevent fibrosis in pathologies involving the liver, lung, and kidney [138–140]. Engineers have also utilized indolamine-2,3-dioxygenase (IDO) which is secreted by dendritic cells to inhibit T cell function [141]. Anchoring IDO to tissue with galectin-3 associates IDO with carbohydrates, specifically  $\beta$ -galactosides within the extracellular matrix [142].

Ultimately, adaptive immunity is an important consideration in material design due to long-lasting memory of adaptive immune cells. Adaptive immune cells themselves are notoriously difficult to target for gene editing and have long-lasting effects on other immune cells and surrounding tissue. Though the innate immune system has been the focus of immune interactions with materials since seminal research in the 1980's, ultimately it is an interaction of the innate and adaptive immune system along with the surrounding tissue that affects the outcome of the implanted device or material.

### 3.4. Beyond biocompatibility: integrating mechanistic biology with material design

Advances in technology for immune cell analysis have led to the ability to analyze many parameters from the level of RNA to protein. These technologies, such as single-cell RNAseq and high-color flow cytometry, have led to new discoveries in the context of immune cell function and subsequent modulation [143–146]. As the immune system is a large network that reaches every organ of the body (even so-called immune-privileged organs have since been discovered to have resident immune cells and lymphatics [147,148]) these high-dimensionality studies can help uncover previously unknown mediators of immune responses in homeostasis and disease. Applications of multi-color flow cytometry to the field of bioengineering have led to notable discoveries and a more thorough understanding of the way immune cells interact with materials used in development of new medical devices and therapeutics [143].

The immune system is a ubiquitous component of all tissues in our body, whether this is through peripheral blood-associated circulating immune cells, tissue-resident immune cells, or the immune activity of parenchymal and stromal cells. Any study of our body in health and disease must take into account actions of immune cells and immune-active proteins. In the context of the development of new materials for tissue engineering and drug delivery, immune cells are often a variable in materials design, and a target for modification of their behavior. In terms of the former, immune cell responses such as production of reactive oxygen species can accelerate the breakdown of polymers used for slow release of drug cargo [8]. They can also induce fibrosis around an implant to block the release and transfer of the drug to the surrounding environment, thereby altering the kinetics of release both by increasing or decreasing the release rate. Through understanding the way that our immune system interacts with engineered materials, we can then hone our development of novel materials to both target immune cells and modulate their phenotype, and account for the action of immune cells in response to those materials (Fig. 5). Next generation materials design is focusing in-part on both developing both novel chemical methods for increasing our library of polymers and their functionalization, as well as leveraging this biologic knowledge for immune-active therapeutics and biocompatible materials.

## 4. New approaches to polymer design

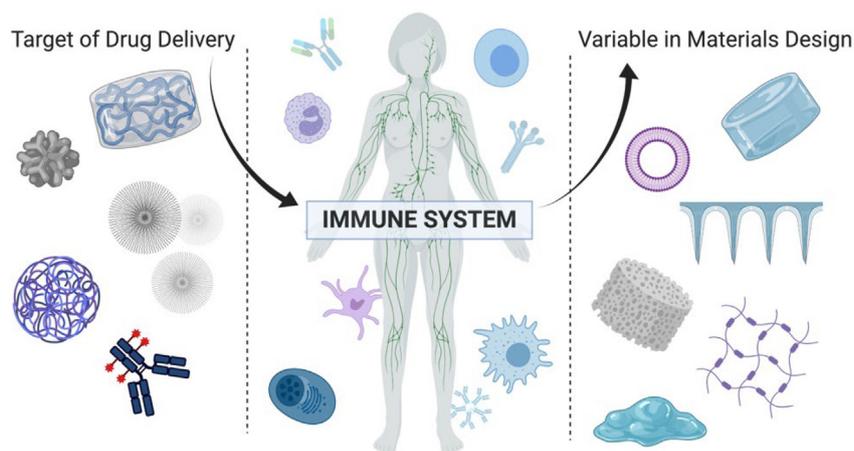
To properly deliver and target drugs, maintain their function within the human body, and ensure safety and biocompatibility, next

