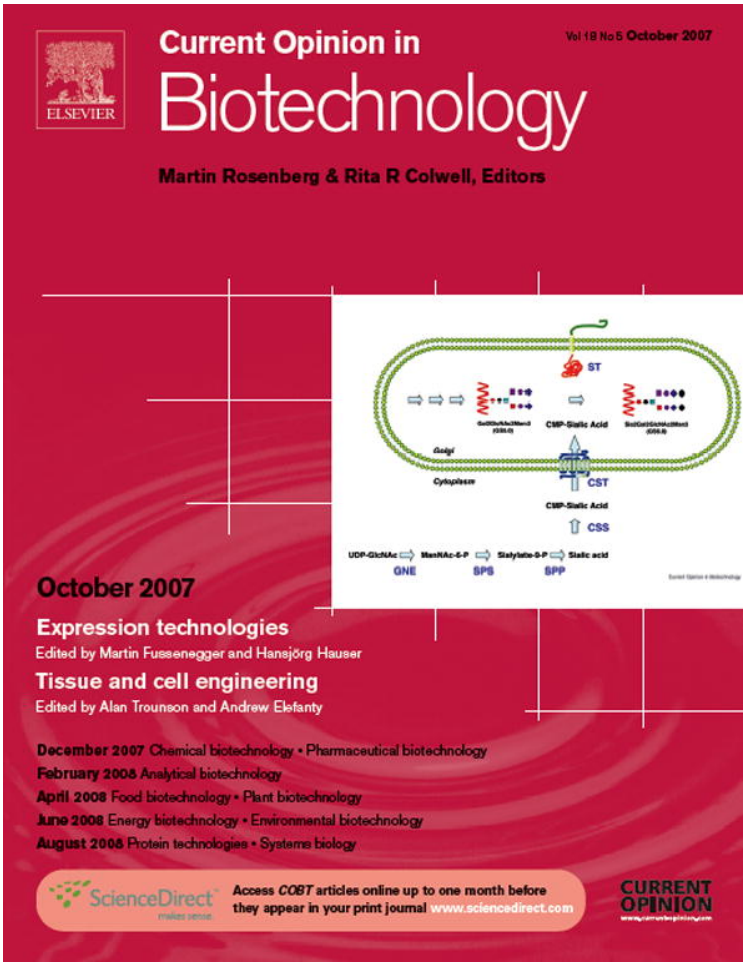

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# Development and therapeutic applications of advanced biomaterials

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Millions of patients worldwide have benefited from technological innovation from biomaterials. Yet, while life expectancy continues to increase, organ failure and traumatic injury continue to fill hospitals and diminish the quality of life. Advances in understanding disease and tissue regeneration combined with increased accessibility of modern technology have created new opportunities for the use of biomaterials in unprecedented ways. Materials can now be rapidly created and selected to target specific cells, change shape in response to external stimulus, and instruct tissue regeneration. Here we describe a few of these technologies with emphasis on targeted drug delivery vehicles, high-throughput material synthesis, minimally invasive biodegradable shape-memory materials, and development of strategies to enhance tissue regeneration through delivery of instructive materials.

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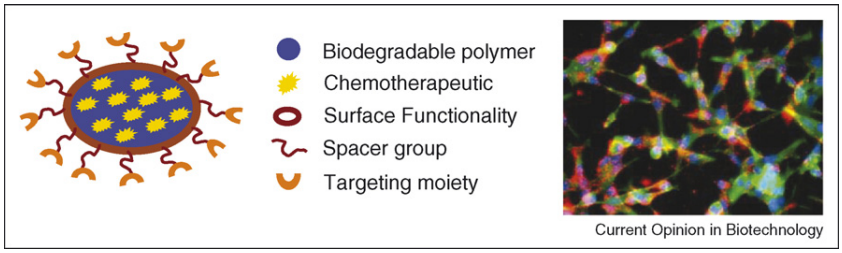
## Introduction

