Phosphoinositide signalling and cardiac arrhythmias

Elizabeth A. Woodcock1*, Peter M. Kistler1, and Yue-Kun Ju2

1Molecular Cardiology Laboratory, Baker IDI Heart and Diabetes Institute, PO Box 6492, St Kilda Road Central, Melbourne, 8008 Victoria, Australia; and 2Department of Physiology, University of Sydney, 2006 NSW, Australia

Received 29 August 2008; revised 13 October 2008; accepted 15 October 2008; online publish-ahead-of-print 20 October 2008

Time for primary review: 33 days

KEYWORDS

PIP2; Ins(1,4,5)P3; K+ channels; Ischaemia/reperfusion; Heart failure; Atrial fibrillation

Arrhythmias arise from a complex interaction between structural changes in the myocardium and changes in cellular electrophysiology. Electrophysiological balance requires precise control of sarcolemmal ion channels and exchangers, many of which are regulated by phospholipid, phosphatidylinositol(4,5)bisphosphate. Phosphatidylinositol(4,5)bisphosphate is the immediate precursor of inositol(1,4,5)trisphosphate, a regulator of intracellular Ca2+ signalling and, therefore, a potential contributor to arrhythmogenesis by altering Ca2+ homeostasis. The aim of the present review is to outline current evidence that this signalling pathway can be a player in the initiation or maintenance of arrhythmias.

Cardiac arrhythmias arise secondary to a complex interplay between the electrophysiological properties of cardiac myocytes and structural substrate providing an obstacle to impulse propagation. Although there have been significant advances in our understanding of the mechanisms of cardiac rhythm disturbance, atrial and ventricular arrhythmias still contribute significantly to morbidity and mortality. Sudden cardiac death due to ventricular fibrillation affects 1 in 1000 people and accounts for 10–20% of all deaths in western society.1 Atrial fibrillation is the most common arrhythmia presenting at cardiology departments worldwide, and the incidence is increasing with the aging of the population.2 A better understanding of the cellular mechanisms will guide future directions and provide new therapeutic options. The phosphoinositides are emerging as potential arrhythmogenic factors, with both membrane phospholipids and soluble signalling molecules potentially involved.

1. Phosphoinositide signalling, a brief overview

The inositol phospholipids form the structural basis for a complex interplay of signalling responses initiated, most commonly, by receptor activation and resulting in changes in Ca2+, protein kinase cascades, and ion channel/exchanger activity. Phosphatidylinositol (PI) itself is a minor phospholipid constituent of all eukaryote plasma membranes. PI is unusual in that it is phosphorylated, most commonly first on the 4- and then on the 5-position to generate PIP2, the central player in inositide signalling5,4 (Figure 1). Phosphoinositide-derived second messengers regulate responses ranging from immediate changes in vascular tone and hormone secretion to more prolonged responses such as cell growth and differentiation that require transcriptional changes. This wide range of downstream responses is made possible, in part, by the multiple signalling molecules generated from phosphoinositides5-9.

Stimulation of appropriate cell surface receptors leads to activation of PI-specific phospholipase C (PLC) enzymes that hydrolyse PIP2 to generate the hydrophilic acidic end-group inositol(1,4,5)trisphosphate (Ins(1,4,5)P3) and the neutral lipid sn-1,2-diacylglycerol (DAG)5 (Figure 1). In cardiomyocytes, this response is most commonly associated with seven transmembrane receptors that bind α1-adrenergic agonists, endothelin, purine nucleotides, or angiotensin, coupled to the Gq family of heterotrimeric G proteins that activate PLCβ isoforms.10,11 In addition, receptor tyrosine kinases can activate PLCγ subtypes in response to growth factors.12 Ins(1,4,5)P3 is well established as a regulator of intracellular Ca2+ by binding its own receptors (IP3-R), intracellular Ca2+ release channels situated on Ca2+ stores in the endoplasmic reticulum, sarcoplasmic reticulum (SR), or nuclear envelope.13 DAG remains within the membrane phase and is a co-activator of conventional protein kinase C subtypes.14 Recent evidence also shows that DAG can activate some of the canonical transient receptor potential (TrpC) channels, independently of PKC.15 In addition to these PLC-generated products, PIP2 is also the precursor of PIP3 following 3-phosphorylation by PI 3-kinases16 (Figure 1). In addition to PLC cleavage and 3-phosphorylation, PIP2 can also be hydrolysed by phospholipase D enzymes, generating phosphatidic acid,17 itself an activator of critical signalling pathways.

