



Review

Cardiac dynamics: Alternans and arrhythmogenesis

Gary Tse, BA Hons MBBS MA PhD^{a,*}, Sheung Ting Wong, BSc^b, Vivian Tse^c, Yee Ting Lee^a, Hiu Yu Lin^a, Jie Ming Yeo, MBBS BSc^b

^a School of Biomedical Sciences, Li Ka Shing Faculty of Medicine, University of Hong Kong, Hong Kong

^b Faculty of Medicine, Imperial College London, SW7 2AZ London, UK

^c Department of Physiology, McGill University, Canada

ARTICLE INFO

Article history:

Received 21 December 2015

Received in revised form

28 January 2016

Accepted 22 February 2016

Available online 28 March 2016

Keywords:

Cardiac arrhythmia

Dynamics

Re-entry

Restitution

Alternans

ABSTRACT

Pre-existing heterogeneities present in cardiac tissue are essential for maintaining the normal electrical and mechanical functions of the heart. Exacerbation of such heterogeneities or the emergence of dynamic factors can produce repolarization alternans, which are beat-to-beat alternations in the action potential time course. Traditionally, this was explained by restitution, but additional factors, such as cardiac memory, calcium handling dynamics, refractory period restitution, and mechano-electric feedback, are increasingly recognized as the underlying causes. The aim of this article is to review the mechanisms that generate cardiac repolarization alternans and convert spatially concordant alternans to the more arrhythmogenic spatially discordant alternans. This is followed by a discussion on how alternans generate arrhythmias in a number of clinical scenarios, and concluded by an outline of future therapeutic targets for anti-arrhythmic therapy.

© 2016 Japanese Heart Rhythm Society. Published by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

Contents

1. Introduction	411
2. Cardiac alternans	412
2.1. APD restitution-dependent mechanisms	412
2.2. APD restitution-independent mechanisms	413
3. Spatially concordant and discordant alternans	414
3.1. Pre-existing tissue heterogeneities	414
3.2. Dynamic factors	414
4. Alternans and arrhythmogenesis in different clinical conditions	415
5. Future therapies	416
Conflict of interest	416
Acknowledgments	416
References	416

1. Introduction

Pre-existing heterogeneities present in cardiac tissue are essential for maintaining the normal electrical and mechanical functions of the heart. However, an increased risk of cardiac arrhythmias can result from the exacerbation of such heterogeneities, which can occur

under pathological conditions or following the administration of cardiotoxic drugs. The emergence of dynamic factors, which can interact with each other as well as with pre-existing tissue heterogeneities, can produce arrhythmogenic repolarization alternans and therefore cardiac arrhythmias. The focus of this review is to illustrate the mechanisms that (i) generate cardiac alternans, (ii) convert spatially concordant alternans to the more arrhythmogenic spatially discordant alternans, and (iii) are responsible for the production of arrhythmias in a number of clinically relevant conditions. This is

* Corresponding author. Tel.: +852 39177548; fax: +852 2817 0857.

E-mail address: gary.tse@doctors.org.uk (G. Tse).

concluded by a discussion on future therapeutic targets for anti-arrhythmic therapy.

2. Cardiac alternans

Cardiac alternans are beat-to-beat oscillations in either arterial pulse or electrocardiographic QRS and T waves. Of these, T-wave alternans (TWAs) have been associated with re-entrant arrhythmogenesis and identified as a good predictor of sudden cardiac death [1]. They are due to alternations in repolarisation time courses (measured as action potential durations, APDs) at the cellular level, which increase in amplitude with faster heart rates. TWAs have been observed in a number of conditions, including electrolyte abnormalities, hypothermia, coronary artery disease, post-myocardial infarction, long QT and Brugada syndromes, vasospastic angina, dilated, hypertrophic, and Takotsubo cardiomyopathies, and end-stage heart failure.

2.1. APD restitution-dependent mechanisms

The relationships between the diastolic interval (DI), APD, and basic cycle length (BCL) are shown in Fig. 1. BCL is the sum of APD and DI. The mechanism of APD alternans was first described by using a graphical method, relating them to APD restitution [2]. This refers to the normal shortening of APD in response to faster heart rates, and is thought to be an adaptive mechanism for preserving diastole at such rates. It can be defined as the dependence of APD on the previous DI. Experimentally, this can be determined by using an S1S2 protocol, which gradually shortens the interval between the S1 and S2 stimuli, or by using a dynamic pacing protocol, which increases the heart rate by progressively reducing the BCL. While both methods can be used to measure APD restitution [3], the S1S2 restitution curve is a measure of the immediate response to a change in BCL, whereas the dynamic restitution curve is a measure of the steady-state response [4]. Fig. 2 shows a typical APD restitution curve obtained from mouse hearts, $APD_{n+1} = f(DI_n)$, where f is the function relating the new APD to its previous DI. The dashed line indicates the gradient of the curve and the gray area refers to values of DIs with gradients greater than one.

The gradient of the restitution curve is a collective measure of the recovery of all the ion channels opened during the cardiac action potential. First, of these channels, sodium channels recover from inactivation rapidly, and therefore their effects on APD restitution occur mainly at short DIs, between 0 and 40 ms in human hearts. However, if the recovery of sodium channels is slowed, which can occur under ischemic conditions [5], their effects on APD restitution would be extended to longer DIs. Second, the L-type calcium channels recover more slowly than sodium channels, and their effects are therefore observed in the short and intermediate DI ranges, between 0 and 100 ms. These calcium channels provide the majority of the inward current during the plateau phase of the action potential and therefore exert major effects on APD restitution. Their inhibition leads to reduced gradients of APD restitution curves. Third, time-dependent potassium channels,

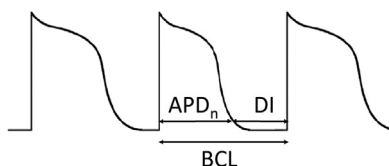


Fig. 1. Voltage trace showing the relationships between action potential duration (APD), diastolic interval (DI), and basic cycle length (BCL).

such as the voltage-gated delayed rectifiers, show the slowest recovery compared to other ion channels and therefore their effects are observed over a much larger DI range beyond 100 ms. In addition, the block of potassium channels shows reverse use dependence, where there is less block with increasing use [6]. Thus, the block increases during phase 4 of the action potential (diastole) and decreases during the plateau phase. Consequently, potassium channel blockers, which prolong APDs, have greater effects at long BCLs (bradycardia) and long DIs (e.g. compensatory pause after an ectopic beat), but have much smaller effects at short BCLs (tachycardia) and DIs [6]. They generally increase the gradients of APD restitution curves. The steep portion of the APD restitution curve is relevant in sinus tachycardia, where the heart rate is increased. It is also relevant in heart failure or the congenital and acquired long QT syndromes. In these conditions, APD is prolonged and therefore the DIs can become short enough to engage the steep portion of the APD restitution curve even at normal heart rates.