Progress in surgical techniques, advancements in computer-assisted surgery, and advanced biomaterials shape all forms of modern medicine. Biomedical innovation is driving more accurate and early diagnoses, less invasive and quicker procedures, fewer hospital visits including shorter stays, and reduced complications. With the development of high-speed computers, establishment of the Internet, and birth of the modern fields of proteomics, genomics, and nanotechnology, we have entered a new era where advanced technology and knowledge are more accessible than ever before. Most laboratories can now easily fabricate microdrug and nanodrug releasing particles, develop new materials, create Biological MicroElectrical Mechanical Systems (BioMEMS) based on soft lithography and interface these technologies with various cell types including adult and embryonic stem cells. Through the convergence of multiple fields including chemistry, materials science, biology, engineering, and medicine into a discipline broadly defined as bioengineering or biomedical engineering, collaborative innovations have produced targeted drug delivery nanoparticles for therapeutics and imaging [1•], the development of materials that respond to physiologically regulated [2•] or external stimuli [3], advanced sensing and imaging modalities [4], high-throughput technologies for developing and discovering new materials [5••] or studying cell–material interactions [6••], immobilization techniques for anchoring biomolecules to surfaces [7], and the biomedical translation of microfabrication and nanofabrication technologies (from the microelectronics industry) which has been used to simulate physiologically and pathologically relevant cellular microenvironments [8••,9]. Millions of patients worldwide have benefited from technological innovation from biomaterial-based products including controlled drug delivery devices, joint replacement and dental implants, endoluminal stents, pacemakers, artificial hearts, contact lenses, surgical adhesives and antiadhesives, vascular grafts, and contrast agents for imaging. New uses of biomaterials continue to be discovered and engineered, however, chronic and degenerative diseases provide a persistent burden to mankind as the population continues to age and as health care costs rise. Although life expectancy continues to increase and converge for most of the world [10], debilitating disease (e.g. cancer, HIV, cardiovascular disease, etc.), organ failure, and traumatic injury (i.e. from car crashes, acts of violence, accidental tissue damage, etc.) continue to fill hospitals and diminish the quality of life. There are many advanced biomaterial approaches on the horizon that are poised to transform the future of patient care. Here we describe a few of these technologies with emphasis on targeted drug delivery vehicles, high-throughput material synthesis, minimally invasive biodegradable shape-memory materials, and development of strategies to enhance tissue regeneration through delivery of instructive materials.

## Development of targeted drug delivery devices for cancer therapy

Treatment of cancer typically involves combinations of drugs, surgery, and/or radiation and its success depends on

**Figure 1**



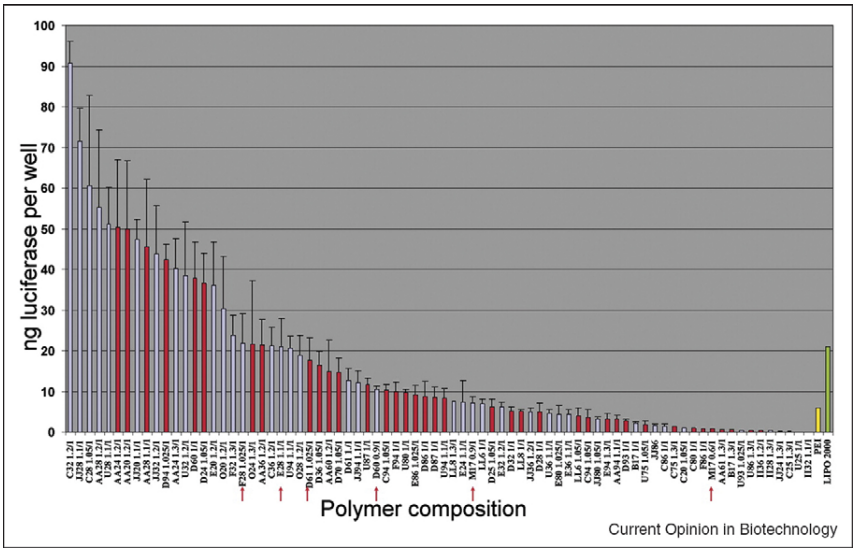

(Left) Targeted drug delivery nanoparticles for achieving high drug doses per biorecognition event. Cytotoxicity to cancer cells is maximized through incorporation of targeting agents that have high specificity for particular antigens on the surface of cancer cells, while minimizing nonspecific cell interactions. Additional functionalities including spacer groups such as poly(ethylene glycol) may be used to enhance the circulating half-life after intravenous administration. (Right) Prostate cancer cells that have internalized fluorescently labeled nanoparticles (shown in red). The cells' nuclei and cytoskeletons are stained blue and green, respectively (image on right courtesy of Omid Farokhzad).

