Special Report

Systems Approach to Understanding Electromechanical Activity in the Human Heart

A National Heart, Lung, and Blood Institute Workshop Summary

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Abstract*—**The National Heart, Lung, and Blood Institute (NHLBI) convened a workshop of cardiologists, cardiac electrophysiologists, cell biophysicists, and computational modelers on August 20 and 21, 2007, in Washington, DC, to advise the NHLBI on new research directions needed to develop integrative approaches to elucidate human cardiac function. The workshop strove to identify limitations in the use of data from nonhuman animal species for elucidation of human electromechanical function/activity and to identify what specific information on ion channel kinetics, calcium handling, and dynamic changes in the intracellular/extracellular milieu is needed from human cardiac tissues to develop more robust computational models of human cardiac electromechanical activity. This article summarizes the workshop discussions and recommendations on the following topics: (1) limitations of animal models and differences from human electrophysiology, (2) modeling ion channel structure/function in the context of whole-cell electrophysiology, (3) excitation–contraction coupling and regulatory pathways, (4) whole-heart simulations of human electromechanical activity, and (5) what human data are currently needed and how to obtain them. The recommendations can be found on the NHLBI Web site at http://www.nhlbi.nih.gov/meetings/workshops/electro.htm. (Circulation*. 2008;118:1202-1211.)**

Key Words: arrhythmia ■ cardiovascular diseases ■ contractility ■ electrophysiology ■ mechanics

The National Heart, Lung, and Blood Institute (NHLBI) convened a workshop of cardiologists, cardiac electrophysiologists, cell biophysicists, and computational modelers on August 20 and 21, 2007, in Washington, DC, to advise the NHLBI on new research directions needed to develop integrative approaches to elucidate human cardiac function. The workshop fits well within the NHLBI Strategic Plan by seeking to integrate understanding of the molecular and physiological bases of health and disease and to develop more effective approaches to cardiac disease diagnosis, treatment, and prevention.

"Systems approach" can be defined as an integrative approach that, in contrast to the reductionist approach of science, assembles the system (in this case, the heart) from its molecular, cellular, and tissue components. The past decade has generated a wealth of information at the genetic, molecular, and cellular scales of the cardiac system. It is timely and important to begin integrating this information within and between scales to the level of the whole heart because electromechanical cardiac function and its alteration by disease (eg, heart failure and arrhythmias) occur at the organ level.

Until the recent advances in genomics, proteomics, metabolomics, and genetic engineering, our ability to understand how the most basic building blocks of life, DNA and the gene, affect the behavior of the living organism was limited to a handful of rare genetic diseases. Identifying the full complement of mRNA transcripts produced in the heart

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Circulation **is available at http://circ.ahajournals.org** **DOI: 10.1161/CIRCULATIONAHA.108.772715**

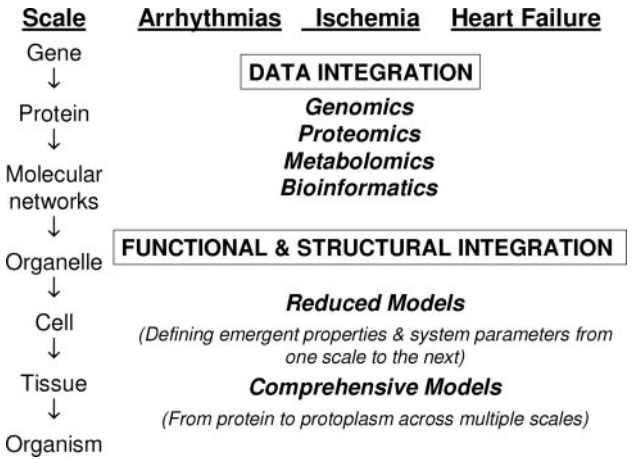

Figure 1. Role of multiscale modeling in the systems approach to understanding electromechanical activity in the human heart.

(the transcriptome), the proteins that they encode (the proteome), and the small metabolites that define the fluid state of the cells (the metabolome) provides a roadmap to eventually link genotype to phenotype.[1,2] With the ability to alter genes through genetic engineering, moreover, it has become possible to directly explore at will the relationship between genotype and phenotype in animal models.[3] This has become a powerful tool for cataloging how individual genes affect phenotype and has illuminated the molecular basis of a number of (mostly rare) monogenic diseases in humans.

However, even in monogenic diseases (such as congenital long-QT syndromes), interactions with modifier genes and environment are critically important in establishing the phenotypic severity (such as arrhythmia risk) among affected family members. Moreover, it has become increasingly clear that most common human diseases, including myocardial ischemia, heart failure, and arrhythmias, are not due to the strong effect of a single defective gene but rather to modest effects of multiple genes combined with modest effects of multiple environmental factors. Therefore, merely cataloging the effects of all possible genetic mutations, even if it were technically feasible, would be unlikely to provide the mechanistic insight required to develop cures for most human diseases. Given this limitation, it is essential to develop new integrative systems approaches to human disease that begin with but go well beyond genotype–phenotype associations to the quantitative prediction of phenotypes from genotypes and their in vivo context in health and disease.