Fig. 3 shows cobweb plots that can be used to determine the stability of APD alternans. As BCL decreases, APD also decreases and the relationship $BCL = APD + DI$ can be shown graphically as a straight line with a gradient of -1 . The equilibrium point of APD for each BCL is the intersection point of the restitution curve and this line, which has the coordinates $[DI_s, APD_s]$. The stability of APD can be determined by perturbing the DI by a small amount, δ ,

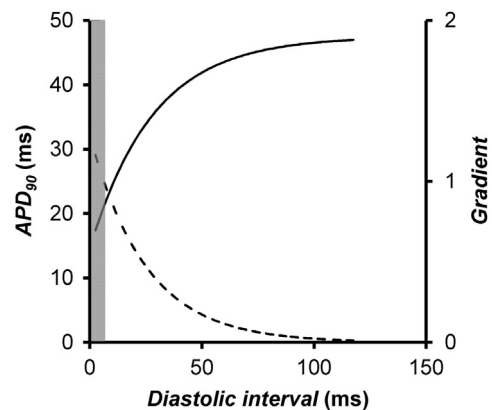


Fig. 2. An APD restitution curve describes the relationship between the APD and the previous diastolic interval (solid line). The gradients of the curve are represented by the broken line. The values of DIs at which such gradients are greater than one are represented by the gray box.

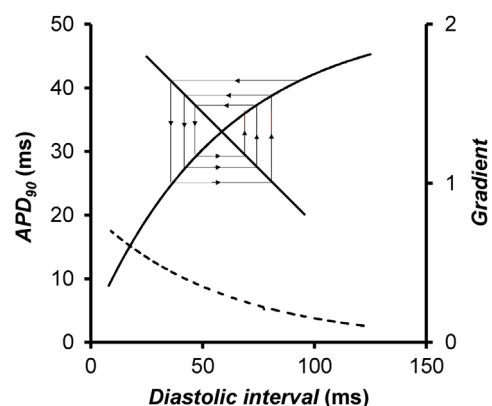


Fig. 3. APD restitution curve plotting APD against the previous DI (solid line) along with their gradients (broken line). The values of DIs with gradients greater than one are represented by the gray box. The cobweb plot shows that when the APD restitution gradient is less than one, a stable equilibrium point is produced on successive beats.

such that $DI_{n+1} = DI_n + \delta$. A negative δ that shortens the DI would move DI_{n+1} to the left. This will in turn produce a shorter APD_{n+2} . However, this will result in a long DI_{n+2} and therefore a long APD_{n+3} .

If the gradient of the APD restitution curve at the intersection with the BCL line is less than one (Fig. 3), then the alternans are

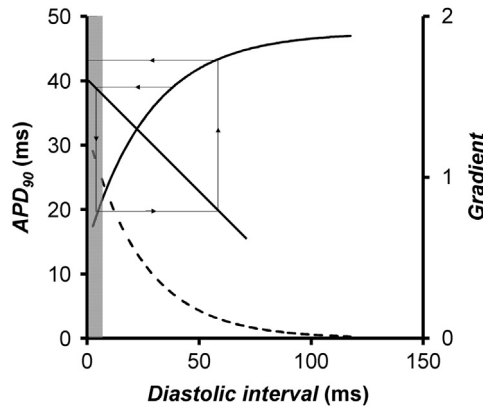


Fig. 4. APD restitution curve plotting APD against the previous DI (solid line) along with their gradients (broken line). The values of DIs with gradients greater than one are represented by the gray box. The cobweb plot shows that when the APD restitution gradient is greater than one, an unstable equilibrium point is produced on successive beats, eventually leading to conduction block.

transient and will return to the stable equilibrium point over subsequent beats. However, if this gradient is greater than one (Fig. 4), then the amplitude of the alternans will increase, eventually leading to 2:1 block in this simplified situation where the APD restitution curve is linear. As indicated in the figure, the slope of the APD restitution curve decreases with longer DIs and therefore alternans will eventually reach a maximum stable value because the flat restitution curve prevents their growth.

2.2. APD restitution-independent mechanisms

Restitution analysis assumes that alterations in DIs produce the changes in APDs, and that the gradient of the restitution curve determines the extent to which alterations in DIs produce APD alternans. However, a feedback-based protocol that permits explicit control of DI independent of APD has subsequently been developed [7,8]. Experiments using such a protocol have demonstrated APD alternans can occur during constant DI pacing [9]. This means that DIs do not necessarily have to change to produce APD alternans, which therefore have APD restitution-dependent and restitution-independent components. Therefore, it can be concluded that other factors influence this situation. Some of these factors that have been identified include cardiac memory, calcium handling dynamics, ventricular effective refractory period (VERP) restitution, and mechano-electric feedback, which will be discussed in turn (Table 1).

Table 1
APD restitution-dependent and APD restitution-independent mechanisms for producing APD alternans.

APD Alternans	Explanation	References
APD restitution-dependent	APD is dependent on the previous DI. Abrupt change in DI leads to engagement in the steep portion of the APD restitution curve.	[2]
APD restitution-independent: Cardiac memory	APD depends on not only the preceding DI but a series of DIs preceding it, i.e. the pacing history is important, which is termed APD accommodation.	[9,71] [3,11]
Calcium handling	Rate-dependent memory, termed hysteresis, results in persistence of alternans despite subsequent slowing of heart rate.	[14–18]
VERP restitution	$Ca^{2+} \rightarrow$ APD coupling	[19,63]
Mechanoelectric feedback	VERP can diverge from APD, e.g. during hypokalemia. Mechano-sensitive ion channels can influence the membrane potential	[21,22]

Table 2
Mechanisms for producing spatially concordant and spatially discordant APD alternans.