the ability to destroy cancer cells with the least collateral damage possible. More effective targeted cancer therapies are clearly needed to develop 'smart delivery vehicles' that will enhance survival and minimize adverse effects. An emerging strategy which holds great promise involves nanoparticle conjugates (Figure 1), also referred to as 'nanovectors' for targeting metastatic cancer through the delivery of drug laden nanoparticles conjugated to targeting moieties [11,12••,13]. Through utilizing various types of biomaterials, chemotherapeutics can be programmed to release with specific profiles including continuous and pulsatile release, and through controlling the size, charge, and presentation of surface moieties, the particles can be programmed to be internalized by the cell. Recently, nucleic acid ligands (referred to as aptamers) that bind to target antigens with high specificity and affinity were used as targeting molecules to deliver chemotherapeutic containing microparticles to tumor-antigens, present on the surface of prostate cancer cells [11,14••]. These bioconjugates successfully and selectively adhered to prostate-specific membrane antigen (PSMA)-positive prostate cancer cells, while PSMA-negative cells were not targeted. Using a xenograft nude mice model of ectopic human prostate cancer, a single intratumoral injection of docetaxel–nanoparticle–aptamer bioconjugate nanoparticles completely eradicated the tumors in five of the seven treated animals, and the remaining animals also had significant tumor reduction, compared to the controls. Targeted drug delivery represents one of the most active areas of advanced biomaterials. Although systemic targeting represents the 'holy grail' in cancer and other disease targeting approaches, nonspecific uptake after systemic administration continues to challenge those in the field to the development of new advanced material-based strategies. Given the complexities involved and relative time-consuming material development approaches currently employed, use of high-throughput strategies is in great need to develop property–performance correlations for nanodevices [15] and new materials.

## High-throughput generation of polymers for specific medical applications

Advanced polymeric materials have become extremely complex in chemistry, structure, and function and this poses significant optimization challenges. Since it is often cumbersome and nonobvious how to determine the best possible material properties for each application, more efficient combinatorial discovery and optimization methods of functional materials, including characterization methodologies, have recently been developed. Combinatorial approaches are useful when precise correlations between the basic design variables and the performance outcomes are not available. Typically these approaches involve the synthesis of polymer libraries followed by development of structure–property–performance correlations [16] that may be aided with the use of computational design and modeling tools [17]. Other viable approaches include advanced high-throughput testing procedures of 'random' libraries of candidate materials using specific outcome measures (i.e. with the hopes of getting 'lucky' and discovering useful materials) [18]. These libraries have been used for a variety of applications including discovery of polymers for nonviral gene delivery where damaged or disease causing genes are replaced with a normal gene, or where cells are transformed to produce certain proteins that may be useful for tissue regeneration. Recently, a library of >500 degradable poly(β-amino esters) polymers created via the conjugate addition of primary or secondary amine monomers to diacrylate monomers was used to develop effective materials for nonviral gene delivery [5••]. Polymers can be optimized through modification of molecular weight, polymer chain end-group, and polymer/DNA ratio, to successfully mediate gene transfer at levels that surpass gold standards including poly(ethyleneimine) and lipofectamine 2000 *in vitro* with less toxicity. These polymers selfassemble with plasmid DNA into nanometer-sized DNA/polymer complexes and have been used to deliver DNA to tumor cells *in vivo* with high efficiency and minimal toxicity while avoiding gene expression in healthy tissues [5••]. Specifically, a particular