In addition to characterizing and analyzing networks of functional interactions between genes and proteins, these approaches should aim to integrate understanding across physical scales of increasing complexity, from molecule to cell, tissue, the whole heart, and ultimately the whole patient (Figure 1). Adequate experimental and computational tools currently exist to allow such an integration; the challenge is to have these approaches accepted and used to achieve mechanistic understanding of human cardiac disease at each of the intermediate scales of biological organization between the genome and the living organism. Such an understanding also will provide a formal and quantitative means of extrapolating mechanisms from animal models of cardiac disease to individual patients.

The workshop convened by the NHLBI recognized limitations in the use of data from nonhuman animal species for elucidation of human electromechanical function/activity and identified what specific information on ion channel kinetics, calcium handling, and dynamic changes in the intracellular/extracellular milieu is needed from human cardiac tissues to develop more robust computational models of human cardiac electromechanical activity. Workshop members specifically reviewed and discussed (1) limitations of animal models and differences from human electrophysiology, (2) modeling ion channel structure/function in the context of whole-cell electrophysiology, (3) excitation–contraction coupling and regulatory pathways, (4) whole-heart simulations of human electromechanical activity, and (5) what human data are currently needed and how to obtain them.

The workshop resulted in specific recommendations that can be found on the NHLBI Web site at http://www.nhlbi.nih.gov/meetings/workshops/electro.htm.

Limitations of Animal (Mouse) Models and Differences From Human Electrophysiology

This section identifies important challenges associated with the study of human cardiac electromechanical activity, elements of uncertainty about human cardiac physiology, and the practicability and limitations of using mice and other animals as means to improve our understanding of cardiac diseases in humans. Potential approaches are suggested that might improve the scientific yield of human cardiac studies that, when systematically integrated with relevant studies in experimental animal models, may lead to the creation of improved computational models and provide deeper insights into cardiac pathophysiology. Some of the proposed data generation is not traditional, hypothesis-driven science, but it is believed to be critical for connecting systems approaches at the molecular and organ levels.

Obtaining Data From Nondiseased Human Hearts

Constructing useful computational models of human cardiac electromechanical activity requires a thorough understanding of the distribution of ion channels, exchangers, pumps, and functional currents in various regions of the heart, as well as how these are modified in disease. Thus, it also is essential to integrate the electrical signals with dynamic changes in cytosolic calcium levels and the use of ATP and other energetic substrates. Coordinated efforts are needed to better characterize regional differences in calcium handling, sodium–calcium exchanger activity, and the function of the full complement of sodium, calcium, potassium, and anion channels expressed in the human heart.

Such a complete characterization represents an ambitious and difficult goal, and for ethical and logistic reasons, it is impractical or impossible to study sufficient numbers of normal human hearts to fully characterize the expression, distribution, and function of all of the ion channels and related proteins. However, better use of relatively normal human myocardium that cannot be used for transplantation, made available through collaborations with organ procure-

ment agencies with appropriate consent from the donor family, would dramatically improve our present knowledge. Developing a consensus-building platform to prioritize what specific fundamental data are most needed for consolidation into an integrated systems approach would also allow this precious resource to be best utilized. Optimal use of this relatively rare resource requires teamwork and advanced planning, as well as the development of standardized procurement and distribution procedures. To enhance viability of the hearts for immediate live tissue and cell studies, they should be procured in a timely manner, with the hearts perfused with cardioplegia solution just before explantation in a manner identical to that used for organ transplantation. Healthy myocytes for cellular electrophysiology and electromechanical studies can be enzymatically dissociated from isolated specific regions of the heart that can be perfused via the coronaries, a procedure shown to improve yield and viability compared with chunk dissociation techniques.[4]

The procedures should permit investigators to evaluate gradients in protein expression (transmural, apex to base, left to right) and tissue-specific distributions (atrial, ventricular, sinoatrial and atrioventricular nodal tissues, and His-Purkinje system). A shared tissue repository for physiological characterization of human heart tissue is needed, as well as creation of a distributed network for collaborative studies on these tissues. Such a resource would constitute a valuable asset for the biomedical community. Efforts should be made to integrate the histological or immunohistochemical analyses of intact, isolated tissues with the results of cellular electrophysiology studies and with available clinical data, including high-resolution, noninvasive structural (computed tomography, magnetic resonance imaging), functional (echocardiography, positron emission tomography) and electrophysiological imaging.[5,6]

Especially important but challenging to obtain are direct functional measurements of electrophysiological, calcium handling, and mechanical properties of nonfailing human cardiac myocytes and tissues. This is complicated by the limited availability of viable tissue and the challenge of storing and transporting live myocytes for functional studies. Functional characterizations are the essential link between measures of protein expression (and modulation) and understanding how myocytes function and interact in the whole heart. It would also be valuable to obtain additional data in intact human hearts with calcium and sarcoplasmic reticulum calcium imaging and electrophysiological mapping (optical and plunge electrodes) to bridge these levels.[7–10]

Strategies to Improve Understanding of Diseased Human Cardiac Electromechanical Activity

A similar strategy should be implemented on a broader scale for the study of cardiac tissues from patients with end-stage heart failure and hearts explanted from those individuals receiving a heart transplant. Studies of muscle function from explanted hearts have identified downregulation of β_1-adrenergic receptor density as a prominent mechanism underlying impaired contractility[11] and provided mechanistic insights into the regulation of coronary artery function in failing hearts.[12]

To provide maximum insight into the underlying causes of cardiac dysfunction and the clinical phenotype of these failing hearts, we must ensure that tissues are obtained from properly consented patients in a Health Insurance Portability and Accountability Act–compliant fashion and linked to deidentified clinical data maintained in secure databases. Establishing such a resource would facilitate the systematic characterization of cardiac and coronary histopathology, gene expression profiles, and limits of muscle function (force–frequency response, active and passive muscle mechanics) and integration of this information.

