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Engineered Muscle Actuators:
Cells and Tissues

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9.1 INTRODUCTION

Muscle tissue mechanical actuators have evolved over millions of years within animals as nature’s premier living generators of force, work, and power. The unparalleled efficiency and plasticity of form of living muscles arise from the properties of biomolecular motors. Muscle cells serve to self-organize, maintain and repair, and control the mechanical actions of large arrays of biomolecular motors. Muscle tissues provide the chemomechanical interface between muscle cells and the environment. It is at the tissue level that muscle becomes a practical, responsive, and robust actuator because of the presence of the critical tissue interfaces: the neuromuscular interface, the myotendinous junction (MTJ), and the vascular bed. Tendon tissue is an extension of the muscle extracellular matrix (ECM) and muscle tendon junctions at the ends of each muscle fiber. The mechanical structures that make up this transition from muscle to tendon are critical for the transduction of force, work, and power between muscle tissue and the external environment. Systematic derangements of these structures at any level result in severe and sometimes lethal disease resulting from the impairment of the contractility of skeletal muscle and the increased susceptibility to contraction-induced injury.

A detailed description of the biology of muscle development and morphology is beyond the scope of this chapter. The interested reader is referred to the definitive text on this subject: Myology (Volume I, chapters 1, 2, 3, and 4, Engel and Armstrong, eds., 1994, McGraw-Hill).

Embedded within the genetic code of naturally occurring muscles lies the potential to build mechanical actuators that are adaptable, self-healing smart materials (with integrated sensors for position, force, and velocity) from the submillimeter to meter size scale in the form of tubes, rods, sheets, hollow spheres, cones, and many other physical configurations. The key to engineering efficient, robust, and practical muscle actuators lies in understanding the mechanisms by which to control muscle phenotype, that is, the size, shape, fiber type, and architecture of the muscle tissue itself. The environmental signals that control muscle phenotype are mediated by the tissue interfaces, and thus it is critical to understand, and to ultimately engineer, adequate tissue interfaces for muscle actuators.

There are four basic approaches (classes) to engineering functional living muscle actuators: the use of whole surgically explanted muscles, recellularized muscle within a muscle-derived ECM, scaffold-based engineered muscle, and self-organized muscle tissue engineered culture. When considering muscle tissue as a functional element in an engineered system it is important to formulate well-defined quantitative Figures of Merit (FoM). It is also important to note that at the time of this writing, practical living muscle actuators are an as-yet unachieved research
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objective. The remainder of this chapter will discuss many of the reasons why living muscle is being given serious consideration for use as a mechanical actuator in hybrid robotic systems, as well as the many special considerations involved when attempting to employ living actuators in an engineered biohybrid system. The incorporation of functional living elements into otherwise synthetic engineered systems is called biomechatronics.

9.2 SYSTEMS ENGINEERING OF LIVING MUSCLE ACTUATORS

Tissue engineering of skeletal muscle could be broadly defined to include any alteration to or enhancement of the musculature of a living organism. This definition, though interesting, would not be specific enough to be useful, as it would include the agricultural use of steroids to rapidly increase the total lean body mass of livestock, the use of resistance training by athletes to induce hypertrophy, and surgical procedures including transplants and flaps in which preexisting skeletal muscle is modified and utilized in clinically relevant procedures (including graciloplasty, cardiomypoplasty, and musculoskeletal reconstructive surgery). Though all of these approaches to the modification and use of skeletal muscle are of interest, this chapter will only address skeletal muscle tissue engineering to generate functional muscle tissues actuators.

Successful tissue engineering must include a focus on the organization of large numbers of cells into higher-order structures that confer emergent properties, which are an important aspect of the tissue-level function. Thus, the engineering of functional tissues is by definition within the domain of “systems engineering.” These living structures may be known as tissues or organs depending on the level of anatomical complexity and structural integration. Though all tissue functions arise from fundamental cellular mechanisms, the organization of tissues and organs confers function that is not possible to achieve with individual cells or masses of unorganized cells in a scaffold. By analogy, a pile of bricks does not provide the functionality of a house, nor does a crate full of car parts function as an automobile. Furthermore, when removed from an organism, muscle tissue in general does not persist for long periods. Isolated from its proper environment, muscle tissue tends to degenerate rapidly. The environment that is required to maintain healthy, adult phenotype muscle is highly complex and incompletely understood, involving many chemical, structural, and mechanical signals. In order to understand both natural and tissue-engineered skeletal muscle, we must have a clear working definition of muscle function and understand how the structure of muscle contributes to the emergence of that function. A major challenge facing the use of muscle tissue as a practical living actuator is the identification of suitable tissue interfaces to allow the application of external cues (such as mechanical forces and growth factors) to guide tissue development and to allow the controlled generation of mechanical power.

9.3 MUSCLE: NATURE’S ACTUATOR

Skeletal muscle accounts for nearly half of the total mass of the average adult human and is unique in its ability to actively modify its mechanical properties within tens of milliseconds to allow animals to rapidly react to their environment. Muscle tissues have evolved over the last several billion years as nature’s premier living generators of force, work, and power. The success of muscle tissue actuators hinges in part upon the very favorable efficiency of biomolecular motors. Biomolecular motors are the mechanically functional units of muscle cells and tissues, providing motility and mobility for organs and organisms. Muscle cells (also known as muscle fibers) serve to self-organize, maintain and repair, and control the mechanical actions of large arrays of biomolecular motors. The tremendous plasticity of form of muscle actuators is first realized at the level of cells: biomolecular motors are added in parallel to allow greater force generation, and are added in series to permit more rapid movements over larger displacements. Damaged biomolecular motors are repaired or replaced by
intracellular mechanisms. Muscle tissues provide the chemomechanical interface between muscle cells and the environment. It is at the tissue level that muscle becomes a practical, responsive, and robust actuator. Tissue level organization provides an ECM for the mechanical support and mechanotransduction of signals to and from the biomolecular motors within each cell. Specialized transmembrane structures transduce force from the arrays of biomolecular motors to the external environment. Failures of these tissue-level structures result in severe pathology of muscle.

Muscle phenotype is known to be a result of a complex interaction between the muscle and its environment. In the absence of the proper signals, muscle will rapidly degenerate. These signals must include chemical signals, mechanical signals, and the activation pattern of the muscle itself. The most important point is that these signals are mediated by the tissue interfaces, and thus it is critical to understand, and to ultimately engineer, adequate tissue interfaces for muscle actuators. The critical interfaces are:

1. **Vascular**: the primary chemical interface, necessary for sections larger than 0.4 mm in diameter. Perfusion of muscle tissue is important for many reasons, including the removal of metabolically-generated heat, delivery of circulating hormones and metabolic substrates, and removal of metabolic byproducts.