Alternans	Mechanisms	Clinical relevance	References
Spatially concordant alternans (SCAs)	Positive $Ca^{2+} \rightarrow$ APD coupling		[40]
Spatially discordant alternans (SDAs):			
Pre-existing tissue heterogeneities	Steep APD restitution	AF, Long QT syndromes, Brugada syndrome, heart failure, hypothermia	[24,62,72]
	Spatial gradients in Ca^{2+} transients	Heart failure	[27]
	Spatial gradients in VERP	Hypokalemia	[40,73]
	Gap junction uncoupling or downregulation	Oculodentodigital dysplasia, Naxos disease	[28,29,74,75]
	Abnormal sodium channel function	Heart failure, Long QT syndrome type 3	[31]
	Fibrosis	Heart failure	[30]
Dynamic factors	CV restitution	Ischemia, sodium channel blockade, hypothermia	[5,34,62]
	Steep VERP restitution	Hypokalemia	[38]
	Steep relationship between sarcoplasmic reticulum calcium release and diastolic sarcoplasmic reticulum calcium load	Heart failure, exercise, catecholaminergic polymorphic ventricular tachycardia, atrial fibrillation	[76]
	Negative $Ca^{2+} \rightarrow$ APD coupling	Heart failure	[40,38]
	Calcium accumulation in the sarcoplasmic reticulum	Heart failure, exercise, catecholaminergic polymorphic ventricular tachycardia	[41]
	Reduced repolarization reserve	Long QT syndromes	[47]
	After-depolarization phenomena	Atrial fibrillation, heart failure	[77,78]
	Ectopic beats	Heart failure, long QT syndromes	[34]
	Sympathetic stimulation	Heart failure, exercise	[61]

Cardiac memory results from the slow recovery of ion channels as well as the gradual accumulation of ions on either side of the cell membrane. Consequently, the pacing history is also important, and so APD depends not only on its immediately preceding DI but also on the series of DIs preceding it, i.e., $APD_{n+1} = f(DI_n, DI_{n-1}, DI_{n-2}, \dots)$. Second, APD depends not only on the preceding DIs but also on the previous APDs. Therefore, in the presence of these short-term memory effects, known as APD accommodation [10], APD restitution gradients greater than one may not predict the onset of APD alternans [3,11]. Nevertheless, better methods have been developed to investigate the rate- and memory-dependent aspects of APD restitution. For example, the restitution portrait was generated by modifying the pacing protocol to include a measurement of the transient response of the APD after a change in BCL [4]. This allows measurements of various aspects of APD restitution simultaneously, including dynamic restitution, S1S2 restitution, and the transient response that arises from short-term memory. Such a protocol has subsequently been used to examine restitution properties in the ventricles of perfused whole hearts [12]. Moreover, rate-dependent memory, known as hysteresis, has been demonstrated for repolarisation alternans observed in humans [13]. This refers to alternans induced by rapid pacing that persist despite subsequent slowing of the heart rate [14]. Theoretical work has shown that both restitution-dependent and independent factors are responsible, and abnormal calcium handling plays an important role [14]. Studies investigating onset of alternans should specify the initial pacing conditions to account for hysteresis.

Calcium handling dynamics, when altered, can also produce APD alternans [15]. Membrane potential and $[Ca^{2+}]_i$ are bidirectionally coupled. $I_{Ca,L}$ is a major determinant of both APD and $[Ca^{2+}]_i$. If APD alternates, $[Ca^{2+}]_i$ will also alternate in response to the alternating $I_{Ca,L}$ amplitude (APD \rightarrow Ca coupling). A longer DI allows a longer recovery time of the L-type calcium channels, which would increase the SR calcium load and therefore the calcium transient amplitude. Conversely, $[Ca^{2+}]_i$ also affects APD through its effects on the calcium-sensitive currents, such as $I_{Ca,L}$, I_{NCX} , I_{NS} , $I_{Cl,Ca}$, that are active during the plateau phase (Ca \rightarrow APD coupling). The situation is complex here because $[Ca^{2+}]_i$ can have opposing effects on APD. It shortens APD by potentiating the calcium-induced inactivation of $I_{Ca,L}$ but prolongs APD by increasing the inward current produced by I_{NCX} . The net effect of $[Ca^{2+}]_i$ on APD depends upon which of the two factors dominates. A larger Ca^{2+}_i transient causing prolonged and shortened APDs is termed positive and negative coupling, respectively [16]. Furthermore, the propensity for pacing-induced calcium alternans increases with increasing mitochondrial dysfunction through either dissipation of the mitochondrial membrane potential or inhibition of ATP synthesis [17]. What is clear is that APD alternans is an emergent property involving both voltage- and calcium-dependent mechanisms. Recent experiments using stochastic pacing demonstrated the use of a novel parameter, λ_{alt} , to reveal the onset of alternans as well as to distinguish between voltage- and calcium-driven APD alternans [18].

In situations where the VERP diverges from APD, as has been shown in hypokalemia, restitution gradients may not accurately predict the onset of alternans [19]. In this case, the gradients of VERP restitution curves may be a better indicator. An additional advantage of VERP over APD restitution is that VERP can be measured without the need to accurately record action potential waveforms from monophasic action potential (MAP) recordings.

Moreover, mechanical contraction can modulate the electrical activity of myocytes, a phenomenon referred to as mechano-electric feedback (MEF). Electrical alternans have been observed during simulated pulsus alternans produced by clamping the aorta on alternate beats [20]. In this situation, the myocytes of the vessel

wall did not actually contract and therefore changes in $[Ca^{2+}]_i$ probably did not cause these electrical alternans. It instead pointed to mechano-sensitive ion channels [21], such as volume- or stretch-activated channels (SACs), being responsible. Indeed, these mechano-sensitive ion channels can influence the membrane potential on a beat-to-beat basis and may therefore influence cardiac dynamics. Thus, preliminary evidence shows that SAC activation can have suppress spatially concordant alternans but exacerbate discordant alternans [22].