Complementary Studies in Animal Models

Although better use of systematic characterization of human cardiac tissues will provide much-needed insight, human tissue studies suffer from a number of unavoidable limitations. These include the great intrinsic variability of individual patients, the inability to perform sequential functional assessments, and the fact that human tissues are most often available only at the 2 extremes: end-stage heart failure and relatively normal. It is therefore difficult at best to fully elucidate processes that drive the transition from normal to hypertrophy or hypertrophy to failure in human studies alone. It is not yet feasible, or ethical, to attempt to modify the expression of ion channels or other proteins in human hearts. Thus, some questions cannot be fully addressed by studies of human heart preparations. Judicious use of experimental animal models can help to fill important gaps in our understanding. Which nonhuman species should be used? In considering this question, it is relevant to consider the question being asked.

Mouse and rat ventricular myocytes are used extensively in studies of cardiac electrophysiology and arrhythmogenesis. Although these rodents express many of the same channel and transporter types as other mammals, they have especially prominent transient-outward potassium currents, so their action potential is extremely short (either lacking a plateau phase or having a very negative plateau). Although depolarizing currents are less species specific than repolarizing currents, calcium and sodium regulation in mouse and rat also differs markedly from that in larger mammals and humans.[13] This makes these species less-than-ideal models for human ventricular myocyte function. However, many fundamental aspects of muscle function are qualitatively similar to humans, and mice currently offer unique advantages for genetic manipulation. Indeed, knockout and knock-in technologies have facilitated the study of long-QT and Brugada phenotypes (via sodium channel mutations), catecholaminergic ventricular tachycardia via ryanodine receptor and FKBP12.6 mutations, and arrhythmogenic right ventricular dysplasia.[3] Thus, it is clear that well-considered questions about cardiac electromechanical activity can still be reasonably addressed in rodent systems. However, it is important that studies also are done in larger mammals in which the electrophysiological and calcium-handling properties more closely resemble those in human.

Species differences in the distribution and kinetics of ion channels are significant. As a result, drugs that lengthen human ventricular action potentials may have significantly