2. **Myotendinous (MTJ)**: the primary mechanical interface, necessary for mechano-transduction in muscle, transmission of force and power to the environment without damage to the muscle cells, and transmission of environmental loads to the muscle cells in such a way that the tissue can respond favorably through functional adaptation. In fact, force and power are transmitted transversely into the ECM surrounding each myofibril as well as directly into the tendon. The ECM extends to meet the tendon and transmit this additional force and power. Derangements of these paths of force transduction at any level in general will lead to pathologies of muscle or tendon or both, often resulting in contraction-induced injury to muscle, as is the case in Duchenne muscular dystrophy.

3. **Neuromuscular (NMJ)**: the primary sensing and control interface, nerve input to muscle plays a dominant role in the control of muscle metabolism and phenotype.

### 9.3.1 Potential Classes of Living Muscle Actuators

There are four basic approaches to the use of muscle as a mechanical actuator: whole explanted muscles, recellularized muscle ECM, muscle engineered in an artificial ECM, and self-organized muscle tissue engineered *in vitro*. Each class of muscle actuator has technical advantages and presents technical challenges:

#### 9.3.1.1 Whole Explanted Muscles

Whole muscles are frequently explanted to *in vitro* test systems to carry out muscle tissue evaluations. This is common practice in the pharmaceutical industry as well as in muscle research laboratories around the world. These preparations do not qualify as muscle actuators, as they generally have no provision to maintain the muscle explant for longer than a few hours, and they are not configured in such a way that the muscle could perform useful external work. Such preparations are a far cry from any practical actuator embodiments. It is possible, however, to remove whole muscles from a variety of animals and maintain their contractile function for long periods of time (weeks). The use of such explants as practical mechanical actuators was the focus of preliminary work in biomechatronics at MIT in the year 2000.

**Advantages**: the tissue interfaces are intact and muscle can often be removed with neurovascular pedicles to allow perfusion *ex vivo*.

**Disadvantages**: architecture is limited to that available in nature. Most natural muscles do not have an architecture suitable for use external to the animal, often due to the tendon geometry or lack of suitable tendons.

**Potential applications**: drug testing, actuator applications limited by natural architectures.
9.3.1.2 Recellularized Muscle Extracellular Matrix

Under ideal conditions in this process, muscle cells are chemically removed from the tissue, leaving the ECM intact. The matrix would then presumably provide a perfect scaffold for the reintroduction of suitable myogenic cells. In preliminary experiments it has been demonstrated that the acellularized muscle matrix is entirely nonantigenic, so scaffolds can be removed from one animal and implanted in another without fear of tissue rejection.

Advantages: The ECM retains much of the complex physical architecture of the tissue interfaces, so currently it is hypothesized that it will facilitate the reformation of suitable myotendinous and neuromuscular junctions and vasculature for the creation of tissues suitable for surgical repair of lost or damaged muscle tissue. Because the ECM is nonantigenic, it will be possible to remove intact muscle structures from cadavers and acellularized them to form scaffolds for the reengineering of living muscle tissue from the intended recipient, using the recipient’s cells (from a biopsy or other method) to preclude subsequent postsurgical tissue rejection. Genetically-engineered muscle cells, cells from established cell lines, and primary cells may be reintroduced, as dictated by the actuator application. The existing ECM structure of the acellularized vascular bed allows the acellular muscle to be directly perfused.

Disadvantages: Like whole explanted muscles, the architecture of these actuators is defined by the ECM, and therefore is limited to those forms available in nature. The acellularization process may damage some of the important chemical messages on the matrix, so this method needs to be optimized with this in mind.

Potential applications: recellularized ECM actuators have the complex architecture of whole muscles in vitro, and can be acellularized using cells isolated from any animal, so they would be perfectly suited for engineering complex muscles for surgical transplantation, such as facial muscles. The acellularization process can be readily carried out on cadaveric muscle, so donor tissue availability should present no difficulties whatsoever, thus this class of muscle actuators presents a very promising approach for engineering muscle for surgical transplantation. The acellularized matrix could be repopulated with cells donated (and subsequently amplified in culture) by the recipient of the transplant, thereby totally eliminating the risk of tissue rejection.

9.3.1.3 Muscle Cultured in an Artificial Matrix

A wide range of matrices are available for engineered tissues, but most are unsuitable for engineered muscle due to their limited ability to tolerate repeated macrostrain (± 15% or more the physiologic range for muscle).

Advantages: This is the simplest class of engineered muscle, typically involving the casting of isolated myogenic precursor cells into a gel. It is still the most commonly employed method for engineering muscle in culture, only because it is the easiest method to carry out with the resources available in a typical molecular biology laboratory.

Disadvantages: These constructs in the current state of the art tend to have very weak mechanical interfaces and are thus prone to damage at their points of attachment. In addition, the cellular density in these constructs tends to be well below that of the other three classes, thus they pose significant challenges when their performance is normalized by tissue volume for any functional metric, including protein production, force generation, or sustained power output. To date, these constructs have failed to perform adequately as mechanical actuators. Finally, the most commonly used matrix materials inhibit myocyte fusion into myotubes, arresting the process of muscle development and thereby limiting force and power output. The synthetic matrix materials tend to mechanically fail (tensile failure at the tissue interface) within approximately 2 weeks in culture, whereas self-organized engineered muscle (see below) will persist in culture for approximately 4 months, or longer.
Potential applications: Drug delivery when used as an implanted device, in vitro model for basic research in cultured muscle cells.

9.3.1.4 Self-Organized Muscle Tissue Engineered In Vitro

Isolated myogenic cells are cultured under conditions to provide cues that promote self-assembly of the cells into functional three-dimensional (3-D) tissues.

Advantages: Self-organizing muscle tissues can take full advantage of genetic engineering combined with the inherent phenotypic potential of all muscle tissues. Thus, a virtually limitless range of tissue architectures are possible. In principle, any myogenic cell type from any species can be employed. The authors (Dennis et al.) have successfully engineered skeletal and cardiac precursor cells into functional 3-D tissues in culture from a range of animal species.

Disadvantages: The cells within the tissues tend to remain at an arrested stage of development (neonatal phenotype), exhibiting low levels of contractility and excitability. The mechanical and chemical environment during development must be emulated in order to promote the formation of adequate tissue interfaces.