generation therapeutics rely on the development of new materials and loading strategies. Polymeric materials, lipids, and implanted devices can address some of the challenges mentioned above, and consequently, it is rare to find modern therapeutics that are administered without modification or formulation with polymeric materials or lipids [151]. Polymers, for example, can increase drug solubility, lengthen blood circulation time, extend and tune drug release kinetics, achieve targeted/stimuli-responsive drug release, and assemble into ordered nanostructures for drug-packaging and delivery [151]. While great progress has been made in therapeutic development, significant drawbacks limit the efficacy of currently available drugs. These include non-specific drug release leading to severe off-target side-effects, inconsistent and ineffective drug release kinetics, the use of non-biodegradable polymers that accumulate in vital organs/tissue, or the degradation of polymers into unwanted by-products. To address these challenges demands the development of new materials which are safe, compatible with all biological systems, have tunable and precisely controlled pharmacokinetic profiles, specific targeting, and can deliver multiple therapeutics simultaneously [152–158]. In this section we highlight new advances in synthetic polymer chemistry, with a focus on living polymerizations, for the preparation of unique drug-polymer conjugates and self-assembled nanostructures that address the limitations of current drug delivery systems and generate new therapeutics to improve human health.

### 4.1. Reversible-deactivation radical polymerization (RDRP)

The development of reversible-deactivation radical polymerization (RDRP) holds immense potential for applications in drug delivery. Over the last two decades, RDRP techniques such as atom transfer radical polymerization (ATRP) [159], reversible addition–fragmentation chain transfer (RAFT) polymerization [160], and nitroxide-mediated radical polymerization (NMP) [161] have been studied in great detail and applied to almost all areas of biomedicine [162–164]. RDRPs achieve controlled polymerization by controlling the equilibrium between active (growing) and dormant (not-growing) polymer chains. Several comprehensive reviews are available describing the mechanism of each RDRP [159–161]. Here, we present select examples to introduce how RDRPs can be used to achieve superior therapeutic effects for applications in drug delivery.

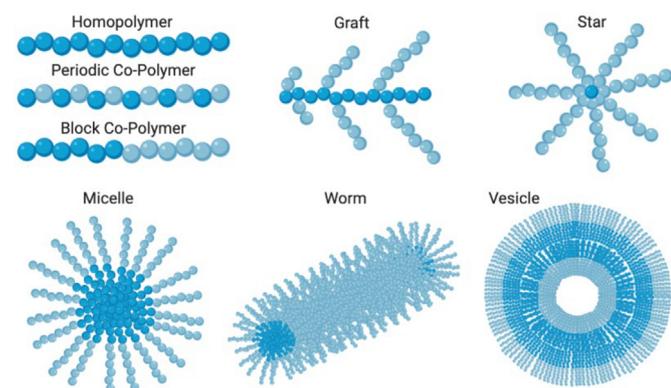
One of the applications of RDRP for drug delivery lies in the ability to prepare precisely designed and functionalized polymers and self-assembled nanostructures. These include single molecular architectures



**Fig. 5.** Immune cells as targets and variables in drug delivery material design. The extent of the immune system makes it a target of drug delivery through nanoparticles [89,135,136], scaffolds [82], biologics [121,149], treating cancer [74,125], infection [27], and tissue damage [86]. Downstream effects of immune responses to materials can also modulate release kinetics or ultimate targets of the drug delivery, through expedited degradation of implants [8], phagocytosis or inflammatory response to nanoparticles [150], and fibrotic encapsulation of devices [58,73].

such as brush/graft polymers, block co-polymers, cyclic polymers, star polymers, and dendrimers as well as self-assembled nanostructures such as micelles, vesicles/liposomes, worms, lamellae, (Fig. 6) [165–169]. By applying RDRPs, the size, number, and nature of the component polymer segments can be controlled at each stage of the synthesis allowing incredibly fine control over the resulting self-assembled structure and enabling the incorporation of functional/stimuli-responsive polymer blocks [170]. Different polymer architectures have different drug release/pharmacokinetic profiles [13,162,170–175], so being able to design and examine precisely prepared structures will enable new drug delivery platforms to be developed.