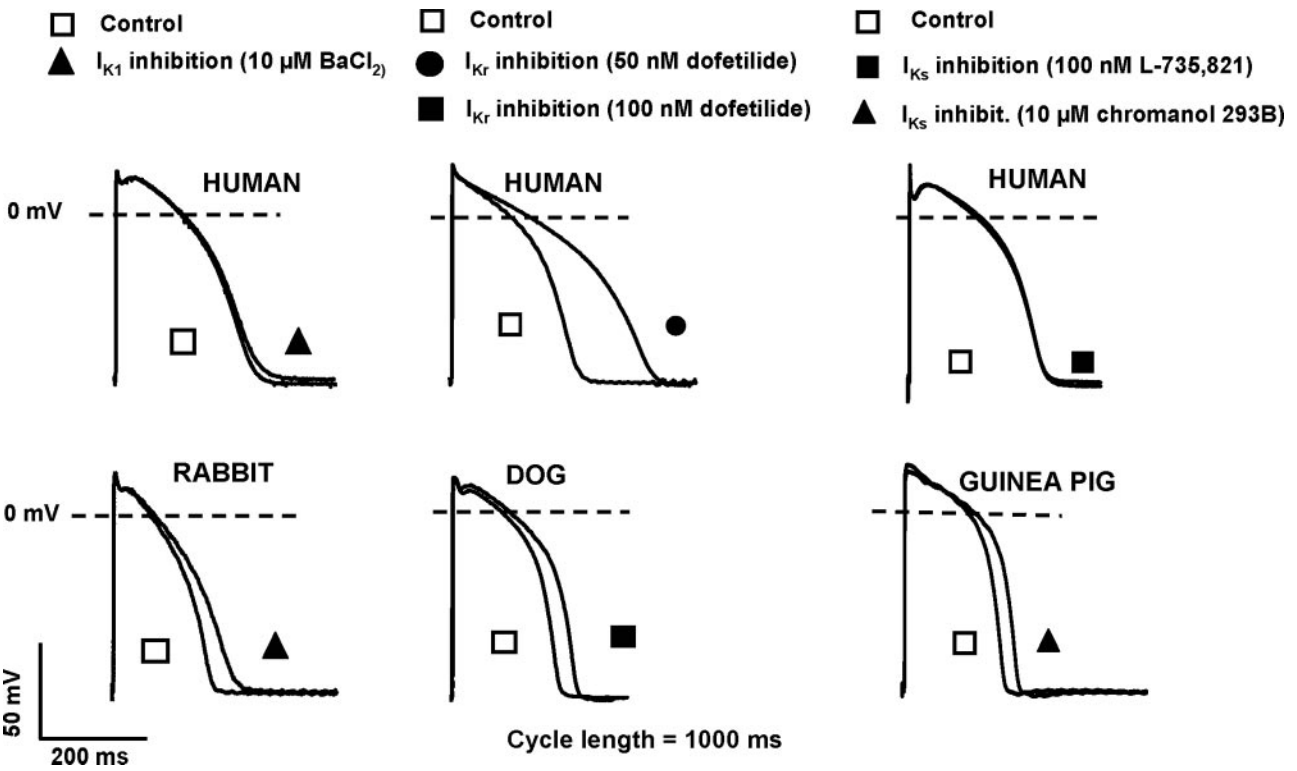

Figure 2. Differential impact of potassium currents I_{K1}, I_{Kr}, and I_{Ks} inhibition on the ventricular repolarization between human, dog, rabbit, and guinea pig measured by the conventional microelectrode technique in right ventricular papillary muscle preparations at 37°C. The figure illustrates the effects on the ventricular action potential of subtle variations in the balance and kinetics of currents underlying repolarization in different species. Note that the baseline morphology and duration of ventricular action potentials are qualitatively similar in each species and that, with respect to predicting human efficacy of drugs that modulate human cardiac repolarization, no species is identical to humans, making the creation of a detailed human-specific model a high priority.

different effects in other species[14] (Figure 2). Thus, interventions with little impact on human action potentials may significantly alter repolarization in other species (eg, blockade of the slow rectifier I_{Ks} in human versus guinea pig ventricle).[14] Development of appropriate computational approaches should better allow investigators to predict how ion current changes created or observed in rodents and other species will affect human cardiac electromechanical activity.

Efforts to develop techniques to manipulate gene expression in larger hearts are of great interest. To this end, it is notable that transgenic rabbits that express either a long-QT syndrome[15] or hypertrophic cardiomyopathy[16] phenotype have been developed. Techniques are evolving to provide local gene delivery to different regions of the heart,[17] and this technology has been used to modify the electrical activity of canine[18] and porcine hearts.[19] In addition to the introduction or overexpression of genes, interfering RNA technologies provide for the selective suppression of specific gene products.[19] Efforts that facilitate selective modulation of gene expression in the hearts of large animals are likely to provide important insights into the biochemical pathways involved in heart failure and arrhythmogenesis, as well as into novel therapeutic strategies. Further development of viral vectors is needed to ameliorate problems of transient transfection and inflammatory responses leading to myocarditis.[20]

Modeling Protein Structure/Function in the Context of Whole-Cell Physiology

Knowledge of the organization at the cardiac myocyte level is a critical focal point in understanding how the heart works and is an absolutely essential intermediate level in the systems biology approach to developing a multiscale understanding and computational models that span from molecules to cells to the whole heart (Figure 3). Knowledge of the organization of relevant ion channels, ion transporters, contractile elements, and signaling complexes within the cell is required to fully understand their function (integrated at the cellular or higher levels). There are substantial quantitative kinetic data for some of these systems in nonhuman cardiac myocytes (especially for ion channels, calcium and other ion transporters, and myofilaments but less so for many signaling complexes).[13,21,22] These data have already helped with the development of a highly mechanistic and quantitatively detailed understanding of myocyte electrophysiology and of calcium handling and contraction in animal models. The same approach is starting to spread to the area of signaling complexes (eg, involved in adrenergic sympathetic signaling and some protein kinase cascades).[23–27] However, there is a paucity of high-quality quantitative data for human cardiac myocytes, which are crucial for extrapolating our detailed knowledge from animal to human myocytes.[28,29] As noted earlier, there are major differences in ion channel, electro-

