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Bioengineering Kidneys for Transplantation

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Abstract

One in ten Americans suffer from chronic kidney disease, and close to 90,000 people die each year from causes related to kidney failure. Patients with end-stage renal disease are faced with two options: hemodialysis or transplantation. Unfortunately, the reach of transplantation is limited because of the shortage of donor organs and the need for immunosuppression. Bioengineered kidney grafts theoretically present a novel solution to both problems. Herein we discuss the history of bioengineering organs, the current status of bioengineered kidneys, considerations for the future of the field, and challenges to clinical translation. We hope that by integrating principles of tissue engineering, and stem cell and developmental biology, bioengineered kidney grafts will advance the field of regenerative medicine while meeting a critical clinical need.

Keywords

kidney; bioengineering; organ engineering; perfusion decellularization; transplantation

HISTORY OF BIOENGINEERING

The field of regenerative medicine started long before its name was coined by hospital administrator Leland Kaiser in 1992 in an article about future medical technologies that could “change the course of chronic disease...and regenerate tired and failing organ systems”¹. In the 1920s, Nobel prize winner Alexis Carrel, a pioneer of cardiovascular and transplant surgery, collaborated with aviator and engineer Charles Lindbergh to create a pump oxygenator for the long-term perfusion of single organs; by 1935, they showed the first successful long-term *ex vivo* perfusion of organs by keeping cat thyroids alive outside the body for several weeks^{2,3}. This perfusion pump would form the basis for bioreactors that

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are now widely used to preserve and grow organs *ex vivo*. However, it took a confluence of factors in the 1990s for regenerative medicine to fully mature into the burgeoning field that it is today. These factors include the discovery of embryonic stem (ES) cells, the more recent development of nuclear transfer technology, new insights into the expansion and differentiation of stem cells, and advances in tissue engineering, in which the principles of biology and engineering are applied to develop functional substitutes for damaged tissue⁴⁻⁶.

In a shift from two-dimensional to three-dimensional cellular adjuncts, in the 1990s, the first living tissue grafts were created based on synthetic polymers onto which cells were seeded⁷. These initial constructs contained hepatocytes, chondrocytes, and enterocytes⁸⁻¹⁰.

Bioengineered trachea, bladders and vessels were the first constructs to be implanted into humans. In 2004, a 30 year old female with end-stage bronchomalacia underwent implantation of a totally bioengineered human trachea constructed from a deceased donor scaffold seeded with autologous cells derived from mesenchymal stem cells and epithelial cells¹¹. In 2006, human bladder matrices were seeded with autologous bladder cells grown from culture and implanted into 10 patients who did well with a mean follow-up of nearly 4 years¹². In 2007, blood vessels engineered from autologous skin and superficial vein cells were also successfully implanted into 10 patients with end-stage renal disease (ESRD) on hemodialysis¹³.

However, to go beyond tubular structures lined with single-cell layers to a more architecturally complex organ requires intra-organ organization of scaffolds, cells, and soluble factors along with intact vasculature for perfusion^{14,15}. A significant advance came in 2008 with the discovery of perfusion-decellularization techniques that led to the development of a whole-heart scaffold with intact three-dimensional geometry and vasculature¹⁶. Using this approach, the native extracellular matrix of whole organs has been isolated from lung¹⁷⁻¹⁹, kidney²⁰⁻²², liver²³, and pancreas^{6,24}. While this most recent vertical step in the field of organ engineering brings us closer to a bioengineered whole organ, this method relies upon donor organs to derive native extracellular matrices. Alternative techniques of fabricating three-dimensional biological structures, such as self-assembly and bioprinting, would avoid this issue^{15,25}.

NEED FOR BIOARTIFICIAL KIDNEY

Chronic kidney disease (CKD) is a major healthcare challenge worldwide. In the United States, about 9.4% of adults suffer from chronic kidney disease, and the prevalence of ESRD is increasing^{26,27}. Medical care for people with ESRD required \$49.3 billion dollars in 2011: \$32,922 Medicare costs per person per year for transplant patients and \$87,945 Medicare costs per person per year for patients on hemodialysis²⁷.