Non-uniform polymers and nano-structures behave differently compared to their uniform counterparts [176–178]. For example, ellipsoidal shaped nanoparticles displaying antibodies on their surfaces were more effective at antigen specific induction of T-cells than the spherical control [178]. In relation to biomimetic structures, novel polymer architectures can more accurately mimic biological functions. Brush polymers often more accurately represent mucus membranes, cilia, and synovial fluid than linear polymers [176,179]. The recent report by Ke et al. highlights the importance of not only size but shape when designing new materials for drug delivery [13,175]. To showcase the therapeutic effectiveness of non-spherical nanostructures, block copolymers composed of a hydrophilic poly(ethylene glycol) (PEG) block and a hydrophobic, drug-loaded polymer block of thioketal-linked camptothecin methacrylate (CPTKMA) and 2-(pentamethyleneimino) ethyl methacrylate (PEMA) were prepared via RAFT polymerization. The thioketal linkage is sensitive to reactive oxygen species which are found in high concentrations in tumor tissue, achieving tumor targeted drug release. The self-assembly of the block copolymer, PEG<sub>113</sub>-b-P(CPTKMA<sub>0.53</sub>-co-PEMA<sub>0.47</sub>)<sub>43</sub>, into spherical micelles (diameter of 40 nm) was achieved by the direct addition of a block copolymer/tetrahydrofuran (THF) solution to phosphate buffered saline (PBS) while long filamentous micelles (filomicelles, diameter of 40 nm and length of 2.5 μm) were prepared by the slow addition of PBS (1 mL/h) to the block copolymer/THF solution. To compare how filomicelle length affects drug delivery, short filomicelles (diameter of 40 nm and length of 180 nm) were prepared by controlled sonication of the long filomicelles. Interestingly, the short filomicelles demonstrated the highest level of cell internalization apparently due to the greater contact area with the cell surface. Additionally, the short filomicelles exhibited longer blood circulation times than the long filomicelles ( $t_{1/2} = 14.4$  h compared to 9.7 h) as well as the deepest penetration and most effective antitumor activity of all three nanostructures (55% tumor diameter reduction as compared with a 38% increase in tumor diameter for the long filomicelles and a 36% reduction with spherical micelles). It was



**Fig. 6.** Controlled polymer architectures enabled through RDRP. Examples include homopolymers of variable lengths, block copolymers comprising two or more distinct polymer blocks, graft or brush polymers with tuneable brush length, star polymers with the desired number and length of polymer arms, and self-assembled structures such as micelles, liposomes/vesicles, and worms.

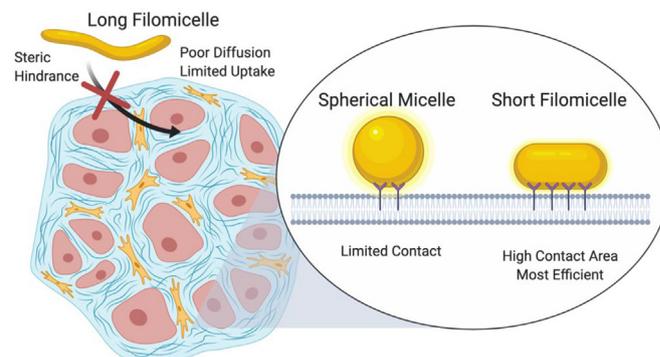
thought that the longer filomicelles perform poorly due to their large size which limits cell uptake and tumor accumulation (Fig. 7). This report highlights the need to investigate exotic, bio-inspired nanostructures capable of being prepared via RDRP and which may hold greater drug delivery capability than common spherical particles.

Beyond filomicelles, to demonstrate how polymer architecture can have a dramatic effect on function, we focus on the high DNA transfection efficiency of graft, star, and branched polymers as compared to their linear counterparts [173,180,181]. In one example, Li et al. prepared a poly(glycidyl methacrylate) (PGMA) polymer functionalized with poly((2-dimethyl amino)ethyl methacrylate) (PDMAEMA) sidechains. ATRP afforded precise control over the length of the sidechains, which were increased from 14 (approximately 2000 Da) to 57 repeat units (approximately 9000 Da), corresponding with an increased ability to complex DNA, likely due to the greater cationic charge on the polymer side chains, and an increased transfection ability when compared to the linear and polyethylenimine controls [181]. Again, the complex and highly controlled polymer architectures achievable via RDRP techniques allow one to explore new polymer structure-function relationships and develop unique materials for biomedical applications.