**Figure 2**



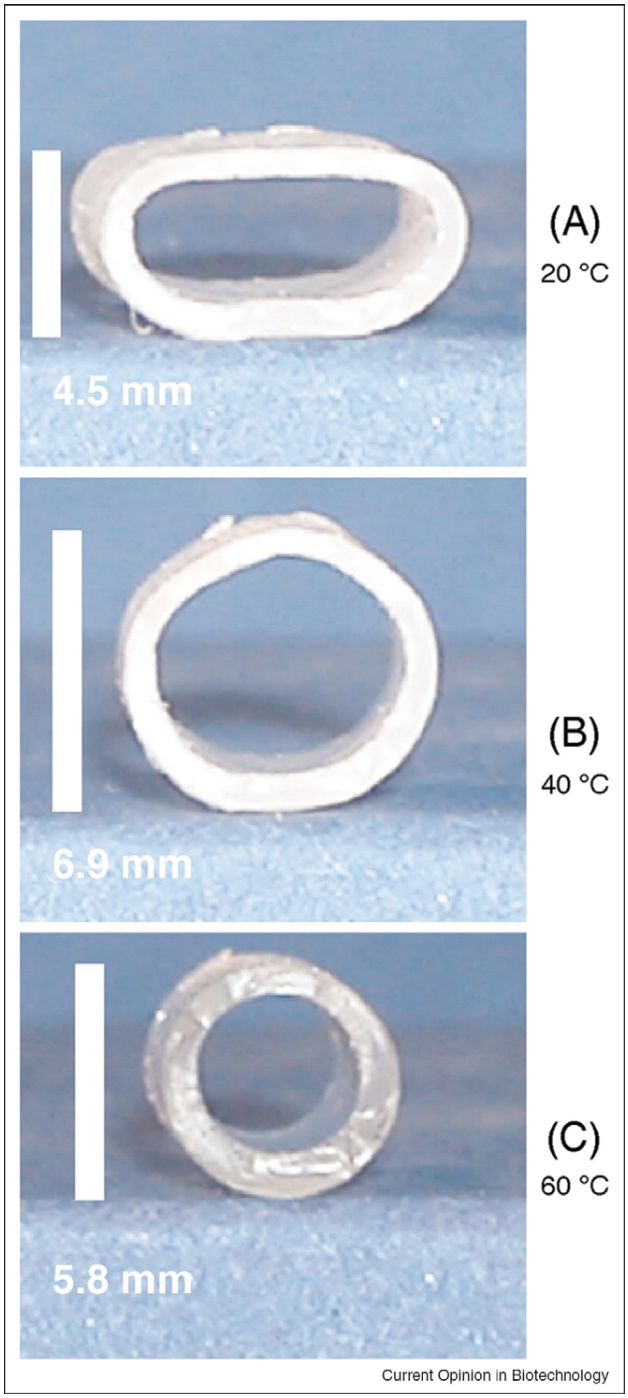

Transfection potential of poly(β-amino esters) *in vitro* presented in descending order with C32, the most effective polymer, shown on the left exhibiting orders of magnitude greater transfection levels compared to gold standard PEI and lipofectamine 2000. Polymers were synthesized at 95 °C in the absence of solvent (blue bars) or at 60 °C in the presence of 2 ml of dimethyl sulfoxide (red bars) (reproduced from [5••]).

polymer named C32 was selected from the polymer library based on its ability to transfect tumors locally at high levels, transfect healthy muscle poorly, and its minimal toxicity. Using this polymer, a DNA construct encoding the A chain of diphtheria toxin (DT-A) to xenografts derived from androgen-sensitive human prostate carcinoma cells (LNCaP) was created that specifically suppressed tumor growth and caused a 40% reduction in tumor size (Figure 2). This strategy was also recently demonstrated to potentially be useful for treatment of benign prostatic hyperplasia [19]. Combinatorial material libraries have also been created with photocrosslinkable degradable polymers to obtain a wide range of material properties [20] and with extracellular matrix proteins to specifically determine combinations that enhance cell function [6••]. Given the complex requirements of materials for advanced biomedical applications, combinatorial material development and discovery can have a significant impact, yet this will require the necessary infrastructure to disseminate these approaches to others in field. In addition to optimization of standard material properties including degradation times, mass loss profiles, mechanical properties, and biocompatibility, advanced materials may require additional design criteria to achieve functions including imaging, sensing, and shape-memory properties.