About 400,000 patients with end-stage renal disease depend on some form of dialysis²⁸. Hemodialysis has revolutionized the care of these patients as a temporizing measure to remove toxic waste products and restore body fluid volume and electrolyte balance. Despite improvements in technology and patient care, one year on hemodialysis is associated with a 6% increase in the relative risk of death²⁹. Adjusted rates of all-cause mortality are 6.5–7.9 times greater for patients on dialysis than for individuals in the general population; this is in

contrast to kidney transplant recipients who have an adjusted all-cause mortality rate of 1.0–1.5 times the general population³⁰. Long-term complications of hemodialysis include hypotension, malnutrition, access site infection, gastrointestinal bleeding, and depression. In addition, traditional dialytic modalities focus on solute clearance and volume management without providing the immunoregulatory, metabolic, and endocrine functions of native kidneys.

To expand the functionality of hemodialysis, cellular components have been added to renal replacement therapy. Development of the first extracorporeal bioartificial kidney (BAK) support systems started in the 1980s, when synthetic scaffolds were combined with cellular components^{31,32}. In these experiments, human proximal renal tubular cells were cultured on hollow fiber scaffolds and then placed in series with a hemofiltration circuit. Phase I/II clinical trials demonstrated that bioartificial kidney systems were safely able to filter urine, improve metabolic parameters, reduce pro-inflammatory cytokine levels, and improve cardiovascular stability^{33,34}. However, a significant impact on survival has been harder to discern as the multicenter, randomized, controlled, open-labeled Phase II clinical trial performed in 2004–2005 was likely underpowered^{35,36}. Of the 58 critically ill patients with acute renal failure who were enrolled in the study, 21/40 completed BAK therapy and 4/18 completed conventional dialysis therapy³⁵. Survival was significantly improved in patients who underwent BAK therapy at 180 days but not 28 days³⁵. Currently, BAKs are limited by the survival of tubular cells and cost-effective manufacturing of the device^{37,38}.

Organ transplantation represents a unique method of treatment to cure people with end-stage organ failure. Since the first successful kidney transplant in 1954, the field of transplantation has made substantial progress. However, transplant surgery still faces one fundamental problem—the number of people requiring organ transplants is simply higher than the number of organs available. In the United States, 18 people die on a transplant waiting list every day due to the critical organ shortage³⁹. There are currently about 120,000 people on the waiting list for an organ; in 2013, there were only about 14,000 organ donors and only about 28,000 transplants performed⁴⁰. In addition, despite an approximately 70% 5-year graft survival after kidney transplantation, patient survival is limited by cardiovascular disease, infection, malignancy, and chronic rejection^{41,42}.

Fully implantable bioengineered kidneys have the potential to address these shortcomings by replacing a diseased organ with a newly functioning one. Bioengineered kidneys represent a new, theoretically inexhaustible supply of organs that could mitigate the ever-growing demand for transplantable organs and reduce waiting list mortality. Furthermore, if generated from patient-derived cells, bioengineered organs could also be transplanted without need for life-long immunosuppression, erasing the heavy burden associated with its side effects (infection, malignancy, drug toxicity) and cost (about \$15,000 to \$20,000 per year)⁴³. With an increased donor pool, patients could be transplanted much earlier, when they have less comorbidities and a lower perioperative risk for adverse outcomes. In addition, regenerative medicine technology could be used to improve the quality of discarded donor grafts (currently about 40% of eligible donor grafts are not used for transplantation in the United States)⁴⁴.

Thus, an ambitious goal for an alternative, efficacious renal replacement therapy is to generate a functional, self-sustaining, safe bioengineered kidney for transplantation (Tables 1 and 2).

CREATING A BIOENGINEERED KIDNEY

Following the principles of tissue engineering, the process of developing bioartificial organ grafts can be broken down into three stages: (1) creating an organ scaffold; (2) seeding the scaffold with cells; and (3) maturing the structure in a bioreactor.

Scaffolds

Initial efforts to engineer kidney tissue combined biological and synthetic components in extra-corporeal renal support systems. BAKs combine a hemofilter used in conventional dialysis with a bioreactor unit containing human primary renal proximal tubule cells derived from discarded donor kidneys to supplement current renal replacement technology with other physiological functions of the kidney. The first clinical trial using BAKs was reported in 2004³⁴ and a subsequent Phase II trial showed improved long-term but not short-term survival in ICU patients with acute renal failure treated with BAKs compared to continuous renal replacement alone³⁵; however, this trial was underpowered and further studies are needed to confirm this result³⁶. Current BAK research is focused on finding the best cell type to use and creating devices that contain growth factors and novel membrane materials to foster the optimal environment for cell viability³⁷. While not implantable by design, these extracorporeal synthetic scaffolds provide very valuable information on tubular epithelial cell biology and function within an engineered construct.

