3 Bioreactor Instrumentation and Control for 3D Cellular and Tissue Systems

Steve Warren

CONTENTS

3.1 Introduction ........................................................................................................... 33
3.2 Bioreactor Embodiments .................................................................................... 36
  3.2.1 Design Considerations .................................................................................. 36
  3.2.2 Thematic Bioreactor Categories .................................................................. 37
    3.2.2.1 Stirred-Flask Bioreactors .................................................................... 37
    3.2.2.2 Rotating-Wall Bioreactors .................................................................. 38
    3.2.2.3 Perfusion Bioreactors ........................................................................ 40
    3.2.2.4 Other Bioreactor Form Factors .......................................................... 40
3.3 Mechanical/Electrical Tissue Stimulation ............................................................... 41
  3.3.1 Mechanical Stimulation .............................................................................. 41
  3.3.2 Electrical Stimulation .................................................................................. 43
  3.3.3 Electromagnetic Stimulation ....................................................................... 43
3.4 Aggregate Component Architecture for Bioreactor Systems ............................... 43
3.5 Sensing and Control in Bioreactor Systems ............................................................ 46
  3.5.1 Parameters and Sensing Methods ............................................................... 46
    3.5.1.1 Offline versus Online Measurement Modes ..................................... 48
    3.5.1.2 Combined Parameter Assessments .................................................. 48
  3.5.2 Controls ....................................................................................................... 49
3.6 Conclusion and Perspective .................................................................................... 51
Acknowledgments ....................................................................................................... 53
References .................................................................................................................. 53

3.1 INTRODUCTION

A tissue bioreactor embodies an artificial environment whose purpose is to maintain physicochemical conditions that promote tissue growth. Tissue bioreactors, also referred to as tissue incubators (Butler et al. 2009; Christen and Andreou 2007), have been used since the early 1980s to provide closed growth environments for human and animal cells (Eibl and Eibl 2009; Plunkett and O’Brien 2010). These
active incubation apparatuses have promoted the growth of small quantities of tissue, such as cultured skin cells used for grafts and wound dressings applied to burn victims or patients with skin ulcers and cancers (Helmedag et al. 2015; Jeong et al. 2014; Prenosil and Kino-oka 1999). Bioreactors have also targeted tissue growth for bone (Bancroft et al. 2003; Cartmell et al. 2003; Gaspar et al. 2012; Rauh et al. 2011), cartilage (Darling and Athanasiou 2003; Popp et al. 2012; Sucosky et al. 2004; Wendt et al. 2005), tendons/ligaments (Ainsworth and Chaudhuri 2005; Laurent et al. 2014; Wang et al. 2013), vessels (Barron et al. 2003; Ferrechio et al. 2009; Niklason et al. 1999), cardiac tissue (Barron et al. 2003; Shachar and Cohen 2003), and heart valves (Amrollahi and Tayebi 2016; Barron et al. 2003).

A bioreactor simulates the physical environment required for the growth and maintenance of viable tissue (Martin and Vermette 2005). As such, a bioreactor should perform at least one of the following functions (Barron et al. 2003; Freed and Vunjak-Novakovic 2000; Partap et al. 2010; Plunkett and O’Brien 2010):

1. Promote cell proliferation and/or uniform tissue growth on a three-dimensional (3D) scaffold.
2. Maintain a culture medium with the correct concentrations of gases and nutrients.
3. Facilitate mass transfer to the tissue constructs.
4. Provide physical stimuli to growing tissue.
5. Offer data regarding the development of the tissue within the bioreactor.

The ideal responsibilities of a bioreactor would include real-time monitoring and control of dynamic physicochemical states and the concentrations of nutrients, metabolites, and cytokine concentrations germane to different kinds of cells (Christen and Andreou 2007; DeBusschere and Kovacs 2001; Xicai et al. 2007). Recent and ongoing bioreactor research themes address the following:

- **Cell/tissue scaling**: The production of tissue in large enough quantities for practical use (Buang et al. 2014; Wu et al. 2014).
- **Environmental management**: The creation of an environment that promotes growth for all cells either suspended in, or surrounded by, the bioreactor medium while minimizing the likelihood of tissue necrosis. This involves maintaining environmental parameters (e.g., pH, temperature, pressure, and osmolarity), nutrient delivery (e.g., oxygen), waste removal (e.g., urea and lactate), and flow in the culture medium (Barron et al. 2003; Cartmell et al. 2011; Martin and Vermette 2005; Pörtner and Giese 2006; Yeatts and Fisher 2011).
- **Mechanical stimuli**: Mechanical actuation to fabricate tissues such as blood vessels, cartilage, bone, tendons, and ligaments that can meet the mechanical rigors of their target environment or, in contrast, to minimize shear stress that cells experience during culture, which can have a detrimental effect on expansion (Barron et al. 2003; Butler et al. 2009; Darling and Athanasiou 2003; Elder and Athanasiou 2009; Laurent et al. 2014; Mauck et al. 2000; Popp et al. 2012; Rauh et al. 2011; Yeatts and Fisher 2011).
• **Electrical/electromagnetic (EM) stimuli:** The use of electromagnetic fields (EMFs) or direct electrical-current injection to stimulate artificial tissues for cardiac and nervous system applications (Dobson et al. 2006; Kim et al. 2011; Radisic et al. 2008; Tandon et al. 2008; Vunjak-Novakovic et al. 2011).

• **Real-time sensing:** Constant monitoring of (a) the constituents that affect the tissue growth process in the entire 3D space and (b) the size/number and viability of the tissue mass itself (Mazzei et al. 2008; Rolfe 2006; Xicai et al. 2008).

• **Real-time control:** Feedback-driven control systems that regulate the growth environment based upon input received from the sensors embedded in the bioreactor space (Christen and Andreou 2007; DeBusschere and Kovacs 2001; Mazzei et al. 2008; Rolfe 2006; Xiang et al. 2015; Xicai et al. 2008).

• **Alternative embodiments:** Portable bioreactors (DeBusschere and Kovacs 2001; Tandon et al. 2013), small complementary metal-oxide-semiconductor (CMOS)-scale, implantable bioreactors (Frantz et al. 2010; Kim et al. 2011), and various bioreactor geometries that support the growth of complex tissues (Atala et al. 2012; Mikos et al. 2006).