Potential applications: With appropriately engineered tissue interfaces and the application of the correct external signals, self-organized muscle actuators can be used in any application for which muscle tissue is needed. This is the most general form of engineered muscle, and has the greatest ultimate potential for many applications. Correspondingly, this class of actuators presents the greatest number of technical challenges.

9.4 BIOMECHATRONICS: WHY USE LIVING MUSCLE IN MACHINES?

The ability to engineer muscle actuators may have significant impact on many areas including: (1) drug testing and screening in in vitro bioreactors, (2) drug delivery when implanted as a living “protein factory,” (3) the ability to construct practical hybrid mechanical actuators and robotic devices using both motile cells and self-organized tissues, (4) the ability to build biohybrid prosthetic devices, (5) engineered tissue for surgical transplantation, including both skeletal muscle (~45% of adult human body mass) and cardiac, (6) the ability to harvest high-quality animal protein for food from a controlled bioreactor environment. The importance of the last application becomes clear in light of recent concerns about prion disease, a growing social pressure to reduce animal suffering, and the need for closed ecosystems for long-duration space flight and exploration.

The focus of this chapter is the use of living muscle as a mechanical actuator in engineered systems. The main reason that living muscle is seriously considered for such use is simply because the performance of muscle tissue as an actuator is generally quite favorable when quantitatively compared with synthetic actuator technologies. Direct quantitative functional metrics of various mechanical actuator systems, including muscle, has been reviewed in detail by Hollerbach et al. (1991). The benchmark for most of the synthetic muscle actuator systems currently under development is living muscle. Muscle has considerable advantages over many synthetic actuator technologies both in terms of quantifiable FoM, as well as in terms of many qualitative features unique to living muscle. The potential qualitative advantages of muscle are many: muscle operates almost silently, generates biodegradable substances when converting fuel to mechanical work, can functionally adapt to changing demands, and can take many forms and sizes unlike any traditional synthetic actuator technology. The potential quantitative advantages of living muscle as a mechanical actuator are principally the high-chemomechanical efficiency when operating at nearly room temperature, and the high power density with peak values ranging from 50 to 150 W/kg, depending
upon the muscle tested and the method of evaluation. There are many synthetic actuator systems with much higher power density, but in these cases often excluded is the external power supply and related hardware that are required to drive the actuator. Examples include hydraulic and pneumatic actuators, as well as some types of electro-magnetic actuator systems.

**9.5 QUANTITATIVE ASSESSMENT OF THE FUNCTION OF LIVING MUSCLE ACTUATORS**

There are many FoM that have been formulated to quantify the performance of muscle actuators to allow comparisons between each class of muscle actuator and synthetic actuators. These standardized FoM may be employed when evaluating a new engineered muscle construct or any living muscle-based actuator system.

### 9.5.1 Efficiency — (Volumetric, Metabolic, Excitatory)

#### 9.5.1.1 Volumetric

Natural muscle tissue is characterized by an extraordinarily efficient packaging of biomolecular motors. Histologic cross-sections of healthy muscle clearly demonstrate that approximately 95% of the muscle CSA is comprised of tightly-packed filaments of biomolecular motors (the contractile proteins *actin* and *myosin*) in a hexagonal lattice. There is little opportunity for improvement upon nature with respect to the volumetric efficiency of the packaging of biomolecular motors into functional macroscopic actuators. Synthetically-organized contractile proteins are likely to have an advantage only in single-molecule or molecular monolayer applications, and are likely to be extremely disadvantaged when compared with natural muscle, in terms of volumetric efficiency. Current cultured muscle tissues suffer from low volumetric efficiency in terms of contractile proteins, typically 5 to 10% of the value of adult phenotype healthy control muscle. Also, muscle actuators do not require external support machinery to operate in the same way that many synthetic actuators do. One could reasonably argue that muscle requires many of the other physiologic systems of the body to operate (pulmonary, cardiovascular, neural, gastro-intestinal, etc.), so consider the relative masses of the actuators and the external support system. In an adult human, approximately half of the body mass is muscle tissue. This is supported entirely by the remaining mass of the body, which comprises all other physiologic systems. Compare this with hydraulic or pneumatic systems, for example, for which the power generation system often weighs many times the total mass of all actuators in the system.

#### 9.5.1.2 Metabolic (Chemomechanical Transduction)

The metabolic efficiency is readily mapped into the most commonly defined form of thermodynamic efficiency: work OUTPUT \( \div \) energy INPUT. In the case of muscle, this would translate simply into the mechanical work done by the muscle actuator divided by the caloric content of the fuel (e.g., glucose) consumed plus the energy required to excite the muscle to contract. Corrections must be made for the glucose stored within the muscle prior to the measurement, and for this and a number of other reasons several indirect measures of metabolism are well advised, such as lactate production. The metabolic efficiency of the muscle actuators will of course be sensitive to many factors, including the mechanical load, muscle phenotype, fuel source, pH, temperature, diffusion distances within the tissues, etc. The sensitivity of the actuator to these factors should be considered, in addition to “peak” or “optimal” efficiency values. For example, certain species of amphibians have muscles that operate relatively efficiently over large temperature ranges.
9.5.1.3 Excitatory (Excitation–Contraction Coupling)

This aspect of efficiency is often overlooked in muscle research, but it can easily be the dominant form of energy loss in engineered muscle actuators. This is because muscle rapidly degenerates when maintained in an inactive state. In addition to loss of mass (atrophy), muscle tissue also experiences a loss of excitability. In order to elicit a contraction, muscle is subjected to electrical pulses characterized by a specified pulse width and pulse amplitude at a specified duty cycle and duration. For any given level of contractile activation, reduced excitability manifests as either increased pulse width, pulse amplitude, or both that are required to elicit the desired force or power output. Based upon our extensive preliminary data with engineered muscle, developing muscle, injured and aging muscle, denervated muscle \textit{in vivo}, and denervated-stimulated muscle both \textit{in vivo} and \textit{in vitro}, we calculate that unless care is taken, muscle tissue can degenerate to the point, where the excitability is reduced by three orders of magnitude, thus requiring approximately 1000 times the electrical energy to elicit any given level of contraction. We have also reported that the excitability of denervated muscle can be maintained at control levels by applying the correct form of electrical stimulation.

9.5.2 Static Contractility

Static measures of contractility are readily made, and allow repeatable quantitative evaluation of living muscle function and normalized comparisons between muscle preparations of vastly differing size and architecture. These metrics include: peak twitch force ($P_t$), peak tetanic force ($P_o$), the force–frequency relationship, specific force ($sP_o$), baseline force ($P_b$), excitability (rheobase, $R_{50}$ and chronaxie, $C_{50}$), and the length–tension relationship.