A broad variety of biologic and/or synthetic materials could be used to create scaffolds for the regeneration of a whole organ. Conventional tissue fabrication techniques relied upon pre-formed acellular scaffolds that incorporated simple cell constructs of a single phenotype by photolithography or layer-by-layer deposition of ionic biopolymers⁴⁵. Another technique distributes cells in patterns determined by laminar fluid flow in microfluidic channels⁴⁶. Currently, fabrication resolution for photolithography is on the submicron scale⁴⁷. Conventional three-dimensional printers have a spatial resolution of about 0.01 to 0.1 millimeters, but more recent technologies can reduce this to 65 nanometers⁴⁸.

Bioprinting is promising method for creating scaffolds that do not rely upon a donor organ to supply the native extra-cellular matrix. It allows for precise spatial placement of different cells in the matrix in a manner that is low-cost and high throughput⁴⁹. Bioprinting involves the use of a printing device that deposits cells and biomaterials into precise three-dimensional arrangements to generate structures that follow along a pre-determined blueprint^{50,51}. The structure can then be matured *in vitro* or *in vivo*. For example, amniotic fluid-derived stem cells were bioprinted onto wound sites and accelerated wound healing in a mouse model⁵². Most recently, complex three-dimensional tissue constructs composed of stem cells, muscle cells, and endothelial cells were printed and retained their function *in vivo*⁴⁹. The unique advantage of bioprinting, aside from its controlled composition and architectural construction, is the fact that both cells and matrix can be layered simultaneously. If further developed, this would allow a multitude of different cell

phenotypes to be assembled in their physiologic three-dimensional relationship. Challenges in applying bioprinting approaches to organ engineering include the limited mechanical stability of the constructs, limited spatial resolution of current bioprinting devices, and the inability to create three-dimensional fiber structures through layering techniques.

Decellularization is an alternative process that generates three-dimensional structures without building an organ from the ground up. Cellular material is removed by mechanical, chemical, or enzymatic methods while the extracellular matrix remains intact⁵³. Perfusion-decellularization, first reported in 2008, takes advantage of the intrinsic vascular structure of any organ (as well as the biliary tree, ureter, and airways) to efficiently and effectively deliver decellularization agents at a constant, low physiological pressure^{16,54}.

Target organs can be harvested up to 4–6 hours postmortem, up to the point at which proteolysis begins. Treating target organs with detergents or acids through the innate vasculature results in a decellularized extracellular matrix composed of proteins and polysaccharides. Following decellularization, ethylene oxide or peracetic acid can be used to sterilize the matrix without destroying the matrix itself. The result is a natural, biocompatible backbone that is an ideal platform for organ bioengineering. These scaffolds are free of significant cellular content, retain major extracellular matrix proteins (such as collagens, laminins, fibronectins, and glycosaminoglycans), maintain tensile strength, and preserve geometric and spatial organization⁵⁵.

Both regulatory signals and physical cues can determine cell phenotype and tissue function⁵⁶. For example, decellularized rhesus monkey kidneys were more easily repopulated in younger donor kidneys compared with older donor kidneys⁵⁷. Specifically engineering microenvironments can guide stem cell differentiation and function⁵⁸. In some experiments, the renal scaffold itself supports embryonic stem cells to proliferate and differentiate into glomerular, vascular, and tubular pathways⁵⁹. Further scaffold modifications can serve to encourage cell differentiation along a preferred pathway. For example, polyethylene glycol hydrogels can be modified with adhesion peptides to influence cellular interactions, such as the osteogenesis of bone marrow stromal cells⁶⁰. Other peptides can be placed in peptide gels to control differentiation, such as the development of stem cells into neurons rather than astrocytes⁶¹. Soluble factors such as growth factors and cytokines can also be delivered in a controlled spatial and temporal fashion depending on how they are incorporated into the scaffold⁶². Conjugating a rodent bladder acellular matrix with basic fibroblast growth factor accelerated the cellularization and vascularization of the bladder after implantation⁶³. Finally, fluid flow through the glomerulus and filtration across tubules generate forces that are essential for proper kidney cell function. Bioreactors that replicate the perfusion-based fluid flow aid in the long-term culture of bioengineered kidneys, especially in promoting the development of appropriate cell phenotypes.