*Corresponding author. Tel: +61 3 8532 1255; fax: +61 3 8532 1100. E-mail address: liz.woodcock@baker.edu.au

Published on behalf of the European Society of Cardiology. All rights reserved. © The Author 2008. For permissions please email: journals.permissions@oxfordjournals.org.
intermediates. Furthermore, PIP2, itself, localizes many central signalling proteins to the plasma membrane, and is a regulator of critical ion channels/exchangers (see below). Any or all of these metabolites have the capacity to influence cardiomyocyte electrical activity and thus could contribute to arrhythmogenesis. However, a specific contribution to arrhythmogenesis has been suggested only for Ins(1,4,5)P3 and PIP2, and therefore these two molecules will be considered here.

2. Arrhythmic activity associated with Ins(1,4,5)P3

Ins(1,4,5)P3 generated at the cell surface binds its own receptors (IP3-R) on intracellular Ca2+ stores generating Ca2+ signals that subsequently activate protein kinase cascades or contribute to Ca2+-induced Ca2+ release (CICR), further increasing Ca2+ responses. Ins(1,4,5)P3 causes Ca2+ release from its own receptors independently of the CICR orchestrated by the ryanodine receptors (RyR). However, IP3-R, like RyR, mediate CICR and so any IP3-R localized close to RyR might be expected to further enhance Ca2+ responses. In atrial myocytes, there is evidence that activation of type 2 IP3-R (IP3-R(2)) contributes to excitation contraction coupling by enhancing Ca2+ transients. Such a secondary mechanism influencing Ca2+ signals could interfere with the highly orchestrated Ca2+ responses mediated by the SR RyR and thereby might predispose to arrhythmia. It has been reported that the IP3-R(2) can initiate ectopic Ca2+ transients, a potential source of ectopic beats. It has also been suggested that any IP3-R(2) located close to the sarcolemma, by causing local Ca2+ signals, could interfere with voltage-regulated Ca2+ channels to shorten action potential duration (APD) or could activate Na+/Ca2+ exchange to enhance Na+ entry. IP3-R and the Ca2+ puff generated following Ins(1,4,5)P3 activation are shown in Figure 2 in relation to RyR and ion channels and exchangers.

IP3-R are expressed at very low level in cardiomyocytes, 1/50 to 1/100 of the RyR that mediate beat-to-beat changes in Ca2+ to sustain rhythm. Furthermore, most studies report that these are concentrated at the nuclear envelope, seemingly distant from the site of generation of Ins(1,4,5)P3 at the sarcolemma and from the RyR on the SR. It is difficult to envisage how nuclear receptors could contribute to RyR-initiated transients or perturb sarcolemmal ion channels/exchangers. Addition of Ins(1,4,5)P3 causes localized Ca2+ puffs in the perinuclear region, but these are small, unlikely to empty Ca2+ stores and unlikely to influence RyR function. Despite this, there have been reports of Ins(1,4,5)P3-mediated enhanced Ca2+ signalling, inotropy, and arrhythmias in ventricular myocytes from some species, although not in others. It is currently not clear whether this reflects an undetectable number of strategically placed IP3-R present on SR Ca2+...
stores in ventricular myocytes of responding species. IP3-R expression is higher in atrial than in ventricular myocytes and the existence of sub-sarcolemmal IP3-R has been reported, leading to the suggestion that atrial rather than ventricular arrhythmias might be associated with perturbations in Ins(1,4,5)P3 signalling. In support of this hypothesis, deletion of the IP3-R(2) gene in murine atrial myocytes prevented endothelin-1-induced Ca2+ transients, which are known to be arrhythmogenic.  