An array of bioreactor designs exists, and many of these are “one-off” designs, meaning that a given design is the only one in existence and is customized to the research group and application for which it was designed. Although these bioreactor manifestations offer different design goals in terms of basic operation, constituent concentrations of interest, and possibly the need for external mechanical/electrical stimulation, etc., they have similar needs in terms of (a) environment and medium management, (b) sensor-based instrumentation that can provide situational awareness regarding parameter levels and tissue viability, and (c) feedback-based control systems that continuously balance the incubation environment. This chapter addresses those general areas of need, with the realization that a specific bioreactor may deviate considerably from the base bioreactor designs, instrumentation, and control options presented here.

Note that the tissue bioreactor literature base is growing rapidly, more so in recent years. A number of excellent bioreactor reviews are available that address the following:


• **Bioreactors for structural tissue growth** (e.g., bone, cartilage, tendons, ligaments) that often incorporate mechanical stimuli (Abousleiman and Sikavitsas 2006; Bancroft et al. 2003; Bilodeau 2004; Carpentier et al. 2011; Elder and Athanasiou 2009; Gardel et al. 2014; Gaspar et al. 2012; Rauh et al. 2011; Sladkova and de Peppo 2014; Yeatts and Fisher 2011).
• **Bioreactors for cardiovascular tissue growth** (e.g., cardiac tissue, vessels, and valves) that may incorporate mechanical and/or electrical stimuli (Amrollahi and Tayebi 2016; Barron et al. 2003; Shachar and Cohen 2003; Vunjak-Novakovic et al. 2011).

Further, a collection of books is also available that addresses the engineering aspects of bioreactor design (Antolli and Liu 2011; Asnaghi et al. 2014; Blose et al. 2014; Chaudhuri and Al-Rubeai 2005; Eibl and Eibl 2009; Haycock 2011; Kasper et al. 2009, 2010; Partap et al. 2010; Wendt et al. 2008). Numerous technical papers speak to application- and tissue-dependent bioreactor design tradeoffs. A synopsis of instrumentation issues related to bioreactor system design, including thematic bioreactor categories, mechanical/electrical actuation, sensor-based monitoring, and environmental control is presented in this chapter.

### 3.2 BIOREACTOR EMBODIMENTS

#### 3.2.1 Design Considerations

While each bioreactor implementation can be unique depending on the tissue type and application, the designer/user of a tissue bioreactor should consider a set of basic design criteria (Lyons and Pandit 2005; Partap et al. 2010; Portner et al. 2005). The bioreactor system should

- maintain a growth atmosphere at 37°C and 90%–100% humidity,
- properly control pH, pO₂, pCO₂, nutrient delivery, and waste removal,
- regulate steady versus pulsatile flow with a pump that can accurately apply small forces,
- control flow rate, volume, shear, pressure, resistance, and compliance,
- support mechanical or electrical stimulation,
- facilitate sampling of the medium, cells, and/or tissue construct,
- be constructed with biocompatible or bioinert materials (e.g., high-quality stainless steel, plastics, or other nonmetal materials),
- allow the user to easily affix the seeded scaffold,
- be easy to clean, sanitize, and maintain,
- incorporate disposable parts and/or reusable parts that are capable of being sterilized,
- offer a simple, robust design that is easy to assemble/disassemble, facilitating harvesting of cell or tissue products and minimizing tissue damage,
- utilize materials that minimize foam generation and allow the medium and tissue constructs to be visualized,
- incorporate flexible tubing and connectors to assist with assembly/disassembly, media exchanges, and microcarrier harvesting in an aseptic fashion,
- lack recesses which can harbor bacteria,
- accommodate multiple ports for various sensing devices employed during the growth phase,
• employ good fluid/gas seals and control headspace and gas exchange, and
• be scalable for the successful production of greater amounts of tissue while making efficient use of the medium.

Bioreactor designs support different types of mass-transfer flow (e.g., static versus turbulent versus laminar [Salehi-Nik et al. 2013]) depending on the cell types and the geometries of the aggregate physical tissues. Further, different physical mechanisms and bioreactor geometries can be used to accomplish a given type of mass transfer. In some cases, external mechanical/electrical stimulation may also be needed to tune the resulting tissues for their intended application environment.

Bioreactors that employ static cultures, meaning they incorporate no flow or mixing of the growth medium, have been useful for cell proliferation at a moderate scale (Georgiev et al. 2013; Martin et al. 2004). However, the resulting tissue structures can be heterogeneous, consisting of healthy cells that surround necrotic cores (Cartmell et al. 2003; Wendt et al. 2008) or poorly populated cores that result from chemotaxis—cell migration to regions of greater nutrient concentration (Goldstein et al. 2001). This is due to reliance on a diffusive mass-transfer mechanism that preferentially provides nutrients to outer cell layers while also harboring waste materials in central regions (Rolfe 2006). This chapter therefore focuses on bioreactor designs that utilize convective mass-transfer mechanisms, where continual mixing and/or flow of the culture medium helps to promote viable tissue growth in greater quantities due to improved nutrient delivery coupled with flow-induced stress. These bioreactors have various physical instantiations but a set of common thematic needs. The following subsections describe the primary categories of bioreactor form factors as well as the types of physical mechanisms that are commonly used to help stimulate tissue growth and enhance tissue viability.

### 3.2.2 Thematic Bioreactor Categories

#### 3.2.2.1 Stirred-Flask Bioreactors

A stirred-flask (a.k.a., spinner flask or stirred suspension) bioreactor vessel design is a commonly used, active bioreactor which incorporates convective flow due to the continual mixing of the culture medium (Ismadi et al. 2014; Salehi-Nik et al. 2013; Sucasosky et al. 2004). A representative solid model for a typical stirred-flask bioreactor is depicted in Figure 3.1a (after Martin and Vermette 2005; Partap et al. 2010; Rauh et al. 2011). In this arrangement, tissue scaffolds are attached to one or more suspension rods, or “needles,” that hang from the top of the flask chamber. The surface of the medium is maintained at a level such that these scaffolds are constantly submerged. Input/output ports are provided at the top of the chamber to refresh the culture medium, sample its constituents, deliver oxygen, and remove carbon dioxide. A magnetic stirring rod constantly mixes the culture medium to provide nutrient delivery to, and waste removal from, cells that occupy the outer layers of the tissue constructs. For cells that occupy interior layers of the tissue constructs, nutrient delivery and waste removal occur primarily by diffusion. While the convective flow offered by this type of design is an improvement over static diffusive approaches, the shear forces generated on the tissue constructs by the eddy currents
induced during the mixing process are heterogeneous, affecting the homogeneity of the resulting samples (Ismadi et al. 2014; Partap et al. 2010; Sucosky et al. 2004). Oxygen delivery provided at the gas–liquid interface (headspace aeration) or by a submerged sparger can be a challenge because of the low solubility of oxygen in the culture medium (Wu et al. 2014). Stirred-flask bioreactors have been useful for the assembly of artificial tissues that include cartilage (Freed and Vunjak-Novakovic 2000; Sucosky et al. 2004) and stem cell aggregates (Ismadi et al. 2014; Wu et al. 2014).

