What $I_f$?

Editorial

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Lead Article

The control of heart rate: the physiology of the sinoatrial node and the role of the $I_f$ current

M. J. Shattock, M. R. Rosen

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Bibliography of One Hundred Key Papers
In 1979, Brown, DiFrancesco and Noble reported in *Nature* a sinoatrial current that was so unusual they felt compelled to christen it the “funny” current ($I_f$). The funny thing about this current, which is generated by the inward movement of sodium and potassium ions, is that it is activated by hyperpolarization of the cell membrane. Some experts argued that this current might play a key role in the control of pacemaker activity, while other experts treated this claim with some suspicion, pointing out that it was a very small current and only one of the 10 or more currents responsible for the propagation of the nodal action potential. It certainly seemed an unlikely that $I_f$ might represent a therapeutic target in the battle against cardiovascular disease. However, nowadays, as explained by Michael J. Shattock and Michael R. Rosen, this curious little channel is known to be able to exert a substantial effect upon heart rate such that its chemical inhibition may reduce cardiac rate by as much as one fifth without altering the profile of the action potential or the inotropic state of the ventricle.

Interest in the possible beneficial effects of reducing heart rate had its origins in studies, also reported in the late 1970s, that demonstrated strong linear relationships between life span, body mass, metabolic rate, and heart rate. This work culminated in a classical paper, published in 1997 by Herbert Levine, showing in mammals an inverse semi-logarithmic relationship between heart rate and life expectancy (see Figure, next page).

Levine’s observation raised the alluring possibility of extending life by cardiac slowing. However, as is very clear from the *Figure*, man appeared to be an exception in this otherwise striking relationship, boasting a life span far greater that that predicted from human heart rate—a phenomenon that might be explained on the basis of the medical and scientific benefit enjoyed by mankind. At present, there is no definitive evidence that reducing heart rate in healthy individuals either by exercise or by drugs results in
an increase in longevity. Although ardent runners might believe that spending 5% of their waking hours acquiring a jogging-induced bradycardia will result in an extended life span, the evidence is just not there, and, in any event, it is perhaps of questionable benefit if they only live 2% longer! By contrast, there is clear evidence that reducing heart rate is associated with an improved survival in a variety of pathologies including cardiovascular disease, and this presents a fascinating therapeutic possibility that is explored in this issue of *Dialogues.* To this end, the superb fail-safe electrical design of the sinoatrial node, together with the advent of *I*ₚ inhibitors that reduce heart rate within a tightly controlled range, should provide a means to test more thoroughly the above associations and perhaps allow us to use our lifetime allocation of $25 \times 10^8$ heart beats most productively.
The control of heart rate: the physiology of the sinoatrial node and the role of the $I_f$ current

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A heart will beat approximately $25 \times 10^8$ times during the course of a human being’s life. The observation that most mammals share the same number of heart beats per lifetime, be it a mouse (600 bpm for 2 years) or a giant tortoise (6 bpm for 200 years), has led people to suggest that if we slowed our heart rate we might live longer! While there may not be a causal relationship in fit healthy animals, there is an impressive literature accumulating suggesting that there is a clear and strong association between a lower heart rate and improved prognosis in a variety of diseases. It is, of course, in cardiovascular disease where the slowing of heart rate with specific bradycardic agents may offer the most advantage. The question as to whether $I_f$ inhibition can specifically improve morbidity and life expectancy in cardiovascular disease is one we only recently have had the tools to answer, and the introduction of ivabradine into the armamentarium offers a unique opportunity to address this question. We will know a lot more when the BEAUTIFUL trial (MorBidity-mortality EvAUltion of the $I_f$ inhibi-tor ivabradine in patients with coronary disease and left ventricULar dysfunction) concludes in December 2007.

We are all born with a finite number of heart beats: I, for one, am not going to waste mine on exercise

Dr A. Glenn Morrow
(attributed by Dr J. Borer)

A fundamental feature of the mammalian heart is its ability to maintain rhythmic contractions in the absence of external stimulation. The mammalian heart contains a myogenic, or muscle-derived, pacemaker, the sinoatrial node (SA)—a small, but complex, conglomeration of specialized tissue located in the outer reaches of the right atrium. The impulses arising from the pacemaker both determine the heart rate and provide a target for its physiological and pharmacological modulation. The node contains a mixture of nodal cells, connective tissue and atrial cells, and an understanding of the complex interplay between these tissues is essential for under-standing how the node works. The specialized nodal cells are themselves not homogeneous—cells from the center of the node being both morphologically and electrophysiologically different from more peripheral cells. However, the defining feature of the nodal cells themselves is that they show inherent pacemaker ac-tivity—that is, their diastolic membrane potential is not stable, but gradually depolarizes to a threshold from which a new action potential is triggered. The ionic currents present during this pacemaker depolarization

**Keywords:** sinoatrial node; heart rate; pacemaker; ion current; action potential; HCN channel; ivabradine

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**SELECTEDABBREVIATIONSANDACRONYMS**

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
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<tr>
<td>BEAUTIFUL</td>
<td>MorBidity-mortality EvAUltion of the $I_f$ inhibitor ivabradine in patients with coronary disease and left ventricULar dysfunction</td>
</tr>
<tr>
<td>HCN</td>
<td>hyperpolarization-activated cyclic nucleotide-gated cation channel</td>
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<tr>
<td>SA</td>
<td>sinoatrial (node)</td>
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phase are many (at least 10 have been identified) and this provides levels of redundancy to the function of the node that make it very hard to stop. Nodal cells also have one other significant feature—they do not have the large time-independent background potassium current ($I_K$) that is present in ventricular cells. This gives the nodal cell a high-input impedance, which, in short, means small currents can exert big effects! Perhaps one of the most important of these small currents is the hyperpolarization-activated inward current—$I_h$. $I_h$ is the target for adrenergic and cholinergic modulation of heart rate: agents that increase $I_h$ accelerate heart rate and those that inhibit $I_h$ slow heart rate. This article reviews the physiology of the SA node and, in particular, the importance of $I_h$ in the regulation of heart rate. Understanding the physiology of the SA node allows the understanding of how such a small current can exert a significant effect on heart rate and the seemingly contradictory observation that even complete blockade of this current is safe and will only slow heart rate by about 30%.

**EXTRANSC VS INTRINSIC PACEMAKERS**

What can we learn from evolution?

As evolution progressed from simple single cells to complex multicellular organisms with discrete specialized tissues, the need for an efficient circulatory system developed in parallel. Simple invertebrates such as the brachiopods (clam-like shellfish) and ascidians (sea squirts) circulate rudimentary blood-like substances around open body cavities aided by inefficient peristaltic “tube” hearts. Higher invertebrates such as the decapods (lobsters) and the cephalopods (octopi, etc) evolved more complex multichambered hearts that require synchronization, coordination, and rhythm (for review see reference 1). To generate the rhythm, nature experimented with two different strategies: (i) extrinsic neuronally derived pacemakers; or (ii) intrinsic specialized muscle tissue. The intrinsic myogenic pacemakers are actually both evolutionarily ancient and widespread—being present in most molluscs, insects, and all vertebrates (apart from the lamprey). Myogenic hearts have their own intrinsic muscle-derived pacemaker, continue to beat when isolated, and contract sequentially with the wave of contraction spreading monotonically from one point to another. Neurogenic hearts are found in the annelids (segmented worms), crustaceans, and arachnids (spiders), receive their pacemaker commands from an extracardiac source (the cardiac ganglion), stop when isolated, and all parts of the heart tend to contract simultaneously.

Both neurogenic and myogenic pacemakers have their advantages and disadvantages. Neurogenic hearts provide instant control, but are easy to stop. The simultaneous contraction of a neurogenic heart works well in a tube heart, but poorly in a multichambered heart. Myogenic pacemakers, conversely, are very hard to stop, but require complex and slow regulation via circulating factors from both endocrine and paracrine sources. The wave-like propagation of contraction in the myogenic heart has mechanical advantages for a single chamber, but does not easily allow for synchronization and coordination between chambers.

While the mammalian pacemaker is essentially myogenic, rather than choosing between these two evolutionary blueprints, the mammalian heart has incorporated the best features of both systems.

**The pacemaker of the mammalian heart: a functional mix?**

The primary pacemaker of the mammalian heart is the SA node, which consists of specialized myocardial tissue (ie, is "myogenic") (see Boyett et al2 for review). Excitation spreads from the node via the atrial muscle before arriving at the atroventricular node. Here the impulse is first delayed and then moved rapidly through the specialized “neuronal-like” conduction pathways (“neurogenic”) of the bundle of His and Purkinje fiber system before spreading in a propagated wave (“myogenic”) through the myocardium from apex to base. In this way, the mammalian pacemaker generates a robust myocardially derived pacemaker, its more organized or "neuronal-like" propagation allows for the necessary coordination of contraction of a multichambered heart, and, within chambers, the contraction wave favors ejection by spreading toward the outflow tracts of the respective chambers.

**LOCATION AND STRUCTURE OF THE MAMMALIAN SINOATRIAL NODE**

**Gross anatomy**

The SA node is a small tear-drop shaped cluster of specialized tissue located in the right atrium at the junction of the superior vena cava, the inferior vena cava and, the crista terminalis (Figure 1). In man, estimates of the size of the node vary from 7 to 20 mm in length4,5 and, in the rabbit, 2 to 4 mm in diameter × 6 to 8 mm in length. The mammalian SA node is a heterogeneous mixture of specialized nodal cells (which
are themselves by no means homogeneous), atrial myocytes and a surprisingly large amount of connective tissue (Figure 1). The amount of connective tissue within the node varies with species and age, but can make up between 50% and 90% of the node. It is now clear that both the preponderance of connective tissue and the mixture of other excitable tissues within the SA node are fundamentally important to normal function.2

The location of the leading pacemaker site in the rabbit SA node is shown in Figure 1 and the isochrones show the propagation of the wave of excitation away from its site of initiation.2 Contrary to the schematics in most textbooks, the wave of excitation actually spreads anteriorly and obliquely (towards the top left of Figure 1) towards the crista terminalis, which it meets as a broad wave front before turning and heading around and down towards the atrioventricular node.2

Figure 1 also shows a region of low conductance (the block zone), which is located close to the leading pacemaker site and provides a region of conduction block preventing excitation from spreading prematurely towards the septum. This anatomical arrangement and the circuitous “up and around” excitation pathway from the node may confer physiological advantages by: (i) ensuring the wave of atrial contraction spreads from the very top of the chamber forcing blood down towards the atrioventricular valve; and (ii) the block zone may then prevent reentry of electrical excitation from the atrium back into the node.

**Microanatomy**

The specialized nodal tissue is itself heterogeneous (Figure 2, next page).3,7 Close to the center of the node, the cells are small (5 to 10 µm in diameter and 25 to 30 µm long), spindle-shaped, poorly differentiated, contain few myofilaments or mitochondria, and have highly convoluted membranes containing many caveolae (Figure 2A). In short, central nodal cells are simply empty membranous bags containing few specialized structures. These central nodal cells (sometimes called P cells) are the site of the primary pacemaker. Moving outward away from the center of the node, in most species there is a gradual transition with cells becoming larger, having a clear myofilament structure, and containing more mitochondria (Figure 2B). Interspersed within the node are a third type of nodal cell termed “spider cells,” which are seen when the node is disaggregated into its component parts (Figure 2). The role of these spider cells is uncertain, but clearly their extensive “dendritic” structure and their large surface-volume ratio may suit them to influence the propagation of the electrical wave within the microdomains of the node and to facilitate electrical coupling between adjacent areas.
The physiology of the SA node and the role of \( I_f \) - Shattock and Rosen

**CELLULAR ELECTROPHYSIOLOGY OF SINGLE NODAL CELLS**

**Regional differences within the node**

The vast majority of cells within the node show the slow diastolic pacemaker depolarization phase that is the defining characteristic of the SA nodal action potential (Figure 3A) and provides the fundamental requisite “clock function.” However, the exact shape of the action potential varies among the differing cell types within the node reflecting an underlying heterogeneity of ion channel expression across the node.\(^2,8\) The small cells, presumed to be from the center of the node, have lower resting membrane potentials and a slower diastolic depolarization phase than the larger peripheral cells.\(^8\) The smaller cells also have a slow action potential upstroke, reflecting a low expression of Na channels and a depolarization phase dominated by Ca inward current. In moving away from the center of the node towards the periphery, the expression of Na channels increases and with this comes a faster, taller action potential.\(^8\) Isolated peripheral nodal cells also show an increase in their spontaneous beating rate (above that seen in situ) and an increased rate of diastolic depolarization—most likely as a consequence of an increase in the pacemaker current \( I_f \).\(^9,10\)

**Figure 2.** Photomicrographs of the four different cell types isolated from the rabbit sinoatrial (SA) node. Cells are stained with hematoxylin and eosin.


**Figure 3.** Spontaneous action potentials recorded from two cells isolated from rabbit sinoatrial (SA) node. A: recording from a small cell (22pF) assumed to be from the center of the node. B: recording from a large cell (57.5pF) assumed to be from the periphery of the node.

This increase in firing rate of the peripheral tissue is probably a feature of isolated cells as, in the intact heart, the electrotonic coupling of these cells to the surrounding atrial tissue is likely to counteract this effect.

The heterogeneous distribution of ion channel expression in the various regions of the node allows the primary pacemaker site to “wander” according to prevailing conditions. Activation or inhibition of a particular channel thus does not inhibit the node altogether, but simply allows the primary pacemaker site to shift to a region of the node where these channels play a more minor role. This spatial heterogeneity is thus a key safety feature of the node making it very hard to stop.

Multiple currents make the node both hard to stop and exquisitely regulatable

Not only does the spatial heterogeneity of ion channel expression provide a fail-safe pacemaker, but within individual cells the multiple ionic currents underlying pacemaker activity provide layers of redundancy. Figure 4 shows a computer simulation (Oxsoft Heart v4.0) of the principal ion currents underlying the SA node action potential. For comparison, these currents have all been plotted on the same vertical scale. At least 10 ionic currents have now been identified as contributing to the SA node action potential. However, Irisawa et al (1993) have pointed out only two currents are required to produce rhythmic activity in nodal cells—a time-independent outward current and the Ca inward current (I\text{Ca}). Thus, the multiple ionic currents within nodal cells provide levels of redundancy that again make the node hard to stop—a very useful safety feature.

Multiple currents make the node both hard to stop and exquisitely regulatable

How can small currents have such a big effect on heart rate?

In ventricular cells, the resting membrane potential is determined principally by the high permeability of the membrane to potassium ions and the intra- and extracellular potassium concentrations. Thus, in ventricular myocytes, the resting membrane potential (i.e., where net current flow is zero) is typically around –85 mV. The relationship between current flow and voltage is described by Ohm’s Law (Voltage = Current × Resistance, or V=IR). Figure 5A (next page) shows this rela-
tionship for the resting membrane of a ventricular myocyte, and this steady-state current through potassium channels is carried by the inward rectifier—\( I_{K1} \).