IP3-R expression in conducting myocytes is higher than in the working myocytes and the IP3-R subtype is different; type 2 in working myocytes and type 1 in conducting tissue. Furthermore, some IP3-R(1) are localized close to the sarcolemma in conducting myocytes. Thus, it is possible that ventricular arrhythmias apparently associated with Ins(1,4,5)P3 derive primarily from the conducting tissue. This is difficult to prove directly, but the generation of mice with knock-outs of the IP3-R subtypes in heart will provide definitive evidence for or against this hypothesis.
Table 1  Cardiac ion channels and exchangers regulated by PIP2

<table>
<thead>
<tr>
<th>Current</th>
<th>PIP2 effect</th>
<th>Channel protein</th>
<th>Gene</th>
<th>Ion</th>
<th>Function</th>
<th>Rel. to arrhythmia</th>
<th>Associated disease</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>I_{ki}</strong> (inward rectifier K current)</td>
<td>Channel opening</td>
<td>K_{ir}2.1</td>
<td>KCNJ2</td>
<td>K⁺</td>
<td>Maintain resting membrane potential</td>
<td>VF</td>
<td>Andersen–Tawil syndrome</td>
<td>123,124</td>
</tr>
<tr>
<td><strong>I_{KACH}</strong> (the acetylcholine-activated K current)</td>
<td>Modify the channel interaction with G protein βγ subunits Decreases the channel affinity for ATP</td>
<td>K_{ir}3.1/4 (GIRK1/4)</td>
<td>KCNJ3/5</td>
<td>K⁺</td>
<td>In response to parasympathetic stimulation</td>
<td>Perpetuation AF and involved in AF remodelling</td>
<td>AF, VF</td>
<td>125</td>
</tr>
<tr>
<td><strong>I_{KATP}</strong></td>
<td></td>
<td>K_{ir} 6.2 and SUR2A</td>
<td>KCNJ11</td>
<td>K⁺</td>
<td>The channel close by intracellular ATP. Metabolic and mechanosensitive repolarization reserve</td>
<td>AF, VF</td>
<td>Sudden cardiac death (vein of Marshall adrenergic atrial fibrillation)</td>
<td>125</td>
</tr>
<tr>
<td><strong>I_{Kr}</strong> (the rapidly activating delayed rectifier K current)</td>
<td>Hyperpolarizing shifts in the voltage dependence of activation and slows deactivation. Also influence responses to cAMP and PKA prevents endogenous channel inhibition and suppress the channel rundown</td>
<td>HERG (Kv 11.1) and mink/ MiRP</td>
<td>KCNH2, KCNE1/2</td>
<td>K⁺</td>
<td>Promote repolarization and maintaining pacemaker automaticity</td>
<td>Polymorphic ventricular tachycardia (torsades de pointes; TdP) ventricular fibrillation (VF)</td>
<td>Type 2 congenital long QT syndrome (LQT-2)</td>
<td>60</td>
</tr>
<tr>
<td><strong>I_{ks}</strong> (the slowly activating delayed rectifier K current)</td>
<td>Repolarization and determine heart rate-dependent shortening of APD and contribute to the slow diastolic depolarization of pacemaker AP</td>
<td>Kv7.1 and minK</td>
<td>KCNQ1, KCNE1</td>
<td>K⁺</td>
<td></td>
<td></td>
<td>Cardiac arrest AV block</td>
<td>LQT-1, LQT5 Jervell and Lange–Nielsen syndrome</td>
</tr>
<tr>
<td><strong>I_{f}</strong> (hyperpolarization-activated cation current)</td>
<td>Shifts the voltage dependence towards depolarized potentials</td>
<td>HCN</td>
<td>HCN4/2</td>
<td>Na⁺, K⁺</td>
<td>Spontaneously diastolic depolarization</td>
<td>yes</td>
<td>AF, sick sinus syndrome</td>
<td>126</td>
</tr>
<tr>
<td><strong>I_{Na/Ca}</strong> (Na⁺/Ca²⁺ exchanger current)</td>
<td>Preventing auto-inhibition by binding to exchanger-inhibitory peptide</td>
<td>NCX</td>
<td>NCX1</td>
<td>Na⁺, Ca²⁺</td>
<td>Ca²⁺ extrusion Ca²⁺ uptake Spontaneously diastolic depolarization Pacemaker current? Stretch activated current?</td>
<td>yes</td>
<td>I/R</td>
<td>75, 76</td>
</tr>
<tr>
<td><strong>SOC/ROC</strong> (store or receptor-operated Ca channels)</td>
<td>Inhibits?</td>
<td>TrpC</td>
<td>Trp (C1-7)</td>
<td>Ca²⁺, Na⁺, K⁺, or others</td>
<td></td>
<td>Not known</td>
<td>?</td>
<td></td>
</tr>
<tr>
<td><strong>NSC_{Ca}</strong> (Ca activated non-selective cation current)</td>
<td>Depletion of PIP2 desensitizes the channel</td>
<td>TrpM4</td>
<td>TrpM</td>
<td>Na⁺, K⁺</td>
<td>Depolarization</td>
<td>Delayed after-depolarization</td>
<td>Cardiac hypertrophy</td>
<td>127, 128</td>
</tr>
</tbody>
</table>
myocytes are exposed to metabolic stress, $K_{ATP}$ channels open, causing action potential shortening and contractile dysfunction.