A commercial bioreactor assembly that utilizes the stirred-flask concept is depicted in Figure 3.2. The assembly consists of an Applikon Biotechnology ez-Control system (Biotechnology 2015) that monitors and regulates a 3 L, single-use Mobius bioreactor (Millipore 2016). The ez-Control unit adaptively controls pH, temperature, dissolved oxygen, foam/level, and agitation—parameters that can be managed via a touch screen interface. The 3 L, plastic, disposable Mobius bioreactor is pre-sterilized with gamma radiation and employs an impeller-like mechanism attached to a central shaft to stir the medium. It incorporates flexible tubing, vent/gas filters, ports for fluid addition/removal, and a drain port. This setup is suited for mammalian cell growth and allows samples to be acquired for offline analyses, for example, to evaluate microcarriers for cell density.

### 3.2.2.2 Rotating-Wall Bioreactors

A rotating-wall bioreactor vessel design (see the depiction in Figure 3.1b) promotes tissue growth in a laminar flow condition, keeping cellular constructs in a state of suspension, or continuous free-fall, by countering the force of gravity with rotational
forces exerted on the medium (Martin and Vermette 2005; Salehi-Nik et al. 2013; Sladkova and de Peppo 2014). This approach, intended to simulate microgravity, was originally developed by Schwarz and coworkers at Krug Life Sciences, Houston, TX (NASA, Johnson Space Center) to help protect cell cultures reserved for space shuttle experiments from the forces incurred during takeoff and landing (Schwarz, Goodwin, and Wolf 1992). The basic design, also known as a slow-turning lateral vessel (STLV), consists of two concentric cylinders that can be independently rotated (Begley and Kleis 2002). Input/output ports at the end of the cylindrical arrangement allow for the circulation of the growth medium, and oxygen is provided via a membrane that surrounds the inner cylinder. To grow tissue, mammalian cells are first seeded onto microcarrier beads (e.g., 100–200 μm polystyrene beads). As these beads accumulate cells, they group together to form larger constructs. On earth, the outer cylinder is rotated at speeds from 15 to 30 rpm. These speeds increase as the tissue structures get larger, in order to keep those constructs suspended, avoiding collisions with the cylindrical enclosure (Begley and Kleis 2002; Martin and Vermette 2005). An STLV variant referred to as a high aspect ratio vessel (HARV) reduces the rotation rate required to keep the tissue construct in suspension and also improves gas exchange. Refer to Freed et al. (1997), Martin and Vermette (2005), and Radtke and Herbst-Kralovetz (2012) for more detailed descriptions of the HARV design.

Rotating-wall bioreactors reduce shear stress levels in comparison to levels incurred in stirred-flask bioreactors. This results in tissue constructs with more homogeneous cell distributions (Goldstein et al. 2001). While arguably not well suited for the production of large tissue masses (Martin and Vermette 2005), rotating-wall bioreactors have shown promise for the growth of various types of tissue, including cartilage (Freed et al. 1997), bone (Granet et al. 1998), liver (Catapano and Gerlach 2007), neuron-like cells (Wang and Good 2001), aortic endothelial
Engineering 3D Tissue Test Systems

3.2.2.3 Perfusion Bioreactors

A perfusion bioreactor vessel (refer to Figure 3.1c) is designed such that the culture medium must flow through a porous scaffold populated with seed cells (Gardel et al. 2014; Yeatts and Fisher 2011). This approach has dual benefit, improving mass transfer of nutrients to all areas of the cell construct while providing flow-dependent mechanical stimuli to the cells (Abousleiman and Sikavitsas 2006; Partap et al. 2010; Rauh et al. 2011; Salon et al. 2009; Salehi-Nik et al. 2013; Yan et al. 2012). Flow is primarily convective in this context but is supplemented with nutrient diffusion to better maintain interior cell viability. Like rotating-wall bioreactors, perfusion reactors have shown promise for the growth of more homogeneous tissues with denser construct cores when compared to stirred-flask bioreactors (Goldstein et al. 2001; Sailon et al. 2009). Perfusion bioreactors have also offered improved cell proliferation and differentiation of osteoprogenitor cells, plus enhanced deposition of the extracellular matrix (ECM; Bjerre et al. 2008; Hofmann et al. 2003; Holtorf et al. 2005; Rauh et al. 2011).

One drawback of this design is the tendency for nutrient flow to follow the path of least resistance as determined by scaffold pore size and geometry. Flow rate optimization is a balance between nutrient delivery, metabolite transfer to/from cells, and shear stress applied to the cells (Wendt et al. 2008), for example, too much flow can result in cell “wash out,” where the forces induced by the fluid remove the cells from the construct (Singh et al. 2007). The flow rate threshold is also influenced by the need for proper O2 delivery to the tissue constructs given, as noted earlier, the poor solubility of O2 in the culture medium. Further, cell harvesting can be difficult to manage and monitor with such a design.

Perfusion bioreactors have been successfully utilized to grow a number of tissue types, including bone (Bancroft et al. 2003; Cartmell et al. 2003; Gardel et al. 2014; Gaspar et al. 2012; Goldstein et al. 2001), cartilage grafts (Carver and Heath 1999; Nesic et al. 2006), cardiac tissue (Radisic et al. 2008; Tandon et al. 2013), intestinal tissue (Kim et al. 2007), vascular grafts (Hoenicka et al. 2010; Radisic et al. 2008; Williams 2003), and stem cells (Bjerre et al. 2008).

3.2.2.4 Other Bioreactor Form Factors

A number of creative bioreactor embodiments exist that are either variations on the themes noted above or are novel designs matched to a specific tissue need. A few selected examples follow:

- In a rocker platform, or wave bioreactor vessel, a flexible bag holding the culture medium is rocked back and forth. The tissue medium is aerated due to agitation from surface wave action, and nutrients are provided to the tissue construct through convective flow (Amrollahi and Tayebi 2016; Blose et al. 2014; Lyons and Pandit 2005; Yuk et al. 2011).
- A hollow fiber bioreactor vessel consists of a closed, medium-filled vessel filled with semipermeable hollow fibers. These fibers emulate blood vessels by providing nutrients and removing waste. Primarily intended for cells (Sanford et al. 2002), kidney cells (Xu et al. 2004), epithelial cells (Radtke and Herbst-Kralovetz 2012), stem cells (Lei et al. 2011), and skin (Lei et al. 2011).
mammalian cell growth, this design offers the advantage of better nutrient delivery to the center of the tissue, but oxygen delivery can be a challenge (Martin and Vermette 2005; Tharakan and Chau 1986). Monitoring cell growth and determining cell removal efficiency at harvest can also be a difficult.