Cells

The basic functional unit of the kidney is the nephron. The kidney contains about 1.2 million nephrons. Each nephron is a tube lined by a single cell layer that can be divided geographically into the renal corpuscle, proximal tubule, loop of Henle, distal tubule, and collecting duct system. The epithelial cells in each segment are highly specialized, varying

in mitochondrial content and membrane properties depending on their function (e.g. proximal tubule cells have many mitochondria and a specialized brush border membrane, whereas cells in the thin loop of Henle do not). This allows for differential filtration, reabsorption, and secretion along the nephron to modulate urine volume and content. The renal corpuscle contains glomerular capillaries containing the afferent and efferent arterioles. The endothelial cells are lined by a basement membrane that in turn is surrounded by epithelial podocytes; together this forms the filtration barrier. Mesangial cells make up the remainder of the renal corpuscle and provide support to the glomerulus by secreting extracellular matrix, producing growth factors/cytokines, and exhibiting phagocytic activity.

Seeding a kidney scaffold requires epithelial cells, endothelial cells, and mesangial cells to make up the complex function of the kidney. Primary cultures of human tubular cells have been successfully integrated into bioartificial devices while continuing to provide metabolic, endocrine, and immunological properties⁶⁴ but are difficult to expand *in vitro* to the numbers that would be required to repopulate a kidney of clinically relevant scale.

In our opinion, pluripotent cells such as ES cells and induced pluripotent stem (iPS) cells expand the repertoire for generating the necessary cell types and numbers for bioartificial organs, and offer the unique advantage of an autologous source. When using pluripotent cells, physiological development must be recapitulated to differentiate the necessary progenitor cells and ultimately the full spectrum of cellular phenotypes of any given organ.

Embryologically, the kidney is derived from two mesodermal structures: the ureteric bud and the metanephric mesenchyme. The ureteric bud gives rise to the calyceal system of the kidney while the rest of the functional components of the kidney including the glomeruli is derived from the metanephric mesenchyme. Following the course of physiologic development, generating metanephric mesenchyme is therefore the first goal in renal regeneration and bioengineering.

ES cells are pluripotent cells with unlimited self-renewal properties that have the capability to generate all cell types of the human body. Single-cell suspensions of murine embryonic cell lines can differentiate into renal epithelial cells and have been used to construct renal organoids *in vitro*^{65–67}. Human ES cells also demonstrate the ability to form kidney-like structures, express genes associated with kidney development, and can be sorted into mesodermal populations enriched for intermediate mesoderm and putative renal progenitors^{68–71}.

Pluripotent stem cells can be induced from somatic cells by the introduction of Oct3/4, Sox2, c-Myc, and Klf4, under embryonic stem cell culture conditions⁷². These iPS cells can be generated from renal cells, including adult mesangial cells⁷³ or renal epithelial cells shed into the urine⁷⁴, or even keratinocytes from patients with ESRD⁷⁵. Delivery of iPS cells into rat kidneys with ischemia-reperfusion injury reduced inflammatory and apoptotic markers and improved the survival of rats with damaged kidneys⁷⁶. The next step is to differentiate pluripotent stem cells into renal lineages. Renal lineage cells have been differentiated from murine ES and iPS cells⁷⁷. Recently, iPS cells have been programmed to differentiate into intermediate mesoderm, the embryonic germ layer that gives rise to the kidneys, by

treatment with a Wnt pathway activator and retinoids found by high-throughput chemical screening⁷⁸.

Mesenchymal stem cells (MSC), found in adult bone marrow, also have multipotent properties and can differentiate into mesenchymal tissues such as osteoblasts, adipocytes, chondrocytes, tendon, muscle, and marrow stroma. MSCs have been used to improve kidney function in models of chronic renal failure by migrating to the site of damaged kidney tissue and exerting immunomodulatory and paracrine effects to restore kidney function⁷⁹.

Understanding and harnessing the renoprotective properties of MSCs could lead to another source of cells for a bioengineered kidney^{80,81}.

Organ assembly

Tracheas, bladders, and blood vessels have been implanted successfully into more than 160 patients without the need for immunosuppressive medication^{82,83}. However, these structures rely on diffusion to satisfy cellular metabolic demand. Without reconnection to a vascular supply, cells can only obtain nutrients and oxygen via diffusion across a distance of 1 to 3 mm⁸⁴, thereby exposing the bioengineered organ to the risk of ischemia and/or graft failure. Indeed, the most proximal 1 cm of the first implanted graft trachea collapsed ventrally 8 months after implantation, likely because of insufficient blood supply¹¹.