Principal FoM (physical units follow definition):
- Peak normalized twitch force: $\frac{P_t}{\text{mass}}$ (kPa);
- Specific force: $\frac{sP_o}{\text{mass}}$ (kPa);
- Specific baseline force: $\frac{sP_b}{\text{mass}}$ (kPa);
- Rheobase: $R_{50}$ = pulse field amplitude to elicit 0.5$P_t$ at wide pulse width (V/m);
- Chronaxie: $C_{50}$ = pulse width to elicit 0.5$P_t$ at field amplitudes $= 2R_{50}$ (s).

9.5.3 Dynamic Contractility

Measures of dynamic contractility are considerably more experimentally challenging than measures of static contractility, however, they provide considerably more insight into the function of living muscle as a practical actuator. For this purpose, it will in general be necessary to develop bioreactors specifically to monitor these values during the extended \textit{ex-vivo} maintenance of each class of living muscle actuator (whole explanted or engineered). Dynamic contractility is generally evaluated using one or more the following metrics: peak power, sustained power, power density ($W/\text{kg}$), maximum velocity ($L_f/s$, where $L_f$ = muscle fiber length), rate of force development ($dp/dt$), fatigue resistance (metabolic), and work loop performance (net power output during cyclic loading).

Principal FoM (physical units follow definition):
- Peak normalized power density: i.e., peak power output/tissue mass (W/kg);
- Sustained power: i.e., power output at 20% duty cycle continuous (W/kg);
- Maximum contractile velocity: $V_{\text{max}}$ = maximum contractile velocity, unloaded ($L_f/s$);
- Rate of force development: $dp/dt$ (where $p$ is relative force, $P/P_{\text{max}}$) (s$^{-1}$)

In general it is necessary to assemble the instrumentation systems that are necessary for quantitative evaluation of muscle actuator function. For larger muscles that generate at least 10 mN of force...
these systems can employ commercially-available components that are typically used in muscle physiology research. For smaller muscles, or muscles at early stages of development, it is in general necessary to build many of the components. The necessary instrumentation and methods have been reviewed by Dennis and Kosnik (2002).

In addition to the quantitative performance FoM above, the following additional evaluation tools can be employed to quantitatively evaluate living muscle actuator systems:

- **Functional resilience**: Total work output capacity per unit mass of actuator over the functional lifetime of the actuator (J/kg)\textsubscript{lifetime}.
- **Cellular function and phenotype**, as determined by quantitative histology and molecular biology. This will include the presence of known adult isoforms of myosin heavy chain, mitochondrial density, prevalence of central nuclei in myotubes and muscle fibers, cross-sectional density of contractile protein lattice, and indications of cellular necrosis or apoptosis.
- **Failure mode analysis** and the demonstrated efficacy of countermeasures that have been engineered into the living actuator system.

### 9.6 PRACTICAL CONSIDERATIONS FOR THE USE OF LIVING MUSCLE ACTUATORS

When considering the use of living muscle in an engineered system it is important to take into account a number of factors that are generally not significant challenges in traditional mechatronic system design.

#### 9.6.1 Fuel Sources

Muscle \textit{ex-vivo} can operate on a range of fuel sources that are inexpensive and readily available. Ultimately, biomechatronic designers envision the ability of a robot to eat while it travels, much like a fish or a horse. It will be necessary to experimentally evaluate each fuel source for use with muscle powered actuators, since each fuel source has practical limitations and advantages. These include specific energy (kJ/kg), solubility, thermal lability, chemical stability, toxicity, transmembrane transport ability in the absence of systemic metabolic modulatory hormones, and second-order effects such as undesired chemical reactivity. The two principal fuel groups utilized by living muscle are fatty acids and sugars. Evaluation criteria should include quantitative comparisons of muscle actuator efficiency and contractility.

#### 9.6.2 Failure Modes

Based upon our experience with each class of living muscle actuators, the following modes of failure have been identified. For each failure mode, the muscle actuator class(es) that are subject to the failure mode is identified, the theoretical basis for the failure is addressed with supporting experimental verification (if available), and corrective actions are proposed that could be implemented in a biomechatronic system.

#### 9.6.2.1 Septic Degradation of Tissue Structure

This mode of failure affects all types of muscle actuators and at room temperature typically results in rapid functional deterioration within 24 h in the absence of countermeasures. Barrier asepsis is probably not practical in the ultimate field applications of living muscle actuators. Chemical countermeasures using broad-spectrum antibiotic or antifungal formulations in the culture media are effective (Dennis et al., 2000, 2001). These are commercially available for tissue and
organ culture and when properly employed are effective for long-term maintenance of living tissue ex vivo.

9.6.2.2 Mechanical Failure within the Tissue (Intracellular, ECM)

Also known as contraction-induced injury, this mode of failure is prevalent in muscle tissue subjected to maximal contractions during forced lengthening, and affects all classes of muscle actuators. The effective countermeasure involves employing control algorithms that prevent repeated eccentric contraction of fully-activated muscle actuators. Living muscle can functionally adapt to tolerate lengthening contractions if the proper maintenance protocols are employed. An attempt can be made to implement such protocols in the muscle actuator bioreactors using feedback control.

9.6.2.3 Mechanical Failure at the Tissue Interface

Less common for muscle in vivo, this is a major failure mode for explanted and engineered tissues in general. For whole explanted muscles, the interface typically involves suture or adhesive applied to the preexisting tendons. Lack of process control in this tissue or synthetic junction leads to unpredictable mechanical failures over time. In engineered tissues the problem is more serious, as tissue failure frequently occurs at the tissue or synthetic interface under relatively mild mechanical conditions. We have extensive experimental data on this failure mode in engineered muscle tissue subjected to external loading. We hypothesize the failure to be due to stress concentration at the tissue or synthetic interface, compounded by inadequate force transduction from the appropriate intracellular force generating machinery to the extracellular synthetic load bearing fixtures, leading to cell membrane damage at the interface with subsequent rapid tissue degradation and necrosis. The best countermeasure requires the engineering of a muscle–tendon interface (MTJ), which is a major objective of current research in muscle tissue engineering. Tendon tissue is 80 to 90% ECM, composed chiefly of parallel arrays of collagen fibers. The tendon-to-synthetic interface, where biology meets machine, is a separate and equally important technical challenge.

9.6.2.4 Metabolic Failure

This failure mode results most frequently from inadequate delivery of metabolic substrates and inadequate clearance of metabolic byproducts, and is exacerbated at elevated temperatures. The best countermeasure for this failure mode is to restrict the muscle actuator cross-section to more than approximately 200 μm diameter, or to provide perfusion through a vascular bed in the case of larger cross-sections. This mode of failure typically initiates at the axial core of cylindrical muscle actuators. For this reason, sustained angiogenesis and perfusion is a major technical objective in current tissue engineering research.