This current-voltage relationship crosses the voltage axis at the resting membrane potential (ie, around –85 mV) and, because the membrane is very permeable to potassium at these voltages, the slope of this relationship is steep (ie, conductance is high). Thus, at around –85 mV, where the “resistance” of the membrane is low, a small current has only a small effect on membrane voltage (Figure 5A inset). This makes the resting potential of a ventricular cell inherently stable and protects the cell against inappropriate excitability and arrhythmias—a useful feature for ventricular cells.

Contrast this with the situation in a SA node cell (Figure 5B). The input impedance (resistance) of the SA node cell is ≈30 times that of a ventricular myocyte\(^1\),\(^2\) (ie, the slope of the I-V relationship is much shallower) and hence a small current can induce a large change in membrane voltage (see inset). The data in B are redrawn from reference 17: Noma A, Nakayama T, Karachi Y, Irisawa H. Resting K conductances in pacemaker and non-pacemaker heart cells of the rabbit. Jpn J Physiol. 1984;34:245-254. Copyright © 1984, Center for Academic Publications Japan.

The “PACEMAKER CURRENT” \( I_f \)

**Characteristics**

In 1979, Brown and colleagues in Oxford identified an inward current in SA node preparations that had the unusual property of being activated on hyperpolarization of the cell membrane.\(^3\)\(^4\) At this time, a hyperpolarization-activated inward current was so unusual it was termed the “funny” current—\( I_f \). Since then, a similar current carried, as it transpires, by the same family of channels, has been identified in neuronal and retina cells and has been termed the hyperpolarization-activated current \( I_h \) (see Pape\(^5\) for review).

Figure 6 shows the principal property of \( I_f \)—that is, it is activated by hyperpolarizations negative to ~50 mV. \( I_f \) can be carried by a mixture of Na and K ions (although Na is the major carrier in physiologic settings) and has all the features necessary to be a primary pacemaker current in the heart.\(^6\)\(^7\) For example, \( I_f \): (i) is activated on hyperpolarization from a threshold of about –50 mV (and is fully activated around –110 mV)
and hence contributes to diastolic 'pacemaker' depolarization; (iii) is increased by \( \beta \)-receptor stimulation; and (iii) is inhibited by acetylcholine (ACh). 21

**Role of \( I_f \) in the control of heart rate**

While it is clear that \( I_f \) may influence pacemaker function, the extent to which it is important in the control of heart rate has been hotly debated. 14,15,23,24 While DiFrancesco and colleagues 14,25 have argued that \( I_f \) is exclusively responsible for pacemaker activity, others have suggested that it plays only a minor role. 15,23,24,26 The roots of some of this controversy are illustrated in Figure 6A. The diastolic depolarization phase of the SA node action potential shows a maximum diastolic potential of –60 to –65 mV, which depolarizes gradually to about –55 mV over about 200 ms. From Figure 6B, however, it is clear that \( I_f \) is both slow to activate and small at these voltages. Thus, 100 to 200 ms into the voltage-clamp steps shown in Figure 6B, at voltages between –65 and –55 mV, \( I_f \) will only reach about 40 pA at most. These observations, and others, have lead to the suggestion that \( I_f \) does not play a major role in cardiac automaticity, but simply is there to prevent excessive hyperpolarization. 2,15,23,24,26

Much of the debate about \( I_f \) and its importance, or lack thereof, in pacemaking relates to the specific details and conditions under which in vitro experiments are performed. 14,15 A simpler and pragmatic answer to the question, "Does \( I_f \) contribute to pacemaker function in man?" is provided by the clinical studies of agents reported to be specific and selective for \( I_f \) inhibition. For example, ivabradine (0.3-3 \( \mu \)M) is a potent inhibitor of \( I_f \) and, at these concentrations, has been shown to reduce heart rate by 10 to 15 bpm in clinical studies. 16 Since ivabradine has been shown to be selective for \( I_f \) at these concentrations, 27 this would suggest that \( I_f \) does indeed contribute to the control of heart rate in vivo. What is clear from this debate is that while \( I_f \) certainly contributes to pacemaker function, it is not the sole determinant of rhythmic activity. This again shows that there are levels of redundancy within the SA node that provide a fail-safe feature such that that cardiac rhythm can be both initiated and regulated in the event of the failure of any given mechanism.

### Regulation of \( I_f \)

The firing rate of the SA node is exquisitely modulated by innervation from the autonomic nervous system. Basal heart rate is under the influence of a maintained vagal tone and sympathetic stimulation can dramatically increase heart rate. In isolated SA node cells, \( \beta \)-receptor stimulation increases, and ACH slows, the rate of SA node cell diastolic depolarization (Figure 7A, next page). The original description of \( I_f \) by Brown et al in 1979 showed that this current is substantially increased by \( \beta \)-receptor stimulation. 19 Figure 7B shows a series of steady-state activation curves for \( I_f \). 28 These curves describe the fraction of channels activated at a given potential and the control curve shows that at –60 mV only \( \approx \)23% of available channels are activated. However, \( \beta \)-receptor stimulation shifts this activation curve to the right such that at –60 mV there is an 87% increase in available channels. Conversely, ACH shifts the activation curve to the left, reducing the available channels at –60 mV by 70%.

These shifts in the activation curve for \( I_f \) are mediated by changes in intracellular concentration of cAMP.
β-Receptor stimulation, through its activation of adenyl cyclase, elevates cAMP, which then binds directly to the cytoplasmic tail of the If channel, shifting its activation curve to the right. Conversely, ACh, via muscarinic receptors, inhibits adenyl cyclase, decreases cAMP, and shifts the If activation curve to the left. DiFrancesco and colleagues have argued that this is the explanation for the negative chronotropic effects of ACh in contrast to the established textbook explanation—that is, ACh activates an ACh-dependent K current (IK,ACh).29 In support of this, these authors have shown that low concentrations of ACh (0.01-0.03 µM) inhibit If without affecting IK,ACh, and an increase in IK,ACh, requires 20 times the concentrations required to inhibit If.29

In addition to the modulation of If by autonomic neurotransmitters, a number of other physiologically-relevant agents have been shown to modulate this current. Activators of If include vasoactive intestinal peptide (VIP),30 thyroid hormone (T3),31 and nitric oxide,32 while If is inhibited by neuropeptide Y (NPY) and adenosine.33

Molecular identity and tissue distribution of If channels

The channel responsible for the cardiac If current is now known to be a member of a family of channels termed the hyperpolarization-activated cyclic nucleotide-gated cation (HCN) channels (for review see reference 22). Four HCN genes have been identified in mammalian tissues, termed HCN1-4. RNA messages for all 4 HCN isoforms have been found in heart,34 but in the SA node HCN4 is the dominant isoform in most species, accounting for ≈80% of the messages.35-37 In mouse, HCN2 makes up the remainder with only very low levels of HCN1.37 This is not the case in rabbit where the remaining 20% is mostly HCN1.36 HCN1 is highly expressed in retinal photoreceptors37,38 and in the brain.39,40 HCNs 2 and 3 are also expressed in brain.40,41 Within the SA node the expression of HCN channels is not uniform. Cells from the center of the node show little If current, while towards the periphery If is larger,9,10 and this correlates with a heterogeneous expression of HCN4 channels.42

Studies in knockout mice have shown that the homozygote deletion of the gene encoding HCN4 is embryologically lethal.43 However, hearts and cardiomyocytes could still be isolated from these embryos and they showed If reduced by 80% and heart rate by about 40%. Importantly, the HCN4-deficient hearts still contracted regularly without arrhythmias, showing that, as suggested above, while If clearly contributes to pacemaker function, it is not the sole determinant of rhythmicity. Interestingly, the contraction rate in HCN4-deficient hearts was unaffected by raising cAMP, suggesting that this channel not only contributes significantly to basal heart rate, but also mediates the chronotropic response to adrenergic stimulation.43

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Figure 7. Effect of β-receptor and muscarinic-receptor stimulation on: (A) spontaneous action potentials recorded from an isolated sinoatrial (SA) nodal cell; and (B) the steady-state activation curve for If. Isoprenaline (iso) accelerates, and acetylcholine (ACh) slows, the spontaneous rate of an isolated SA node cell by altering the rate of diastolic depolarization. The shaded area in both panels indicates the diastolic voltage range from -45 to -70 mV. Panel B shows that at -60 mV only 23% of the available channels are activated. However, the fraction of channels available increases by 87% in isoprenaline (10 nM) or decreases by 70% in response to ACh (3 nM).

Consequences of $I_f$ block

An elevated resting heart rate has been shown to correlate strongly with increased mortality in angina, heart failure, and even cancer. It is possible that it is an epiphenomenological indicator of an underlying increase in sympathetic tone. In recent years the pharmaceutical industry has searched for pure bradycardic agents that can lower heart rate without the negative inotropic consequences of β-blockade or Ca antagonism. The list of such specific bradycardic agents (SBAs) now includes alnididine, zatebradine, and ivabradine. Figure 8A shows that ivabradine (0.3 µM) slows the rate of diastolic depolarization in an SA node cell, and hence slows the firing rate of this cell (in this case by approximately 18%), without changing the shape of the subsequent action potential. The inhibitory action of ivabradine on $I_f$ is shown in Figure 8B where a higher concentration (3 µM) reduces $I_f$ by ≈78%. Ivabradine blocks the channel when it is in its open state and hence its block is use-dependent—that is, in isolated tissues its ability to block the channel increases as the beating rate increases. While such use-dependence sounds an attractive feature (ie, the drug should exert a larger effect when heart rate is high) evidence from Borer et al (2003) suggests that in man the negative chronotropic effect of ivabradine in absolute terms is comparable at rest and during peak exercise.

Figure 8C shows a computer simulation (Cellular Open Resource; COR - Oxford University) of the effects of the complete blockade of $I_f$ on the SA node action potential. This model predicts that, under steady state conditions, complete blockade of $I_f$ will reduce SA node rate by approximately 20% to 30% in an isolated cell. This demonstrates the fail-safe feature of SA node cells discussed earlier—that the intrinsic pacemaker activity of the node is not dependent on a single ionic current and hence intrinsic pacemaker activity at the level of a single cell or, when integrated across the entire node, is hard to stop.

Figure 9A (next page) shows the dose-response relationship for ivabradine in isolated SA node cells with a half-maximal inhibitory concentration of 2.2 µM. Figure 9B shows a dose-titration study in patients where the dose of ivabradine has been increased stepwise from 5 to 20 mg bid. In Figure 9B, it is interesting to note that the major bradycardic effect is seen on going from control to 10 mg (bid). Further increases in dose above 10 mg result in only a small further drop in heart rate, indicating there appears to be a “plateau effect” such that the maximal drop in heart rate seen in this study plateaus at about 30%. This is in accordance with the in vitro and simulation data shown in Figure 8, suggesting that even complete block of $I_f$ will not cause...

severe bradycardia or sinus arrest. Thus, assuming ivabradine is indeed selective for the $I_f$ channel, then, at least in terms of cardiotoxicity, this agent is likely to be safe and of potential therapeutic benefit in pathologies where lowering the heart rate is desirable.

**CAN “FAIL-SAFE” GO TOO FAR?**

**THE PLUSES AND MINUSES OF INTERACTIVE BACKUP SYSTEMS**

As detailed above, one of the positive aspects of a pacemaker whose function is supported by multiple ion currents is that in the event of malfunction of part of the system, other limbs may come into play. For example, excess hyperpolarizing action of outward K currents may be counteracted by the activation of the inward $I_f$ pacemaker current. The rapid rates that may be achieved with excess increases in net inward current or decreases in repolarizing current may be attenuated by enhanced vagal tone to decrease $I_f$ and increase $I_{K,Ach}$. The latter current provides yet another means whereby hyperpolarization and a decrease in diastolic depolarization and pacemaker rate may be achieved.

A further fail-safe mechanism is provided by the dispersion of pacemaker cells within the SA node. The dissemination of anatomically and electrophysiologically defined pacemaker tissue within the region of the SA node is such that more than one site can function as the pacemaker, with the most rapid site of impulse initiation overdrive-suppressing the slower sites. Autonomic tone and other less-well defined interventions also influence both site and rate of cardiac pacemaking.

A problem that arises is in the setting of SA nodal dysfunction, where bradycardias and tachycardias may occur often in seemingly haphazard juxtaposition, although the initiation of tachycardia after a period of bradycardia has been well defined. Treatment is often difficult and may involve the use of SA nodal ablation and device therapy. One might question whether this syndrome, or family of syndromes, arises from a malfunction in the primary pacemaker current, $I_f$, or in one of the other currents that contribute to the pacemaker potential. Alternatively, does it come about as a result of structural abnormalities in the nodal region, such as excess uncoupling of pacemaker cells from one another and the surrounding tissues? These are issues for which there are currently no answers and which must be solved if we are to contend with this important cardiac rhythm disturbance.

**CONCLUSION**

In summary, the mammalian SA node is a complex heterogeneous mixture of nodal cells, atrial tissue, and connective tissue. The nodal cells themselves are both morphologically and electrophysiologically heterogeneous and it is this macro- and micro-diversity that is key to the functioning of the node. The heterogeneous ion channel expression in different regions of the node and the multiple ion channels within a single cell that contribute to pacemaker activity make the node fail-safe and very hard to stop. The multiple currents contributing to the pacemaker depolarization, and the high
input-impedance of the nodal cells, mean that not only is the node hard to stop, but it is also exquisitely regulatable. That is, small currents can exert significant effects on heart rate. One such small current that appears to play a crucial regulatory role is the hyperpolarization-activated cyclic nucleotide gated cation channel that conducts the “funny” current \( I_f \). Prevailing evidence suggest that it is this current that is the target for both the adrenergic and cholinergic modulation of heart rate. \( I_f \) is also the target for a new class of specific bradycardic agents such as ivabradine that have the potential to safely and selectively lower heart rate without the risk of profound bradycardia or sinus arrest. Results of the BEAUTIFUL trial (MorBidity-mortality EvAlUaTion of the \( I_f \) inhibitor ivabradine in patients with coronary dis-ease and left ventricUlar dysfunction), which concludes in December 2007, should shed more light on the clinical benefits expected from this promising agent.

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Can \(I_f\) inhibition help in angina?

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The case for a rate-lowering approach to the prophylaxis of ischemia, the underlying cause of angina, was always firmly grounded in physiology and therapeutics. What it lacked, until the research effort sparked in 1979 by the discovery of the \(I_f\) current, the central sinoatrial determinant of heart rate, was pharmacological embodiment. Ivabradine, approved in 2005 for angina prevention, displays the efficacy and safety profile of a specific and selective \(I_f\) inhibitor that was predicted on pharmacological first principles: noninferiority versus atenolol and amlodipine in times to ST-segment depression and limiting angina, with none of the extraneous and negative hemodynamic, inotropic or metabolic effects associated with \(\beta\)-blockade or calcium channel blockade. First principles also predict improved survival, which awaits confirmation in a clinical trial.