- In an airlift reactor vessel, oxygen is supplied to the tissue through the delivery of air at the bottom of the vessel. A draft tube is needed to prevent sparging due to direct air contact with the developing tissue. Nutrient availability can be a challenge due to the lack of mixing of the glass spheres or polystyrene beads upon which cells are seeded, since they tend to settle on the bottom of the vessel (Al-Mashhadani et al. 2015; Martin and Vermette 2005).

- A double-chamber bioreactor vessel is employed for the growth of larger, more complex tissue constructs such as osteochondral grafts and tracheal allografts. Two chambers, for example, a chondral compartment and a bone compartment, each with their own mixing apparatus, are separated by a membrane, and the tissue scaffold spans the membrane so that it resides in both chambers (Chang et al. 2004; Haykal et al. 2014; Wendt et al. 2005).

- Microfluidic bioreactors based on the integration of electronics at, for example, the CMOS level are becoming available for portable and implantable applications (Christen and Andreou 2007; DeBusschere and Kovacs 2001; Kim et al. 2011).

### 3.3 MECHANICAL/ELECTRICAL TISSUE STIMULATION

Hydrodynamic forces such as the shear stress that fluid flow exerts on tissue constructs in stirred-flask, rotating-wall, and perfusion bioreactors can be healthy for tissue development by improving cell proliferation, expression of osteogenic marker genes, and ECM mineralization (Rauh et al. 2011). For musculoskeletal tissues (e.g., bone, cartilage, ligaments, and tendons) and cardiovascular tissues (e.g., cardiac tissue and vessels), additional pulsatile and steady-state mechanical forces created by bioreactor actuators can further improve the viability of those constructs. In the case of cardiovascular tissues, electrical/electromagnetic supplements provide additional improvements. This section addresses the tissue-stimulation role of bioreactor instrumentation as represented by mechanical actuation and electrical/electromagnetic excitation.

#### 3.3.1 MECHANICAL STIMULATION

The incorporation of mechanical forces into tissue bioreactors is essential for the growth of viable load-bearing tissues, such as articular cartilage, which will atrophy in the absence of mechanical stimulation (Darling and Athanasiou 2003). External mechanical stimuli can also improve the homogeneity, structural viability, growth rate, and functionality of cultured tissue (Carver and Heath 1999; Mauck et al. 2000), in part because those stimuli encourage the production and mineralization of ECM, the noncellular tissue component that provides “not only essential physical scaffolding for the cellular constituents but also initiates crucial biochemical and biomechanical cues that are required for tissue morphogenesis, differentiation and homeostasis”
Mechanical stimulation can also influence cell differentiation. One study illustrated that a combination of translational and rotational strain could differentiate mesenchymal progenitor cells from the bone marrow (embedded in a collagen gel) into aligned collagen fibers with a density indicative of ligament cells but with no cell markers indicative of bone or cartilage (Altman, Horan et al. 2002). As noted in Section 3.1, the types of engineered tissues that benefit from mechanical stimulation include bone, cartilage, tendons, ligaments, and vessels. (Refer to that section for citations associated with mechanical stimuli as well as citations for reviews that address structural tissue engineering.)

Bioreactors that incorporate mechanical stimulation take three primary forms:

1. **Compression bioreactor:** This type of bioreactor generally employs a computer-controlled linear actuator and a type of flat plate to deliver dynamic loading to the tissue scaffold(s), where the amplitude and rate of the displacement depend on the tissue type and size. For a tissue like cartilage, this compressive loading can help to improve fluid flow in the center of the scaffold and result in a more homogeneous construct with an elastic modulus that is close to that of native cartilage (Mauck et al. 2000). Load cells can be used to deliver real-time feedback regarding the force applied to the engineered tissue and scaffold (Popp et al. 2012). Compressive strains of 1–10% at a rate of 0.5–1 Hz over durations of several hours per day are typical for cell differentiation studies (Sladkova and de Peppo 2014).

2. **Tensile strain bioreactor:** The design of a strain bioreactor is similar to that of a compression bioreactor, but the scaffold is clamped in such a way that the linear actuator delivers a tensile strain instead of a compressive force. Rotational strains can also be applied (Altman, Horan et al. 2002). This type of bioreactor is a suitable match for tissues such as ligaments and tendons, which can exhibit a substantive increase in Young’s modulus when compared to nonloaded control tissues (Garvin et al. 2003). Tensile strains of 0–30%, forces up to 200 N, and rates of 0–2 Hz are typical (Riehl et al. 2012).

3. **Hydrostatic pressure bioreactor:** A hydrostatic pressure chamber can be implemented with a medium-filled container and a piston driven by a computer-controlled linear actuator. Here, either the piston itself is sealed but in direct contact with the medium, or an impermeable membrane separates the two (Darling and Athanasiou 2003; Elder and Athanasiou 2009). Alternatively, the entire vessel containing the growth medium and tissue scaffold can be placed in a water-filled pressure chamber, where an impermeable membrane separates the two types of fluid media (Watanabe et al. 2005). Tissues subjected to hydrostatic forces are typically cultured separately and then placed in the hydrostatic chamber to be loaded for a predetermined treatment period using either constant or slowly varying pressure. For hydrostatic pressure systems applied to chondrocytes for cartilage tissue engineering, static pressures of 1–10 MPa have been applied for time intervals from a few hours to a few weeks. Dynamic systems have cycled at rates of 0.25–1 Hz using similar pressure ranges and time intervals. Refer to Tables 1 and 2 in Elder and Athanasiou (2009) for a good comparative
3.3.2 Electrical Stimulation

Electrical stimulation has been primarily employed in bioreactors designed for cardiac tissue growth, where the end goal is to replace tissue that has been destroyed by myocardial infarction: a “patch for a broken heart” (Vunjak-Novakovic et al. 2011). Pulsed electrical excitation (e.g., a 1 Hz square wave at 5 V/cm (Radisic et al. 2008); a 2 ms pulse at 3 Hz and 3 V/m (Tandon et al. 2011)) has been demonstrated to help induce contractile properties of engineered cardiac tissue, where the safe range of operation appears to be 0–8 V/cm (Tandon et al. 2008). The stimulation amplitude and frequency, as well as the electrode type, have a significant bearing on the quality of tissue produced (Tandon et al. 2011). Electrical stimulation can also be combined with mechanical stimulation in a hybrid bioreactor. Liao and coworkers illustrated that synchronized electromechanical stimulation (electrical stimulation coupled with mechanical cues) can improve the alignment and elongation of skeletal myoblasts as well as the upregulation of cardiac proteins (Liao et al. 2008).