$K_{IR}$ channels are tetramers, with each subunit comprising two transmembrane segments and a pore loop, that together form a transmembrane pore. The four cytoplasmic loops from each subunit in the tetrameric channel form a girdle around the central cytoplasmic pore.48 This structure forms a flexible diffusion barrier between the cytoplasm and the transmembrane pore46,49 (Figure 3). Opening of $K_{IR}$ channels requires PIP2 binding to basic and polar amino acids in cytoplasmic domains, whereas depletion of PIP2 acts to close the channel.50 Mutations in $K_{IR}$ 2.1 channel proteins resulting in lowered PIP2 binding affinity are a cause of Anderson’s syndrome, a condition associated with ventricular arrhythmias,51 demonstrating the importance of PIP2 in cardiomyocyte electrophysiology and arrhythmogenesis.

$K_{IR3}$ channels ($K_{ACh}$) in atria and sino-atrial node belong to the G protein regulated inward rectifying K+ channel family (GIRK),49,52 which are regulated via activation of the heterotrimeric G protein, Gi, causing release of Gbγ subunits. Regulation of $K_{ACh}$ channel activity is crucially dependent on PIP2.53 Blockade of PIP2 binding to channels retards the stimulatory effects of Gbγ or Na+ ions on channel activity. Such effects can be reversed by restoring PIP2. Mutant channels that interact weakly with PIP2 do not open under control conditions, but can be activated when the interaction with PIP2 is strengthened by adding Gbγ.53

PIPs dramatically decreases the apparent affinity of $K_{ATP}$ channels ($K_{IR6}$) for ATP and thus influences responses to metabolic challenge.54 Most $K_{IR}$ channels are relatively specific for PIP(4,5)P2 over other positional isomers of PIP2, but the $K_{ATP}$ channels are relatively non-specific, responding to 3,4- and 3,5-PIP2 as well as the 4,5-isomer and even to PIP3.55 The relative non-specificity of the $K_{ATP}$ channels means that they can be activated by lipids other than the inositol phospholipids, particularly long chain fatty acyl CoA derivatives55,56 and these act similarly to PIP2.57 This may be of considerable functional importance as the content of these fatty acid derivatives can be manipulated by dietary lipid intake and can change under different metabolic or pathological conditions. Thus, long chain fatty acyl CoA derivatives, as well as ATP may serve as metabolic regulators of $K_{ATP}$ channels, and this regulation will be influenced by the availability of sarcolemmal inositol phospholipids. Long chain fatty acyl CoA derivatives are elevated in type 2 diabetes and the mechanism outlined may contribute to cardiac complications of this disease.57 It has also been reported that $K_{ATP}$ channels are less selective than GIRK channels in terms of the fatty acid residues constituting the PIP2 molecule, with GIRK channels, but not $K_{ATP}$ channels, showing strong preference for arachidonyl, stearyl PIP2.56 Again, this suggests the possibility of subtype-selective dietary influence on channel activity, as it is known that fatty acid intake can alter the lipid composition of PIP2.58