Coronary artery disease (CAD) is characterized by obstructions to myocardial blood flow that limit maximal oxygen delivery to the myocardium. In settings of heightened demand for flow, as with physical exertion or emotional stress, the flow limitation can result in myocardial ischemia, a pathophysiological condition in which myocardial oxygen demand exceeds available supply. This condition is often accompanied by angina pectoris, a stereotyped chest discomfort that develops reproducibly when the inciting stress is incurred and is typically relieved within \(\leq\) 20 minutes of stress cessation, and is occasionally associated with symptoms such as dyspnea and lightheadedness. Angina is the most common clinical manifestation of CAD. Throughout the world, angina is a major cause of debility, particularly among men above 55 years and women above 65 years. Indeed, it is estimated that, in Europe and the United States, 30,000 to 40,000 persons per million suffer from angina.\(^1\)\(^-\)\(^3\) The association between exertional chest discomfort and CAD, and the association between CAD and “premature” death, are both relatively well recognized among nonphysicians. Consequently, angina is generally frightening for the patient. The combination of discomfort and fear associated with angina results in loss of productivity at the workplace and limitation of the capacity for leisure activity among afflicted individuals. Therefore, prevention of angina is a legitimate therapeutic objective, despite the fact that most patients with angina are at relatively low imminent risk of death or myocardial infarction from their underlying CAD.\(^1\)\(^-\)\(^4\)

Control of angina can be achieved either by masking the symptom with analgesia or by eradicating ischemia, the underlying cause. The former strategy may allow continuation of the inciting stress without the warning provided by the symptom, potentially leading to severe and irreversible myocardial damage. Thus, angina prevention can be achieved with acceptable safety only by preventing ischemia, a concept well recognized by governmental drug regulatory authorities, which require relief of ischemia as well as angina by therapeutics (drugs or devices) approved for marketing.\(^3\)

However, angina is not the only clinical sequel of coronary occlusion. If both sufficiently abrupt and sufficiently severe, occlusion can lead to myocardial infarction, arrhythmia, and sudden death. If myocardial injury during infarction involves an area of sufficient size, heart failure can ensue. This can occur acutely or can develop late after the acute event, when abnormal mechanical loading of the noninfarcted myocardium results in dysfunction of these areas, adding to the effect of the infarcted zone. Development of myocardial infarction and other sequelae of coronary occlusion involves a sudden reduction in flow

Keywords: coronary heart disease; heart rate; \(I_f\) current; ivabradine

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through an artery that may already be occluded sufficiently to cause angina with effort. Therefore, drugs that simply reduce ischemia may be sufficient to prevent angina, but may or may not impact significantly on the risks of infarction, subsequent heart failure, or sudden death. As noted later in this article, heart rate slowing may be uniquely suited to mitigate not only angina, but also these other sequelae of CAD.

**PATHOPHYSIOLOGY OF MYOCARDIAL ISCHEMIA: THE ROLE OF HEART RATE**

Heart rate is a primary and direct determinant of myocardial oxygen demand. Thus, its modulation is a reasonable strategy for angina prevention. However, heart rate can mitigate ischemia by enhancing myocardial oxygen supply as well as by reducing demand (Figure 1).

Heart rate determines the duration of diastole, the portion of the cardiac cycle during which most coronary flow occurs. As diastolic duration increases with constant perfusion pressure, coronary flow increases. Moreover, diastolic duration and heart rate are not related linearly: relatively small decreases in heart rate result in proportionally greater increases in diastolic duration and flow. Some coronary flow occurs during systole. However, because of the anatomy and conduction sequence of the left ventricle, intramyocardial systolic pressure is particularly high in the subendocardial region, virtually precluding flow throughout all of systole and rendering the subendocardium particularly vulnerable to ischemia when epicardial coronary artery obstruction is present. Thus, decreases in heart rate, increasing diastolic duration, are particularly advantageous for enhancing subendocardial flow.

Given the reduction in myocardial oxygen demand and increase in supply associated with heart rate slowing, it is not surprising that the development of β-blocking agents for angina prevention was undertaken largely because of the heart rate-slowing capacity of these drugs. Electrical stimulation of the carotid sinus, developed almost 40 years ago for angina treatment, is based on the same concept. However, both these modalities, as well as calcium channel-blocking drugs with heart rate-slowing capacity, have additional direct hemodynamic and/or negative inotropic effects that may also be important in ischemia prevention and may directly affect heart rhythm and heart failure development, or may have other adverse consequences. Thus, assessment of the therapeutic effects of isolated heart rate slowing requires a therapy highly specific for this action, with no other potentially confounding actions. Moreover, to define the role of pure heart rate slowing for ischemic heart disease, it is necessary to determine not only the capacity for angina prevention, but also the impact on acute myocardial infarction, heart failure, and sudden death.

**I$_f$ INHIBITION AND HEART RATE SLOWING: EXPERIMENTAL CONSIDERATIONS**

A quarter century ago, the I$_f$ current was discovered and was recognized as a central physiological determinant of heart rate. Thus, inhibition of this cyclic-nucleotide-gated, voltage-dependent, mixed sodium-potassium current is a legitimate basis for ischemia prevention. More than a decade elapsed from discovery of the current to synthesis of a molecule, ivabradine, capable of achieving sufficient I$_f$ current inhibition to prevent angina by heart rate slowing, while devoid of unacceptable adverse effects at antianginal doses. Preclinical testing to demonstrate potential benefits and to identify possible risks, followed by clinical testing to demonstrate...
efficacy and acceptable safety for angina prevention, required more than another decade. This work culminated in 2005 with regulatory authority approval of ivabradine by the European Medicines Evaluation Agency (EMEA) as the first \( I_{	ext{h}} \) current inhibitor marketable for angina prevention. Preclinical evaluation and early clinical studies suggest that pure heart rate slowing with this drug may be beneficial in preventing other sequelae of coronary occlusion as well, and, particularly, that pure heart rate slowing with ivabradine is devoid of negative inotropic effects.\(^{10-13}\) As yet, however, the benefits expected as a result of these findings have not been proven by adequate and well-controlled clinical trials.

The following section reviews the clinical evidence that pure heart rate slowing as demonstrated with the \( I_{	ext{h}} \) current inhibitor ivabradine is effective in angina prevention.

**\( I_{	ext{h}} \) INHIBITION AND HEART RATE SLOWING: CLINICAL TRIALS FOR ANGINA PREVENTION**

Controlled clinical trials and associated studies to evaluate the benefit of ivabradine for angina prevention have involved almost 5000 patients with CAD and chronic stable angina. Trials generally continued active treatment for at least 3 months, ie, long enough to allow emergence of drug effects and to evaluate the likelihood of effect persistence during chronic therapy, based on empirical evidence with drugs indicated for prevention of angina. No other antianginal has been studied so intensively. The drug has been assessed as monotherapy (ie, compared with placebo in the absence of any background therapy); it has been compared with placebo on a background of amlodipine, the calcium channel blocker most commonly used for angina prevention in Europe, and it has been compared directly with atenolol, the most commonly employed \( \beta \)-blocker in Europe. These trials have resulted in considerable information about the efficacy of ivabradine and, together with additional observational and comparative experience, have also provided data about the drug’s safety and benefit-to-risk relationship.

**Ivabradine as monotherapy**

Within the regulatory and research communities, the primary evidentiary standard for antianginal efficacy is improvement in exercise tolerance on standard treadmill or bicycle ergometric testing.\(^{5,14,15}\) Reduction in exercise-inducible symptoms must be supplemented by reduction in associated ischemia to demonstrate that treatment is not “masking” angina by an analgesic effect, which might allow patients to exercise to severe and potentially lethal ischemia without the warning of symptoms. Angina frequency in daily living, recorded in diaries, is considered adjunctive evidence, but is not accepted as primary evidence of drug efficacy because the intensity of stress inciting angina in daily living cannot be determined from diary reports. Drug effectiveness needs to be demonstrated at the end of the prescribed interdose interval (“trough”), though the effect also should be demonstrated at the time of maximal drug effect (“peak”).

The first large trial to assess pure heart rate slowing with ivabradine, carried out according to the principles noted above, involved 360 patients from multiple centers throughout several European nations.\(^{16}\) Patients had documented evidence of coronary occlusive disease, ambient angina, and \( \geq 0.1 \text{ mV} \) (ie, \( \geq 1 \text{ mm} \)) downsloping or horizontal electrocardiographic ST-segment depression induced by upright bicycle ergometry. These patients were randomly assigned to either placebo or to ivabradine 2.5 mg orally twice daily, 5.0 mg twice daily, or 10 mg twice daily, using a double-blind, parallel design (Figure 2, next page).\(^{16}\) The initial treatment assignment was continued for 2 weeks (“dose ranging”) after which, during an open-label extension, all patients were invited to use ivabradine 10 mg twice daily. Inclusion in this extension was voluntary and depended in part on appropriately permissive local regulations. After 2 to 3 months on the open-label regimen, patients were randomized to continue ivabradine 10 mg twice daily or to be withdrawn to placebo, under double-blind conditions. After 1 week of double-blind withdrawal, a final exercise tolerance (ETT) assessment was performed.

Of the 360 patients initially included (intention to treat [ITT] population), voluntary withdrawals or protocol violations affected 103 patients, leaving 257 patients treated according to protocol. These patients were included in the primary (per protocol [PP]) analysis. With all doses of ivabradine, time to limiting angina was nominally greater than with placebo (Figure 3, page 25).\(^{17}\) This difference reached statistical significance when ivabradine 10 mg twice daily was compared with placebo.\(^{16}\) In addition, a dose-effect relationship was apparent when all doses were considered, and a between-group comparison was statistically significant (\( P=0.049 \)). For the ITT population, in which protocol violators were included, ivabradine 10 mg bid also was superior to placebo at trough, but the difference was minimally outside the statistically significant range, reaching the level of “statistical trend” (\( P=0.058 \)).
Ivabradine 10 mg bid was administered open-label to 173 study participants. Compared with prior, blinded, regimens, those who increased to 10 mg bid nominally increased time to limiting angina in bicycle ergometry, while no change was apparent in the group already receiving 10 mg bid. However, among those who then were withdrawn to placebo, time to limiting angina fell significantly, but remained unchanged in patients maintained on ivabradine 10 mg bid (between-group difference: \(P=0.018\)). Consistent with an anti-ischemic as well as an antianginal effect, time to 1-mm ST-segment depression was significantly reduced by ivabradine 5 mg bid and 10 mg bid, a significant dose-response relationship was seen across all doses for this effect. Diary recordings and tabulation of

**Figure 2.** Protocol and patient distribution through all 4 phases of the first large placebo-controlled trial of ivabradine as monotherapy for angina prevention.

**Abbreviations:** AE, adverse event; CAD, coronary artery disease; ETT, exercise tolerance test; PP, per-protocol population.

short-acting nitroglycerin use confirmed the antianginal effect by revealing a significant reduction in angina attack rate and nitroglycerin use during routine daily living at the end of the protocol compared with pretreatment among patients receiving ivabradine. At the conclusion of the randomized withdrawal, ambient angina increased among those randomly withdrawn to placebo, but was unchanged among those who continued on ivabradine 10 mg twice daily.

This trial also indicated the relative safety of ivabradine use during a 3-month treatment interval. Adverse events were relatively few and generally similar in frequency and distribution compared with placebo, except for relatively minor visual symptoms. Visual symptoms were dose-related, were rarely sufficiently bothersome to cause voluntary withdrawal from the drug, and were invariably reversible during treatment or with drug cessation, consistent with the absence of irreversible retinal effects reported in preclinical studies. Visual side effects occurred in approximately 15% at 10 mg twice daily during the dose-ranging phase and 18% during the open-label phase. (The visual effects are presumably due to the similarity heart rate slowing with ivabradine (a problem theoretically associated with this pharmacological effect). Note, of course, that ivabradine would be ineffective in patients with chronic atrial fibrillation, since the drug does not importantly affect the atroventricular node and, therefore, cannot modulate heart rate in these patients.

**Direct comparisons with approved antianginal drugs at commonly employed doses**

These comparisons were designed to test the “noninferiority” of ivabradine versus standard antianginal therapies. Optimally, noninferiority trials are constructed to demonstrate that a certain portion of the effect of an established drug is likely to be retained by a new agent in settings in which placebo cannot be employed. The requisite portion retained, conventionally accepted as at least 50% of the effect of the comparator, is believed to indicate the likelihood both of “clinical utility” and of equivalence, within an acceptable uncertainty based on the variability of outcome measures in clinical trials. For this concept to be rigorously tested, the effect size of the established active comparator must be precisely defined from earlier placebo-controlled trials. Unfortunately, such information is seldom available for antianginal drugs; historically, these have been studied in relatively small trials, resulting in relatively unstable point estimates of absolute effect. (Also, of course, the use of historical placebo-controlled data assumes the persistence of the background conditions in place at the time of the earlier trials, an assumption that may not be correct.) In the absence of precise historical information, other approaches, fundamentally based on the consensus of experts in the field, are employed to define the

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**Figure 3. Effect of ivabradine, at trough of drug activity, on time to angina sufficient to limit continued bicycle exercise among 257 protocol-compliant patients with coronary artery disease. Measurements were obtained after 2 weeks of double-blind randomized therapy. A dose-response relation is apparent and the dose of 10 mg bid individually is significantly more effective than placebo.**

difference in outcome between new agent and established comparator that can be inferred to indicate that the effect of the new agent is greater than that of the placebo and not inferior to that of the established comparator. The latter approach was employed in defining the “margins” for declaring noninferiority between ivabradine and atenolol and between ivabradine and amlodipine.

In 1195 patients randomized to ivabradine 7.5 mg twice daily or amlodipine 10 mg daily, a 3-month multicenter, multinational, double-blind study demonstrated that ivabradine was indistinguishable from amlodipine in its effects on total exercise duration to angina onset, time to 1-mm ST-segment depression, and time to angina onset, and time to 1-mm ST-segment depression. Formal statistical testing of data from this as yet unpublished trial revealed ivabradine to be noninferior to amlodipine (P<0.0001 for this conclusion).

In a 4-month double-blind study, 939 patients were randomized, first, to ivabradine 5 mg twice daily or atenolol 50 mg daily for 2 weeks; then, doses were uptitrated to ivabradine 7.5 mg or 10 mg twice daily or to atenolol 100 mg daily. No statistically significant differences were found when various outcomes were compared among the tested regimens at each stage, and ivabradine’s noninferiority to atenolol was significantly established at the doses employed (P<0.001). However, despite the fact that atenolol 100 mg daily resulted in slightly greater heart rate reduction than with either ivabradine 7.5 mg or 10 mg twice daily, ivabradine was nominally superior to atenolol in enhancing time to angina onset, time to limiting angina, and total exercise duration at all doses, while time to 1-mm ST-segment depression was virtually identical among the regimens.

This finding suggests that pharmacological effects of β-blockers other than heart rate slowing may impact negatively on the pathophysiology of angina. This hypothesis remains to be rigorously tested. Importantly, this trial also demonstrated that unacceptable fatigability was more common with atenolol than with ivabradine.

Finally, ivabradine was studied in a randomized, blinded modified dose-response trial comparing ivabradine 5 mg twice daily versus ivabradine 7.5 mg twice daily in 386 patients for 1 year. At the end of study, both doses were associated with a substantial reduction in resting heart rate compared with pretreatment measures, consistent in magnitude with that found in earlier studies. Also, compared with the pretreatment interval, significant reductions in angina attack rate were associated with each dose. Visual symptoms, previously described, were present, but led to treatment cessation in only 1% of patients. The QT interval was rigorously assessed in this study and, when corrected for heart rate, was not altered by ivabradine administration, nor were other ECG abnormalities noted.