3. Spatially concordant and discordant alternans

APD alternans can be either spatially concordant or discordant (Table 2). As the pacing rate is increased, spatially concordant alternans are observed, in which APD is long throughout the cardiac tissue on one beat and short on the next beat, i.e., APDs in different regions alternate in phase with each other. When the pacing rate is increased further, spatially discordant alternans are produced, in which APD is long in one region but short in an adjacent region, and changes phase on the next beat; i.e., APD in different regions alternate out of phase with each other. A number of mechanisms have been identified as being responsible for the production of spatially discordant APD alternans. These can involve pre-existing heterogeneities, which often interact with dynamic factors to produce them. However, pre-existing tissue heterogeneities may not be necessary; the presence of dynamic factors alone may be sufficient for producing spatially discordant alternans [23].

3.1. Pre-existing tissue heterogeneities

Pre-existing tissue heterogeneities are normally present in the heart, such as spatial gradients in repolarization between the endocardium and epicardium or the base and apex. A steep APD restitution gradient can convert spatially concordant alternans to discordant alternans [24]. Like APD, Ca^{2+}_i handling in the ventricle also exhibits apex-base [25] and endocardium-epicardium [26] gradients. Since Ca^{2+}_i affects APDs, heterogeneity in Ca^{2+}_i handling can produce spatially discordant APD alternans via either differential $Ca^{2+}_i \rightarrow$ APD coupling or spatial heterogeneities in the phase of the Ca^{2+}_i alternans [27]. Moreover, electrotonic coupling between cardiomyocytes via gap junctions attenuates differences in properties between individual cells. When gap junctions are uncoupled, pre-existing heterogeneities such as spatial gradients in APDs, CVs, and Ca^{2+}_i can become amplified, leading to discordance [28,29]. Finally, under conditions where cardiac conduction is abnormal, e.g. in heart failure [30], the threshold for inducing spatially discordant alternans is lower. This may be due to fibrosis, altered expression of gap junctions, or delayed recovery of sodium channels from inactivation [31].

3.2. Dynamic factors

A number of dynamic factors can convert spatially concordant to discordant APD alternans. First, CV restitution describes the relationship between the CV and the preceding DI (Fig. 5). It is almost entirely dependent on the recovery of sodium channels because the upstroke of the action potential is mainly determined by these channels [32]. Under normal conditions, these channels recover from inactivation rapidly, and therefore CVs only slow at very short DIs, between 0 and 10 ms (Fig. 5, solid line). However, if their recovery is slowed, CVs vary over a broader range of DIs (Fig. 5, dashed line). Fast pacing can engage the steep portion of the CV restitution curve to convert pre-existing spatially concordant alternans to discordant APD alternans when the tissue size is

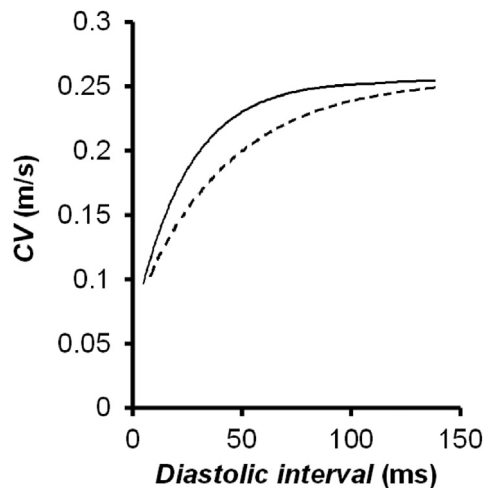


Fig. 5. CV restitution curve. CV is plotted against its previous DI in which sodium channel recovery is normal (solid curve) or slowed (dashed curve). The latter is observed in clinical situations such as tissue ischemia, after application of sodium channel inhibitors and hyperthermia.

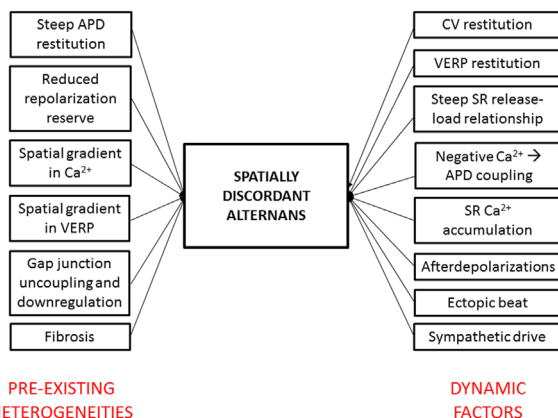


Fig. 6. Pre-existing heterogeneities or dynamic factors can convert spatially concordant APD alternans to spatially discordant APD alternans.

sufficiently large [33]. Initially, at a relatively low heart rate, spatially concordant APD alternans are observed. When the heart rate increases, a beat with a long APD has a sufficiently short DI after it. This engages CV restitution, causing the CV of the subsequent beat to decrease. The conduction slowing allows the DI to increase slightly, which in turn allows APD to increase slightly. This process amplifies itself with subsequent beats, eventually producing discordant alternans [34]. This mechanism may be important during ischemia or sodium channel blockade where sodium channel recovery is slowed [5,35]. This would extend the range of DIs over which CV varies, producing spatially discordant APD alternans even at normal heart rates [36], thereby increasing arrhythmogenic risk [32,37] in both of these conditions. Alternatively, CV restitution can also interact with steep VERP restitution or altered $[Ca^{2+}]_i$ cycling dynamics to produce discordant alternans [38] (Fig. 6).