3.3 Repolarizing K+ Channels

The repolarization phase of the action potential is mediated by voltage-regulated K+ channels (Kv), in particular by Kv11.1 (human ether a go, HERG) responsible for the rapid phase of repolarization and Kv 7.1 (KCNQ1/KCNE1), which causes slow repolarization. Both Kv11.1 and Kv 7.1 are activated by PIP2, although its interaction is less well studied than for the $K_{IR}$ channels.59–61 Mutations in either of these channels have been shown to be responsible for inherited arrhythmias, particularly long Q–T syndrome (LQT), and one mutant in the Kv 7.1 channel causes short QT.62–64 At least in Kv 7.1, some of these mutants are in residues likely to be important for PIP2 interactions.63 Both Kv11.1 and Kv 7.1 channels have six trans-membrane spanning regions that form an ion pore, together with a long C-terminal tail and a relatively short cytosolic N-terminal tail (Figure 3). HERG channels are regulated by PKA phosphorylation of C-terminal residues and by direct cAMP binding to motifs present in the C-terminal tail. The phosphorylated protein associates with 14-3-3 proteins and this interaction is central to the heightened activity.65 There are potential PIP2 interaction sites on both sides of the cAMP-interaction domain,66 and it is likely that PIP2 can influence responses to cAMP and PKA. In the HERG channel, PIP2 causes hyperpolarizing shifts in the voltage dependence of activation and also slows deactivation,66.
and loss of PIP2 can explain channel deactivation, following activation of PLC-coupled receptors.67

The slow phase of repolarization, the IKS current, is mediated by Kv7.1 channels comprising a heterodimer of a six transmembrane spanning protein, KCNQ1, and the smaller KCNE1 molecule with one membrane spanning domain. The functional channel also requires A-kinase anchoring protein 9 (AKAP9), also known as yotiao, that modulates activation by protein kinase A.68 PIP2 is required for Kv7.1 activity, and the PIP2 binding site is in the N-terminal cytosolic region of the protein. The PIP2-binding sequence is part of an endogenous inhibitory region on KCNQ1, and PIP2 binding prevents this inhibition (Figure 3).69 Mutations in critical arginine residues involved in PIP2 binding that cause reduced PIP2 affinity have been shown to be a cause of inherited LQT.70 In patch clamp studies, addition of excess PIP2 reversed the lowered activity of the mutant channels and returned channel activity to normal, further confirming the importance of PIP2 binding for the optimal functioning of the channel71 and raising the possibility that changes in PIP2 availability could initiate arrhythmia.

3.4 Pacemaker channels
PIP2 also regulates the pacemaker (Ih) current by regulating the hyperpolarization-activated cyclic nucleotide gated channels (HCN). PIP2 shifts the voltage dependence of the pacemaker IKh channels towards depolarized potentials and thus increases the spontaneous firing rate,49,70 put the molecular basis for this is not yet known. These channels have been suggested to be important in the development of AF,71 but there is currently no evidence that this involves PIP2.

4. Arrhythmic responses possibly involving Ins(1,4,5)P3 or PIP2
4.1 Na+/Ca2+ exchanger
The Na+/Ca2+ exchanger NCX1 is a large membrane protein with nine transmembrane sequences and a long cytosolic loop between transmembrane sequences 4 and 5, which include a sequence, known as the exchanger inhibitory peptide (XIP), that serves to auto-inhibit channel activity (Figure 3).72 NCX1 contributes to arrhythmia under a number of pathological circumstances. Reverse mode (Ca2+ entry) NCX1 is activated following Na+ influx during ischaemia/reperfusion73 and NCX activity is required for arrhythmias in early reperfusion.74-76 Increased expression of NCX1 has been reported in heart failure in some studies involving clinical samples and in some experimental models.75,77 PIP2 activates NCX1 activity by binding the XIP sequence and preventing auto-inhibition.72,78 Importantly, long chain fatty acyl CoA derivatives can substitute for PIP2 as activators of NCX1.79 As saturated fatty acyl CoA derivatives are most effective NCX1 activators, this suggests another possible mechanism whereby diet could influence predisposition to arrhythmia. In addition to activation by PIP2, possible effects of localized Ca2+ signals on sarcolemmal NCX1 are often suggested as a mechanism of Ins(1,4,5)P3 arrhythmogenesis, as noted above.