9.6.2.5 Cellular Necrosis and Programmed Cell Death

Several controllable circumstances can lead to this general mode of failure in all classes of muscle actuators. Cellular hypercontraction and hyperextension in muscle results in rapid necrosis. This mechanism will occur more or less uniformly across the muscle cross-section, but will theoretically occur more frequently in areas with reduced physiologic cross-sectional area or inhibited sarcomeric function. This failure mode can be prevented by control of the internal mechanical compliance and stroke of the muscle actuator. Muscle maintained at an inappropriate length, either too short or too long, will deteriorate, even if the muscle is quiescent. In explanted muscles, maintenance at lengths greater than the plateau of the length–tension curve appears to be the most damaging over time.
9.6.2.6 Fatigue (Mechanical and Metabolic)

These failure modes apply to all classes of living muscle actuators. For metabolic fatigue the preferred countermeasures will include genetic engineering of the muscle to promote fatigue-resistant fiber types, the provision of adequate perfusion of the tissue actuator, and the development of protocols for actuator control that optimize total work output, such as the intermittent locomotory behavior of both terrestrial and aquatic animals. It is in terms of mechanical fatigue that living actuators have an enormous advantage over fully synthetic actuators. By monitoring the state of health of the actuator and modifying the mechanical demands accordingly, it is possible to promote functional adaptation of the living component of the actuator as well as the tissue or synthetic interface. It will be necessary to identify biomarkers of mechanical fatigue, such as reduced or altered contractility, to actively detect these markers, and to respond with appropriate modifications of the embedded excitation and control algorithms to allow tissue functional adaptation. In principle a properly monitored and controlled living muscle actuator will exhibit improved dynamic performance and structural resilience with use over a period of decades, unlike any synthetic actuator technology currently available.

9.6.2.7 Toxicity

A serious problem for all classes of living muscle actuators, the best countermeasure is barrier exclusion of exogenous toxic agents, the use of biocompatible materials in the fluid-space of the hybrid actuator assembly, and the clearance of toxic metabolic byproducts via a perfusion and filtration system integrated with the living actuator.

9.6.2.8 Electrochemical Tissue Damage

This failure mode affects all classes of living muscle actuators when exposed to chronic electrical stimulation. The single best countermeasure is to promote and maintain tissue phenotype exhibiting very high excitability. In addition to vastly improving the excitation efficiency of the tissue, adult muscle phenotype excitability can yield as much as a 99.9% reduction in electrical pulse energy requirements for any given level of muscle activation, when compared with chronically denervated or tissue engineered muscle tissue arrested at early developmental stages. For this reason, the development of electro-mechanical muscle bioreactor systems and maintenance stimulation protocols form a core component of all current research on muscle tissue engineering. Additional countermeasures include the selection of appropriate electrode materials, the use of minimally energetic stimulation protocols, the use of pure bipolar stimulation pulses with careful attention to charge balancing, and the use of high-impedance outputs to the electrodes when not stimulating.

9.6.2.9 Damage from Incidental Mechanical Interference

The living actuator will require electrodes to be placed in contact with the tissue, the presence of tubing for perfusion, and other structures required within the hybrid actuator. Lateral mechanical contact between these synthetic objects and the living muscle tissue can result in a range of mechanical failures, including abrasion, incision, and chronic pressure atrophy. The appropriate countermeasure for this is careful mechanical design of the hybrid actuator assembly, with these considerations explicitly included in the system Design Specification.

9.6.2.10 Retrograde or Arrested Phenotype ("Failure to Thrive")

Effective countermeasures for this failure mode have been reported for denervated whole muscles in vivo, employing a long-term electrical stimulation protocol (Dennis et al., 2003; Dow et al.,
2004). This failure mode is most prevalent in engineered muscle maintained in culture. There are two approaches to dealing with this in engineered muscle: (1) genetic enhancement and (2) development of electromechanical tissue maintenance protocols. In the case of genetic enhancement, the approach is to forcibly express desired genes in an attempt to promote the desired tissue phenotype. The effectiveness of this approach is the core issue in gene therapy for diseases of muscle, but this approach has not yet been demonstrated to be effective for engineered muscle \textit{ex vivo}. Optimal tissue maintenance protocols are a much more natural and subtle approach, based upon the fact that all viable muscle cells contain the necessary genetic machinery to develop any desired muscle phenotype, if the correct signals and growth conditions prevail. In addition to genetic engineering of myocytes to enhance performance of tissue-based actuators, other potential countermeasures include: (1) development of appropriate tissue interfaces to permit signal transduction to the cellular machinery, (2) development of tissue and organ culture bioreactors to allow the experimental determination of optimal control and maintenance protocols for \textit{ex vivo} muscle tissue, (3) use of these protocols to guide tissue development (cell phenotype and tissue architecture), and (4) implementation of this technology into the hybrid actuator system. This topic is currently an area of very active research. Success in terms of counteracting this failure mode in engineered muscle will constitute an extraordinarily significant scientific contribution, as well as providing the key enabling technology to the further development of practical living actuators.

### 9.7 SELF-ORGANIZING MUSCLE TISSUES

Self-organization within developing animals gives rise to an enormous array of muscle actuator architectures. Each myogenic precursor cell contains the genetic potential to self-organize into muscle tissue with the desired phenotype and tissue interface. The ability to guide the development of self-organizing muscle tissues in culture will provide the systems engineer with the greatest level of design flexibility, since it will in principle be possible to start with a small population of muscle progenitor cells and guide them to self-organize into a muscle actuator of any imaginable geometry. It will also be possible to construct hybrid actuators not found in nature, containing regionally organized tissue structures, perhaps even consisting of fundamentally different types of muscle tissue (skeletal, cardiac, or smooth), depending upon the functional requirements of the actuator system. It is implicit in most muscle tissue engineering research programs that skeletal muscle self-organization and development can be guided by the application of the correct external cues. The general method of guided tissue self-organization in culture (Figure 9.1) briefly is:

- Isolate and coculture the desired cells. The cells may be primary or from cell lines;
- Engineer a cell culture substrate with controlled adhesion properties for the cells;
- Provide permanent anchor points and surfaces to guide tissue architecture formation;
- Culture the cells to permit the formation of a cohesive monolayer;
- Induce monolayer delamination from the substrate at the appropriate point in cell differentiation (the monolayer remains attached to the anchor points);
- Promote tissue self-organization and further development by applying external signals: chemical, electrical, mechanical.