#### 4.2. RDRP for drug-polymer conjugation

In addition to precise control of polymer architecture, RDRP has also been applied with great success to drug-polymer conjugation [182]. The conjugation of therapeutic agents to polymeric carriers, e.g. polyethylene glycol, offers several advantages including increased blood circulation time, reduced immune response, targeted drug delivery, and increased solubility and safety which is evident by the numerous conjugated therapeutics translated into clinical use [183]. Before the advent of RDRP, polymer conjugation to drug molecules was limited to relatively simple systems and the dispersity of polymer molecular weights, composition, and structure would often limit translational success [182]. Through RDRP we now can prepare complex and precisely tuned drug-polymer conjugates incorporating functional monomers leading to improved therapeutic effect [182].

Traditionally focused on small molecules [182], recent developments in RDRPs that can be performed under biologically benign conditions made possible the synthesis of well-defined drug-polymer conjugates where the drug of interest is a biomolecule (DNA, RNA, proteins, and peptides) [184–186]. Polymer-biomolecule conjugates combine the therapeutic traits of the biomolecule with the functional ability of the synthetic polymer [184]. The Maynard Group has used ATRP and RAFT polymerization to prepare well-defined insulin–trehalose glycopolymer conjugates which show enhanced stability and



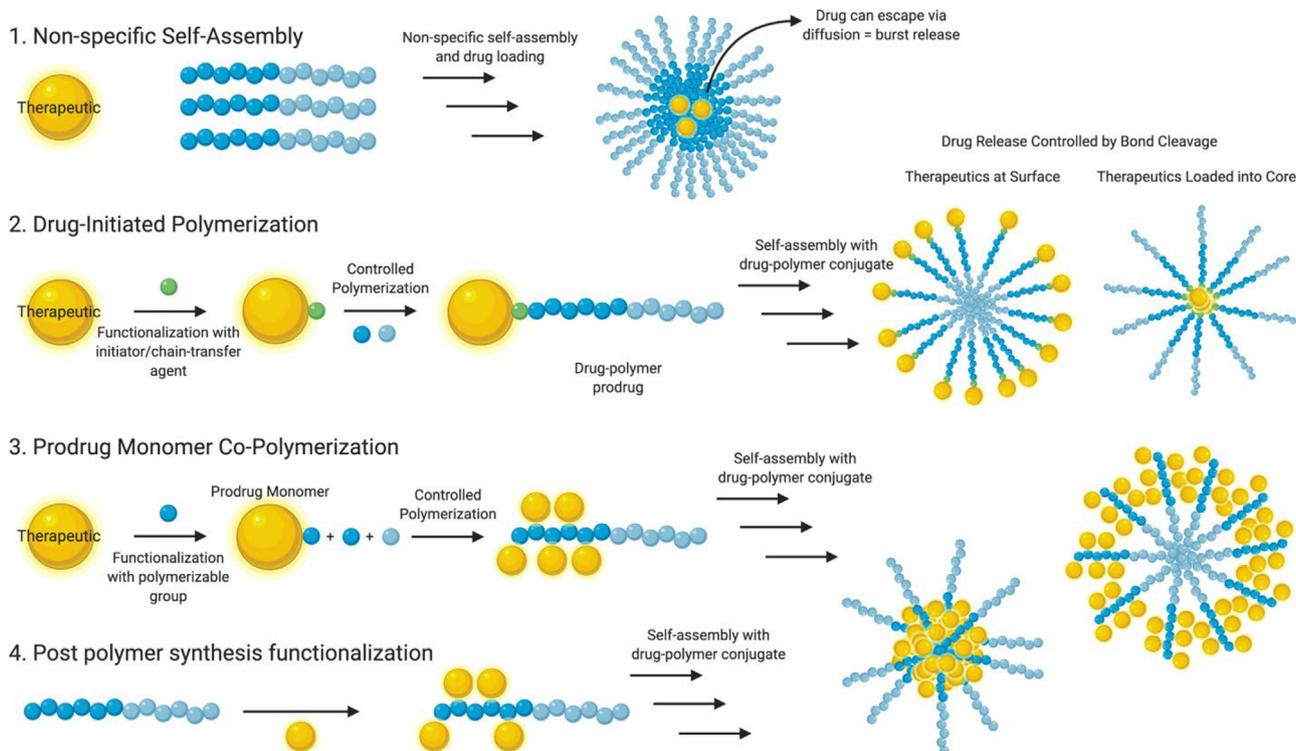
**Fig. 7.** Anti-tumor nanoparticle efficacy is determined by size, shape, and morphology. Large filomicelles cannot penetrate the tumor tissue due to their size, spherical micelles have limited contact with the cell surface, but short filomicelles have increased contact area and are not burdened by steric hindrance.