4.2 Canonical transient receptor potential channels
TrpC channels are low conductance, relatively non-selective cation channels activated by receptors coupled to PLC.80-82 TrpC channels are regulated by stretch, DAG, and IP3-R-mediated Ca2+ store depletion.80 TrpC3, 6, and 7 are activated directly by DAG,83,84 whereas TrpC4 and 5 are regulated by PLC, independently of DAG formation.85 TrpC4 can be inhibited by PIP2 binding86 and this may explain the activation by PLC as this would be expected to cause PIP2 depletion. On the other hand, TrpC7 and human TrpC6 channels are activated by PIP2, which enhance store-operated Ca2+ entry.87 In these cases, PLC-induced depletion of PIP2 would oppose any stimulatory effect of Ins(1,4,5)P3-induced Ca2+ store depletion. A number of interesting properties of this channel family suggest a potential role in causing electrophysiological imbalance. TrpC3 is physically associated with NCX1 in cardiac sarcomplasma and Ca2+ entry via TrpC3 may partly depend on reverse mode NCX1.88,89 TrpC6 is activated directly and selectively by α1A-adrenergic receptors and these have previously been implicated in arrhythmogenesis.90,91 TrpC1 is a stretch-activated channel in heart,92 and stretch is associated with arrhythmia.93,94 Furthermore, TrpC channels, by causing localized Ca2+ increases in the subsarcomemral region, might interfere with the functioning of the voltage-regulated Ca2+ channels or NCX1, as proposed earlier for Ins(1,4,5)P3-induced Ca2+ release. TrpC3 protein is expressed in the surface membrane of single pacemaker cells from mouse heart and a store-operated Ca2+ influx in pacemaker tissue has been demonstrated. Blocking this Ca2+ influx slowed pacemaker firing rate.95 These studies suggest that TrpC3 could be an important pacemaker current modulated by Ins(1,4,5)P3 or PIP2, and could contribute to arrhythmias. In addition, it has been reported that the closely related TrpM4 channel is a calcium-activated non-selective cation channel, which is activated by Ins(1,4,5)P3 in mouse sino-atrial node.96

4.2.1 Ischaemia and reperfusion
A number of laboratories have reported that ischaemia and post-ischaemic reperfusion are associated with heightened Ins(1,4,5)P3 generation compared with responses under normoxic conditions.97-102 Evidence for an association between this Ins(1,4,5)P3 response and arrhythmogenesis was provided by use of inhibitors of PLC to reduce Ins(1,4,5)P3 and arrhythmia, most importantly ventricular fibrillation (VF), in parallel.103,104 Such studies can be questioned because all inhibitors of PLC are notoriously non-specific and in particular the aminoglycosides bind PIP2 and interfere with channel regulation as well as Ins(1,4,5)P3 generation.105 However, the PLC inhibitor U-73122 was found to inhibit Ins(1,4,5)P3 generation only when this was caused by thrombin receptor (PAR1) activation. Importantly, only thrombin-induced VF was prevented by U-73122, providing support for PLC activity as being critical for arrhythmogenesis under these conditions.104 The heightened Ins(1,4,5)P3 generation during post-ischaemic reperfusion would be expected to be accompanied by a lowering of PIP2 in the vicinity of the PLC. Such loss of PIP2 could cause changes in the activity of critical ion channels and exchangers as described above. However, our studies showed that PIP2 actually increased, rather than decreased, in early
post-ischaemic reperfusion and that this increase was confined to caveolar fractions. Such increases in PIP₂, if they occur in the vicinity of repolarizing K⁺ channels, potentially could lead to reduced APD.

The situation in ischaemia is more complex. Enhanced PLC responses have been reported, but our studies showed that ischaemia caused degradation of inositol phosphates, including Ins(1,4,5)P₃ in both intact heart and cardiomyocyte models. However, despite this, Ins(1,4,5)P₃ responses to agonist were enhanced, rather than diminished. Ischaemia also causes rapid loss of PIP₂ and its immediate dephosphorylation product PI(4)P. It remains to be established whether changes in Ins(1,4,5)P₃ or PIP₂ contribute to arrhythmia under ischaemic conditions.