Ca^{2+} alternans can be induced in cardiac cells by making sarcoplasmic release of calcium strongly dependent upon the SR Ca^{2+} content [39]. They can become discordant if $Ca^{2+} \rightarrow APD$ coupling is changed from positive to negative [40]. Recently, a novel mechanism for the production of spatially discordant alternans was demonstrated despite positive $Ca^{2+} \rightarrow APD$ coupling [41]. Here, rapid pacing led to Ca^{2+} accumulation in the sarcoplasmic reticulum over many beats, leading to Ca^{2+} alternans that were out-of-phase with each other [41]. Compelling evidence suggests

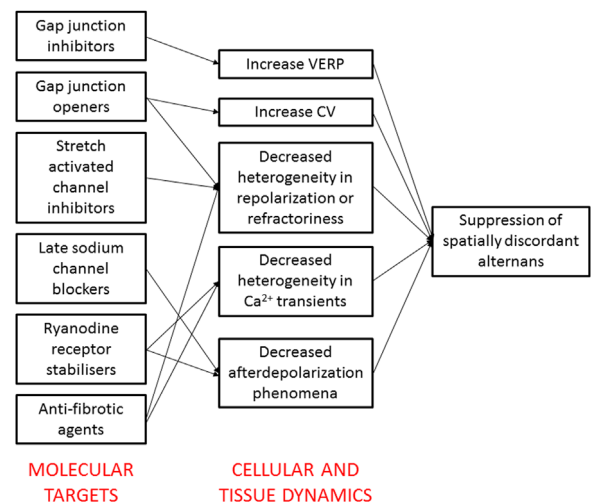


Fig. 7. Potential molecular targets for anti-arrhythmic therapy, by suppression of spatially discordant APD alternans by influencing cellular and tissue dynamics.

that APD alternans, whether in the atria or the ventricles, are driven by Ca^{2+} ; alternans [42], which are attributable to ryanodine receptor refractoriness [43].

The concept of repolarisation reserve was introduced to explain the occurrence of arrhythmias, positing that redundancy in repolarization exists [44,45]. The idea is that elimination of one repolarizing current need not necessarily lead to failure of repolarisation because of compensation by other repolarizing currents [46]. Recent experiments in transgenic LQT1 rabbits have shed light on the mechanistic link between repolarisation reserve and alternans [47]. Interestingly, in this model, reduced repolarization reserve per se was paradoxically associated with a shallower gradient of APD restitution and the development of alternans. This in turn has been attributed to abnormal calcium handling, in which I_{Ks} downregulation led to discordant Ca^{2+} alternans. Under conditions of prolonged repolarization or abnormal calcium handling, afterdepolarization phenomena are observed and these can produce spatially discordant alternans. Where the voltage change brought about by such phenomena are sufficiently large, an ectopic beat can be generated, which can cause dispersion of the diastolic interval, thereby converting spatially concordant alternans to discordant alternans [34].

4. Alternans and arrhythmogenesis in different clinical conditions

APD alternans, whether spatially concordant or discordant, can produce arrhythmias. For example, spatially concordant alternans themselves can produce 2:1 conduction block, thereby initiating re-entry [48]. Nevertheless, discordant alternans are considered to be more arrhythmogenic. They can produce large spatial gradients in repolarisation and refractoriness, which can result in local conduction block of a premature extrasystole (such as a premature ventricular complex, PVC [37]), thereby facilitating re-entry [23,49]. They can also promote phase 2 re-entry involving either a fixed or a variable I_{to} , allowing antegrade and retrograde phase 2 re-entry, respectively [50].

These mechanisms for producing spatially discordant alternans are important in a number of clinical conditions, potentially causing lethal arrhythmias. In heart failure, there is extensive ion current remodeling with a lower threshold for inducing APD

alternans [51]. APDs are prolonged due to downregulation of potassium currents and increased late sodium current [30]. This would allow the steep portion of APD restitution to be engaged, and therefore generation of spatially concordant alternans. These can become discordant in the presence of conduction abnormalities [30], cardiac fibrosis [52], or abnormal Ca^{2+}_i handling dynamics, such as increased SR Ca^{2+} leak [53], decreased SERCA pump activity [54], increased NCX currents [55], or steeper fractional release of SR Ca^{2+}_i content [56]. In catecholaminergic polymorphic ventricular tachycardia (CPVT), mutations in the ryanodine receptor lead to diastolic calcium leak and generation of discordant Ca^{2+}_i alternans [57,58]. Long QT syndromes are characterized by APD prolongation, a reduction in repolarization reserve, and increased APD restitution gradients leading to the production of APD alternans. Spatial heterogeneities in repolarization are exacerbated due to differences in ion channel expression and heterogeneities in restitution across the myocardial wall.

Other pro-arrhythmic conditions are clearly associated with a flatter APD restitution curve, but discordant alternans can be generated when regional differences in restitution lead to spatial heterogeneities in APDs [59]. In myocardial ischemia and sodium channel blockade, CV restitution may be more important in the conversion of concordant alternans to discordant alternans [60]. Increased beta adrenergic drive, which can occur in heart failure or exercise, can increase the maximum gradients of APD restitution and produce discordant alternans [61].

Regardless of the mechanisms generating these arrhythmogenic alternans, the final common pathway involves wavebreak, conduction block, and the initiation and maintenance of re-entrant arrhythmias [62]. It should be recognized that alternans are only one factor in determining arrhythmogenesis. An anti-arrhythmic state can occur even in the presence of both steep APD restitution and discordant alternans, as exemplified by hypokalemia [19]. Heptanol, a gap junction uncoupler, was shown to exert anti-arrhythmic effects in hypokalemia by influencing VERP alone [63]. This finding is perhaps surprising, given that reduced electrotonic coupling should exacerbate dispersion in APDs and promote arrhythmogenesis.