activity when compared to unfunctionalized insulin [187,188]. Initially, ATRP of a methacrylate-trehalose functional monomer was performed under biologically compatible conditions (23 °C) with complete conversion occurring in 4 h. The trehalose-polymer displayed excellent excipient stabilization to heating (90 °C, 30 min) with  $93 \pm 3\%$  intact insulin compared to 45% for the unmodified control, demonstrating that the polymer prevented heat-induced aggregation of insulin [188]. The free amine groups of insulin were then modified to possess a brominated-initiator from which the trehalose-polymer chains could be directly grown via ATRP. Polymer chains of 8.7 kDa (PDI 1.21) were grown from the insulin biomolecule and the bioactivity of the conjugate was compared to both free insulin and insulin conjugated to 10 kDa PEG. The trehalose- and PEG-insulin conjugates performed similarly, requiring a 3-fold smaller dose to achieve the same change in blood glucose concentration relative to insulin alone (48 vs 16  $\mu\text{g}/\text{kg}$ ).

As described in Sections 3.2 and 3.3, targeting drugs to specific immune cells holds great potential for immune therapies, vaccine development, and cancer treatment. However, targeted drug delivery to immune cells is highly complex and remains a significant challenge [189]. To address this, the Hubbell group has used RAFT polymerization to prepare glycosylated polymeric antigens which are able to target dendritic cells and T cells to stimulate a robust and highly specific immune response (Fig. 8) [14,190]. Polysaccharide modifications can be both protective (as seen with trehalose) and used as targeting agents to immune cells that bear sugar-sensing receptors on their surface. In one instance, a vaccine platform based on a highly functional methacrylamide polymer was prepared and found to improve the immunogenicity of protein subunit vaccines [14]. Copolymerization of a mannose-functionalized methacrylamide monomer, able to target mannose-binding receptors on dendritic cells, together with a methacrylamide monomer functionalized with a Toll-like receptor (TLR) 7 agonist which induces dendritic cell activation was performed using RAFT polymerization. Precise control over the polymer length and ratio of the functional monomers could be achieved as well as

incorporation of an active chain-end functionality allowing the polymer to be conjugated to the antigen of interest via highly efficient and biorthogonal click chemistry. The final polymer had a molecular weight of 18 kDa and was composed of a 1:2.1:3.5 mol. Ratio of TLR7:mannose monomer:HPMA (N-(2-hydroxypropyl) methacrylamide). Importantly, owing to the targeting and stimulation afforded by the functional polymer, the antigen-conjugate was found to promote a strong CD8+ and CD4+ T-cell response through the amplification of antigen presentation on the surface of dendritic cells, expand neutralizing antibodies and memory B-cells, and avoid a systemic inflammatory response [14]. The benefits of RAFT polymerization (tolerance to a variety of functional groups, precise control of polymer length and co-monomer ratio, and control over end-group functionality) are displayed here and without such a technique the complex and highly tuned material design could not have been achieved.