3.3.3 Electromagnetic Stimulation

EMFs have long been known to assist with bone growth during fracture healing and bone stretching (Bassett et al. 1977; Eyres, Saleh, and Kanis 1996). In an EMF bioreactor, the tissue scaffold is placed in the electric field generated by a pair of Helmholtz coils. Typical flux densities in recent research are 0.3–2 mT, with pulse durations of 0.3–60 ms at pulse frequencies of 15–75 Hz (see Table 1E in (Rauh et al. 2011)). Field strengths can be verified with Hall-effect probes (Fassina et al. 2006). Benefits of EMF stimulation include improved osteogenic cell differentiation, enhanced osteoprogenitor cell proliferation, and increased mineralization of osteogenic cells (Rauh et al. 2011).

3.4 Aggregate Component Architecture for Bioreactor Systems

Component architectures, or system block diagrams, have been presented in the literature for various types of tissue bioreactor systems, where the components employed depend on the type of bioreactor vessel, the fluid flow requirements, and the means for nutrient (e.g., oxygen) delivery and waste removal. This section presents an aggregate component architecture that lays out a superset of these system components, or building blocks, with a goal of identifying the sensing and control needs that are relevant within this overall design space. These needs drive the remaining discussion in this chapter.

The aggregate component architecture is depicted in Figure 3.3. This layout attempts to incorporate the collective components and functionality noted in the block diagrams and the accompanying text presented in a selected collection of
Engineering 3D Tissue Test Systems

technical papers and reviews: Rauh et al. (2011, Figure 3), Singh et al. (2007, Figure 2), Kim et al. (2007, Figure 1), (Lyons and Pandit 2005, pp.18–19), Barron et al. (2003, Figures 1, 2, and 4), Wang et al. (2013, Figures 1, 4, and 5), Portner et al. (2005, Figure 1), Portner et al. (2005, Figures 2.2 and 2.3), Chen and Hu (2006, Figures 1 and 2), Bilodeau (2004, Figure 2.1), Bancroft et al. (2003, Figure 5), Yeatts and Fisher (2011, Figure 3), Gaspar et al. (2012, Figures 1 and 2), and Amrollahi and Tayebi (2016, Table 1 and Figure 3). Note that block diagrams for systems geared toward cell seeding will be different, as they have a short-term goal of populating a scaffold with cells just before that scaffold is removed and placed in a bioreactor growth chamber (Wendt et al. 2005).

The lower panel in Figure 3.3 displays five general embodiments of the Bioreactor Vessel component in the center of the diagram. Each of these was addressed in a prior section. In the flow diagram, blue lines with arrows denote the direction of fluid flow, and black lines denote gas flow. While the aggregate diagram appears complex, it may be helpful to note that several operational modes are depicted, labeled as green letters “A” through “E,” where the modes depend on the type of bioreactor and the related nutrient delivery requirements. For modes A through D, the diagram makes the assumption that the collection of components (medium/waste reservoirs, bioreactor vessel, pump, valves, and tubing) all reside within a closed incubator space maintained at 100% humidity, 37°C, 5% CO₂, and 21% O₂ (Chen and Hu 2006; Christen and Andreou 2007; Wang et al. 2013). Prior to operation, all components to

FIGURE 3.3 Aggregate component architecture representative of multiple classes of tissue bioreactor systems.
be operated within the incubator space are sterilized, for example, using an autoclave and ethylene glycol gas (Kim et al. 2007). These operational modes are characterized as follows:

- **Mode A**: Incubator air is fed directly to the bioreactor vessel for either headspace, airlift, or inner tubing aeration (Al-Mashhadani et al. 2015; Martin and Vermette 2005). Note in the lower panel of Figure 3.3 that only the stirred-flask and rotating-wall bioreactors depict an input port for that air (black, hashed triangle). The other bioreactors typically employ a flow perfusion model where the fluid is aerated externally prior to reaching the bioreactor vessel.

- **Mode B**: Incubator air is fed directly to the medium reservoir for headspace aeration.

- **Mode C**: Tubing is employed that is permeable to both carbon dioxide and oxygen, ensuring gas equilibrium with the incubator space (Bancroft et al. 2002).

- **Mode D**: An Oxygenator provides nutrients to the medium before it is pumped into the bioreactor vessel (Chen and Hu 2006), which is common when employing flow perfusion bioreactors that may have an added mechanical or electrical stimulation role (Janssen et al. 2006; Kim et al. 2007).

- **Mode E**: In this mode, the system components are not assumed to reside within an incubator. Rather, they reside in open air, and the environmental chamber heats the medium to 37°C and employs its own internal bubble humidifier and aeration scheme to provide dissolved oxygen to the medium (Wang et al. 2013).

With regard to other figure details:

- The **Pump** in many systems is, for example, a six-roller peristaltic pump capable of delivering fluid at various computer- or manually-controlled rates. Flow rates can vary, for example, from 1.5 mL/min up to 3 L/min depending on the vessel/tissue type and the number of pump channels (Kim et al. 2007; Lyons and Pandit 2005). Flow types can be steady state, oscillatory (less effective for bone), or pulsatile, where dynamic flow becomes less potent for tissue growth as frequency increases (Jacobs et al. 1998).

- The generic **Bioreactor Vessel** (container, single/dual chamber, or cartridge) offers input ports for incubator air, the fluid medium, and a cable that contains lines for mechanical and/or electrical stimulation (e.g., to drive the stirred-flask needle, the rotating-wall cylinders, linear actuators, etc.). It offers output ports for waste (exhaust) gases and the fluid medium. A tissue port offers a means to insert/extract the tissue scaffold(s) or to sample the cells within the medium.

- The lower left portion of the diagram depicts a means to sample the flow medium for offline analysis.

- A **Filter** offers a means to remove particles and other contaminants from the flow medium, and an impedance unit ($Z_{RC}$) provides the desired resistance and compliance for the flow “circuit” (Barron et al. 2003).
• The Waste Reservoir can function as a repository for a spent or contaminated medium, or it can be part of the active flow loop, occasionally offering a role as an exchange tank to refresh part of the medium while the system is in operation. Some systems do not incorporate this element, choosing instead to recirculate the medium from the bioreactor vessel back into the Medium Reservoir.