**Placebo-controlled therapy with amlodipine background**

Among 728 patients receiving amlodipine 10 mg daily throughout the study, ivabradine (5 mg twice daily or 7.5 mg twice daily) was compared with placebo during a 3-month interval in a randomized, double-blind multicenter, multinational trial. These data are not yet published and, therefore, cannot be presented in detail. Among patients receiving 5 mg twice daily for both doses at peak drug effect, no statistically significant superiority of ivabradine versus placebo was found during a 3-month interval in a randomized, double-blind multicenter, multinational trial. These data are not yet published and, therefore, cannot be presented in detail; preliminary analysis revealed a statistically significant superiority of ivabradine versus placebo for both doses at peak drug effect, with nominal superiority at trough.

**CONCLUSIONS**

A growing body of data indicates that angina and the underlying ischemia can be effectively minimized using selective sinoatrial node I current inhibition to slow heart rate. These data also suggest that, when this effect is achieved with ivabradine, effective prevention of angina is associated with acceptable safety and tolerability.

Because of the relatively high prevalence of angina, the availability of a new approach to angina prevention has important public health implications. Currently available drug therapies with single or multiple agents are often inadequate to provide complete or acceptable angina prevention. According to one recent study, most patients with angina receive combination antianginal therapy involving at least two drugs, but, nonetheless, continue to experience approximately two angina attacks per week. A new agent, with pharmacological effects different from currently available drugs, may enhance the rate of acceptable angina relief. Also, since drugs with different pharmacological profiles are likely to differ in frequency, character, and distribution of adverse effects, tolerability of treatment across large populations is likely to be enhanced by the availability of a new drug that may be better accepted by some patients for whom other agents are associated with unacceptable adverse effects. For example, β-blockers may increase the symptoms of peripheral arterial occlusive disease (commonly associated with peripheral arterial occlusive disease) or obstructive pulmonary diseases and can increase the risk of hypotension or symptomatic conduction block in patients with intrinsic atrioventricular node disease. Management of diabetes mellitus or hyperlipidemia can be confounded by
β-blockade Certain calcium channel blockers\textsuperscript{27} can precipitate or potentiate heart failure or atrioventricular node dysfunction. Calcium channel blockers (particularly the dihydropyridines) also frequently cause unacceptable peripheral edema, while all calcium channel blockers often are associated with constipation. Long-acting nitrates\textsuperscript{28} can cause severe headaches or can result in lightheadedness or even syncope (direct results of what are presumed to be its beneficial pharmacological effects); intermittent use of nitrates may result in rebound angina and vasoconstriction. These adverse effects are not expected with I\textsubscript{f} current inhibition and have not been associated with ivabradine use. Moreover, as inferable from preclinical studies (and as yet unpublished experience in patients with heart failure and with left ventricular ejection fraction <40%), I\textsubscript{f} current inhibition with ivabradine does not reduce myocardial inotropy\textsuperscript{29,30} (sometimes a problem with β-blockers\textsuperscript{31}) and does not result in potentially lethal "rebound" effects (reported with abrupt cessation of β-blockers\textsuperscript{22,23}). Also, from results of the randomized withdrawal after 2 to 3 months of treatment during the placebo-controlled trial of ivabradine as monotherapy, described above, it can be confidently inferred that pharmacological tolerance to its therapeutic effects does not occur when ivabradine is administered chronically. Pharmacological tolerance was identified as an important problem when long-acting nitrates are employed without drug-free intervals.\textsuperscript{32}

Finally, in addition to symptom reduction and consequent quality of life enhancement that can be expected from ivabradine-mediated I\textsubscript{f} current inhibition, it is possible that heart rate reduction may improve survival. Though the latter hypothesis remains to be tested in a large clinical trial, clinical observations suggest at least one basis for such a benefit. Specifically, heart rate slowing appears to minimize the likelihood of disruption of atherosclerotic plaques in the coronary arteries.\textsuperscript{33} The pathophysiology presumed to underlie most myocardial infarctions. The putative mechanism is reduction in mechanical perturbation of the plaque caused by foreshortening and twisting of large epicardial arteries during systole. Perhaps more importantly, when results of multiple clinical trials for survival among patients with heart failure are considered as a group, drug-induced heart rate changes have been inversely related to survival: survival has improved in proportion to the extent of heart rate slowing and survival has worsened when drugs have been associated with increases in heart rate.\textsuperscript{34} Similarly, in trials of β-blockers administered after myocardial infarction, reduction in mortality has been directly proportional to the extent of heart rate slowing.\textsuperscript{35} In a recent late follow-up of patients entered into the Coronary Artery Surgery Study (CASS) registry (and, thus, with angiographically-proven CAD), survival among those whose heart rate at entry was <62 beats per minute was 30% greater than survival of those whose entry heart rate exceeded 83 beats/min\textsuperscript{16}; by Cox model analysis, the effect of heart rate was independent of other risk factors. Other, earlier epidemiological and actuarial studies involving more than 150 000 free-living persons without specified diseases also reveal a significant relation between casually measured heart rate and survival.\textsuperscript{37} The intriguing potential for survival enhancement with ivabradine in patients with CAD and, indeed, in other settings as well, remains to be explored.

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Can $I_f$ inhibition help in congestive heart failure?

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Ivabradine is a selective inhibitor of the $I_f$ pacemaker current, and as such decreases heart rate without inducing any negative inotropic effects. Experimental and clinical evidence suggests that the beneficial effects of β-blockers in heart failure are mediated by a reduction in heart rate. Long-term heart rate reduction with ivabradine in a rat model of heart failure elicited an improvement in left ventricular function and a positive effect on cardiac remodeling, leading to a decrease in collagen density and an increase in capillary density. In addition, recent evidence indicates that the $I_f$ current may be reexpressed in animal and human ventricular myocytes from failing hearts and may have an arrhythmogenic role. $I_f$ inhibition would thus exert an antiarrhythmic effect in heart failure, but this hypothesis remains to be proven in the clinical setting.

**Keywords:** heart failure; heart rate; $I_f$ inhibition; electrophysiology; ventricular myocyte; electrophysiological remodeling; ivabradine

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The importance of heart rate reduction per se was evaluated in post-hoc analyses of the major clinical trials testing the effects of β-blockers in heart failure. In the MERIT-HF, two dosage subgroups were compared, one receiving more than 100 mg metoprolol CR/X twice daily (n=1202) and the other receiving 100 mg or less (n=412) during the first 3 months of follow-up. Heart rate was reduced to a similar degree in both groups. The reduction in total mortality with metoprolol CR/XL versus placebo was similar in both groups (38%). This may indicate a higher sensitivity to β-blockade in the low-dose group, but it may also suggest that the main benefit of β-blockade resulted from its effect on heart rate.

The Second Cardiac Insufficiency Bisoprolol Study (CIBIS II) evaluated the relationship between baseline heart rate changes 2 months after the beginning of treatment and outcomes (mortality and hospitalization for heart failure). Multivariate analyses showed that, in addition to β-blocker treatment, both baseline heart rate and changes at 2 months significantly correlated with survival and hospitalization for worsening of heart failure. Furthermore, the lowest baseline heart rate and the greatest heart rate reduction were associated with better survival and a greater reduction in hospital admissions. These results were interpreted as confirming the hypothesis that heart rate reduction per se in patients with heart failure is associated with improved survival. This result is possibly the consequence of the induced reduction in ischemia, which is present to different degrees in dilated ventricles even in the absence of coronary artery disease. The benefit of bisoprolol was not significant in patients with atrial fibrillation. It was observed that among these patients, those treated with β-blockers and who subsequently died, had a more important decrease in blood pressure, suggesting that the marked decrease in blood pressure induced by β-blocker treatment may be deleterious. This effect is not expected with drugs inhibiting the \( K_C \) channels.

Conflicting results have been reported in studies investigating the effects of carvedilol in heart failure patients. Post-hoc stratification of patients in the US–Carvedilol heart failure program into groups above and below the mean baseline heart rate showed that the mortality benefit of carvedilol compared with placebo was only significant in the group with a high baseline heart rate. Others have not found such a relationship. In a retrospective, observational, single-center study aimed at determining whether the process of reverse remodeling in response to carvedilol in heart failure patients was dependent on baseline heart rate and on β-blockade–induced heart rate reduction, no relationship was found between left ventricular size and left ventricular function and heart rate, either at baseline or after β-blockade. This suggests that the remodeling process may also be controlled by mechanisms other than heart rate.

Recent experimental and clinical findings consistently indicate that the heart rate reduction induced by ivabradine may be beneficial in patients with left ventricular dysfunction. The effects of long-term heart rate reduction on left ventricular function and remodeling was investigated in a rat model of heart failure. The animals randomized to ivabradine, exhibited long-term (90 days) heart rate reduction. In these animals, treatment with ivabradine...
β-blockers are contraindicated in blind placebo-controlled clinical trials. In a 3-month randomized double-blind clinical trial, the relative hypoxia in the subendocardial layers can also induce endothelial dysfunction, triggering the release of cytokines and free radicals. Moreover, elevated heart rate shortens the diastolic intervals during which the coronary blood flow perfuses the myocardium and provides the myocytes with nutrients and oxygen and carries away the terminal products of cellular metabolism. Relative hypoxia in the subendocardial layers can also induce endothelial dysfunction, triggering the release of cytokines and free radicals.

In a 3-month randomized double-blind placebo-controlled clinical study, ivabradine was administered at a dosage of 10 mg twice daily on top of conventional therapy (except β-blockers) to 56 patients with coronary artery disease and mild-to-moderate congestive heart failure. Patients with the highest degree of left ventricular systolic dysfunction (echocardiographic left ventricular ejection fraction [EF] <35%) demonstrated a mean 5% increase in EF with ivabradine, compared with a mean 0.5% decrease in patients receiving placebo.

Several data support ivabradine’s safety in the elderly population. A pharmacokinetic analysis performed in elderly patients showed that there were no differences in pharmacokinetics (AUC and Cmax) between elderly (>75 years) and very elderly patients (>85 years) and the overall population. In contrast, β-blockers are contraindicated in patients with obstructive lung disease, decompensated conditions, hypotension, and atrioventricular conduction disturbances, as listed by a retrospective study assessing β-blocker utilization patterns in clinical practice setting. Similar findings were observed in a nationwide observational study performed in Italy. Moreover, in patients in whom β-blocker therapy needed to be uptitrated to the maximum tolerated dose, the dose administered was frequently lower than the recommended dose. β-Blocker use may also be hampered by the occurrence of fatigue, hypotension, dizziness, and dyspnea, requiring discontinuation of treatment. Thus, though β-blockers are very effective in heart failure patients, a number of contraindications and poor tolerability may in practice restrict their use at the full dosage proven to be effective in clinical trials.

Ivabradine can be combined with all the drugs currently recommended for use in heart failure patients without any risk of untoward drug interactions developing. Thus, no kinetic or dynamic interactions with ivabradine were observed in patients of pivotal phase 3 studies receiving angiotensin-converting enzyme inhibitors, angiotensin II receptor antagonists, diuretics, aspirin, digoxin, amiodarone, or cholesterol-lowering agents. No clinically significant changes in atrioventricular conduction, QT interval, and myocardial contractility have been reported. Therefore, combination of ivabradine and β-blockers or other heart rate-lowering agents does not pose specific problems. Thus, it appears that patients with coronary artery disease and left ventricular systolic dysfunction may benefit from heart rate reduction with ivabradine whatever the background therapy, including or not β-blockers. Ivabradine is currently being tested in a large international phase 3 trial—the MorBidity-mortality EvAlUaTion of the If inhibitor ivabradine in patients with coronary disease and left ventricular dysfunction (BEAUTY).

**POTENTIAL CLINICAL EFFECTS OF IF INHIBITION IN NONPACEMAKER CARDIAC CELLS**

In addition to the benefit of If inhibition in regard of the pacemaker cells of the atrioventricular node, discussed above, If inhibition may also be of benefit in heart failure through a different mechanism of action, by acting on the reexpression of the If current in the ventricular cells of the failing heart.

Whereas the role of If in the generation of spontaneous sinus node activity and in the control of heart rate is widely documented and has been known for a long time, the presence of If in ventricular myocytes is a more recent and quite intriguing finding. First of all, it must be stressed that the presence of If in ventricular myocytes relates to pathophysiology rather than physiology. The first recording of If in ventricular cardiomyocytes was reported in 1993 in the normal guinea pig. The characteristics of this If current were such that they ruled out any possible physiological role in the ventricle, because it was much smaller than the If current recorded in pacemaker cells, and was activated at negative voltages far below the values of normal resting potentials, (ie, more negative than -100 mV). However, in the setting of heart disease, the situation is very different. This is because, in various animal models of cardiac hypertrophy and failure, If has been shown to be upregulated, to the extent that it may assume a functional role. It was found that during the recording of the intracel-
lular electrical activity of papillary muscles isolated from 18-month-old spontaneously hypertensive rats (SHR), ie, rats with a severe cardiac hypertrophy, the diastolic phase was not flat but a “sort” of diastolic depolarization could be detected between two driven action potentials. The papillary muscles isolated from old SHR were also particularly sensitive to the arrhythmic action of isoprenaline, a β-adrenergic agonist. It seemed obvious to conclude that one of the reasons for this enhanced susceptibility to the arrhythmic action of the catecholamines could be due to the “unusual” presence of that “unusual” diastolic depolarization. The presence of If was subsequently documented in ventricular myocytes isolated from the heart of animals with different degrees of cardiac hypertrophy. The degree of hypertrophy was positively correlated with an increased If density, and changes in expression levels were most pronounced in those cardiac regions with the highest overload, indicating that the processes leading to hypertrophy directly affected the level of hyperpolarization-activated cyclic nucleotide-gated cation channel (HCN) expression, which are the molecular components of native f-channels. Four HCN genes were identified, encoding different proteins, which assemble to form tetrameric (homo- or, likely, heteromeric) compounds.

Figure 2A shows If current recordings obtained in ventricular myocytes isolated from normal human heart (human donor heart not used for transplantation because of technical problems) and in myocytes isolated from a human heart explanted because of terminal failure. Figure 2B summarizes the relative increase in “current density,” comparing diseased vs control ventricular cardiomyocytes, in several rat models of cardiomyopathies and in human ischemic and dilated cardiomyopathies. “Current density” designates a current amplitude that is normalized with respect to cell size: since the cardiomyocytes are “hypertrophic,” ie, enlarged, in cardiomyopathies, it is absolutely necessary to correct the current amplitude for cell dimensions. If density is markedly increased in left ventricular cardiomyocytes from rats with moderate or severe cardiac hypertrophy (LVH) caused by pressure overload (PO), and is even greater in rats with overt heart failure (HF) consequent to high blood pressure (PO) or post–myocardial infarction (PMI). In addition to electrophysiological data, molecular biology techniques allowed to demonstrate a parallel upregulation of the HCN2 and HCN4 mRNA levels, which are the predominant isoforms underlying ventricular If.
that mislocalized expression and/or overexpression of cardiac HCN channels may represent an example of a general phenomenon, called cardiac remodeling, which consists namely in the reexpression of fetal proteins: in fact, f-channels are present in fetal or neonatal ventricular myocytes, which lose their capacities of generating spontaneous activity during electrophysiological maturation toward adult phenotype. However, from a clinical point of view, the most interesting aspect of this phenomenon is that $I_h$ may represent an arrhythmogenic mechanism in heart failure, which is a condition associated with high risk for sudden cardiac death. Further support for this hypothesis comes from the evidence of $I_h$ expression in the failing human ventricle from explanted hearts. Correlation with the severity of cardiac disease was obviously impossible in this setting, as all patients had terminal heart failure. However, a fascinating finding was that changes in $I_h$ density correlated with the etiology of the disease, and, for example, $I_h$ overexpression was greater in ischemic cardiomyopathy (ICM) than in idiopathic dilated cardiomyopathy (DCM) (Figure 2). Recent molecular findings have shown that HCN2 and HCN4 expression at both mRNA and protein levels is markedly increased in samples obtained from hearts explanted from patients with ICM, providing a molecular explanation for the functional upregulation of $I_h$ in heart failure. The signaling pathways leading to enhancement of $I_h$ in nonautomatic regions of the heart are not fully clarified, but the renin-angiotensin system seems to play a pivotal role. Overall, these studies represent the first evidence in favor of an altered $I_h$ expression in cardiac diseases and suggest its potential role in the associated abnormal electrical activity.