The preparation of polymer-drug assemblies (micelles, liposomes, etc) with tunable loading, controlled released kinetics, and coordinated delivery of multiple therapeutics remains a formidable challenge in drug delivery. In some cases, drug encapsulation in nanoparticle systems is achieved through self-assembly of component drug and polymer molecules in solution. This leads to significant disadvantages including low loading levels, difficulty in loading poorly soluble/miscible drugs, and the often-seen “burst release”, which involves a significant and immediate release of a large fraction of the adsorbed drug resulting in drug loss and toxic side-effects. Many current therapeutics suffer from the burst release, including DOXIL, the FDA-approved liposomal formulation of doxorubicin (described in Section 2.2) [191,192]. By preparing stimuli-responsive, drug-conjugated monomers, which are polymerized into a polymer-drug conjugates that self-assembly into polymer-drug assemblies, the drug loading can be precisely controlled and the burst release effect can be minimized [193–196]. RAFT polymerization and NMP have been successfully used to prepare drug-polymer nanoparticles via “drug-initiated” polymerization [193,197–199] (where the drug is located at the end of the polymer



**Fig. 8.** Approaches to drug loading in polymeric materials. Several approaches have been identified and optimized to load drugs into polymeric delivery devices, either non-specifically, or through polymer functionalization and organized particle assembly.

chain) and main-chain polymerization (where the drug is located along the polymer sidechains) [194–196,200,201].

In one case, Son et al. employed RAFT polymerization to prepare prodrug polymers carrying the chemotherapeutic, camptothecin, and the kinase inhibitor, dasatinib [195]. The drug molecules were first linked to a methacrylate monomer via a hydrolysable ester bond to form the prodrug monomer. Using RAFT polymerization, the prodrug monomers were directly incorporated into the polymer backbone and the dose of each drug could be independently tuned by changing the monomer feed ratio. Initially, polymer prodrugs comprising 24 mol% camptothecin and 27 mol% dasatinib were prepared as random copolymers together with PEG methacrylate (49 mol%). The authors went on to prepare the diblock copolymer version composed of a hydrophilic PEG block and a hydrophobic drug-polymer block. Owing to the amphiphilic nature of the diblock copolymer, self-assembly into polymer nanoparticles was observed with the drug-loaded polymer block being localized within the hydrophobic core. As a result, drug release from the block copolymer assemblies was significantly slower than from the random copolymer (50% drug release after 10 days for the random copolymer which increased to 20 days for the assembled block copolymer). Importantly, due to the direct conjugation of the drug to the polymer chain, very minimal burst release was observed. Instead, a constant rate of drug release was achieved for both polymer architectures. Here, just by changing the polymer structure from a random, unoriented copolymer to a self-assembled block copolymer, one can achieve significantly different drug release profiles from the same constituent material.

#### 4.3. New polymer architectures

Looking to alternative synthetic techniques for the preparation of novel self-assembled structures formed from drug-polymer conjugates, the Johnson group used ring opening metathesis polymerization (ROMP) to prepare brush-arm star polymers (BASPs) which self-assemble into unique polymer nanoparticles [202–206]. The highly specific BASPs are prepared by ROMP of functional drug-monomer conjugates. The drug is conjugated to the norbornene-monomer through a variety of stimuli responsive bonds to achieve controlled drug release in response to water (hydrolysis) [202], light [202,206], or specific enzymes [207]. Importantly, by altering the monomer feed ratio, precise and tunable drug loading can be achieved. In one instance, a single BASP nanoparticle was designed to carry a precise dose of camptothecin (2.07 equiv.), doxorubicin (0.83 equiv.), and cisplatin (3.0 equiv.) with drug release achieved in response to culture media or UV light. The cytotoxic activity of the triply loaded BASPs was found to be higher than BASPs prepared with only one or two for the formulation with cisplatin and CPX or  $IC_{50} = 93 \pm 11 \mu\text{g BASP/mL}$  ( $14 \pm 1 \mu\text{M drug}$ ) for the formulation with cisplatin and DOX) of the drugs when tested against OVCAR3 human ovarian cancer cells [202].