5. Future therapies

The question of how understanding the cardiac dynamics can enable us to devise better pharmacotherapy for arrhythmia management persists. Numerous studies using animal models have demonstrated that the anti-arrhythmic actions of many drugs are in part mediated by their effects on cardiac dynamics. These include traditional agents such as beta-blockers as well as novel drugs such as late sodium current blockers and gap junction openers [64–66] (Fig. 7). Gap junction inhibitors can exert anti-arrhythmic effects by prolonging effective refractory periods [63,67]. Moreover, mild loss of gap junction function in non-uniform tissue may paradoxically increase CV and improve the safety margin of conduction [68]. This in turn could remove unidirectional conduction blocks, converting them into bilateral conduction [69]. In contrast, gap junction enhancers can improve conduction and reduce the spatial heterogeneities in repolarization and refractoriness, thereby suppressing discordant alternans and arrhythmogenesis. Their effects on calcium dynamics are complex, depending on the nature of $\text{Ca}^{2+} \rightarrow$ APD coupling. The differences in Ca^{2+} transient between adjacent cells are amplified in the case of negative coupling, but reduced with positive coupling [40]. Activation of stretch-activated channels exerts opposing effects on alternans, suppressing those that are concordant whilst exacerbating those that are discordant [22]. SAC inhibitors could potentially exert anti-arrhythmic effects by suppressing discordant alternans. Discordant alternans can also be inhibited by late

sodium channel blockers [65], ryanodine receptor stabilizers [57] or anti-fibrotic agents [70], which would prevent conduction block and inhibit arrhythmogenesis. Some of the examples described above illustrate the difficulty in predicting the overall electrophysiological effects of a drug, and some may exert their anti-arrhythmic effects without influencing restitution [71,72]. Future efforts therefore require a computational strategy in which modeling of the heart can be achieved at different levels of biological organization to take into account of the complex spatiotemporal properties of cardiac dynamics.

Conflict of interest

All authors declare no conflict of interest related to this study.

Acknowledgments

GT received a BBSRC Doctoral CASE Studentship at the Department of Biochemistry, University of Cambridge, in conjunction with Xention Discovery, for his Ph.D studies.

References

- [1] Rosenbaum DS, Jackson LE, Smith JM, et al. Electrical alternans and vulnerability to ventricular arrhythmias. *N Engl J Med* 1994;330:235–41.
- [2] Nolasco JB, Dahlen RW. A graphic method for the study of alternation in cardiac action potentials. *J Appl Physiol* 1968;25:191–6.
- [3] Tolkacheva EG, Schaeffer DG, Gauthier DJ, et al. Condition for alternans and stability of the 1:1 response pattern in a "memory" model of paced cardiac dynamics. *Phys Rev E Stat Nonlinear Soft Matter Phys* 2003;67:031904.
- [4] Kalb SS, Dobrovolsky HM, Tolkacheva EG, et al. The restitution portrait: a new method for investigating rate-dependent restitution. *J Cardiovasc Electrophysiol* 2004;15:698–709.
- [5] Pu J, Boyden PA. Alterations of Na^+ currents in myocytes from epicardial border zone of the infarcted heart. A possible ionic mechanism for reduced excitability and postrepolarization refractoriness. *Circ Res* 1997;81:110–9.
- [6] Hondeghem LM, Snyders DJ. Class III antiarrhythmic agents have a lot of potential but a long way to go. Reduced effectiveness and dangers of reverse use dependence. *Circulation* 1990;81:686–90.
- [7] Patwardhan A, Moghe S. Novel feedback based stimulation protocol shows hysteresis in cardiac action potential duration restitution. *Biomed Sci Instrum* 2001;37:505–10.
- [8] Wu R, Patwardhan A. Restitution of action potential duration during sequential changes in diastolic intervals shows multimodal behavior. *Circ Res* 2004;94:634–41.
- [9] Wu R, Patwardhan A. Mechanism of repolarization alternans has restitution of action potential duration dependent and independent components. *J Cardiovasc Electrophysiol* 2006;17:87–93.
- [10] Zhang H, Ueyama T, Wang J, et al. Short-term memory and electrical restitution in the canine transmural ventricle. *Physiol Meas* 2011;32:207–22.
- [11] Fox JJ, Bodenschatz E, Gilmour RF. Period-doubling instability and memory in cardiac tissue. *Phys Rev Lett* 2002;89:138101.
- [12] Pitruzzello AM, Krassowska W, Idriss SF. Spatial heterogeneity of the restitution portrait in rabbit epicardium. *Am J Physiol Heart Circ Physiol* 2007;292:H1568–78.
- [13] Narayan SM, Smith JM. Exploiting rate-related hysteresis in repolarization alternans to improve risk stratification for ventricular tachycardia. *J Am Coll Cardiol* 2000;35:1485–92.
- [14] Walker ML, Wan X, Kirsch GE, et al. Hysteresis effect implicates calcium cycling as a mechanism of repolarization alternans. *Circulation* 2003;108:2704–9.
- [15] Pruvot EJ, Katra RP, Rosenbaum DS, et al. Role of calcium cycling versus restitution in the mechanism of repolarization alternans. *Circ Res* 2004;94:1083–90.
- [16] Shiferaw Y, Sato D, Karma A. Coupled dynamics of voltage and calcium in paced cardiac cells. *Phys Rev E Stat Nonlinear Soft Matter Phys* 2005;71:021903.
- [17] Florea SM, Blatter LA. The role of mitochondria for the regulation of cardiac alternans. *Front Physiol* 2010;1:141.
- [18] Prudat Y, Madhavi RV, Angelini M, et al. Stochastic pacing reveals the propensity to cardiac action potential alternans and uncovers its underlying dynamics. *J Physiol* 2016. <http://dx.doi.org/10.1113/jp271573>.