CONCLUSIONS

The molecular identification of HCN subunits and their functional/molecular detection in the diseased ventricle have resulted in an unforeseeable surge of interest for pacemaker channels. Based on these new findings, it appears that selective f-channel inhibitors such as ivabradine, in addition to selective heart rate reduction (which in itself may be beneficial in patients with left ventricular dysfunction and heart failure), may also block the $I_h$ current expressed in ventricular myocytes of the failing heart (where it may represent an arrhythmogenic trigger), and thus reduce the risk of sudden death—an exciting prospect that still awaits clinical confirmation.

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Can if inhibition help in congestive heart failure? - Tavazzi and Mugelli


Can $I_f$ inhibition help after myocardial infarction?

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Ivabradine is a selective and specific $I_f$ channel inhibitor with proven antianginal and anti-ischemic properties, but without effects on blood pressure, myocardial contractility, and atrioventricular (AV) node conduction. These advantages place ivabradine in a unique position to reduce heart rate in unstable patients. Contraindications to the use of $\beta$-blockers in patients with acute MI represent additional potential indications for ivabradine. Moreover, ivabradine improves myocardial stunning following ischemia better than $\beta$-blockade. Ivabradine may also be helpful in patients with residual angina or ischemia in whom $\beta$-blockers should be avoided and in those not tolerating them well. In the longer-term follow-up, evidence suggests that reducing heart rate could prevent progression of atherosclerosis and plaque rupture. Ivabradine may therefore become very helpful in the management of patients after myocardial infarction.

**HEART RATE, MYOCARDIAL INFARCTION, AND MORTALITY**

Many large observational studies have shown a link between increased heart rate and all-cause mortality and cardiovascular events in patients with hypertension, with metabolic syndrome, and in the elderly.\(^1\)\(^3\) Recently, we have assessed the relationship between resting heart rate and future cardiovascular events in a population of 25,000 patients with suspected or proven coronary artery disease (CAD).\(^4\) Over a median follow-up of 14.7 years, resting heart rate was a predictor of overall and cardiovascular mortality, independent of other known risk factors such as hypertension, diabetes, and smoking. The size of the study also allowed adjustment of the multivariable model for the extent of CAD and left ventricular ejection fraction. Resting heart rate was an independent risk factor for total and cardiovascular mortality, even after adjusting for such covariates. A high resting heart rate (283 bpm) was indeed a strong predictor of total and cardiovascular mortality (hazard ratios of 1.32 and 1.31, respectively). In addition, resting heart rate was a risk factor for time to cardiovascular hospitalization.

Several studies conducted before the thrombolysis era have demonstrated that a high heart rate during hospitalization increases overall mortality up to 2 years after myocardial infarction.\(^5\) In patients with no or mild heart failure, mortality was more than doubled ($P<0.001$) for patients with heart rate $>90$/min versus those with $<90$/min. Disegni et al.\(^6\) reported similar results in 1044 patients who survived an acute myocardial infarction. Overall mortality was 4.3%, 8.7%, 11.8% for patients with discharge resting heart rate $<70$/min, 70 to 90/min, $>90$/min, respectively. For patients without clinical signs of heart failure during hospitalization, 1-year mortality was...
2.8%, 5.1%, 6.6% for patients with discharge resting heart rate <70/min, 70 to 90/min, >90/min, respectively. A retrospective analysis of data from the Gruppo Italiano per lo Studio della Sopravvivenza nell’Infarto Miocardico–2 (GISSI-2) study in patients thrombolysed for acute myocardial infarction was also published. The overall 6-month mortality was 0.8% for those with a discharge heart rate below 60 beats/min and 14.3% for those with a heart rate above 100 beats/min. Resting heart rate remained an independent prognostic factor in a multivariable analysis.

These data demonstrate a strong link between heart rate and cardiovascular morbidity and mortality. In addition, whereas agents that induce tachycardia have been found to have neutral or deleterious effects on mortality, several of those that reduce heart rate have been shown to improve survival. The relationship between reduction in heart rate and decrease in mortality has indeed been well established with β-blockers after myocardial infarction and in patients with heart failure.

**I<sub>1</sub> CHANNEL INHIBITION DURING IN-HOSPITAL COURSE AFTER MYOCARDIAL INFARCTION**

Patients with acute myocardial infarction generally benefit from heart rate reduction both through a decrease in myocardial oxygen requirement and a shortening in the duration of diastole and myocardial perfusion. However, the negative inotropic and hypotensive effects of β-blockers contraindicate their use in patients with pulmonary congestion, borderline blood pressure, overt pulmonary edema, or cardiogenic shock. Ischemic left ventricular dysfunction is also often accompanied by diastolic dysfunction. Because excessive tachycardia has deleterious consequences on diastolic function, heart rate reduction is important to achieve. However, the negative lusitropic effects of β-blockers may represent another disadvantage in this setting. As previously explained in this issue, ivabradine provides selective, reversible use-dependent channels inhibition. The I<sub>1</sub> current located in the sinus node has a major influence on slow diastolic depolarization. Ivabradine therefore offers pure heart rate reduction without affecting hemodynamic parameters such as ejection fraction, blood pressure, contractility, atrioventricular conduction, or coronary vaso-motion. The absence of effects on myocardial contractility and blood pressure places ivabradine in a unique position to control heart rate in unstable patients.

Although β-blockers have several side effects, bronchospasm and atrioventricular (AV) block constitute the most relevant adverse reactions that limit their use in the emergency setting. While asthma or chronic obstructive pulmonary disease (COPD) represent only relative contraindications to β-blockade, some patients clearly develop bronchospasm and wheezing with β-blockers, which require dose reduction or abrupt withdrawal. Such patients who require heart rate reduction would clearly benefit from the lack of this side effect with ivabradine.

**IVABRADINE POST-DISCHARGE AFTER MYOCARDIAL INFARCTION**

The role of ivabradine in the treatment of patients discharged following a myocardial infarction must take into account several considerations: (i) the prognostic value of resting heart rate and the relationship between heart rate reduction and decrease in mortality after myocardial infarction shown with β-blockers; (ii) several patients have residual angina and/or ischemia after myocardial infarction even if they undergo revascularization; (iii) side effects such as fatigue, depression, and sexual dysfunction may limit long-term compliance to β-blockers; and (iv) ivabradine has shown antianginal and anti-ischemic properties that are not inferior to those of the β-blocker atenolol.

It is particularly relevant to note that more than half of the 939 patients involved in the InternaTional Trial of the Antianginal effects of Iivaradine compared to atenolol (INITIA-TIVE) (see other article in this issue) had previously suffered a myocardial infarction.

Ivabradine shares with β-blockers the property of decreasing heart rate and oxygen demand from the ischemic heart, which is presumably fundamentally important in mediating anti-ischemic effects. In addition, Colin et al compared the effects of ivabradine, atenolol, and placebo on oxygen consumption and supply during exercise in dogs. For a given heart rate, left ventricular ejection time was longer with atenolol than with ivabradine because of the negative inotropic effects of atenolol. As a consequence, the increase in diastolic time and coronary blood flow was greater with ivabradine than with atenolol (Figures 1 and 2, next page). Furthermore, in contrast to β-blockers, ivabradine has also been shown not to limit the decrease in coronary resistance induced by exercise. Given the absence of cardiac effects other than exclusive heart rate lowering, ivabradine is suitable after myocardial infarction for patients with residual angina or ischemia in whom β-blockers should be avoided (those with AV block, peripheral vascular disease, and COPD).
for those not tolerating well β-blockers or calcium antagonists. Unlike β-blockers, ivabradine may be used in vasospastic angina because it does not increase coronary vaso-motor tone.

Ivabradine may therefore be very useful in such patients. The fact that asthma and COPD represent relative contraindications to β-blockade has been mentioned previously. However, some patients with both CAD and COPD develop angina when treated with inhaled β-adrenergic agonists because of the resulting tachycardia. The heart rate reduction obtained with ivabradine could be very helpful in this setting. Patients can also have variable degrees of AV block after myocardial infarction that develop or are exacerbated with β-blockers. The need for selective heart rate reduction in patients with myocardial ischemia and AV-node conduction abnormalities represents another excellent indication for ivabradine. This is particularly relevant for older patients with a prolonged PR interval.

**CURRENT INHIBITION AND LIMITATION OF MYOCARDIAL STUNNING**

Postischemic myocardial dysfunction, or myocardial stunning, can result in systolic and/or diastolic ventricular dysfunction in different settings, such as at the time of reperfusion after myocardial infarction, but also after periods of ischemia in patients with chronic angina. Monnet et al.13 demonstrated the benefit of ivabradine on myocardial stunning in dogs compared with atenolol and placebo. Ivabradine is better than placebo and atenolol for time to recovery of myocardial contractility after exercise-induced stunning, and this holds true whether the drugs were infused before or after exercise (Figure 3).13 Under atrial pacing, the negative effects of atenolol on systolic function during stunning were amplified (Figure 4).13 These results demonstrate that ivabradine enhances recovery of myocardial stunning after exercise-induced ischemia in dogs.
whereas atenolol has deleterious effects, explained by its negative inotropic properties. Thus, ivabradine has the potential to improve myocardial stunning in patients with ischemic heart disease.

HEART RATE ANDATHEROSCLEROSIS PROGRESSION: A TARGET FOR $I_f$ INHIBITION?

Secondary prevention is central to the therapeutic strategy after discharge following myocardial infarction. Prevention of atherosclerosis progression and stabilization of plaque are indeed of paramount importance to prevent recurrent clinical events in the long-term follow-up. Heart rate is significantly correlated with the severity and progression of atherosclerosis on coronary angiography among men who had developed myocardial infarction at a young age. Experimental data have also demonstrated that a reduction in heart rate can delay the progression of coronary atherosclerosis in monkeys. Beere et al showed that male cynomolgus monkeys subjected to sinus node ablation or those with innately low heart rates had significantly less coronary atherosclerosis than did animals with higher heart rates. Atherosclerosis was also inhibited when cardiac responses to stress were inhibited in monkeys fed an atherogenic diet. Stress-related tachycardia has been used to investigate the mechanisms of enhanced atherogenesis at high heart rates. The proportion of dysfunctional coronary artery endothelial cells in monkeys exposed to behavioral stress was much higher in untreated, tachycardic animals than in those treated with a $\beta$-blocker. This coronary artery endothelial dysfunction associated with high heart rates, which was also observed by Skantze et al, may represent an important mechanism of increased atherogenesis. These observations are supported by results from the Beta-blocker Cholesterol-lowering Asymptomatic Plaque Study (BCAPS)—a randomized trial that showed that a $\beta$-blocker reduced the rate of progression of carotid-intima thickness in asymptomatic patients. More recently, positive associations have been identified between plaque disruption, an elevated left
ventricular mass, and a mean heart rate >80 beats/min, while a negative association with the use of β-blockers was also shown. Specifically, a mean heart rate >80 beats/min was an independent predictor of atherosclerotic plaque disruption with an odds ratio of 3.19 in a multivariable analysis. 20 I toReturnadine may therefore improve patient outcomes after myocardial infarction by reducing atherosclerotic progression and by preventing plaque rupture.

CONCLUSION

A high resting heart rate is a strong predictor for cardiovascular mortality and morbidity in patients with CAD. Heart rate reduction is an integral part of an optimal pharmacological strategy to restore the balance between myocardial oxygen demand and supply, both through a decrease in myocardial oxygen requirement and a lengthening in the duration of diastole and myocardial perfusion. Early after myocardial infarction, the negative inotropic and hypotensive effects of β-blockers contraindicate their use in patients with pulmonary congestion, borderline blood pressure, overt pulmonary edema, or cardiogenic shock. Following discharge after myocardial infarction, β-blockers have traditionally been considered as a first-line therapy, but their use may be limited by side effects including fatigue, depression, and sexual dysfunction. Bronchospasm and atrioventricular block represent other limitations of β-blockers. ITOpradine is a selective and specific β₂ inhibitor with proven antianginal and anti-ischemic effects that have been shown to be noninferior to those of the β-blocker atenolol. Unlike β-blockers, ITOpradine is devoid of intrinsic negative inotropic effects and does not affect coronary vasomotion; it has no effects on blood pressure, myocardial contractility and AV node conduction. These advantages place ITOpradine in a unique position to reduce heart rate in unstable patients. In addition, a whole range of patients with CAD may benefit from exclusive heart rate reduction with ITOpradine, including those with contraindications or intolerance to the use of β-blockers, such as patients with bronchospasm and AV block, and patients that are insufficiently controlled by β-blockers or calcium channel blockers. ITOpradine has also been shown to improve myocardial stunning after myocardial ischemia more effectively than β-blockade. Furthermore, evidence suggests that reducing heart rate could prevent the progression of atherosclerosis as well as plaque rupture. ITOpradine for the reduction of hard cardiovascular events in patients with ischemic heart disease will be tested in the MorBidity-mortality EvAlUaTion of the Iβ inhibitor ITOpradine in patients with coronary disease and left ventricular dysfunction (BEAUTyUL) trial, an ongoing large-scale, multicenter, placebo-controlled, randomized trial.