Note that computerized measurement and control connections are not specified in this connectivity diagram to avoid clutter. In many commercial systems (e.g., the Applikon system depicted in Figure 3.2), the incubation parameters, medium concentration/pH, pump, valves, and mechanical/electrical stimulation mechanisms are computer controlled.

The variety of needs met by the numerous instantiations of bioreactor systems is understandable, as each tissue construct is unique in terms of its cell distribution, nutrient, mechanical/electrical conditioning, and application requirements. In response to this reality, some investigators have moved toward the development and use of more modular, componentized systems, where interchangeable parts can be more rapidly combined to construct systems well matched to the needs of the target tissue, avoiding “one-off” designs that are typical in this research arena (Illa et al. 2014; Lovett et al. 2010; Mazzei et al. 2010; Orr and Burg 2008; Vinci et al. 2011).

3.5 SENSING AND CONTROL IN BIOREACTOR SYSTEMS

An “instrument” can be defined as any tool that helps one accomplish a task or measure its progress. To this point, this chapter has addressed bioreactor instrumentation issues that relate to bioreactor design requirements, chamber embodiments that address those needs, flow/medium management, actuators, and mechanical/electrical stimulation. The following sections complete that instrumentation overview with a summary of the sensing and control mechanisms that come into play when designing a bioreactor system. Here, the notion is that sensors are employed to measure (a) parameters that a researcher wishes to quantify, as a means to gauge the operational viability of the system and its resulting tissue products and (b) parameters that may also need to be controlled to maintain system viability.

3.5.1 PARAMETERS AND SENSING METHODS

The number of bioreactor system parameters that warrant attention is potentially myriad. Parameters of interest for a given system (refer to Figure 3.3) depend on the type of tissue, the bioreactor vessel design, the approach to medium flow, the methods used to deliver nutrients to the medium/tissue, the presence of active components (e.g., mechanical actuators), the relative effects of waste/contamination, and the desire to track those parameters occasionally (e.g., manually) versus in real time. Table 3.1 lists the more common parameters tracked during bioreactor system operation, along with representative parameter ranges and sensing approaches. Some of these parameters are tracked offline versus online, and some are simply measured
**TABLE 3.1**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Typical Range or Value</th>
<th>Sensing Method(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Incubator/medium temperature</td>
<td>25–39°C</td>
<td>Thermistors or platinum resistance thermometers—see Section 4.4 in Najafpour (2015)</td>
</tr>
<tr>
<td>pH</td>
<td>6–8</td>
<td>Commercial blood gas analyzer (Ramaswamy et al. 2014) such as the NOVA Bioprofile 400 (Nova Biomedical), which can measure 14 different components in one pass; pH probes (e.g., AppliSens probes supported by the Applikon system in Figure 3.2); Fiberoptic sensors and variations on a standard hydrogen electrode—see Xu et al. (2006) Section 4.6 in Najafpour (2015); CMOS chip (Hammond et al. 2004)</td>
</tr>
<tr>
<td>pO₂</td>
<td>0%–20%</td>
<td>Commercial blood gas analyzer (Ramaswamy et al. 2014); Electrochemical sensor with polymeric membrane; optical fiber sensors (Rolfe 2006)</td>
</tr>
<tr>
<td>pCO₂</td>
<td>0%–8%</td>
<td>Commercial blood gas analyzer (Ramaswamy et al. 2014)</td>
</tr>
<tr>
<td>Dissolved oxygen</td>
<td>0–50,000 mg L/h</td>
<td>Tubing method, mass spectrometer probes, or electrochemical detectors—see Section 4.5 in Najafpour (2015); dissolved oxygen probes (e.g., AppliSens probes supported by the Applikon system in Figure 3.2)</td>
</tr>
<tr>
<td>O₂ transfer rate</td>
<td>0–0.5 × 10² s⁻¹</td>
<td>Sulfite oxidation method and others—see Table 2 in Salehi-Nik et al. (2013); Table 1 in Garcia-Ochoa and Gomez (2009)</td>
</tr>
<tr>
<td>Flow rate (steady state, oscillating, or pulsatile)</td>
<td>1.5 mL–3 L/min</td>
<td>Particle image velocity and others—see Table 3 in Salehi-Nik et al. (2013); see also Janssen et al. (2006), Kim et al. (2007), and Lyons and Pandit (2005)</td>
</tr>
<tr>
<td>Flow profile</td>
<td>N/A</td>
<td>Particle image velocity and others (Salehi-Nik et al. 2013; Singh et al. 2007)</td>
</tr>
<tr>
<td>Compressive strain</td>
<td>1%–10% at 0.5–1 Hz over hours</td>
<td>Load cells, tomography, ultrasound, and others (Popp et al. 2012; Sladkova and de Peppo 2014; Wang et al. 2013)</td>
</tr>
<tr>
<td>Tensile strain/displacement</td>
<td>0%–30%; 0–200 N; 0–2 Hz</td>
<td>Actuator displacement and others (Popp et al. 2012; Rauh et al. 2011)</td>
</tr>
<tr>
<td>Surface shear stress</td>
<td>5 × 10⁻⁵ Pa</td>
<td>Particle image velocity (Rauh et al. 2011)</td>
</tr>
<tr>
<td>Hydrostatic pressure</td>
<td>1–10 MPa; hours to weeks; 0.25–1 Hz</td>
<td>Diaphragm-based commercial pressure sensors, load cells, and others—see Tables 1 and 2 in Elder and Athanasiou (2009) for studies</td>
</tr>
<tr>
<td>Tissue cell count</td>
<td>cells/mL; cells/cm²</td>
<td>Upon cell removal from the bioreactor: hemocytometer, electronic cell counter, cell image analysis, or a metabolic activity assay (Divieto et al. 2013)</td>
</tr>
</tbody>
</table>

(Continued)
to quantify a process, whereas others play important roles for system control. These parameters are an aggregation of parameters noted in a number of bioreactor papers and reviews, some of which are cited in the table.