- [19] Osadchii OE, Larsen AP, Olesen SP. Predictive value of electrical restitution in hypokalaemia-induced ventricular arrhythmogenicity. *Am J Physiol Heart Circ Physiol* 2010;298:H210–20.
- [20] Murphy CF, Horner SM, Dick DJ, et al. Electrical alternans and the onset of rate-induced pulsus alternans during acute regional ischaemia in the anaesthetised pig heart. *Cardiovasc Res* 1996;32:138–47.
- [21] Kohl P, Bollensdorff C, Garny A. Effects of mechanosensitive ion channels on ventricular electrophysiology: experimental and theoretical models. *Exp Physiol* 2006;91:307–21.
- [22] Galice S, Bers DM, Sato D. Stretch activated channel activation can promote or suppress cardiac alternans. *Biophysical Journal* 2015;108:106a.
- [23] Qu Z, Garfinkel A, Chen PS, et al. Mechanisms of discordant alternans and induction of reentry in simulated cardiac tissue. *Circulation* 2000;102:1664–70.
- [24] Pastore JM, Girouard SD, Laurita KR, et al. Mechanism linking T-wave alternans to the genesis of cardiac fibrillation. *Circulation* 1999;99:1385–94.
- [25] Katra RP, Pruvot E, Laurita KR. Intracellular calcium handling heterogeneities in intact guinea pig hearts. *Am J Physiol Heart Circ Physiol* 2004;286:H648–56.
- [26] Laurita KR, Katra R, Wible B, et al. Transmural heterogeneity of calcium handling in canine. *Circ Res* 2003;92:668–75.
- [27] Aistrup GL, Kelly JE, Kapur S, et al. Pacing-induced heterogeneities in intracellular Ca^{2+} signaling, cardiac alternans, and ventricular arrhythmias in intact rat heart. *Circ Res* 2006;99:65–73.
- [28] Poelzing S, Akar FG, Baron E, et al. Heterogeneous connexin43 expression produces electrophysiological heterogeneities across ventricular wall. *Am J Physiol Heart Circ Physiol* 2004;286:H2001–9.
- [29] Poelzing S, Rosenbaum DS. Altered connexin43 expression produces arrhythmia substrate in heart failure. *Am J Physiol Heart Circ Physiol* 2004;287:H1762–70.
- [30] Tse G, Yeo JM. Conduction abnormalities and ventricular arrhythmogenesis: the roles of sodium channels and gap junctions. *IJC Heart Vasc* 2015;9:75–82.
- [31] Zang Y, Dai L, Zhan H, et al. Theoretical investigation of the mechanism of heart failure using a canine ventricular cell model: especially the role of up-regulated CaMKII and SR $Ca(2+)$ leak. *J Mol Cell Cardiol* 2013;56:34–43.
- [32] Qu Z, Karagueuzian HS, Garfinkel A, et al. Effects of $Na(+)$ channel and cell coupling abnormalities on vulnerability to reentry: a simulation study. *Am J Physiol Heart Circ Physiol* 2004;286:H1310–21.
- [33] Qu Z, Xie Y, Garfinkel A, et al. T-wave alternans and arrhythmogenesis in cardiac diseases. *Front Physiol* 2010;1.
- [34] Watanabe MA, Fenton FH, Evans SJ, et al. Mechanisms for discordant alternans. *J Cardiovasc Electrophysiol* 2001;12:196–206.
- [35] Pu J, Balsler JR, Boyden PA. Lidocaine action on $Na+$ currents in ventricular myocytes from the epicardial border zone of the infarcted heart. *Circ Res* 1998;83:431–40.
- [36] Qian YW, Clusin WT, Lin SF, et al. Spatial heterogeneity of calcium transient alternans during the early phase of myocardial ischemia in the blood-perfused rabbit heart. *Circulation* 2001;104:2082–7.
- [37] The Cardiac Arrhythmia Suppression Trial [CAST] Investigators. Preliminary report: effect of encainide and flecainide on mortality in a randomized trial of arrhythmia suppression after myocardial infarction. *N Engl J Med* 1989;321:406–12.
- [38] Cao JM, Qu Z, Kim YH, et al. Spatiotemporal heterogeneity in the induction of ventricular fibrillation by rapid pacing: importance of cardiac restitution properties. *Circ Res* 1999;84:1318–31.
- [39] Diaz ME, O'Neill SC, Eisner DA. Sarcoplasmic reticulum calcium content fluctuation is the key to cardiac alternans. *Circ Res* 2004;94:650–6.
- [40] Sato D, Shiferaw Y, Garfinkel A, et al. Spatially discordant alternans in cardiac tissue: role of calcium cycling. *Circ Res* 2006;99:520–7.
- [41] Sato D, Bers DM, Shiferaw Y. Formation of spatially discordant alternans due to fluctuations and diffusion of calcium. *PLoS ONE* 2013;8:e85365.
- [42] Kanaporis G, Blatter LA. The mechanisms of calcium cycling and action potential dynamics in cardiac alternans. *Circ Res* 2015;116:846–56.
- [43] Wang L, Myles RC, De Jesus NM, et al. Optical mapping of sarcoplasmic reticulum Ca^{2+} in the intact heart: ryanodine receptor refractoriness during alternans and fibrillation. *Circ Res* 2014;114:1410–21.
- [44] Roden DM. Taking the "idio" out of "idiosyncratic": predicting torsades de pointes. *Pacing Clin Electrophysiol* 1998;21:1029–34.
- [45] Roden DM. Repolarization reserve: a moving target. *Circulation* 2008;118:981–2.
- [46] Varro A, Baczko I. Cardiac ventricular repolarization reserve: a principle for understanding drug-related proarrhythmic risk. *Br J Pharmacol* 2011;164:14–36.
- [47] Lau E, Kossidas K, Kim TY, et al. Spatially discordant alternans and arrhythmias in tachypacing-induced cardiac myopathy in transgenic LQT1 rabbits: the importance of IKs and Ca^{2+} cycling. *PLoS One* 2015;10:e0122754.
- [48] Fox JJ, Gilmour RFJ, Bodenschatz E. Conduction block in one-dimensional heart fibers. *Phys Rev Lett* 2002;89:198101.
- [49] Qu Z, Garfinkel A, Weiss JN. Vulnerable window for conduction block in a one-dimensional cable of cardiac cells, 2: multiple extrasystoles. *Biophys J* 2006;91:805–15.
- [50] Maoz A, Krogh-Madsen T, Christini DJ. Instability in action potential morphology underlies phase 2 reentry: a mathematical modeling study. *Heart Rhythm* 2009;6:813–22.