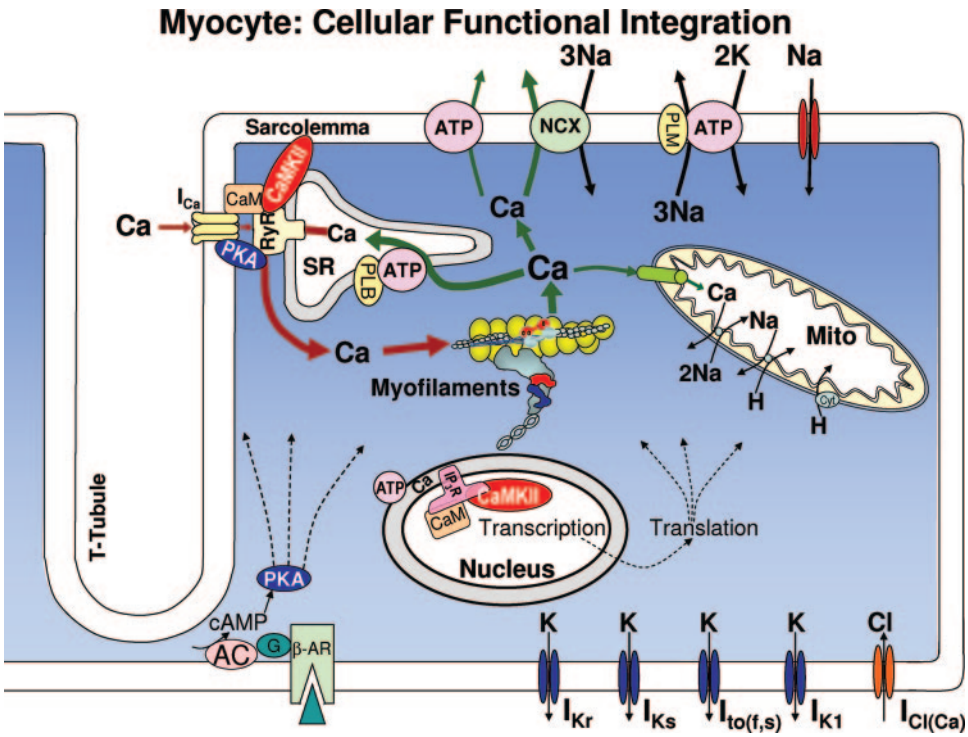

Figure 3. Cellular functional integration in a cardiac myocyte. Understanding the many systems (eg, ion channels, ion transporters, signaling cascades, mechanical function, energy metabolism, transcription, translation, and targeting of proteins) is a practical intermediate stage in multiscale modeling and understanding of the heart. We can integrate knowledge about molecules and their complex networks at this level and use it as building blocks for understanding and modeling at more integrative levels. RyR indicates ryanodine receptors; SR, sarcoplasmic reticulum; Mito, mitochondria; CaMK, calcium-calmodulin–dependent protein kinase; NCX, Na/Ca exchanger; PLM, phospholemman; PLB, phospholamban; ATP, ATPase; PKA, protein kinase A; β-AR, β-adrenergic receptor; I_{Kr}, I_{Ks}, I_{K1}, potassium channels; and $I_{Cl(Ca)}$, a calcium-activated Cl current.

physiological, and calcium handling systems between the common animal models and the human heart.

How Do Genetic Mutations Alter Protein Function to Cause Disease?

Advances in large-scale genetic screening have linked a predisposition for various forms of cardiac dysfunction to specific polymorphisms and mutations.[30,31] Understanding how arrhythmias and other cardiac disorders arise from aberrations in ion channel genes is an important and complex area of research. It is not yet possible to reliably extrapolate from a specific missense mutation to the precise clinical presentation of cardiac abnormalities in a given host.[31] This leads to a critical challenge in basic biomedical research: How do we predict altered physiological and pharmacological responses of ion channels and cells in the human heart that result from mutations and polymorphisms?

Basic research must be encouraged, facilitated, and supported to establish interrelationships among the kinetics and structure of human cardiac ion channels, transporters, and sarcomeric proteins using compendiums of disease-producing mutations and presumed "normal" genetic variations to link structure to function. Exploring the link between channel genotype and phenotype also requires systems approaches that combine appropriate clinical and experimental data to build computational models.[23,32] These representational models can then be used to simulate and elucidate the mechanisms linking channel defects to arrhythmogenesis and contractile pathologies.[23,33–36]

An important step is the development of sufficiently detailed mathematical models of ion channels that have significant predictive abilities. Such models are based on structural and functional studies of the biophysics of channel gating and can be expanded to incorporate the effects of mutations and molecular modification of channels into the native human cell. Computational model development is a multistage process that begins with the development of models that predict and improve understanding of the relationship between ion channel structures and their kinetic behaviors. Recent progress in x-ray crystallography has provided snapshots of gating processes, but many are still controversial.[37,38] At this juncture, a reasonable experimental approach is the measurement of channel function in well-controlled reduced systems using cloned channels aimed at measuring the permissible movements of the core domains.[39] As the structure and composition of the macromolecular assemblies of subunits and other regulatory proteins are better defined, the principles of relating structure to kinetic function can be expanded to apply to more complex transitions and interactions.

Even once the relationships between rare pathogenic mutations, common polymorphisms, and channel behavior are understood well enough to form a predictive model, realizing the consequences of a given kinetic modification or change in expression level on higher-order cardiac dynamics at the cell and tissue levels will require additional approaches.[32,33] Thus, systems biology approaches across scales (cell- and tissue-

level models) need to be developed.[40–42] Simulations in these models can help define and stratify the risk associated with a particular mutation in terms of well-defined arrhythmia susceptibility parameters.[43]

What Are the Mechanisms of Regulation and Modulation of Human Cardiac Ion Channels?

In addition to the unpredictability of ion channel function when mutations and polymorphisms are present, ion channels in the heart are targets of multiple modifications that permit normal cardiac function, drive or suppress disease states, and induce or respond to processes like aging.[44–47] Such regulation occurs on many temporal and spatial scales, resulting in targeting of channels to specific membrane locations and subsequent regulation by second messengers, hormones, neurotransmitters, humoral factors, kinases, phosphatases, and structural modulation by subsidiary subunits. Improving our understanding of the regulation of ion channels (and other cardiac proteins) is a critical future research direction. We must better determine how ion channels and other cardiac proteins are regulated in the native environment to construct a complete picture of electromechanical functioning in the human heart.