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Icons of Cardiology

Maude Elizabeth Abbott: bringing order to congenital malformations of the heart

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The woeful state of knowledge of congenital heart disease at the beginning of the 20th century can be appreciated by examining two major textbooks. Osler's *The Principles and Practice of Medicine* devotes 4 pages to this subject, which is described as of only limited clinical interest, as in a large proportion of the cases the anomaly is not compatible with life, and in others nothing can be done to remedy the defect or even to relieve the symptoms.1

Mackenzie's *Diseases of the Heart* has even less to say; a single page on "Congenital Affection of the Heart" includes statements such as:

- The most characteristic symptom is cyanosis, which is present in a great number of patients. [and] Murmurs are usually present, almost invariably systolic in time.2

Lack of interest in these conditions was due in part to the fact that, with the notable exception of patent ductus arteriosus that has a characteristic "machinery" murmur, virtually none of these conditions could be diagnosed in living patients. In 1931, Paul White wrote that the possibility of identifying a particular defect in a living patient had seemed so remote some years ago that a diagnosis more detailed than that of "congenital heart disease" was considered foolhardy, and even this diagnosis was ventured often with uncertainty.3

In 1944, commenting on the "great progress [that] has occurred in the past twelve years," White acknowledged the "pioneer work of enthusiastic students of the subject, especially Maude Abbott of Montreal."4 More recently, Acierno described Abbott's contribution as a comprehensive, organized, and systematized knowledge of congenital heart disease, [which] provided insight into the pathophysiologic pathways [that] enabled surgeons to devise a rational surgical method for correction or amelioration of the altered physiology.5

MAUDE ELIZABETH ABBOTT

Maude Abbott (Figure 1),5-7 the second daughter of the reverend Jeremia Babin and Elizabeth Abbott, was born in March 1869, in a small village in eastern Quebec. Her father had deserted his family before Abbott was born and her mother (like seven of her eight siblings) died at a young age of tuberculosis, as a result, Abbott was adopted and raised by her maternal grandmother. She was initially educated at home by a governess, and from 1884 to 1886 attended a private school in Montreal. The diary she wrote as a child portrays a gentle, introspective young woman with a strong desire to become educated at a time when women's access to a university education was severely limited. Thanks in part to a recent gift to support higher education for women, Abbott won a scholarship to McGill University, but because a smallpox epidemic in Montreal caused her grandmother to fear for her safety, she did not begin her studies until 1886. Four years later, having been class president every year and editor of the college's only publication, she graduated as valedictorian, having earned the Lord Stanley Gold Medal for academic excellence. Abbott ap-
pears to have decided on a career in medicine during her second year in college when a childhood friend asked about her plans and she replied “if I weren’t an artist I would be a doctor. It’s a lovely life. So human and full of people.” This desire to help others was apparent after Abbott graduated from college, when she and other women graduates worked in a soup kitchen for factory and working girls.

Abbott applied to study medicine at McGill, where she had strong family ties, but was met with an adamant refusal because she was a woman; the Registrar wrote: “I am sorry to inform you that the Faculty of Medicine can hold out no hope of being able to comply with your request.” This led Abbott and a few other women to initiate a campaign that drew the support of several newspapers and leading physicians, but which eventually failed. In 1890, therefore, Abbott enrolled in the Medical College of Bishop’s University, a small school in Montreal. When she attempted to attend the lectures of the summer session of McGill Medical School, she was again rebuffed; as noted by the Registrar:

The Faculty…were almost unanimously opposed to allowing young ladies to attend the course in the Medical building, even during the summer session, because it would be a dangerous precedent, inasmuch as the Faculty have already declared themselves opposed to co-education in medicine.

The women medical students at Bishop’s College were subsequently told they could not attend rounds on the wards at the Montreal General Hospital, an independent entity where teaching was supervised by McGill Faculty. Abbott and her fellow female students fought this refusal and created such a storm that several Governors of the hospital refused to pay their subscriptions and a local newspaper mocked the “poor little men students [who needed to be protected] from the great big lady students.”

As a result, Abbott received the “ticket” needed to study on these wards.

In 1894, having won the senior anatomy prize and the Chancellor’s prize for the best examination in her final subjects, Abbott graduated from Bishop’s with honors. She immediately went to Europe for further training, spending 9 months attending clinical rounds and operations in England, Germany, and Switzerland, after which she reached Vienna where for 2 years she studied mainly obstetrics and gynecology. She returned to Canada by way of the United Kingdom where she passed licensing examinations in Edinburgh, which allowed her to serve as a locum tenens at a women’s hospital in Glasgow and as a clinical assistant at the city asylum in Birmingham. She opened a practice in Montreal in September 1897, but made little mention of this in her memoirs; a contemporary described Abbott at that time as “a feminine misfit in an exclusive male environment.”

Abbott’s future opened in 1898, when she was appointed assistant curator of the Medical Museum at McGill and began cataloguing a collection of pathological specimens that dated back almost 75 years. At this time Abbott came under the influence of William Osler, who for the next 20 years was to provide both encouragement and guidance as she became the world’s authority on congenital heart disease. Her demonstrations for the medical students at McGill received such high praise that they became a required part of the curriculum.

Figure 2. Three examples of cor biaatriatum triloculare (single ventricle). (A) The “Holmes Heart” (anterior view) from a 22-year-old man with cyanotic heart disease showing the single large ventricle with no septum and a rudimentary cavity that gives rise to the pulmonary artery. (B) Embryonic mammalian heart (cross-section) showing a single ventricle that receives blood from both the right atrium (RA) and left atrium (LA). (C) Adult turtle heart (dorsal view) whose single ventricle receives blood from both the right atrium (RA, shown in outline as this structure was removed) and the left atrium (LA).

of the Pathology course. During and after World War I, in addition to work at the Medical Museum, she brought a method to measure basal metabolism that she had learned in Boston to Montreal, gave a lecture on Florence Nightingale at Harvard, was acting Editor of the Canadian Medical Journal, and helped prepare (and pay for) a memorial to William Osler. Harold Segall, a museum assistant who went on to become one of Canada’s leading cardiologists, described Abbott as having about a dozen irons in the fire at the same time, and all seemed in confusion. Moreover, she took on any new thing that came up with the greatest of enthusiasm, indeed as if she had nothing else to do. In her demonstrations with students she would speak rapidly, showing specimen after specimen, with an enthusiasm which never faltered. (Cited by reference 6.)

Abbott also began a catalog of this museum that Osler described as: “one of the best pieces of work ever done at the School... She evidently has a genius for this sort of thing.”

CONTRIBUTIONS TO THE UNDERSTANDING OF CONGENITAL HEART DISEASE

Abbott’s interest in congenital heart disease appears to have been triggered by the “Holmes Heart,” a specimen of cor biaatriatum triloculare (single ventricle) in the McGill collection that dated back to 1824 (Figure 2A). Because this heart had been labeled “ulcerative endocarditis,” which was obviously an error, Abbott wrote to Osler who had used this collection when he taught at McGill. Osler replied that he remembered this specimen as he had often shown it to students as a special form of a three-chambered heart. Osler, impressed by Abbott’s energy and skills, asked her to write a section on congenital heart disease for his new System of Medicine. He subsequently described Abbott’s section, the only one in his book written by a woman, as “…far and away the best thing ever written on the subject in English—possibly in any language.” (cited by reference 6). First published as a 22-page description of 412 hearts, Abbott’s section grew to 200 pages that described 1000 hearts and led to her Atlas of Congenital Heart Disease, which provided a pathophysiological and statistical analysis of the clinical and anatomical abnormalities in these patients.

In bringing order to what had previously been a disorganized array of fascinating, but poorly understood, congenital malformations of the heart, Abbott divided these patients into three groups: Those with no abnormal communications, those with a left-to-right shunt (acyanotic), including patients with shunt reversal later in life (cyanose tardive), and those with permanent right-to-left shunt (cyanotic). For each she developed a diagram of the circulation, one example of which is depicted in a mural at the Instituto Nacional de Cardiologia in Mexico City (Figure 3).

In 1910, after Abbott gained her reputation as an authority on congenital heart disease, McGill awarded her an honorary MD degree and, in addition to her title as Curator of the Medical Museum, she became Governor’s Fellow in Pathology from 1905 to 1909 and lecturer in pathology from 1910 to 1918. In recognition of her work Abbott was made a Miembro Fundador of “El Colegio Nacional.” Through the pages of this book we see the impetus for our modern understanding of the etiologies of congenital heart disease.

Table 1.

QUESTIONS PROPOSED BY ABBOTT TO HELP EXPLAIN THE ETIOLOGY OF CONGENITAL CARDIOVASCULAR MALFORMATIONS

<table>
<thead>
<tr>
<th>Question</th>
<th>Answer</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Did the abnormality result from a fault in the germ plasm?</td>
<td>Yes</td>
</tr>
<tr>
<td>2. Was the abnormality inherited?</td>
<td>Yes</td>
</tr>
<tr>
<td>3. Did the abnormality occur because of a change in the environment of the embryo in utero?</td>
<td>Yes</td>
</tr>
<tr>
<td>4. Was the abnormality a result of maternal disease?</td>
<td>No</td>
</tr>
<tr>
<td>5. Was the abnormality due to fetal trauma or a uterine disorder?</td>
<td>No</td>
</tr>
<tr>
<td>6. Did a disease affecting the early embryo cause the abnormality?</td>
<td>No</td>
</tr>
</tbody>
</table>
Maude Elizabeth Abbott: bringing order to congenital malformations of the heart - Katz

1923. In 1923 she went to Philadelphia as chair of the Pathology Department at the Women’s Medical College of Pennsylvania, but after a stay of 2 years during which she significantly raised the status of this department, she returned to McGill where she remained as assistant professor of medical research until her retirement in 1936. Her enthusiasm continued unabated, as evidenced by the following story, told by William C. Gibson who met Abbott in 1993 when he was a student:

One day a heavy grey haired woman came into the library where I was working, and recognizing [that I was examining] the Osler notebooks, asked if I was an out-of-town researcher in medical history. I replied that I was a first year medical student at McGill and was interested in Osler. That was my fatal mistake! I was at once whisked downstairs by this bustling human dynamo who seemed only to cling to the stair rail, letting her feet find the steps if they could. Within a few minutes I was seated in the midst of a sea of charts, book, and pictures. (Cited by reference 6.)

This encounter began a five-year collaboration on a new edition of the Osler Bibliography that continued when Gibson left Canada to study in Oxford, from where he pleaded that because of “the broad expanse of the Atlantic Ocean” he would find it difficult to continue working on this project. However, Abbott insisted, and when Gibson’s work on the bibliography hit a snag, Abbott wrote “as if she might come over and scalp me as a low grade procrastinator.” Happily, the bibliography was published in 1939.

Abbott retired in 1936 at the prescribed age of 67 with the remarkably inappropriate rank of assistant professor of medical research, considering that in addition to her many scholarly achievements she was a Charter Member of the Royal College of Physicians of Canada and a Fellow of the Royal Society of Medicine, London, England. Shortly after her retirement, however, McGill did award her a honorary LLD, the highest academic honor they could confer. She remained active, continuing to publish and lecture (Figure 4) until July 1940 when she had a cerebral hemorrhage; she died 2 months later, on September 2.

CONCLUSIONS

Maude Abbott is remembered today largely for her seminal work on congenital heart disease, which is described in more than 40 of her 116 publications,6 and her Atlas of Congenital Heart Disease,9 which summarized her major academic contribution and provided the foundation of our modern understanding of these malformations. However, her written work is only a part of her contribution to medicine. Those who knew Abbott place even greater emphasis on her remarkable personal qualities, describing her as an energetic, enthusiastic woman who was warm, unselfish, and completely natural. MacDermot, whose biography captures her indomitable personality, wrote that even though Abbott was a pioneer who clarified the once obscure field of congenital heart disease, her character was itself “a contribution, and one which was probably more important, if less tangible.”6 Paul White, whose 3rd Edition of Heart Disease4 includes a foldout table showing Abbott’s classification, wrote:

...far more important than any of her written works was her vital stimulus to others. Her spirit was indefatigable... many of the contributions... to our knowledge of congenital heart disease made by others are due directly to Maude Abbott’s influence... it is not simply as the world’s authority on congenital heart disease that Maude Abbott will be best remembered, but as a living force in the medicine of her generation. Hers was a great spirit. (Cited by reference 6.)

Abbott, who was born 10 years after publication of Darwin’s Origin of the Species and 2 years before his Descent of Man, clearly documented the practical importance of human evolution by basing her analysis of congenital heart disease on the many similarities between the malformations, the structure of embryonic mammalian hearts, and the architecture of reptilian hearts (Figure 2). She observed that knowledge of the successive stages through which the mammalian heart passes in its evolution from its primitive tubular state to the completely divided four-chambered organ conducting a double circulation allows the most bizarre combinations of defects to be interpreted quite simply, as due to early arrests of development, marked as it may be by ingenious structural adaptations of growth.

In describing the importance of comparative anatomy, she noted the “truly extraordinary way in which [the] various orders in the ascending vertebrate scale mirror the successive stages through which the human heart passes in very early intrauterine life,” and how this ontogeny presents “a complete review of this organ’s evolution...
down to the closure of the cardiac septa in the eighth week of fetal life.”
Abbott also highlighted the importance of a detailed study of the individual patient, as well as of large groups, when she observed:

It is… a well known fact that, in clinical medicine, the intimate knowledge of a relatively small number of individual cases is likely to yield a richer harvest of understanding of diseased conditions than wider generalizations covering a more vast material.”

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Membrane currents in the rabbit sinoatrial node cell studied by the double microelectrode method

A. Noma, H. Irisawa

Pflugers Arch. 1976;364:45-52

This paper constitutes a landmark in the study of mechanisms underlying generation of sinoatrial nodal (SAN) activity. Hitherto, membrane potential measurement had offered a means of measuring SAN spontaneous activity; however, in order to understand the underlying basis for this, direct measurements of membrane currents were needed. This required the use of the “voltage-clamp” technique, a methodology that allows control of cellular transmembrane potential, and measurement of corresponding ion flow.

Cardiac tissue presents particular challenges in terms of the study of ionic currents. Electrical coupling between adjacent cardiac cells makes it difficult to control adequately the membrane potential of multicellular preparations using the voltage-clamp method. This is undesirable because a lack of adequate spatial control of membrane potential introduces unwanted measurement errors. Ultimately, this problem was to be solved by the advent of single-cell preparations and the use of the patch-clamp technique. In a carefully executed series of experiments, it was shown that membrane potential was not homogeneous in preparations with dimensions of 1.7x0.3 mm and 0.7x0.3 mm, but that at 0.3x0.3 mm such inhomogeneity was minimal.

It is in the last three figures of the nine in this paper that we see the first SAN ionic current data from this preparation. The transient inward current elicited on depolarization has a voltage dependence now known to be characteristic of L-type calcium current, while outward current “tails” observed following depolarization and outward rectification of the current-voltage relationship at positive voltages involve delayed rectifier potassium channels. A low membrane conductance was observed between -60 and -20 mV and is now accepted to be important for SAN function as it means that comparatively small ionic currents can have significant effects on membrane potential over this voltage range. Importantly, on hyperpolarization to negative membrane potentials, an inward current was observed that showed a clear time-dependent increase throughout the voltage command. The profile of this current is now clearly recognizable as that of If, though at the time the basis for the inward current changes during and following hyperpolarization was difficult to explain. What is clear, however, is that many of the cellular electrophysiological properties of the SAN that were later to be studied in detail are visible in this study, which played a major role in setting the scene for subsequent investigations.

263 US citizens are evacuated from Beirut to avoid becoming entangled in the Lebanese civil war; East German swimmers break 14 world records at their national championships; and six Palestinians hijack an Air France Airbus in Greece, taking 250 passengers hostage.
How does adrenaline accelerate the heart?