## Development of materials for minimally invasive surgery

Minimally invasive surgery (also known as 'keyhole' surgery), often supplemented with image-guided surgical navigation technology, has become routine within nearly all surgical disciplines. These procedures are typically performed through a small incision, often using specialized surgical instruments to reduce trauma, risk of infection, pain, and recovery times associated with open surgery. With the advent of minimally invasive surgery, it is possible to place biomaterial-based devices with laparoscopes including small endoluminal stents or to place large implants through small incisions including hip implants. Given the increasing need to develop degradable materials which do not require subsequent removal, and to increase the scope of minimally invasive procedures, new degradable shape-memory materials that change shape on demand in response to a stimulus were recently introduced. These materials can be programmed to respond to temperature, light, and magnetic fields. In addition to contact initiated changes, noncontact triggering of shape changes in polymers has been realized through incorporating magnetic nanoparticles within the shape-memory polymers followed by inductive heating achieved through alternating magnetic fields [21]. This was realized through the use of a polyetherurethane and a biodegradable multiblock copolymer with poly (*p*-dioxanone) as hard segment and poly(epsilon-caprolactone) as soft segment which were doped with nanoparticles consisting of an iron(III)oxide core in a silica matrix. Shape-memory materials have also been used to create materials with the capacity for multiple shape changes [22••] which may be useful for first, insertion into the body; second, expansion at a target site; and third, removal at a later point in time which may be necessary even with degradable materials (Figure 3).

**Figure 3**




Through increasing the temperature from 20 °C **(A)** to 40 °C **(B)** an initial shape change was induced followed by a second shape change through increasing the temperature to 60 °C **(C)** (reproduced from [22••]).

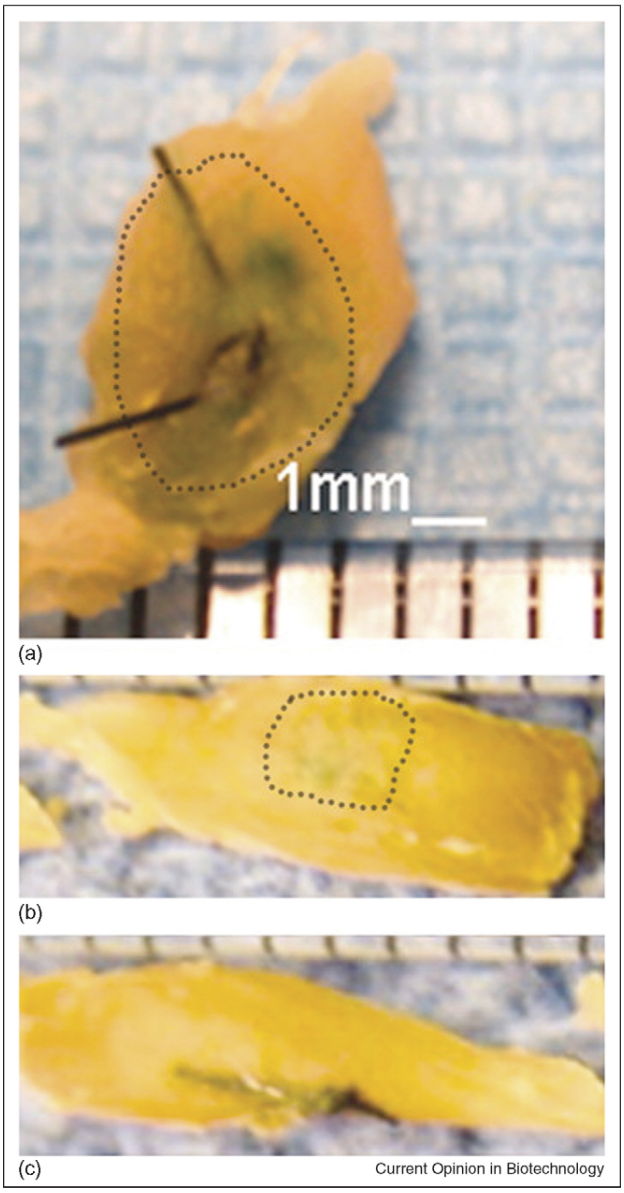
These materials have also been used to create a smart degradable suture that can tie itself in conjunction with minimally invasive surgical approaches [23]. Through providing additional design parameters such as shape change, these advanced materials remove challenging barriers which may ultimately increase the scope of application of many biomaterial-based strategies including perhaps tissue engineering constructs.