Progress in genetics, molecular biology, protein techniques, and imaging has markedly improved identification of human genes and the proteins they encode. Using these techniques also permits study of the structure and function of human ion channels in simplified cellular expression systems. This approach is critical for relating channel structural domains to specific kinetic transitions.[35] However, the benefits of studying human proteins in isolation are not yet readily translatable to the investigation of ion channel regulation and assembly with protein partners as they occur in vivo. In the myocyte environment, the proteins are in their normal membrane, mechanical couplings, and cellular milieu; experience the appropriate energetic and redox environment; and are physically localized near partner proteins that are part of very local signaling complexes. Therefore, characterization of key proteins (ion channels or transporters, myofilaments, and signaling networks) in the more complex myocyte setting is essential for building an appropriate understanding and quantitative models that may be scaled up to more integrated levels of tissue, organ, and organism. The simplicity of isolated systems has another inherent disadvantage: Ion channel modulation by protein partners must be known before it can be reconstituted in simplified systems. Unfortunately, partial reconstitution in heterologous systems may not reproduce the function of the protein in its native environment. This importance of the cellular environment is becoming more apparent as experimental studies increasingly reveal that ion channels exist as part of extensive macromolecular complexes that allow rapid modulation in response to a variety of extrinsic factors.[44,48,49] This is underscored further by the recent discoveries of novel long-QT syndrome and Brugada syndrome susceptibility genes involving channel interacting proteins like caveolin-3 (LQT9), a sodium channel β-subunit (LQT10), yotiao (LQT11), and glycerol-3-phosphate dehydrogenase 1-like protein (BrS2).[50–53] As indicated previously, an important objective is to obtain human data for the determination of kinetics, protein partners, and regulation in both normal and diseased human cardiac myocytes across the age spectrum. Determination of human ion channel regulation requires experimental studies in human cells and tissue or in animal or culture systems that approximate the native human system well. Related challenges include increasing our limited understanding of genotype–phenotype relationships (ie, a given disease-susceptibility mutation does not necessarily forecast a particular resulting phenotype), variability in response to pharmacology, neural hormonal mediation, rate dependence of arrhythmias, and gender and age as arrhythmia risk factors. A primary recommendation for improving our understanding of ion channel regulation is the collection of electrophysiological data, protein levels, and mRNA in cardiocytes derived from humans across the spectrum of age and disease states. Such experiments will allow the identification of ion channel protein partners in the human heart and functioning of channels in their native environment. Once pathways of modulation and their constituents are identified, the interactions of multiple modulatory pathways and perturbations to the pathways also must be studied. Development of novel cell expression systems will allow the recapitulation of the molecular complexes and regulatory function of human cardiac channels. Because of the enormous number of parameters involved in such investigations, computational methods must play a vital role in integration of experimentally obtained data.[32–34] The development of models and theory will yield quantitative predictive criteria for the functional/clinical relevance and outcomes of genetically based and acquired perturbations to cardiac channels, especially proarrhythmia susceptibility.[43]

Why Is Improving our Understanding of Protein Structure, Function, and Regulation so Important to the Patient?

The elucidation of cardiomyopathic and channelopathic genotypes has provided an abundant supply of naturally occurring perturbations that suggest a variety of structure-function relationships to be examined at the molecular, cellular, whole-organ, and whole-organism levels. However, although numerous variants have been identified and topological locations throughout critical cardiac ion channels have been identified, we currently have a very limited ability to use that information to make reasonable predictions about the effect on channel function. In addition to the identification of mutations and polymorphisms that increase disease susceptibility, a large number of other seemingly innocuous genetic variants have been identified. These nonpathological variants are often found in close proximity to, or even in the same position as, disease-causing variants, which tend to be scattered throughout the channel (Figure 4).

At the present time, our lack of understanding of channel structure and its relationship to channel kinetics and function prevents us from making robust predictions about manifestation of genetic defects (harmful or not). A vital research direction is to determine what constitutes structure and function of a "normal" channel. Furthermore, we must elucidate the mechanisms underlying incomplete penetrance

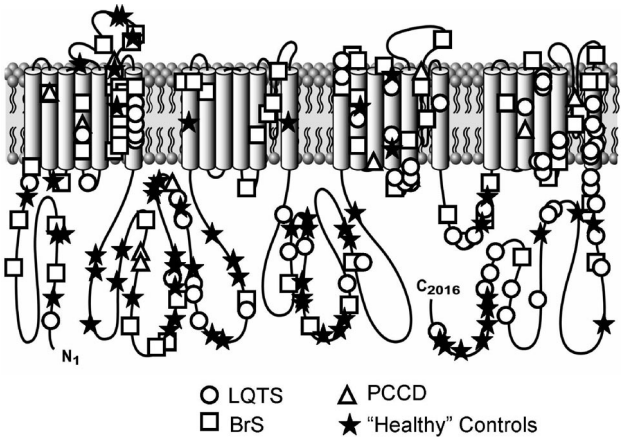

Figure 4. Location of disease-susceptibility mutations and "healthy" variants in the *SCN5A*-encoded NaV1.5 α-subunit. Shown is the linear topology for the *SCN5A*-encoded NaV1.5 α-subunit containing 2016 amino acids. Circles indicate the location of published missense mutations associated with long-QT syndrome (LQT3 specifically); squares, Brugada syndrome (BrS)–associated missense mutations; triangles, missense mutations associated with progressive cardiac conduction disease (PCCD); and stars, the location of rare nonsynonymous single nucleotide substitutions found among >1000 ostensibly healthy volunteers. The overlap most evident in the cytoplasmic interdomain linkers (DI–DII and DII–DIII) underscores the complexity and challenge in distinguishing between causative mutations and so-called background genetic noise.

and variable expressivity of polymorphisms or mutations.[47,54] This is especially important given that one of the current therapeutic interventions for the patient who is genotype positive for a heritable channelopathy or cardiomyopathy is an implantable cardiac defibrillator.[55] Such intervention is costly, both financially and psychologically. To avoid unnecessary interventions, systems analysis should be used to develop predictive criteria for disease predisposition and to develop a reliable functional readout.