Through the highly specific and rationale design of polymer-drug conjugates, new treatments for currently untreatable conditions can be developed. Telmisartan, a small-molecule antihypertensive drug, was recently incorporated into BASP nanoparticles to treat chronic liver fibrosis, a condition which has no FDA approved treatment [207]. Currently used to treat high blood pressure, telmisartan also displays antifibrotic activity but has limited clinical use because it causes systemic hypotension. Golder et al. overcame the unwanted systemic effects of telmisartan by preparing telmisartan-conjugated BASP nanoparticles [207]. Telmisartan conjugation through an esterase-sensitive linker achieved targeted drug release in the fibrotic liver microenvironment with minimal drug release observed in the blood (< 15% after one week). In this way, reduction of liver fibrosis could be achieved without causing systemic hypotension. Importantly, the rate of telmisartan release could be increased threefold by changing the structure of the ester bond which links the telmisartan to the BASP and the synthesis could be performed on large (100 g) scale. The ability

to achieve controlled, targeted drug release from BASPs highlights how new the development of new polymer-drug conjugates will be critical when designing the next generation of drug delivery systems and how they can be used to overcome severe side effects which traditionally limit the use of clinical therapeutics.

Solid polymeric depots (such as Gliadel wafers and stents reviewed in Section 2.3) can deliver a large dose of therapeutic over a prolonged period. However, these drug eluting depots require surgical or interventional implantation at the site of interest and have limited drug loading and release kinetics. To avoid surgical implantation, injectable hydrogels have been developed which are minimally invasive and are offer new therapeutic treatments for complex, sensitive, and small areas of the body, such as the eye and ear [208,209]. Injectable hydrogels spontaneously gelate after injection through a chemical reaction, temperature change, or self-assembly [210]. One class of injectable hydrogel, based on non-covalent polymer-nanoparticle interactions, has been developed by the Appel group [211,212]. These hydrogels are shear-thinning allowing for flow under applied stress (during injection) with complete recovery upon stress relaxation (post injection). The hydrogels are composed of amphiphilic, biodegradable materials (hydroxypropyl methylcellulose and PEG-PLA nanoparticles) allowing for the loading of both hydrophilic and hydrophobic molecules with drug release controlled by either diffusion or surface erosion. This new class of material offers a simple means of preparing injectable hydrogels capable of delivering a range of therapeutics, but work remains to achieve drug release over longer periods of time. Interestingly, the Appel group applied their supramolecular polymer-nanoparticle hydrogel system to the treatment of post-operative pericardial adhesions with very promising results [213], indicating the necessity of investigating new polymer architectures and their wide translational potential.

#### 4.4. Approaches to improve the biocompatibility of injected/implanted materials and devices

Although used throughout biomedicine, a major health concern of materials prepared via free radical polymerization, RDRP, and ROMP is the non-biodegradable polymer backbone which can lead to bioaccumulation in specific organs and stimulation of the immune system. Currently, ring-opening polymerization (ROP) of cyclic monomers is the most common way to achieve completely biodegradable polymers which possess hydrolytically degradable polyester or polyamide backbones (common examples include poly(lactic-co-glycolic acid) and polycaprolactone) which has made ROP-synthesized polymers the material of choice for biomedical applications [214,215]. A major drawback of ROP-synthesized polymers is their narrow tolerance to different functional groups which can affect the polymerization and severely limit the materials application. Considering this, work is focused on developing new synthetic methods to incorporate biodegradability into ATRP, RAFT, and ROMP-based polymers [216–218].

Recently, Shieh et al. prepared a new cyclic monomer incorporating a hydrolytically degradable silyl-ether bond [15]. The biodegradable monomer was compatible with ROMP thereby imparting a biodegradable linkage into traditionally non-biodegradable polymer backbone, solving one of the largest problems associated with this family of ROMP-based polymers and opening them up to new biomedical applications. The hydrolytic degradation of the silyl ether bond can be tuned by increasing the hydrophobicity of the groups bound to the silicon atom, increasing the lifetime and blood circulation of the ROMP-based polymer. At pH 5.5, the lifetime of the most hydrolytically unstable silyl ether was less than 1 h which increased to around 15 days for the most hydrolytically stable. To demonstrate the application of this new material for biomedical applications, fluorescently labelled silyl-ether-based ROMP polymers were intravenously injected to BALB/c mice and the biodistribution monitored for 10 weeks. Compared to the non-degradable control, the silyl-ether based ROMP-polymer was found in six-fold less amounts in blood circulation (% ID was approximately 3% for the non-degradable control and 0.5% and