Bioengineering a viable kidney graft is more challenging because of the complex architecture and functionality of the kidney. The adult human kidney weighs between 115 to 170 grams, has a volume of about 200 cm³, and receives 25% of the total cardiac output (1.25L/min). The kidney functions as a filtration unit, endocrine organ (blood cell production, bone metabolism), immune regulator and modulator of cardiovascular physiology. Simplified kidney organoids, largely composed of a single cell type in a three dimensional matrix, have been used to study kidney disease, drug nephrotoxicity, and kidney development^{85,86}, but are far from accomplishing the varied functions that the kidney must provide.

Whole kidney scaffolds have been derived through decellularization of cadaveric rat, pig, and rhesus monkey kidneys^{20,21}. Renal extracellular matrices produced from porcine kidneys were implanted into pigs *in vivo* as proof-of-concept and demonstrated the ability to withstand physiologic blood pressure without extravasation; however, despite preservation of renal architecture at 2 weeks, the naked vasculature was thrombosed by 24 hours after implantation⁸⁷. As further proof-of-principle, our group reported the regeneration of functional rat kidneys that were seeded with epithelial and endothelial cells and produced rudimentary urine when transplanted orthotopically in rats²². Future advances depend on further understanding the developmental biology of the kidney, including the role of growth factors, the regenerative/repairative properties of the kidney, the role of the extracellular matrix, and identification of potential renal progenitor cells^{88,89}.

CHALLENGES AND OPPORTUNITIES IN TRANSLATING TO CLINICAL MEDICINE

Scaffolds

Perfusion-decellularization techniques have been successful in various organs from large animal models (swine, non-human primate) and humans, but these protocols need to be standardized and include steps to ensure clinical-grade quality complete with sterilization and preservation for future use. However, the current clinical use of decellularized bone, dermal, and heart valve allografts demonstrates the feasibility of scaling production of decellularized matrices to meet clinical demand.

Cells

The ideal clinically feasible cell source to generate progenitor cell populations on a large scale has yet to be identified. Cells from fetal tissues can properly differentiate and function; however, this source may have limited expansion capabilities and may meet with ethical concerns⁴. These same barriers are faced by human ES cells, use of which is surrounded by controversy. However, human ES cells also carry a theoretical oncogenic risk; for example survivin (BIRC5), an anti-apoptotic oncofetal gene, is highly expressed in human ES cells and may lead to teratoma formation⁹⁰. The ectopic expression of transcription factors such as OCT4, SOX2, KLF4, c-MYC, NANOG, LIN28 can generate iPS cells for organ replacement and circumvent ethical concerns; however, these cells have tumorigenic traits since reprogramming is often accompanied by genetic and epigenetic alterations⁹¹. These autologous iPS cells provide a unique advantage in that they may potentially provide an inexhaustible source of patient- and tissue-specific stem cells

In addition, repopulating a scaffold requires an adequate number of viable cells (delivered during initial seeding or *in situ* expansion). *In vitro* expansion with the signals necessary to drive appropriate differentiation of multiple cell types into a primordial kidney has been accomplished such that transplanted embryonic metanephrons can grow and secrete concentrated filtrate, but this approach is limited in terms of scalability⁹². To circumvent the difficulties of large-scale cell culture, one approach could be to rely on repopulation by host cells *in vivo*, a process which has been successful in implanted dermal matrices and trachea⁴.

Xeno-bioengineered organs

An intriguing prospect is to combine the strengths of both xenotransplantation and bioengineering to generate “semi-xenografts” where the scaffold would be animal-derived and the repopulated cells would be human-derived³. This would make it easier to control the quality of the scaffold, as pathogen-free herds and post-harvest processing would eliminate most known pathogens⁴. For example, bioartificial human tissue with an innate vascularized network was created using a porcine small bowel platform^{83,93}. Porcine small bowel was decellularized with preservation of vasculature, which was then reseeded with human endothelial cells and then implanted into the arm of a patient. After 1 week, the construct was viable and the vasculature was patent⁹³. Though the majority of proteins in the extracellular matrix are highly conserved across species⁹⁴, the immunological barriers

facing xenotransplantation still pertain, such as the antigenicity of the Gal epitope on swine⁹⁵.

Immune response

Surgical meshes—acellular dermal allografts—were first developed in the 1990s in order to treat full-thickness burn injuries⁹⁶. These allografts were able to modulate tissue repair without antigenicity generating a specific immune response⁴. The host immune response to bioengineered organ constructs requires further study. What is known is that both the innate and acquired immune system are involved⁹⁷, that the early response involves activation of PMNs and Th-2 pathways as a remodeling response rather than a rejecting one⁹⁸, and that eventually a state of chronic inflammation is reached, where the foreign body is accepted and usually surrounded by a fibrous capsule⁶. The scaffold itself may be minimally immunogenic. Complete decellularization mitigates the immunogenicity of the scaffolds by removing cellular material containing antigenic epitopes⁹⁷. Decellularized native extracellular matrix products currently in clinical use, such as dermal matrices, bone allografts, and heart valves, are fully biocompatible and are repopulated by host cells after implantation. However, implantation itself is accompanied by inflammatory, wound healing, and remodeling responses typical of any surgery.