- [51] Wilson LD, Jeyaraj D, Wan X, et al. Heart failure enhances susceptibility to arrhythmogenic cardiac alternans. *Heart Rhythm* 2009;6:251–9.
- [52] Tse G. Mechanisms of cardiac arrhythmias. *J. Arrhythm* 2015. <http://dx.doi.org/10.1016/j.joa.2015.11.003>.
- [53] Shannon TR, Pogwizd SM, Bers DM. Elevated sarcoplasmic reticulum Ca^{2+} leak in intact ventricular myocytes from rabbits in heart failure. *Circ Res* 2003;93:592–4.
- [54] O'Rourke B, Kass DA, Tomaselli GF, et al. Mechanisms of altered excitation-contraction coupling in canine tachycardia-induced heart failure, I: experimental studies. *Circ Res* 1999;84:562–70.
- [55] Xu L, Chen J, Li XY, et al. Analysis of $Na(+)/Ca(2+)$ exchanger (NCX) function and current in murine cardiac myocytes during heart failure. *Mol Biol Rep* 2012;39:3847–52.
- [56] Shannon TR, Wang F, Bers DM. Regulation of cardiac sarcoplasmic reticulum Ca release by luminal $[Ca]$ and altered gating assessed with a mathematical model. *Biophys J* 2005;89:4096–110.
- [57] Lehnart SE, Terrenoire C, Reiken S, et al. Stabilization of cardiac ryanodine receptor prevents intracellular calcium leak and arrhythmias. *Proc Natl Acad Sci USA* 2006;103:7906–10.
- [58] Yang PC, Moreno JD, Miyake CY, et al. In silico prediction of drug therapy in catecholaminergic polymorphic ventricular tachycardia. *J Physiol* 2015;594:567–93.
- [59] Taggart P, Sutton PM, Boyett MR, et al. Human ventricular action potential duration during short and long cycles. Rapid modulation by ischemia. *Circulation* 1996;94:2526–34.
- [60] Wu TJ, Lin SF, Weiss JN, et al. Two types of ventricular fibrillation in isolated rabbit hearts: importance of excitability and action potential duration restitution. *Circulation* 2002;106:1859–66.
- [61] Ng GA, Brack KE, Patel VH, et al. Autonomic modulation of electrical restitution, alternans and ventricular fibrillation initiation in the isolated heart. *Cardiovasc Res* 2007;73:750–60.
- [62] Hsieh Y-C, Lin S-F, Lin T-C, et al. Therapeutic hypothermia (30 °C) enhances arrhythmogenic substrates, including spatially discordant alternans, and facilitates pacing-induced ventricular fibrillation in isolated rabbit hearts. *Circ J* 2009;73:2214–22.
- [63] Tse G, Tse V, Yeo JM. Ventricular anti-arrhythmic effects of heptanol in hypokalaemic, Langendorff-perfused mouse hearts. *Biomed Rep* 2015;4:313–24.
- [64] Hsieh YC, Lin JC, Hung CY, et al. Gap junction modifier rotigaptide decreases the susceptibility to ventricular arrhythmia by enhancing conduction velocity and suppressing discordant alternans during therapeutic hypothermia in isolated rabbit hearts. *Heart Rhythm* 2015;13:251–61.
- [65] Alves Bento AS, Bacic D, Saran Carneiro J, et al. Selective late I_{Na} inhibition by GS-458967 exerts parallel suppression of catecholamine-induced hemodynamically significant ventricular tachycardia and T-wave alternans in an intact porcine model. *Heart Rhythm* 2015;12:2508–14.
- [66] Kharche SR, Stary T, Colman MA, et al. Effects of human atrial ionic remodeling by beta-blocker therapy on mechanisms of atrial fibrillation: a computer simulation. *Europace* 2014;16:1524–33.
- [67] Tse G, Tse V, Yeo JM, et al. Atrial anti-arrhythmic effects of heptanol in Langendorff-perfused mouse hearts. *PLoS One* 2016;12:e0148858.
- [68] Shaw RM, Rudy Y. Ionic mechanisms of propagation in cardiac tissue. Roles of the sodium and L-type calcium currents during reduced excitability and decreased gap junction coupling. *Circ Res* 1997;81:727–41.
- [69] Rohr S, Kucera JP, Fast VG, et al. Paradoxical improvement of impulse conduction in cardiac tissue by partial cellular uncoupling. *Science* 1997;275:841–4.
- [70] Bathgate RA, Leggabe ED, McGuane JT, et al. Adenovirus-mediated delivery of relaxin reverses cardiac fibrosis. *Mol Cell Endocrinol* 2008;280:30–8.
- [71] Tse G, Wong ST, Tse V, Yeo JM. Restitution analysis of alternans using dynamic pacing and its comparison with S1S2 restitution in heptanol-treated, hypokalaemic Langendorff-perfused mouse hearts. *Biomed Rep* 2016 [in press].
- [72] Tse G, Hothi SS, Grace AA, Huang CL. Ventricular arrhythmogenesis following slowed conduction in heptanol-treated, Langendorff-perfused mouse hearts. *J Physiol Sci* 2012;62:79–92.
- [73] Agarwal A, Patwardhan A. A new approach to measure the contribution of restitution to repolarization alternans. *Conf Proc IEEE Eng Med Biol Soc* 2009;2009:4516–8.
- [74] Nishii N, Nagase S, Morita H, et al. Abnormal restitution property of action potential duration and conduction delay in Brugada syndrome: both repolarization and depolarization abnormalities. *Europace* 2010;12:544–52.
- [75] Bishop MJ, Connolly A, Plank G. Structural heterogeneity modulates effective refractory period: a mechanism of focal arrhythmia initiation. *PLoS One* 2014;9:e109754.
- [76] Kaplan SR, Gard JJ, Protonotarios N, et al. Remodeling of myocyte gap junctions in arrhythmogenic right ventricular cardiomyopathy due to a deletion in plakoglobin (Naxos disease). *Heart Rhythm* 2004;1:3–11.
- [77] Kalcheva N, Qu J, Sandeep N, et al. Gap junction remodeling and cardiac arrhythmogenesis in a murine model of oculodentodigital dysplasia. *Proc Natl Acad Sci USA* 2007;104:20512–6.
- [78] Chang KC, Bayer JD, Trayanova NA. Disrupted calcium release as a mechanism for atrial alternans associated with human atrial fibrillation. *PLoS Computational Biology* 2014;10:e1004011.