5. Heart failure

Heart failure is the endpoint of a number of cardiac pathologies and thus can be considered to embody a number of different diseases. The failing heart is often enlarged, dilated, and fibrosed and all of these factors will predispose to arrhythmia by increasing the likelihood of re-entry mechanisms and facilitating rotor initiation and perpetuation. However, in addition, some of the cellular changes associated with heart failure may also contribute to arrhythmogenesis. Increased expression of NCX1 is common in human heart failure and in arrhythmogenic animal models. As outlined above, increased NCX1 would be expected to exacerbate any arrhythmic activity of either Ins(1,4,5)P₃ or PIP₂, as both of these can directly or indirectly influence exchange functioning. Increased expression of IP₃-R has also been reported in human heart failure and in a rabbit model. Furthermore, the failing ventricle undergoes a loss of t tubules, becoming, in that respect, more atrial-like, opening up the possibility of a greater contribution of Ins(1,4,5)P₃ and IP₃-R to Ca²⁺ regulation, as outlined earlier. Myocytes from failing hearts show AP prolongation that to some extent mimics LQT, associated with alterations in repolarizing K⁺ currents. However, there is currently no evidence that the altered channel activity involves PIP₂.

6. Atrial fibrillation

AF is associated with a number of different cardiac pathologies and can be initiated and sustained by a range of different mechanisms. Relatively little is known about the mechanisms involved at the cellular level and there is currently no direct evidence for an involvement of Ins(1,4,5)P₃ or PIP₂. Heightened IP₃-R expression in right atrial tissue from patients with AF was reported in one study, but in contrast, another study investigating patients with VHD reported reduced IP₃-R expression in right atrial tissue. Mutations in KCNQ1 can cause AF, but the mutations described to date do not involve PIP₂ binding. The recent development of mouse models of AF should help in defining mechanisms and identifying any involvement of Ins(1,4,5)P₃ as an anti-arrhythmic strategy has theoretical advantages. In practice this may be difficult. IP₃-R blockers are notoriously non-specific and currently known PLC inhibitors are only minimally effective, have many off target effects and are often poorly tolerated. However, recent studies in our laboratory have shown that responses to Gq-coupled receptor agonists in cardiomyocytes involve primarily only one splice variant of one PLCβ subtype (PLCβ1b). As PLCβ1b is the nuclear PLC subtype in many other cell types, this raises the possibility of inhibiting PLC in a cardiac-specific manner, by interfering with the binding of the C-terminal of PLCβ1b to the sarcolemma.

The many different functions that depend on PIP₂ make this an unlikely candidate for drug development. However, a number of reports suggest ways in which the interaction between PIP₂ and ion channels might be a start point for targeted anti-arrhythmic agents. As noted earlier, acyl-CoA derivatives of long chain fatty acids can compete with PIP₂ for binding Kᵢ channels, especially Kir 6.0, or NCX1. Such association reverses any effect of PIP₂. Therefore, while manipulation of PIP₂, itself, would be impossible or inadvisable, it might be possible to design molecules that specifically reduce PIP₂ binding to particular channels/exchangers. However, such approaches would require knowledge of the underlying defect. Another possible approach would be to target specific subtypes of PI(4,5)P₂ if the subtype responsible for generating the appropriate PIP₂ pool can be identified.

Acknowledgements

We thank Theresa Filtz (Department of Pharmacology, Oregon State University, USA) for critical reading of the manuscript.

Conflict of interest: none declared.

Funding

This work was supported by the Australian National Health and Medical Research Council nos 418935 and 317803 and by an award from the Baker IDI Heart and Diabetes Institute.

References


Downloaded from https://academic.oup.com/cardiovascres/article-abstract/82/2/286/276356 by guest on 30 April 2018
Phosphoinositides and arrhythmias


35. Mao YS, Yin HL. Regulation of the actin cytoskeleton by phosphatidylinositol 4-phosphate 5-kinases. Pflugers Arch 2007;455:5–18.


60. Bian JS, McDonald TV. Phosphatidylinositol 4,5-bisphosphate interactions with the HERG K⁺ channel. Pflugers Arch 2007;455: 105–113.


121. Wu J, Takeo T, Suga S, Kanno T, Osanai T, Mikoshiba K et al. 2-aminoethoxypipdihenyl borate inhibits agonist-induced Ca\textsuperscript{2+} signals by blocking inositol trisphosphate formation in acutely dissociated mouse pancreatic acinar cells. *Pflugers Arch* 2004; 448:592–595.