H. F. Brown, D. DiFrancesco, S. J. Noble


Modulation of the activity of the sinoatrial node (SAN) underpins the automatic regulation of heart rate, and this paper provides a wonderful early insight into the mechanism(s) by which adrenaline modulates SAN activity. Previous studies had shown that adrenaline increased the slow inward (ie, calcium) current in other areas of the heart, it was possible that this might mediate or contribute to the effects of adrenaline on the SAN. The small multicellular SAN preparation developed by Noma and Irisawa (see preceding Summary) provided a means by which the effects of adrenaline on both spontaneous activity and ionic currents could be studied.

Possibly due to the concise *Nature* paper format or to a stylistic choice of the authors, detailed methodological information is not included in this report, though it is clear that the small rabbit SAN preparation of Noma and Irisawa was used together with a two-microelectrode voltage-clamp. Initial measurements were made in the absence and presence of adrenaline of both membrane potential and of the rate of change of voltage during SAN spontaneous activity. At a physiologically relevant concentration of 50 nM, adrenaline increased both action potential amplitude and upstroke velocity. These effects were consistent with an increase in the slow inward current, particularly because they were made in the presence of tetrodotoxin, which would have inhibited any fast sodium current in the preparation. In addition to its effects on the action potential upstroke, adrenaline also produced a slight hyperpolarization of the maximum diastolic potential (the most negative membrane potential reached following action potential repolarization) and accelerated the rate of action potential repolarization. This suggested that both inward (depolarizing) and outward (repolarizing) conductances were modulated by adrenaline.

Voltage-clamp measurements provided a direct demonstration that the transient inward current was increased by adrenaline (500 nM). Significantly, the outward potassium current (\(I_K\)) observed in the same experiments was also increased, confirming the notion that adrenaline exerted dual effects on inward and outward conductances. These observations raised the question as to how adrenaline accelerates rate as, while an increase in the slow inward current might accelerate the latter part of the pacemaker depolarization, the authors reasoned that the concomitant increase in \(I_K\) might act to decelerate the pacemaker depolarization. This led to an examination of the effect of adrenaline (100 nM) on time- and voltage-dependent current activated by hyperpolarizing voltage-clamps—making this the first report of the importance of \(I_K\) in the adrenergic modulation of heart rate. A significant and reversible increase in \(I_K\) was observed, which would be expected to accelerate the pacemaker potential under membrane potential recording conditions.

The importance of this observation is clear: it constitutes the first evidence that modulation of \(I_K\) contributes to the positive chronotropic effects of adrenaline. This is a remarkable contribution in itself, but doubly so in the context that at this time the nature of \(I_K\) was unclear. It was considered possible that deactivation of a potassium current called \(I_{K1}\), as opposed to a distinct inward conductance, could underlie \(I_K\). As the authors commented in discussing their findings, testing the identity of \(I_K\) under voltage-clamp was difficult at that time, as the small SAN preparation did not readily tolerate the large voltage excursions necessary to examine the ionic basis of the current. Nevertheless, this study constitutes a pivotal step in identifying \(I_K\) as an important means of modulating SAN activity. Added to this, it is a clearly written and very readable report, and this makes its findings accessible to the specialist and nonspecialist alike.

1979

Bjorn Borg defeats Roscoe Tanner to win the Wimbledon men’s singles title; Nicaraguan president Gen. Anastasio Somoza Debayle resigns and flees into exile in Miami; and Marxist philosopher Herman Marcuse, writer of *One-Dimensional Man* and *Eros and Civilization*, dies, aged 81
Functional and morphological organization of the rabbit sinus node

W. K. Bleeker, A. J. Mackaay, M. Masson-Pevet, L. N. Bouman, A. E. Becker

Circ Res. 1980;46:11-22

In order to gain an integrated understanding of the generation and conduction of sinoatrial node (SAN) activity, two types of experimental data are required: first, those data that provide insight into the ionic basis of impulse generation at the cellular level and, second, those indicating how impulse propagation occurs through the intact node and between this and adjacent atrial tissue. Morphological investigations before this study had shown the SAN to be a heterogeneous structure, but morphology alone was insufficient to provide insight into the location of the dominant pacemaker site within the SAN or to predict how emerging excitation would spread. This study constitutes an important landmark, as it sought to both map excitation within the node and to correlate this with structural properties of the SAN.

In the 25 years since this study, technical advances have been made in recording techniques such that nowadays multielectrode arrays or optical mapping with voltage-sensitive dyes offer ways of studying excitation spread at the tissue level. At the time of this study, however, such options were not available. Instead, using a preparation from the rabbit heart comprising the SAN, intercaval region, and roof of the right atrial appendage, the authors of this study employed single microelectrode recordings to map excitation within the SAN area. This is a painstaking and technically challenging approach: to produce an excitation map, such recordings would both need to be relative to a time-reference value and to allow proximate recordings to be made with a high degree of spatial resolution. The time-reference was provided by an electrogram obtained from a surface electrode placed on the inferior part of the crista terminalis. A high spatial resolution in control of position was obtained by mounting the micromanipulator controlling the microelectrode onto a mechanical stage with lateral movements accurate to 0.01 mm. Initial measurements during each experiment located the area of preparations where electrical propagation was slowest. Then a series of 150 to 300 successive impalements was painstakingly made over 3 to 5 hours over an area approximating 4x6 mm. The spatial resolution between impalements in the pacemaker region was 0.2 mm.

The site of earliest activation was found to lie within the intercaval region, between 0.5 and 2 mm from the medial border of the crista terminalis. Excitation then propagated preferentially towards the crista terminalis in an oblique cranial direction, but was markedly slower in other directions and was blocked toward the interatrial septum. Within the intercaval region, the SAN was identified by distinct staining properties from atrial cells. Within the SAN region, distinct zones could be identified. The central compact node was characterized by cells showing a compact, interweaving structure, while in the nodal periphery cells lay in a parallel fashion. The leading pacemaker site was located as a group of ≈5000 cells within the compact node. Central nodal cells were smaller in size than those in the periphery of the node. Electron microscopy showed gap junctions to be present throughout the node, but to be larger and more frequent towards the periphery. Unlike atrial muscle, where they were found preferentially at intercalated discs, the gap junctions were dispersed randomly at the cell surface.

This report set the scene for subsequent studies in a number of respects: it laid the foundations for investigation of heterogeneity of cellular electrophysiology and also of connexin isoforms that underlie gap junctions within the SAN, for the development of models of SAN regional heterogeneity (the mosaic and gradient models), through to the present when 3-dimensional combined anatomical and functional SAN models lie within reach.

1980

American writer Henry Miller, author of Tropic of Cancer and Tropic of Capricorn, dies, aged 87; nuclear physicist Andrei Sakharov is sent into exile in Gorky after speaking out against the Soviet presence in Afghanistan; and Robert Mugabe returns to Rhodesia (Zimbabwe) after five years in exile.
Prior to this study, the ability of acetylcholine (ACH) to increase potassium conductance of pacemaker tissue had already been recognized. Indeed, fluctuation analysis had provided evidence that this effect resulted from activation of a distinct potassium channel coupled to muscarinic ACh receptors (mACh-R) in the pacemaker cell membrane. The importance of this conductance was considered to be that, by decreasing nodal cell excitability, it would reduce the rate of sinoatrial node (SAN) spontaneous activity and thereby heart rate. What had not been reported prior to this study were direct measurements of single channel behavior of the mACh-R-activated conductance.

Today, a key difference between pacemaker and nonpacemaker tissues is considered to be a strong role for an inwardly rectifying K\(^+\)-current called \(I_{K1}\) in setting the resting potential of ventricular and atrial cells; in pacemaker tissues, \(I_{K1}\) channels are sparse or absent. Importantly, this study showed that the ACh-activated K\(^+\) current in pacemaker cells is mediated by a distinct channel, which was to be reinforced later when the molecular basis of the channel was also found to be distinct from that of other inwardly rectifying K\(^+\) currents in the heart.

Two key technical advances made this investigation possible. First, enzymatic and mechanical dispersion methods had been developed to allow the isolation of single cardiac cells for electrophysiological study. Second, the development of the patch-clamp technique allowed recordings from small areas of cell membrane. In this, the application of suction to low-resistance glass pipettes placed against the cell membrane forms a high-resistance seal, and current flow in response to voltage changes across the small "patch" of membrane occurs through any ion channels in this area of the membrane. Channel openings in such recordings are visible as rectangular deflections during which tiny currents (in the order of \(10^{-12}\) A) flow. The impact of this technique on increasing understanding of ion channel function cannot be overstated: it's developers (one of whom is the first author of this study) were later (1991) to win the Nobel Prize for Physiology or Medicine.

The experiments in this paper were performed on single cells isolated from both the SAN and the atioventricular node (AVN). Initial measurements were made of ACh-activated current at the whole-cell level, this was larger at negative than at positive voltages (a property termed "inward rectification") and depended on the external K\(^+\) concentration as expected for a K\(^+\) current. Single channel measurements were then made, and by changing the voltage across the patch the authors obtained single channel conductance values for the ACh-activated current: this was greater at negative than at positive voltages, consistent with the inwardly rectifying nature of the whole-cell current. Single channel currents were strongly K\(^+\)-dependent and also dependent on the ACh concentration applied.

Cells that responded to ACh showed a second interesting feature: in the absence of ACh, they exhibited spontaneous channel openings of similar size and duration as the ACh-activated current, but at a lower frequency. It was concluded that this resting K\(^+\) channel activity could represent spontaneous openings of the ACh-activated channels. In nodal cells without spontaneous activity, and in atrial cells, a distinct type of resting K\(^+\) channel (with much longer channel openings, resembling the resting K\(^+\) channels of ventricular cells) was visible, suggesting that resting K\(^+\) channel activity differed between pacemaker and nonpacemaker cells. The study concludes with a study of the gating behavior of the ACh-activated channel current based on a two-step reaction underlying channel activation.

Konrad Kuja, a calligrapher from Stuttgart, admits to faking the Hitler diaries; the Iranian Communist Tudeh party is dissolved; and the South African air force bombs ANC bases in Maputo in retaliation for a car bombing in Pretoria that killed 18 and injured 190.
Why doesn’t atherosclerosis affect all human arteries equally? This is an important question as areas around arterial branch points and curves are particularly susceptible, and the coronary arteries, the abdominal aortic segment, carotid bifurcation, and vessels supplying lower extremities tend to be affected in a much worse way by atherosclerosis than do other major arteries.

While an intuitively obvious answer might be that high shear stress predisposes toward plaque formation, in fact the evidence at the time of this study by Beere and colleagues was that, in the case of the carotid bifurcation, it is low shear stress and oscillations in shear stress direction (which occur during pulsatile flow) that are the major hemodynamic risk factors associated with lesion formation.

Coronary arteries are particularly predisposed towards severe atherosclerosis, with plaque formation tending to localize at branch points. Coronary arterial blood flow undergoes several alterations during each cardiac cycle: during the isovolumetric contraction and rapid ejection phases of systole it decreases; it increases as peak systolic aortic pressure exceeds intracoronary pressure, then decreases during the remainder of systole. During isovolumetric relaxation in diastole, coronary flow is greatest, while it gradually decreases during ventricular filling. The authors of this study reasoned that if hemodynamic changes are important to coronary arterial plaque formation, a high heart rate should exacerbate coronary atherosclerosis, while a low rate might have a protective effect.

This idea was investigated using surgical ablation of the sinoatrial node (SAN) in cynomolgus monkeys (a species of macaque originally from South-East Asia), with a sham procedure being carried out as a control and an additional group that did not undergo surgery. An atherogenic diet (incorporating 25% peanut oil and 2% cholesterol) was given to each group for 6 months and heart rate was monitored routinely by telemetry. When atherosclerotic lesion area and average and maximum stenosis were subsequently compared between control and SAN ablation groups, there was no significant difference. However, when animals that had undergone surgery were grouped according to whether heart rate was less than or greater than the preoperative mean (classified as ‘low heart rate’ and ‘high heart rate’ groups, respectively), the values of these indicators of coronary atherosclerosis in the high heart rate group were greater (by more than twofold) than those from the low heart rate group. Comparisons of blood pressure, weight, and cholesterol and triglyceride levels showed no differences in these between low and high heart rate groups. Therefore, heart rate itself appeared to be influencing the severity of atherosclerotic lesions.

Since this study, the association between a high heart rate and atherosclerotic plaque formation has been reinforced, while a high rate has also been associated with increased coronary plaque disruption. What then are the implications of such associations? First, regular aerobic physical activity is associated both with a reduction in resting rate in humans and with a protective effect against coronary artery disease. Second, regular stress-induced heart rate rises during either prolonged psychological stress or in association with ‘Type A’ personality traits would be anticipated to elevate resting rate for prolonged periods and, in turn, this might link to coronary atherogenesis. Third, if an elevated resting heart rate is an independent risk factor for coronary atherosclerosis, then heart rate reduction should be expected to reduce the rate of lesion development in at-risk groups. Results that emerged from the BCAPS (β-Blocker Cholesterol-lowering Asymptomatic Plaque Study) trial in 2001 are consistent with this notion.

Indian Prime Minister Indira Gandhi is assassinated by two members of her personal bodyguard; China announces a program of economic reforms giving factory owners greater autonomy; and Italy indicts three Bulgarians and four Turks in the conspiracy to assassinate the Pope.
Mechanism of beneficial effect of beta-adrenergic blockade on exercise-induced myocardial ischemia in conscious dogs

B. D. Guth, G. Heusch, R. Seitelberger, J. Ross Jr

_Circ Res._ 1987;60:738-746

Guth et al performed this study against a background in which, while a number of drugs were known to be beneficial in treating effort-induced angina pectoris, precise mechanisms underlying the beneficial effects had not been established. Cardioselective β-blockade might be expected to have a number of effects that would mitigate the effects of restricted blood flow in local ischemia. For example, in a setting of restricted coronary inflow, lowering of heart rate would increase time for tissue perfusion during diastole. Also, the negative inotropic effect of β-adrenoceptor blockade would reduce the oxygen demand of nonischemic tissue and possibly also any discrepancy between oxygen supply and demand of such tissue. A reduction in left ventricular pressure consequent upon β-blockade could also contribute to improved function in the ischemic region. However, while this range of possibilities was appreciated, the relative importance of heart rate reduction compared with other consequences of β-adrenoceptor blockade was not known.

The authors addressed this question using a canine model in which regional ischemia and dysfunction were induced by using an aneroid constrictor to produce coronary stenosis of the left circumflex artery. Following recovery from surgery, regional ischemia was induced in conscious dogs by treadmill exercise. Blood flow and ventricular wall dysfunction were assessed under differing conditions: a control run (no treatment), following administration of the β-adrenoceptor blocker atenolol, and in the presence of atenolol, but with atrial pacing to prevent the reduction of heart rate. This neat experimental design allowed the authors to determine, at a matched workload, the relative importance of heart rate reduction compared with other consequences of β-adrenoceptor blockade was not known.

The results are striking. In the absence of atenolol, wall thickening of the ischemic zone during systole was somewhat compromised, but atenolol administration improved this (nearly doubling the measured value) along with producing the expected reduction in heart rate. However, when the negative chronotropic effect of atenolol was prevented by pacing, no improvement of wall thickening during systole in the ischemic zone was seen, rather this fell to a value that was lower than that in the absence of atenolol, possibly because β-blockade may have unmasked α-adrenergic vasoconstriction in the ischemic myocardium.