Quality control and cost

Practically, bioengineered organs must meet standards of quality despite their patient-specific customization. Tools such as imaging, in vitro assays, and bioinformatics can aid in assessing the quality of the construct and the state of the tissue and cells within it. To date, the technology has not been successfully scaled up to a clinically relevant size, but first milestones towards clinically relevant graft dimensions have been met. In addition, post-operative care of the bioengineered organ to ensure long-term viability, and graft longevity will have to be determined. Lastly, reducing cost of personalized organ engineering will depend on developing cost effective reprogramming, differentiation, and regeneration strategies⁹⁹.

CONCLUSIONS

The demand for innovative and personalized renal replacement therapy is substantial. The generation of autologous bioengineered kidneys for transplantation is a promising concept for patients suffering from ESRD. This requires, in general, three main building blocks: an organ scaffold, cells for repopulation, and bioreactors for maturation (Figure 1). An overview of the current status of bioengineering organs demonstrates the exponential progress that has been made, with several regenerated constructs reaching clinical application. Recellularized kidney scaffolds are successfully transplanted into large animal models, and in small animal models, regenerated kidneys produce rudimentary urine. Major challenges such as derivation of all necessary cellular phenotypes from patient-derived cells, refined seeding strategies, and culture techniques to fully mature function remain. As a multidisciplinary community, we will be able to develop innovative solutions to overcome all of these hurdles, and bring bioengineered kidneys to clinical translation.

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Figure 1. Current strategies for engineering a bioartificial kidney for transplantation

Current strategies to create a bioartificial kidney can be divided into three stages: (1) generating a scaffold by bioprinting or decellularization; (2) populating the scaffold with cells from embryonic stem cells or induced pluripotent stem cells; (3) maturing the organ *in vitro* or *in vivo*. After the kidney has been matured, it can be transplanted into humans.

Table 1
Minimum design criteria to generate a transplantable bioengineered kidney graft that can produce a filtrate

This table outlines basic components needed to generate a single kidney that is capable of filtering.

Structure	<i>Scaffold</i>	• Biocompatible (non-toxic to cells and recipient) • Outline basic organ architecture (artery, vein, ureter, parenchyma) • Maintain integrity during culture and post-implantation
	<i>Cells</i>	• Viable • Basic function
	<i>Organ assembly</i>	• Enable perfusion, filtration, and drainage of filtrate during culture and post-impantation
Function	<i>Filtration</i>	• Excretion and reabsorption to maintain balance of – electrolytes – acid/base – volume
	<i>Implantation</i>	• Surgical anastomoses and tissue handling comparable to donor organ • Vascular integrity to prevent hemorrhage and coagulation
Safety	<i>Minimize antigenicity</i>	• Scaffold • Cells
	<i>Sterility</i>	
	<i>Clinical monitoring</i>	• Durability of the construct

Table 2
Optimal design criteria to generate a fully functional transplantable bioengineered kidney graft

This table outlines the ideal components of a bioengineered kidney that would be fully functional and clinically efficacious for use in humans.

Structure	Scaffold • Biocompatible (non-toxic to cells and recipient) • Outline basic organ architecture (artery, vein, ureter, parenchyma) • Maintain integrity during culture and post-implantation • Provide niche for site-specific cell engraftment, differentiation, and function
	Cells • Autologous • Viable • Basic function • Self-renewing
	Organ assembly • Enable perfusion, filtration, and drainage of filtrate during culture and post-implantation • <i>In vivo</i> remodeling and maturation
Function	Filtration • Excretion and reabsorption to maintain balance of – electrolytes – acid/base – volume • Macromolecular sieving • Glucose reabsorption
	Hormonal • Erythropoietin • Calcitriol • Renin production
	Other • Gluconeogenesis • Peptide hormone catabolism
Safety	Implantation • Surgical anastomoses and tissue handling comparable to donor organ • Vascular integrity to prevent hemorrhage and coagulation

Minimize antigenicity
• Scaffold   • Cells
Sterility
Clinical monitoring
• Durability of the construct