Self-organization of tissues in culture is one effective way to produce small functional tissue constructs from a range of tissues. Examples include:

- \textit{Cardiac myocytes} cocultured at confluence with fibroblasts will self-organize into long cylinders and tapered cones in culture in 340 to 400 h. These constructs are electrically excitable and also spontaneously contract as a syncytium to continuously generate significant mechanical work cycles. Such constructs could be engineered to power cell-scaled implantable pumps, pumps for
stand-alone hybrid tissue actuators, or to engineer cardiac tissue for surgical transplantation in cardiac reconstructive surgery.

- **Tendon (Ligament)** tissue will self-organize in culture under the appropriate conditions. The fibroblasts within the tissue produce a prodigious amount of ECM material, with collagen fibers that are oriented along lines of tensile stress, particularly at locations within the tissue where mechanical interfaces are present (such as suture anchor materials, metal posts, etc.). Self-organization is driven by loss of substrate adhesion and the generation of internal tensile stress by the action of the fibroblasts on the order of 0 to 6 Pa, which can be experimentally controlled by external factors such as the presence of ascorbic acid, serum concentration in the cell culture medium, pH, etc.

- **Muscle Chimeras:** One additional interesting technical possibility is the in vitro fusion of myogenic precursor cells from different tissue sources to form chimeric self-organized engineered muscles. Preliminary experiments demonstrate that skeletal muscle satellite cells from differing species will fuse to form multinucleated myotubes with desirable contractile function. In addition, isolated cardiac myocytes will fuse into preexisting myotubes in culture, to produce a skeletal–cardiac muscle hybrid. Such chimeric muscle tissues are not known to exist in nature, but our preliminary data indicate that they are both stable and functional in culture. The contractile function of such chimeric cells and tissues could potentially be engineered to produce tissue-based actuators with combinations of desired characteristics that would be advantageous for use in hybrid bioactuator applications.

### 9.8 ACELLULARIZED–RECELLULARIZED ECM ENGINEERED MUSCLES

The native ECM of muscle tissue occupies approximately less than 5% of the tissue volume, yet it contains information about the complex architecture of muscle and the corresponding soft tissue...
interfaces. The cellular components of muscle can be chemically removed while retaining the detailed architecture of the muscle ECM. Preliminary results indicate the success of the reintroduction of myogenic cells into these natural ECM scaffolds. This approach to engineering muscles as actuators has several advantages, among these are that heterogenic cells can be introduced into the preexisting matrix. For example, skeletal–cardiac chimeric muscles could be employed or myogenic precursors from an entirely different species. The main advantage of the use of natural ECM scaffolds is that the fine architecture of the entire muscle organ is retained by the acellularized ECM scaffold. It is possible to perfuse the scaffold using the remnant vascular bed ECM to reintroduce cells and later to provide perfusion to the reengineered muscle organ. The acellularized muscle ECM also has matrix architecture specific to the MTJ and tendon, which may be advantageous in the development of this very critical tissue interface. The principal disadvantage of this approach is that the ECM scaffold architecture is limited to those architectures that are available in nature.

### 9.9 TISSUE INTERFACES: TENDON, NERVE, AND VASCULAR

For any type of muscle actuator, it will be essential to provide appropriate tissue interfaces. In some cases, the tissue interfaces are already in place and specific measures must be taken to maintain them properly. In other cases, their formation must be guided and facilitated. Based upon our in vivo work, we have demonstrated that muscle phenotype can be controlled and maintained in the absence of innervation via electrical stimulation. A considerable volume of published research has been directed toward the promotion of adult phenotype in muscle tissue in culture directly by electrical stimulation, in the absence of nerve-derived trophic factors or depolarization via the neuromuscular junction and related synaptic structures. It remains to be demonstrated, however, that muscle can be guided through the necessary developmental stages in the absence of innervation to achieve adult phenotype. Adequate and functional vascular and tendon interfaces to muscle engineered in vitro are also yet to be demonstrated, although they are the topic of intensive research.

#### 9.9.1 Vascular Tissue Interface

Nutrition and oxygen delivery in static culture conditions always limit the cross-sectional area, particularly for tissues with high metabolic demand, such as muscle. Therefore, a 3-D organ culture system with perfusion of a vascular bed within the muscle tissue is a core objective of current research. Cell types associated with angiogenesis, such as endothelial cells are also crucial players in organ development (Bahary and Zon, 2001). Endothelial progenitor cells from peripheral blood are readily isolated, and have been shown to incorporate into neovessels (Asahara et al., 1997) and also have potential to expand to more than $10^{10}$-fold in vitro (Lin et al., 2000). Furthermore, functional small-diameter neovessels can be created in culture by using endothelial progenitor cells (Kaushal et al., 2001).

#### 9.9.2 Strategies for Engineering Functional Vascularized Muscle Tissue

There are three strategies for generating vascularized muscle constructs:

1. **Recellularization** of an acellular muscle construct;
2. **Coculture** of myoblasts with endothelial cells and growth factor stimulation for induction of the endothelial cells to form capillary like structures;
3. **Induction** of sprouting of microvessels into temporarily implanted tissues or from vascularized and perfused tissue explants (such as adipose) cultured adjacent to the engineered muscle.
The strategies for generating functional muscle tissue can be broadly divided into *in vitro* and *in vivo* strategies, the ultimate outcome of which would be a vascularized muscle construct. In any case, once a vascular bed is established, the constructs need to be maintained in a bioreactor to provide further electrical, mechanical, and chemical stimulation, thus guiding both the phenotype and resulting in the development of a fully functional muscle construct.

### 9.9.2.1 Recellularization of an Acellular Muscle Construct

This experimental approach involves harvesting muscle tissue from any natural source and using chemical acellularization to remove myoblasts and fibroblasts leaving behind an intact ECM. The ECM should be evaluated for structural integrity and immunogenic behavior and its ability to support myoblast growth and differentiation. The ECM should then be used as scaffolding material for seeding primary myoblast and the construct will be placed in a perfusion bioreactor allowing formation of functional skeletal muscle tissue (Hall, 1997). Immunohistochemical studies should be performed to determine which ECM components are present in the acellular construct, such as collagen types I and IV, fibronectin, laminin, vitronectin, entactin, heparin sulfate, proteoglycan, and elastin. The acellular muscle can be repopulated by obtaining a purified sample of myogenic precursor cells, which may be injected or perfused into the acellular muscle. Although some initial success has been reported with this general approach, it has not yet been possible to maintain perfusion of the tissue samples in culture for a period sufficiently long to promote and maintain full cellular infiltration into the acellular scaffold.