Whole-Heart Simulations

A key strategy in integrating from individual building blocks to the whole heart is to characterize the emergent properties that arise at each successive scale in the hierarchy of biological complexity. With the increase in complexity at each scale, new and often unexpected properties arise from the cooperative interactions between individual components; that is, the whole becomes greater than the sum of the parts. Because these properties are often not intuitive, detailed knowledge of the individual components themselves may provide little mechanistic insight into their integrated function. Therefore, purely reductionist approaches do not suffice to explain biological complexity; rather, integrative approaches hold the most promise, wherein the greatest value of computational modeling and nonlinear dynamics lies.

Models can be either comprehensive and detailed (high dimensional), to realistically represent as many of the known biological interactions at the scale under investigation as possible, or reduced (low dimensional), typically using nonlinear dynamics approaches to capture phenomenologically and explain conceptually the essential emergent behaviors under investigation. Both methods complement experimental approaches in that a large number of parameters can be followed simultaneously and interactions between them can be revealed. Specific perturbations to 1 or more parameters also can be exactly applied to determine their effects at multiple scales of the system. A caveat with either form of modeling is that the boundaries and limits of interpretation of simulations must be specified.

Reduced (dynamic) models are most useful for understanding how new properties emerge from one scale to the next. For example, the action potential has no meaning at the scale of the proteome but emerges as a property at the scale of the cell because of relatively simple cooperative interactions between the ion channel proteins. Similarly, reentry has no meaning at the scale of the cell but emerges as a property at the scale of tissue. Reduced models, in which concepts such as excitability, refractoriness, electrical restitution, and other important electrophysiological characteristics are represented phenomenologically at the scale of the cell (so that they can be easily manipulated), have led to important insights into how action potential features control the stability of reentry.[43] Their limitation, however, is that phenomenologically represented parameters (such as refractoriness or action-potential-duration restitution slope) do not directly correspond to actual physical biological entities such as ion channel proteins.

In contrast, *comprehensive (detailed) models* are designed to span multiple scales to delineate the molecular basis of complex behaviors. They attempt to integrate from the protein scale of ion channels to the tissue/organ scale of reentry but are computationally challenging because they combine functionally integrated systems models with structural complexities at each scale.[33] The ideal approach combines both modeling strategies; it takes advantage of detailed structural data and physical principles to constrain structure–function predictions and uses nonlinear dynamics analysis to delineate the system parameters that control emergent behaviors at each scale.

Arguably, simulating cardiac electromechanical function is one of the most striking examples of a successful integrative multiscale modeling approach applied to a living system directly relevant to human disease. Today, thanks to nearly 50 years of research in the field and the rapid progress of high-performance computing, we stand at the threshold of a new era. Anatomically detailed, tomographically reconstructed models are being developed that integrate functions from the ion channel or sarcomere to the electromechanical interactions in the intact heart.[56,57] Such models hold great promise for enhanced interpretation of clinical and physiological measurements in terms of cellular mechanisms and for improving the basic understanding of the mechanisms of dysfunction in disease conditions such as reentrant arrhythmias, myocardial ischemia, and heart failure. Recently, these models also have been extended beyond electrical and mechanical mechanisms to include regulatory processes such as energy metabolism[58,59] and signal transduction.[23–27]

Although this progress has been encouraging, substantial room for further improvement remains. The limitations implicit in reductionist experimental approaches also apply to theoretical approaches in which model structures are specific

to the problem to be solved; for example, an action potential model may not be useful for understanding ionic homeostasis or contractile dysfunction. In addition, model parameters are only as good as the data they fit. For example, if an experimental approach is insufficiently detailed (eg, providing a single value for ATP concentrations that vary with location in ischemic tissue, or incapable of recording small changes in channel open probability of ATP-sensitive potassium channels), the model simulations may not be accurate in predicting associated phenomena. Detailed quantitative data and reconstructions of structure and physical properties at critical mesoscales such as those of subcellular organelles and multicellular tissue-matrix organization remain sparse, although new techniques are emerging for characterizing structure and function at these scales. Similarly, although models of short-term regulatory mechanisms such as responses to acute ischemia or adrenergic stimulation have been developed, mechanistic models of long-term remodeling associated with aging, development, and the pathogenesis of disease are still in their infancy. Efforts at developing comprehensive human heart models have lagged well behind those aimed at simulating animal models. And finally, new strategies developed by systems biologists using simple model organisms such as bacteria and yeast for systematic analysis of genome scale measurements have remained largely unexplored in cardiac biology.[60]

In implementing the integrative multiscale modeling strategy outlined above, we face a number of challenges, especially if our goal is to relate the findings to human disease. Specifically, we need to make better use of available human data to develop integrative, physiologically realistic multiscale models of the human heart. We also need to combine multiscale models of the human heart with patient-specific clinical information toward the goal of developing new tools and technologies that can inform clinical decisions and improve healthcare delivery.