### 3.5.1.1 Offline versus Online Measurement Modes

Some parameters can be measured occasionally (e.g., every few hours or days) and are therefore often acquired manually, resulting in an *offline* measurement (e.g., refer to the Sample valve and the Bioreactor Vessel Tissue Port in Figure 3.3). Other parameters must be tracked more closely, or *online*, perhaps in real time, as a means to obtain continuous feedback regarding the viability of the tissue growth process and the surrounding support system. Such measurements are facilitated with computer analysis and control. As an example of an offline mode, a traditional method to track the viability of cell cultures is to incorporate flow injection analysis, where sensors are not in contact with the culture medium. Rather, a sample is removed from the bioreactor and analyzed offline, as is the case with the cell cultures related to the work depicted in Figure 3.2. These “spot checks” represent, at best, a time-averaged view of bioreactor activity and the viability of the physicochemical environment, even if the environmental variables themselves (e.g., temperature and flow) are continuously controlled. An active area of research and development relates to the real-time tracking of bioreactor parameters (e.g., tissue construct development) and continuous control of the bioreactor environment based upon these assessments (Christen and Andreou 2007; Popp et al. 2012; Xicai et al. 2008). This leads to the notion that one could optimally tweak processes such as tissue growth or cell differentiation based on immediate or just-in-time feedback.

### 3.5.1.2 Combined Parameter Assessments

Parameters are rarely monitored on their own. Rather, groups of parameters must often be assessed in order to obtain a sensible snapshot of the process of interest. For example, Xicai et al. note that the collection of pH, oxygen tension, carbon...
dioxide tension, and temperature are the most important physicochemical parameters, as they affect the cell expansion rate and therefore the cell population within the chamber medium (Xicai et al. 2008). They also note that cell growth, differentiation, and cell death are affected by four nutrient and metabolite parameters: glucose, glutamine, lactate, and ammonia. Likewise, Rauh and coworkers note that temperature, oxygen concentration, pH, nutrient concentration, and biochemical and mechanical stimuli all modulate bone growth in complex bioreactors (Rauh et al. 2011). Combined parameter assessments are also helpful in determining the status of components within the bioreactor system. For example, the functional viability of an oxygenation unit, as depicted in Figure 3.3, can be assessed by tracking the pH, pO₂, and pCO₂ of the culture medium over a range of incubator CO₂ concentrations (Bioprofile 400 Analyzer, NovaBiomed [Kim et al. 2007]).

### 3.5.2 Controls

The purpose of a control system is to regulate processes that might otherwise exhibit unsatisfactory static or dynamic behavior if left alone. The regulation process might entail parameter stabilization (i.e., keeping one or more parameters constant) or process implementation (i.e., allowing parameters to change according to a prescribed protocol). Consistent with other authors, Lyons and Pandit note several bioreactor parameters that warrant control (Lyons and Pandit 2005):

- **Biochemical parameters**: Temperature (37°C), humidity (100%), pH (7.2–7.4), nutrients (e.g., oxygen or glucose), partial pressures of dissolved gases (e.g., pO₂ at 21%, pCO₂ at 5%), and waste products.
- **Biomechanical parameters**: Flow rate, shear, pressure, volume, resistance, and compliance.

Note that these parameters can be interdependent on one another in a closed, fluid-filled system. In the context of controls, oxygen must be provided at the same rate it is consumed to avoid cell apoptosis; medium flow at a suitable rate must be present and often pulsatile to provide nutrients, shear stress, and proper mechanical loading to cell constructs; pH must be balanced in order to main cell homeostasis; heat must be maintained at a normal body temperature of ~37°C (the optimal growth temperature for most mammalian cells); humidity must be maintained at 100% to avoid water evaporation and related changes in medium concentration; and CO₂ concentrations can be heightened to help maintain the proper medium pH (Barron et al. 2003; Christen and Andreou 2007; Lyons and Pandit 2005).

Bioreactor control techniques have existed for a few decades, where early implementations were applied to fermentation bioreactors (Royce 1993; Sonnleitner et al. 1991). These techniques have been extended to tissue bioreactors (Cout et al. and Mantovani 2012; Scutcher 2011), which in large part take advantage of control theory developed for a number of application domains. Bioreactor controls take multiple forms, from easy-to-implement manual/mechanical methods up to complex, adaptive, computer-controlled approaches that utilize real-time feedback. The paragraphs below briefly describe a few of these approaches.
As indicated by the bubble humidifier in Figure 3.3, humidity control within an incubator can be relatively straightforward to implement by allowing dry gas to rise through a fluid medium. This is a control system in its most basic form—a simple mechanical process. Temperature control as related to incubator/medium temperature is also well known in the bioreactor domain and is often implemented with a proportional-integral-derivative (PID) controller (Christen and Andreou 2007). This type of controller is well suited to parameters such as temperature and fluid level that are likely to change slowly and in small proportion. A PID controller, easily implemented on a microprocessor platform (DeBusschere and Kovacs 2001) or a personal computer (e.g., running LabVIEW [Xicai et al. 2007]), calculates the sum of the present (proportional) error, past (integral) error, and anticipated (derivative) error as they relate to a parameter value versus a targeted set point. The controller then adjusts a control variable (e.g., a heater position control) in an attempt to minimize the total error over time. Sources of information regarding this type of control are numerous. A reasonable summary is included in Astrom and Murray (2008). Other information regarding bioreactor temperature control can be found in Section 4.4 of Najafpour (2015).

Bioreactors employ pH control in an effort to maintain a medium pH between approximately 7.2 and 7.4. If the measured pH (see Table 3.1) falls below a prescribed value, the control system initiates the release of, for example, acetic acid, into the medium, whereas if pH is too high, an alkali pump engages—refer to Section 4.6 of Najafpour (2015). pH control can be implemented with an analog controller (Lee et al. 1987), PID controller, or first-order closed-loop control system (Griswold 2004), where the set point is the target pH and the controller drives the acid/base pumps. Flow control and mechanical stress/strain control are a coupled problem. Medium flow can be important for both nutrient delivery and waste removal; yet, any type of flow results in surface shear stress, which can either be healthy for tissue development or result in surface-cell loss if flow rates or eddy currents are excessive. As noted earlier, tissue bioreactor systems also incorporate active flow profiles (steady state, oscillating, or pulsatile flow via a peristaltic pump) as a mean to induce mechanical stress/strain stimuli to tissue constructs and therefore increase their viability. Flow control systems must therefore specify the process for the physical flow profile (e.g., through voltage-level changes as input signals for the peristaltic pump) as well as confirm the amount of resulting flow (or applied stimulus, often via load cells [Popp et al. 2012]) and then offer the data as feedback to the computer that is driving the pump and/or actuator. Such systems can therefore incorporate relatively complicated control schema. For example, Xiang and coworkers designed a dual-frequency mechanical system that relied on the method of friction compensation to deliver linear mechanical loads to cartilage tissue (Xiang et al. 2015). Low-frequency pulsations (0–3 Hz) were designed to mimic human walking, whereas higher frequency pulsations (20–50 Hz) were designed to mimic muscle and ligament loading. This work employed a programmable multi-axis controller with a feed-forward closed loop. Translational controls can also be coupled with rotational controls, as demonstrated by Altman and colleagues in an effort geared toward engineered anterior cruciate ligament (Altman, Lu et al. 2002). Rotational controls are also traditional in the bioreactor domain for controlling the rate of...
rotating-wall vessels that must vary their rotation speed as a function of tissue construct size (Freed et al. 1997).