Eliminating the heart rate reduction produced by atenolol would not have removed its other possible beneficial actions—for example, the potential metabolic benefits of preventing the inotropic effect of increased sympathetic drive during exercise. So the inference that can be drawn is that heart reduction per se was the key player in the beneficial effect of β-blockade, likely by increasing diastolic perfusion time, resulting in improved diastolic flow to the ischemic myocardium. A link here to If is not hard to see: β-adrenoceptor blockade would be expected to prevent/limit augmentation of If resulting from increased sympathetic drive (see Summary of paper by Brown et al, page 51), with a consequent limitation on heart rate increase.

The results of this study also set the scene for the investigation of other means of heart rate reduction. Indeed, the authors suggest that specific bradycardic agents could be beneficial for exercise-induced ischemia, producing heart rate reduction without the negative-inotropy (or indeed the range of side-effects) of β-adrenoceptor blockade. Since that time, specific If inhibition has been seen to produce heart rate reduction without limiting beneficial stroke volume changes, with positive effects in terms of angina prevention, and with minimal side effects—the main ones being transient and reversible visual disturbances due to the role of an If-like current in visual transduction.

1987

Iraqi missiles kill 37 in an attack on the US frigate Stark in the Persian Gulf; the trial of nazi war criminal Klaus Barbie opens in Lyon; and US film actress Rita Hayworth dies after a long battle with Alzheimer’s disease
Muscarinic modulation of cardiac rate at low acetylcholine concentrations

D. DiFrancesco, P. Ducouret, R. B. Robinson

Science. 1989;243:669-671

By the time this study was conducted, it was known that acetylcholine (ACh) could modulate multiple ionic currents: activation of a K+ conductance (I_{KACh}, see summary of paper by Sakmann et al, page 53), reduction of the slow inward current (I_{si}), and inhibition of I_{f} had all been reported. However, the question arose as to whether activation of I_{KACh} or inhibition of I_{f} is the predominant mediator of the negative chronotropic effect of ACh at physiologically relevant concentrations. Therefore, this study was performed in order to make a direct comparison of the effects of ACh on I_{f} and I_{KACh}, using the rabbit isolated sinoatrial node (SAN) cell preparation and whole-cell patch clamp technique. The key here was that by studying both currents in a single study, it would be possible to determine the relative potency of ACh effects on the two currents in a more reliable fashion than would be possible by comparing data from different investigations.

In the first experiments shown, the authors observed that inhibition of I_{f} occurred at ACh concentrations an order of magnitude lower than that necessary for I_{KACh} activation. Precise quantification of the difference was not possible in these initial experiments in which both currents were studied concomitantly, because at higher concentrations effects on the two currents overlapped. Therefore, the authors proceeded to quantify the difference by studying each current under selective recording conditions. Barium ions block I_{KACh}, so selective I_{f} measurements were made with Ba^{2+} present, while I_{KACh} was measured in a voltage range at which I_{f} channels would not have been active. For inhibition of I_{f}, the half-maximal ACh concentration was found to be 13 nM, while that for activation of I_{KACh} was 260 nM. Thus, there was a 20-fold difference in the potency of the two effects of ACh. A limited number of measurements of effects of ACh on I_{f} were performed, in which a low (10 nM) ACh concentration failed to alter I_{f}, whereas at higher concentrations (100 nM and 1 \mu M) I_{f} was inhibited. Thus, under the conditions of this study, the predominant effect of low ACh concentrations was to attenuate I_{f}. When spontaneous activity was measured, the majority of the slowing of action potential rate with ACh occurred below 100 nM, consistent with a dominant role for I_{f} in mediating ACh’s negative chronotropic effect. A role for I_{KACh} was not excluded, but this was considered to play a role at high ACh concentrations. A similar argument applied to I_{si}.

The study is an elegant one and the message of the paper is clear: I_{f} is likely to mediate the negative chronotropic effect of ACh at low concentrations, with two major mechanisms (I_{f} inhibition + I_{KACh} activation) likely responsible at higher concentrations. So, is that it—job done—in terms of understanding vagal control of SAN rate? Well, not quite. This study sparked further investigation of the relative roles for I_{f} and I_{KACh} with Boyett and colleagues studying both small multicellular and single cell SAN preparations and presenting evidence in 1995 for a dominant role for I_{KACh} over I_{f} in mediating the effect of ACh. This exemplifies the fact that, over the years, the pacemaking field has been dogged with conflicting experimental findings, leading to controversy as to the relative importance of I_{f} and other currents in mediating particular aspects of control of SAN rate.

Iran’s Ayatollah Khomeini declares author Salman Rushdie’s book The Satanic Verses offensive, and sentences him to death; the last Soviet troops leave Afghanistan, bringing and end to the 9-year war; and Czech playwright Vaclav Havel is jailed for dissident activities.

1989
Molecular characterization of the hyperpolarization-activated cation channel in rabbit heart sinoatrial node

T. M. Ishii, M. Takano, L. H. Xie, A. Noma, H. Ohmori

J Biol Chem. 1999;274:12835-12839

Although, historically, the patch-clamp technique was pivotal in establishing the ion channel concept, few would argue against the view that the development of molecular cloning techniques was the next technical advance to revolutionize the study of ion channel function. This enabled genes responsible for ion channel proteins to be identified, it led to the ability to determine ion channel gene mutations associated with ion-channel related diseases (‘channelopathies’) and, by targeting mutations to particular parts of ion channel proteins, it became possible to relate ion channel structure to function.

This paper identified a molecular candidate for the cardiac $I_f$ channel protein. In the lead-up to this study, three full-length and one partial cDNA clones encoding $I_f$-like channels had been found in mouse brain, with a further clone obtained from the sea-urchin testis. However, the molecular identity of channels responsible for $I_f$ in the heart was not clear.

Visiting the paper 6 years after its publication prompts a digression into ion channel nomenclature, for without this some of the terminology used may seem a little confusing. As for some other ion channel genes, those responsible for channels carrying $I_f$-like currents have been given more than one name. In the case of $I_f$, this was probably always on the cards, as even before candidate genes had been identified, the current had several names, being variously labeled $I_f$ (funny current), $I_H$ (hyperpolarization-activated current), or simply pacemaker current.

Nowadays, the family of channels of which cardiac $I_f$ is a member is generally known as the “hyperpolarization-activated cyclic nucleotide-gated cation channel” (HCN channel) family and the four genes that have been identified in mammalian tissues are termed HCN1-4. This paper uses an alternative terminology, with HAC (hyperpolarization-activated channel) in place of HCN. In turn, the members of the HAC family isolated from mouse brain corresponded to cyclic nucleotide-gated (CNG) channels, and so one also finds the term BCNG (Brain-CNG) used in the text.

By screening a rabbit sinoatrial node (SAN) cDNA library, the authors identified a cDNA encoding a protein of 1150 amino acids that showed a high degree of homology in the transmembrane region to the previously identified mouse partial clone, BCNG-3. They named the SAN clone HAC4 (now HCN4) and studied the distribution of its messenger RNA. This showed that HCN4 was most highly expressed in the SAN over other cardiac regions and that it was not significantly expressed in cerebellum, forebrain, kidney, or skeletal muscle. By expressing the HCN4 gene product in a mammalian cell line, the authors found that HCN4 channels exhibited characteristics concordant with those of $I_f$: time-dependent activation on membrane hyperpolarization, sensitivity to cyclic-AMP (a positive shift in voltage-dependent activation, as had been seen previously for $I_f$), sensitivity to block by Cs+ ions, and Na/K ion permeation properties.

The findings of this study have pretty much withstood the test of time. HCN1 and HCN4 have been shown to be able to form heteromeric channels in vitro and the rabbit SAN has been shown also to express message for HCN1 and HCN2, but that for HCN4 is by some margin the most abundant, implicating it as at least a key component if not the sole basis of SAN $I_f$. Identification of the HCN4 clone has also enabled its distribution throughout the SAN to be mapped and, as we shall see in the summary of the paper by Stieber et al (next page), it has also led to the targeted knockout of HCN4 in order to investigate the effect of its absence on native $I_f$ and control of heart rate.

1999

Students Eric Harris and Dylan Klebold storm Columbine High School, killing 12 other students, a teacher, and then themselves;

Libya hands over two suspects wanted for the 1988 Pan Am jet bombing; and

the new Inuit territory of Nunavut officially joins the Canadian Federation
The hyperpolarization-activated channel HCN4 is required for the generation of pacemaker action potentials in the embryonic heart

J. Stieber, S. Herrmann, S. Feil, J. Loster, R. Feil, M. Biel, F. Hofmann, A. Ludwig

Proc Natl Acad Sci U S A. 2003;100:15235-15240

It is one thing to demonstrate the presence of an ion channel candidate in a particular tissue type, but it is quite another to elucidate its contribution to that tissue's functional activity. Selective pharmacology offers one possible approach, but is of limited value when several channel isoforms exist, as in the case of the HCN family, but isoform-selective blocking agents do not.

A more elegant strategy is to eliminate expression of the channel isoform of interest, either in isolated cells or in tissues, by using antisense oligonucleotides, or short interfering RNA, or in the intact animal by the generation of knockout mice. None of these approaches is without potential problems: cell or tissue culture can change electrophysiological properties, whereas targeted gene knockout can result in compensatory changes to other genes and also relies on the suitability of the mouse for the question to be investigated. However, despite such caveats, gene knockdown strategies are currently the best means available of eliminating particular ion channel isoforms.

This paper builds upon accumulating evidence for HCN4 being the predominant HCN isoform both in the adult sinoatrial node and in mouse embryonic hearts. The authors generated HCN4 knockouts, as well as animals with cardiomyocyte-specific deletion of HCN4. The first consequence of this was that both global and cardiomyocyte-specific HCN4 deletion resulted in animals that died between embryonic days 9.5 and 11.5, in the absence of any cardiac structural abnormalities. Consequently, the data come from embryonic, rather than adult, hearts and cells.

Homozygote knockout (HCN4−/−), but not heterozygote (HCN4+−) embryonic hearts showed a significantly reduced spontaneous contraction rate compared to wild-type (WT) hearts. HCN4−/− hearts also showed a reduced sensitivity to pharmacological I_{f} blockade, suggesting that the bulk of drug-sensitive I_{f} was carried by HCN4. However, the fact that I_{f} inhibitors still exerted some bradycardic effect on HCN4−/− heart beating indicates that HCN4 was not exclusively responsible for I_{f}. Moreover, the fact that −/− hearts treated with I_{f} blocker could still beat spontaneously suggests that some pacemaking was possible independent of I_{f}, albeit at a markedly reduced rate. Electrophysiological investigation of cardiomyocytes from mutant embryos showed a 70% to 90% attenuation of I_{f}, and cAMP application failed to alter heart beating rate or I_{f} activation. These data all point toward a dominant (albeit not exclusive) role for HCN4 in mediating I_{f} and to a pivotal role for HCN4 in mediating the chronotropic effect of cAMP.

The skeptic might point out that in contrast to the adult heart, in the embryonic heart spontaneous activity is widespread and that the cardiomyocytes used for single cell electrophysiology in this study could not be described as sinoatrial. This study attempts to address this potential problem in two ways. First, the distribution of HCN4 was highly localized to a specific area of the developing heart that is likely ultimately to develop into the adult sinoatrial node. Second, the authors identified 4 distinct action potential profiles from embryonic cardiomyocytes, including “mature pacemaker-like action potentials.” These were present in a small proportion of WT embryonic cardiomyocytes, but were completely absent in those from HCN4−/− hearts. Considered collectively, therefore, the findings of this study provide compelling evidence that significant attenuation of I_{f} prevents normal development of normal embryonic cardiac activity and implicates HCN4 as a major player in this.

American troops find Saddam Hussein in hiding near Tikrit, who surrenders without a fight; Muammar Qaddafi announces that Libya will give up its nuclear, biological, and chemical weapons programs; and the Nobel Prize for Physiology or Medicine goes to Paul Lauterbur and Sir Peter Mansfield for their discoveries leading to magnetic resonance imaging.
Perhaps it is fitting to save the most recent study selected for comment until last. A number of epidemiological studies in the literature now point toward the significance of a high resting heart rate in humans, with recognized associations between resting heart rate and mortality in patients with hypertension, metabolic syndrome, and in elderly subjects. The importance of heart rate reduction in preventing angina is also now well recognized. However, until this report from Diaz and colleagues, there had been little information available regarding the prognostic value of resting heart rate in the setting of coronary artery disease (CAD).

This study, focusing on subjects with stable CAD, was made possible by the existence of the Coronary Artery Surgery Study (CASS), a multicenter program involving a large registry of patients who underwent coronary angiography for investigation of suspected or proven CAD and subsequent medical or surgical treatment. The total pool of patients was large (24,999, consisting of 18,894 men and 6,065 women) and of these 24,913 were included in this study, with a median follow-up time close to 15 years. For analysis, resting heart rate was divided into quintiles with the top and bottom quintiles being ≤62 beats/min and ≥83 beats/min, respectively. Patients in the top heart rate quintile were found to be at significantly greater risk for both all-cause mortality and cardiovascular mortality (hazard ratio [HR], 1.32 and 1.31, respectively) than those in the bottom quintile. Furthermore, high resting heart rate was found to have a significant association with cardiovascular rehospitalization.

What is particularly notable is that the prognostic value of high heart rate for mortality was maintained when other risk factors/variables were taken into account (hypertension, diabetes, smoking, number of diseased coronary vessels, ejection fraction, recreational activity, body mass index [BMI]—in the case of cardiovascular mortality—and treatment with antiplatelets, diuretics, β-blockers, and lipid-lowering drugs). It is also noteworthy that in this study the predictive relationship between heart rate and mortality was similar for men and women. With such a large cohort, it is reasonable to conclude, therefore, that in CAD patients a high resting heart rate is an independent risk factor for both cardiovascular and overall mortality.

If an abnormally high resting heart rate merits treatment to bring the rate into an acceptable range, the question arises as to where the upper limit for a normal resting heart rate range stops and the lower limit of “too high” starts? Of course, this question presupposes the existence of such a cutoff as opposed to a continuum of risk. However, from the cutoff value used for the top quintile by Diaz and colleagues, 83 beats/min would clearly appear too high, and Palatini, in an excellent editorial accompanying the paper (pages 943-945 of the same journal), comments that this is concordant with previous results found in different clinical settings and suggests that resting values >80-85 beats/min should be considered abnormal.

It is too early to label this study a “classic,” but it surely has the potential to become that. The ability to make treatment decisions based on accurate risk assessment is clearly of paramount importance, and this study provides compelling evidence that in stable CAD, resting heart rate, which is a straightforward and inexpensive variable to measure, has powerful predictive value and should therefore not be overlooked.

Mark Felt, a former top FBI official, admits to having leaked information to the Washington Post about the 1972 Watergate break-in; Russian billionaire Mikhail Khodorkovsky, founder of the Yukos oil company, is sentenced to 9 years in prison for fraud, embezzlement, and tax evasion; and French Prime Minister Jean-Pierre Raffarin steps down days after voters reject a new constitution for Europe in a referendum.
### Bibliography of One Hundred Key Papers

selected by Michael J. Shattock,* PhD; Michael R. Rosen,† MD, PhD

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