Conclusions

In reviewing the limitations of available animal models and computational models based on data from them in understanding human cardiac electromechanical activity, the workshop identified the following challenges: (1) There is limited availability of human tissues, especially from those with no cardiovascular disease, for electromechanical characterization; (2) there is a lack of standardized protocols for procuring, preserving, and storing human cardiac tissues for study, and characterization of clinical parameters, cardiac function, genetics, and biomarkers is frequently inadequate; (3) studies with human myocardium thus far have provided limited information on regional differences in the human heart in cell physiology, histology, or gene expression; (4) available animal models do not adequately recapitulate the progression of heart failure or common arrhythmias in people; and (5) there is a limited understanding of the genetics and electromechanical activity in large-animal models (dogs, pigs, sheep, goats, and primates).

The workshop participants identified a critical need for a truly coordinated and integrative systems approach to understanding electromechanical activity in the human heart. To this end, the specific recommendations of the workshop participants are that the NHLBI should:

- Improve the quality and reliability of human cardiac electromechanical data by:
  - — Supporting research that identifies optimal conditions and methodologies for human cardiac tissue procurement, handling, and storage.
  - — Developing new tools and protocols for the perfusion and preservation of explanted human hearts that are unsuitable for transplantation but suitable for physiological studies.
  - — Improving protocols for the isolation and short-term culture of human myocytes.
  - — Making all of these protocols broadly available to the scientific community.
  - — Supporting studies of electrophysiology, calcium handling, and sarcomeric properties (and regulation) of the human myocardium at more institutions while promoting data sharing between investigators and institutions.
- Improve the understanding of mechanisms underlying the normal and abnormal activity of the human heart by supporting:
  - — Basic research to establish interrelationships among kinetics and the structure of human cardiac ion channels, transporters, and sarcomeric proteins using compendiums of disease-producing mutations and presumed “normal” genetic variations to link structure to function.
  - — Comparative studies of human versus animal cardiac gene expression, electrophysiology, and excitation–contraction coupling while also supporting model-based strategies for extrapolating information from animal to human models.
  - — The development of large animal models modified by genetic manipulation (eg, by viral infection and use of siRNAs) to identify the mechanisms underlying atrial and ventricular arrhythmogenesis and the development of heart failure in longitudinal studies of aging, remodeling, and disease progression.
  - — The study of electromechanical activity in normal human hearts and in specific disease states (ie, heart failure, ischemia/infarction, hypertrophy, specific genetic disorders that adversely affect cardiac activity) across life stages (fetal, pubertal, adult, senescence). Such studies should involve the development of novel cell systems for the recapitulation of human cardiac molecular complexes and regulatory function; the identification and characterization of cardiac channel macromolecular complexes and their electrophysiology, participation in calcium handling and sarcomere function; the determination of the effects of age, disease states, and remodeling on regulatory control (by phosphorylation, oxidation, etc) of human ion channels, calcium handling proteins, and sarcomeric proteins; and the examination of the intracardiac heterogeneities at each life stage and disease state to determine regional

myocyte properties (right ventricle versus left ventricle, base to apex, epicardial to endocardial, and atria versus ventricles).
— The development of novel noninvasive technologies for assessing detailed structural and functional properties of the human heart.

- Support the development and experimental validation of integrative multiscale computational models of the normal human heart and of specific cardiac diseases (arrhythmias, heart failure, and myocardial ischemia/infarction) and their progression that:
  - — Integrate multiple subsystems specific to the pathogenesis of the disease state and characterize the dynamics of their interactions at each scale. This will also require support to develop and maintain advanced computational tools and technologies needed to ensure computational tractability and robust and efficient models of human electromechanical activity.
  - — Incorporate the structural alterations associated with specific diseases at each scale.
  - — Characterize the adaptive and maladaptive responses that underlie the progression of disease.
  - — Integrate across scales to predict the electromechanical outcomes of genetically based and acquired perturbations.
  - — Inform clinical diagnosis and guide the selection of appropriate therapies in a patient-specific manner. Molecular/cellular investigations of explanted tissues, including histological and/or immunohistochemical characterization, should be combined with clinical data and high-resolution, noninvasive structural (computed tomography, magnetic resonance imaging), functional (echocardiography), and electrophysiological imaging obtained before surgery.

- Convene a series of workshops to build consensus and to improve communication among investigators working at the same horizontal level (those measuring and modeling individual ion channels, transporters, or myofilament properties in myocytes) and at different vertical levels (genomic/proteomic to cellular, and cellular to more integrative levels). This is part of a broader aim to support collaboration and cooperation among molecular and cellular experimental, computational modeling, bioengineering, and clinical investigators in a true systems approach to understanding electromechanical activity in the normal and diseased human heart.

Disclosures

Dr Rudy is a member of the scientific advisory board and holds equity in CardioInsight Technologies, Inc. Dr Ackerman is a consultant for PGxHealth with respect to their FAMILION genetic tests for cardiac ion channel mutations and also serves as a consultant to Medtronic and Pfizer. Dr London has served as a consultant to and/or on the advisory board of Medtronic. Dr McCulloch is a cofounder of Insilicomed, Inc, a licensee of University of California San Diego software developed in his research. Dr Solaro is a member of the scientific advisory board of Cytokinetics, Inc, and has received honoraria from Eisai Pharmaceuticals. The other authors report no potential conflicts.

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Circulation. 2008;118:1202-1211
doi: 10.1161/CIRCULATIONAHA.108.772715
Circulation is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231

Print ISSN: 0009-7322. Online ISSN: 1524-4539