Dissolved oxygen control has also received attention in the bioreactor design community for over 20 years (Lee et al. 1991; Najafpour 2015), as this parameter is a direct indicator of nutrient availability to the engineered tissues. Variants of a linear PID controller have been used to regulate dissolved oxygen in stirred-tank bioreactors, where the stirrer speed was the actively controlled element (Akesson and Hagander 1997). Scutcher noted that such PID controllers are adequate when the assumption is that oxygen is the only contributing factor (Scutcher 2011). This tuning can be precise, as demonstrated by Whiffin and colleagues through the design of a “starvation-based” transient controller that tracked dissolved oxygen as a mean to control the delivery of glucose to fed-batch cultures of recombinant *Escherichia coli* (Whiffin, Cooney, and Cord-Ruwisch 2004).

### 3.6 CONCLUSION AND PERSPECTIVE

This chapter presented an overview and thematic description of various types of bioreactors and the engineered tissues they are designed to support. These systems employ active flow, possibly supplemented with mechanical and/or electrical stimuli to aid tissue growth and increase the viability of the resulting constructs for their intended application environments. This overview was followed by the presentation and description of an aggregate component diagram that can be a suitable means to compare different classes of bioreactor systems. Finally, the chapter provided an overview of the types of environmental and tissue parameters that are tracked with bioreactor-based sensors, followed by an overview of the types of control systems used to stabilize these parameters or drive tissue-development processes. A number of technical papers and reviews are cited in support of this overview.

While advancements in this domain are substantive, future work remains in various areas of bioreactor development. The first broad area of need relates to the real-time tracking and dynamic control of bioreactor parameters and components, including the ability to easily change nutrient delivery profiles and hardware component operations mid-experiment. This relates to what is measured and/or controlled at (1) the vessel (chamber) level (e.g., cell counts, differentiation status, and 3D structure development) using new sensing and imaging techniques, coupled with appropriate software and display support, as well as (2) the system level, which speaks to integrated online sensors for fluid flow/status, valve operation, etc., much the same way as a good vehicle dashboard allows one to view the system status, and ideally history, at a glance.

For example, as a driver for their own work, Xicai et al. note that, “there is no compact measurement system that records a variety of physicochemical parameters (such as pH, dissolved oxygen, tension, nutrient, and metabolite concentrations) simultaneously and in real-time” (Xicai et al. 2008). This is, in part, driven by the need for new types of sensors, as mentioned by Yan and coworkers: “it is also noted that the information (such as oxygen and glucose distribution) provided by model simulation may not be readily accessible using current experimental techniques” (Yan et al. 2012). Similarly, Martin and coworkers note that, “the presence of urea
is rarely monitored in reactors, in part because on-line sensors are difficult to find” (Martin and Vermette 2005). Scuteri noted that the inability to monitor cell count online presented a significant obstacle in the ability to design the control system for a vascular bioreactor (Scuteri 2011).

These sensing needs are coupled with mid-experiment control needs in terms of how nutrients are delivered during different phases of the tissue-development process. Xicai et al. state this well when they note that, “in the future, the ability to supply cells with dynamic profiles of nutrients, oxygen and growth factors in an optimal manner” will alter the tissue engineering paradigm for the better (Xicai et al. 2008). Spatial and temporal information regarding the physicochemical well-being of the bioreactor system/medium, coupled with information regarding the development of the related tissue constructs, is an important step to this end. This is consistent with a statement by Scuteri, who noted that, “one key area is how process control can be designed to accommodate changes in cellular metabolic demand as immature cell types differentiate into more mature cell types” (Scuteri 2011).

These sensing and control needs supplement a more common statement of needs that is mentioned more often in the literature and is twofold: (1) the need for bioreactors that support the development of more complicated 3D tissue structures, possibly with layered or encapsulated tissues (Atala et al. 2012; Mikos et al. 2006), coupled with (2) the need for cell and tissue production at a much greater scale in order to create tissue constructs that are clinically useful (Garcia-Ochoa and Gomez 2009; Prensosil and Kino-oka 1999; Tharakan and Chau 1986). Some authors also note the need for portable, small-scale (even implantable) bioreactors that add the mobility goal to the desired set of bioreactor requirements (Christen and Andreou 2007; DeBusschere and Kovacs 2001; Kim et al. 2011).

From a system-design perspective, this research and development community can benefit from modular systems as improvements on current modular approaches (Illa et al. 2014; Lovett et al. 2010; Mazzei et al. 2010; Orr and Burg 2008; Vinci et al. 2011; Vorstius et al. 2011). The following system features apply:

- Interchangeable components designed for rapid system-level reconfiguration: plug-and-play components that can be rapidly reconfigured to create a bioreactor system well matched to the needs of the tissue constructs.
- Bioreactor system components that can be easily altered to support new tissue and sensing needs, including chambers with flexible insertion/extraction ports, adaptability for different types of scaffolds, and support for different types of mechanical or electrical stimuli.
- Proper software/app support, including customizable software interfaces for various component configurations (e.g., as an update to existing LabVIEW VI-based systems [Xicai et al. 2008]). This includes phone- and tablet-level support for personal apps that allow communication with wireless bioreactor components that may, for example, reside in sealed vessels where wired access can be difficult.
- Planning and assembly tools that offer intelligent advice to researchers based on lessons learned, in much the same way that existing board-layout tools aid electrical designers.
Such a toolset will promote the development and use of a variety of unique but robust bioreactor systems, each assembled in a “Lego-like” manner to support the growth and monitoring needs of an increasingly diverse array of cellular constructs.

ACKNOWLEDGMENTS

The author acknowledges Charles Carlson, Electrical Engineering doctoral student at Kansas State University, Manhattan, KS for the solid-model renderings of the bioreactor form factors in Figures 3.1 and 3.3. Appreciation is also offered to Dr. Mark Weiss, Professor, Kansas State University, Department of Anatomy and Physiology for his early feedback on the chapter content as well as the images depicting the commercial bioreactor in Figure 3.2.

REFERENCES