### 9.9.2.2 Coculture Systems

Since the early 1990s, there have been reports of the use of various coculture systems to study cell–cell interactions and the formation of tissue interfaces. For vasculogenesis, the cells in question are presumed to be myoblast and endothelial cells. Although promising initial reports have been published, a truly successful demonstration of a vascular bed self-organizing within a tissue construct has yet to be demonstrated. The design of bioreactors for such a technology must stimulate the myoblasts to form functional muscle tissue and simultaneously guide the endothelial cells to form capillary-like structures within the newly forming muscle tissue, while providing perfusion during development. The environment, which the bioreactor provides together with soluble growth factor stimulation will presumably allow formation of a functional muscle construct (Vernon, 1999).

### 9.9.2.3 Induced Microvessel Sprouting

This approach can be attempted either *in vivo* or *ex vivo* using small vascularized tissue explants which are cannulated and perfused while adjacent to an avascular tissue such as engineered skeletal muscle. This is an active area of current research. For the *in vivo* approach, it is necessary to mechanically support the muscle tissue while implanted to prevent hypercontraction and subsequent tissue damage. It is also necessary to take measures to prevent tissue rejection to implantation into syngenic animals, or the use of immune-suppressive agents, is required. Otherwise, this method is relatively quite simple and often yields satisfactory results. In addition to vascularization of the implanted muscle tissue, there are collateral effects, as yet not fully understood, that also tend to drive the muscle phenotype toward an adult phenotype, with enhanced contractility. For this reason, it is likely that the future of tissue engineering will see increasingly common application of the approach where the intended recipient is used as a ready-made bioreactor vessel. The engineered tissues would be implanted within the person, presumably along with means to enhance tissue development and to prevent tissue degeneration or resorption while implanted. The tissue need not
be implanted at the ultimate site for which it is intended, however, it is essential to consider the morbidity of the site at which the disuse will be initially developed.

9.9.3 Engineered Tissue Interface: Tendon

The MTJ is critical for the ability of muscle tissue to transduce force to and from the external environment, and to produce maximal power without subsequent injury to the muscle cells in the contractile tissue. The MTJ contains specialized structures at the cell membrane which facilitate transmembrane transmission of force from the contractile proteins (biomolecular motors) within the cell to the surrounding collagen fibrils in the ECM (Trotter, 1993). These structures include a large number of infoldings of the muscle cell membrane at the MTJ, increasing the membrane surface area and acting to transfer stress from the cytoskeleton to the ECM in the tendon. These structures have also been demonstrated to occur when myotubes are cocultured with fibroblasts concentrated near the ends of the muscle constructs \textit{in vitro} (Swasdison and Mayne, 1991). In the case of whole explanted muscle actuators, the MTJ already exists, and it is necessary to maintain this structure \textit{in vitro}. In all other classes of muscle actuator it is necessary to generate or regenerate the MTJ and tendon structures. Currently, attempts to engineer tendon-like structures and muscle–tendon junctions in culture follow one of three distinct approaches:

1. Scaffold-based tendon, used as an anchor material for engineered muscle;
2. Self-organizing tendon and muscle-tendon structures in co-culture;
3. Direct laser transfer of muscle and tendon cells into defined 3-D structures.

9.9.4 Nerve–Muscle Interfaces

Skeletal muscle phenotype is defined largely by the motor nerve which innervates each muscle fiber. Adult muscles may be either fast- or slow-twitch, but in general in humans muscles are mixed, containing significant populations of both fast- and slow-twitch fibers. Denervated muscle rapidly loses tissue mass and the adult phenotype, with contractility eventually dropping to essentially zero. Although it is possible to maintain adult phenotype of adult skeletal muscle in the absence of innervation, it is not yet clear whether it is possible to guide skeletal muscle tissue development to an adult phenotype in an entirely aneural culture environment. For that reason, nerve–muscle synaptogenesis in culture is an area of active research in tissue engineering. Putative synaptic structures \textit{in vitro} have been reported for decades (Ecob et al., 1983; Ecob, 1983, 1984; Ecob and Whalen, 1985), in some cases axon sprouting from nerves to muscle tissue in culture is clearly visible (Figure 9.2) and verified upon histologic examination, however, functional nerve–muscle \textit{in vitro} systems that result in advanced tissue development have yet to be demonstrated.

9.9.5 Tissue–Synthetic Interfaces

Another key challenge is to develop means to mechanically interface living muscle cells and tissues to synthetic fixtures in such a way that the tissue development and function will not be inhibited. The technical challenge is to provide a transition of mechanical stiffness and cell density in the region between the contractile tissue and the synthetic fixture, to reduce stress concentrations at the tissue interface and provide mechanical impedance matching. Several approaches are currently under investigation, including the chemical functionalization of synthetic surfaces to bind collagen, and the use of porous scaffolds to promote tissue in-growth at the desired tissue or synthetic interface.
The engineering of complex functional tissues such as skeletal muscle is by definition a systems engineering problem. Functional muscles are composed of a number of highly integrated tissue systems, none of which is known to function in isolation for any significant period of time without massive deterioration in performance. Any attempt to engineer a functional muscle tissue system ex-vivo, and to employ that muscle system as a source of motility in robots or prostheses, will by necessity require the development of bioreactor technologies to (1) guide the tissue development to the desired phenotype ex-vivo, (2) maintain the tissue at the desired phenotype while it is performing its function, and (3) control the mechanical output of the tissue through electrical stimulation. Critical to these three objectives are bioreactor technologies that are capable of monitoring and controlling a muscle’s mechanical and electrical environment.

In Figure 9.3, a muscle bioreactor is shown that can implement muscle identification, control, and maintenance protocols under generalized boundary conditions while also providing flexible feedback control of electrical stimulation parameters (Farahat and Herr, 2005). These features are accomplished by having two real-time control loops running in parallel. The first loop, or the mechanical boundary condition (MBC) control loop, ensures that the mechanical response of the servo simulates the dynamics of the associated muscle boundary condition. For example, if the desired boundary condition is a second order, mass–spring–damper system, the MBC control loop controls the motion of the end points of the muscle–tendon unit as if the muscle–tendon were actually pulling against physical mass–spring–damper mechanical elements. The MBC control loop allows for a whole host of boundary conditions, from finite (but nonzero) to infinite impedance conditions. Clearly, to understand muscle tissue performance, muscle dynamics and the dynamics of the load for which the muscle acts upon must be taken into consideration. Examples of finite-impedance boundary conditions include loads such as springs, dampers, masses, viscous friction, coulomb friction, or a combination thereof. Such loads prescribe boundary conditions that are generally defined in terms of dynamic relationships between force and displacement. Under these loading conditions, it would be expected that the dynamics of the load will interact with the contraction dynamics of the muscle, leading to a behavior that is a resultant of both. This is
primarily because the force generated by muscle is dependent on its mechanical state, namely its length and velocity.