0.8% for two types of degradable BASPs at 3 weeks which reduced to 1.5% for the non-degradable control and 0.15% and 0.4% for the degradable BASPs at 6 weeks). Furthermore, significantly less accumulation of the degradable BASP was observed in the liver and spleen after 3 and 10 weeks when compared to the non-degradable control. This exciting advance may lead to the application of ROMP-based polymers in a variety of different biomedical applications and highlights how innovative monomer design can expand classic materials to new applications.

As discussed in Section 3.1, the foreign body response can prevent the function of certain drug delivery implants as the body produces a dense fibrotic matrix to surround and isolate some implanted materials. Efforts to treat type-1 diabetes, a condition of unregulated blood glucose concentration which affects millions of people worldwide, has been severely hindered by the foreign body response. Patients with diabetes frequently use implanted devices such as continuous glucose monitors and insulin pumps.

In light of this, significant research has been focused on materials that minimize the foreign body response or are “invisible” to the immune system and therefore do not promote fibrotic encapsulation or biofouling. In one example, Xie et al. used RAFT polymerization to prepare a library of zwitterionic polymers, then coated the surface of implanted glucose monitors to reduce biofouling and increase the performance of the device [219]. RAFT polymerization realized the preparation of controlled polymers with tunable molecular weights and monomer compositions. The most effective polymer had a molecular weight of 10 kDa and was composed of 2-(methacryloyloxy)ethyl 2-(trimethylammonio)ethylphosphate and lipoic acid methacrylate which reduced biofouling and improved the performance of the implanted glucose sensor.

To decrease the dependence on frequent injections and needlesticks, one potential treatment of type-1 diabetes involves the implantation of encapsulated pancreatic islet cells to restore the regulation of normal blood glucose concentrations. In some cases, pancreatic cells are encapsulated in hydrogel microspheres which allow the exchange of nutrients between the encapsulated cells and the environment and creates a physical barrier to separate the foreign cells from the body's immune system. While promising for the treatment of diabetes and other conditions, the foreign body response has limited clinical translation due to the production of a fibrotic capsule around the implanted cells which prevents nutrient exchange causing cell death. To address this, Vegas et al. performed a high throughput combinatorial screen of modified alginate to identify compounds which elicit no foreign body response when implanted in the body [50]. Over seven hundred alginate analogues were prepared via esterification, amidation, or click chemistry with three lead candidates sharing a short PEG-azide linker with either a triazole-thiomorpholine dioxide, a triazole-tetrahydrofuran, or a triazole-aniline end group. These data may indicate that the triazole group could be effective in mitigating foreign body response, but further research would be necessary to make any conclusions. The use of high throughput screen together with highly efficient chemistry has allowed for the preparation of large numbers of modified polymer analogues which show promise as a platform material for the mitigation of the foreign body response.

It is through interdisciplinary collaboration that some of the greatest challenges facing biomedicine and human health will be solved. The above discussion aims to introduce new synthetic methods for polymer synthesis to scientists not familiar with them and to inspire the collaboration between materials scientists, biologists, engineers, and clinicians. To translate front edge science to the clinic we must learn from current clinical products, consider new developments in biological research, and develop new synthetic techniques which overcome the limitations of current materials.

## 5. Conclusions

Understanding of the reactions of our body with materials that we use for drug delivery, and how those ultimately affect the function of

those materials, can be integrated with new advances in materials and polymer chemistry to drive forward medical innovation. Clinical applications of drug delivery devices have led to advances in the treatment of cancers and infectious disease, through chemotherapeutic delivery and immunomodulatory drugs. Modulation of the immune system has become central to many diseases and is also an important factor in development of devices that are not directly intended for immune engineering. Approaches to modifying immune responses and paving way for new therapeutics are also advanced by new techniques in polymer chemistry that increase our abilities to load and modify polymers, at the same time taking into account biocompatibility and the effect of the patient's immune response to those materials.

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