## Development of advanced materials for tissue engineering

Approximately half of the medical spending in the U.S. is attributable to tissue loss or organ failure with about 8 million surgical procedures and 40–90 million hospital days per year required to treat these disorders [24,25]. In 2006, about 29 000 patients received organ transplants, while almost 100 000 patients remain on the waiting list [26], and many will probably die without treatment. The demand for tissue and organ replacement or regenerative strategies following tissue damage (e.g. bone fractures, kidney failure, and severe burns) or diseases (e.g. diabetes, cancer, and cardiomyopathy) is expanding. Thus, the scientific and medical communities are working together to develop engineered tissues or regenerative approaches utilizing various combinations of stem cells, biomaterials, growth factors, and gene therapy. However, after three decades, the potential to provide tissues and organs to millions of patients suffering from trauma, congenital defects, and chronic diseases has yet to be fully realized [24,27]. Although this is partly due to uncertainly and difficulties with clinical markets, typical results in preclinical animal models remain highly variable with poor rates of success in larger defects and in higher animal species, probably because of poor survival of the transplanted cells [28••,29]. Although it is not surprising that the effectiveness of cell-based therapies rely on the retention of cell viability after implantation [30,31], little attention has been focused on this issue. Use of transplanted cells offers great potential to augment tissue healing and regeneration, especially in elderly patients who have reduced stem cell numbers or in those who have been subjected to irradiation for cancer treatment.

Recently, an advanced cell-instructive tissue engineering approach was successfully employed that utilized first, high-density arginine, glycine, aspartic acid (RGD)-containing cell adhesion ligands; second, an exogenous cell source; and third, growth factors to enhance the regenerative capacity of the transplanted cells through promoting their survival, preventing their terminal differentiation, and promoting their outward migration (Figure 4) [32••]. Specifically, cells were delivered on porous alginate/calcium sulfate scaffolds that contained both hepatocyte growth factor (HGF) and fibroblast growth factor-2 (FGF-2) which were employed to maintain the cells in an activated, proliferating, but nondifferentiated state. Whereas control groups had only modest effect on muscle regeneration, a combination strategy employing controlled release of HGF and FGF-2 in combination with scaffolds and cells dramatically enhanced the participation of transplanted cells leading to significant tissue regeneration.

**Figure 4**




Photographs of muscles treated with **(a)** scaffolds delivering cells and releasing HGF and FGF2, **(b)** scaffolds containing only HGF and FGF2, and **(c)** scaffolds containing only myoblasts (dotted lines outline positively stained tissue). Size bars are shown on the photomicrographs (reproduced from [32••]).

Despite the relatively small size of the scaffolds employed here (50 $mm^3$) and the uncertainties in translating this strategy into larger clinically relevant defects, the work demonstrates a proof of concept for advanced materials that can be designed to direct tissue regeneration.

## Conclusions

Advances in understanding disease and tissue regeneration combined with increased accessibility of modern technology have created new opportunities for the use of biomaterials in unprecedented ways. Materials can now be rapidly created and selected to target specific cells, change shape in response to external stimulus, and instruct tissue regeneration. Despite the rapid advancement of state of the art medically driven technologies, numerous challenges still exist for translating technology to the clinic including long and often undefined regulatory approval pathways, high translational costs, potential safety concerns with nanomaterials, assessing appropriate risk/benefit and cost/benefit ratios, and appropriate matching of technology and application. The latter may be one of the most difficult tasks faced by the biomedical engineer. Although development of general platform technologies can lead to major breakthroughs across multiple disciplines, conventional wisdom suggests rapid clinical translation requires a systematic approach that involves first identifying and understanding a problem followed by engineering a focused solution. As technology rapidly advances, the bioengineer has more tools at their disposal than ever before, yet only if they are able to select the right tools and technologies, can society benefit.

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