The second control loop for the bioreactor design of Figure 9.3 implements the electrical stimulus (ES) control based on measurements of the muscle’s mechanical response. This loop, referred to as the ES control loop, offers simultaneous real time modulation of pulse width, amplitude, frequency, and the number of pulses per cycle. There is increasing experimental interest in real time control of muscles, primarily in the context of functional electrical stimulation (FES) (Chizeck et al., 1988; Veltink et al., 1992; Eser et al., 2003; Jezernik et al., 2004). In these investigations, attempts were made to control the response of muscle(s) and associated loads to a desired trajectory by varying electrical stimulation parameters as a function of time. Electrical stimulation patterns are typically square pulses characterized by frequency, amplitude, pulse width, and number of pulses per trigger (considering the cases of doublets, triplets, or more generally N-lets). For testing a variety of FES algorithms, the ES control loop is designed for real time modulation of these stimulation parameters as a function of a muscle’s mechanical response, including tissue length, contraction velocity and borne muscular force.

9.11 CASE STUDY IN BIOMECHATRONICS: A MUSCLE ACTUATED SWIMMING ROBOT

Biomechatronics is the integration of biological materials with artificial devices, in which the biological component enhances the functional capability of the system, and the artificial component provides specific environmental signals that promote the maintenance and functional adaptation of the biological component. Recent investigations have begun to examine the feasibility of using animal-derived muscle as an actuator for artificial devices in the millimeter to centimeter size scale

Figure 9.3 (See color insert) Muscle Bioreactor Technology. Muscle identification, control and maintenance apparatus is shown with the primary sensors and actuators noted. The coarse positioning stage is adjusted at the beginning of the experiment to accommodate different tissue lengths, but is typically kept at a constant position during a particular contraction. The primary stage provides the motion that simulates the boundary condition force law with which the muscle specimen pulls against. The vertical syringe has a suction electrode at its tip that is connected to the stimulation electronics in the background. The encoder and load cell measure muscle displacement and force, respectively, and are employed as sensory control inputs during FES control experimentation. Silicone tubing recirculates solution via a peristaltic pump, while oxygen is injected in the loop.
Although a great deal of research has been conducted to develop an actuator technology with muscle-like properties, engineering science has not yet produced a motor system that can mimic the contractility, energetics, scalability, and plasticity of muscle tissue (Hunter et al., 1991; Meijer et al., 2003). As a demonstratory proof of concept, Herr and Dennis (2004) designed, built, and characterized a swimming robot actuated by two explanted frog semitendinosus muscles and controlled by an embedded microcontroller (Figure 9.4). Using open loop stimulation protocols, their robot performed basic swimming maneuvers such as starting, stopping, turning (turning radius ~ 400 mm) and straight-line swimming (max speed > 1/3 body lengths/sec). A broad-spectrum antibiotic or antimycotic ringer solution surrounded the muscle actuators for long term maintenance, *ex vivo*. The robot swam for a total of 4 h over a 42-h lifespan (10% duty cycle) before its velocity degraded below 75% of its maximum. The mechanical swimming efficiency of the biomechatronic robot, as determined by a slip value of 0.32, was within the biological efficiency range. Slip values increase with swimming speed in biological swimming, ranging from 0.2 to 0.7 in most fish (Gillis, 1997, 1998).

The development of functional biomechatronic prototypes with integrated musculoskeletal tissues is the first critical step toward the long term objective of controllable, adaptive, and robust biomechatronic robots and prostheses. The results of the swimming robot study of Herr and Dennis (2004), although preliminary, suggest that some degree of *ex vivo* robustness and controllability is possible for natural muscle actuators if adequate chemical and electromechanical interventions are supplied from a host robotic system. An important area of future research will be to establish processes by which optimal intervention strategies are defined to maximize tissue longevity for a given hybrid-machine task objective. Another important research area is tissue control. It is well established that natural muscle changes in size and strength depending on environmental workload, and when supplied with appropriate signals, changes frequency characteristic or fiber type (Green et al., 1983, 1984; Delp and Pette, 1994). Hence, an important area of future work will be to put forth strategies by which muscle tissue plasticity can be monitored and controlled. Still further,
strategies must also be devised to control the force and power output of muscle, in the context of robotic systems, through the modulation of electrical pulses to the muscle cell. Also, for the development of controllable, adaptive and robust biomechatronic systems, feedback control systems that monitor and adapt the mechanical, electrical, and chemical environment of muscle actuators are of critical importance.

9.12 CONCLUDING REMARKS

Muscle tissue as a mechanical actuator has great, though as-yet unrealized potential for use in engineered systems. Synthetic technologies such as electroactive polymers are rapidly emerging as quantitatively functional equivalents to muscle tissue, and it is likely that the technological evolution of EAP muscles will soon out-pace the natural functional evolution of living muscle tissue. This means that the quantitative performance advantages that muscle tissue has over some forms of synthetic actuators in terms of efficiency, power density, and so forth are not likely to remain the case for very much longer. One then invariably must ask why it is advantageous to even consider the use of living muscle tissue as a mechanical actuator. It is easy to point out that the many disadvantages of muscle outweigh the few performance advantages it may have. The answer lies chiefly in the qualitative differences between muscle and competing synthetic actuator technologies, among these are those qualities that arise from muscle being a living tissue: its ability to functionally adapt and to potentially integrate seamlessly with other living structures. So it is likely that living muscle actuators will only be employed in practical systems where their qualitative advantages as living tissue can be exploited to maximum benefit, such as in hybrid biomechatronic prosthetic systems and implants, and perhaps in bioreactors where their biological products (such as edible proteins) are of primary importance. Certainly though, living muscle tissue serves as the explicit benchmark against which the performance of synthetic actuator technologies will be evaluated for many decades to come.

FURTHER READING

The following list of papers and book chapters comprise a set of useful references for further work in this area. These were not referenced directly in the text, but have been included because the authors have found them to be useful during the course of the development of the technology discussed in this chapter.


Engineered Muscle Actuators:


REFERENCES


Engineered Muscle Actuators:


WEB SITES

http://www.bme.unc.edu/~bob/
http://biomech.media.mit.edu/

Author Queries

[AQ1] Au: Changed as in list of References.
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