DDT Vol. 8, No. 4 February 2003

A novel approach to identifying antiarrhythmic drug targets

Robert F. Gilmour, Jr

Sudden cardiac death, secondary to ventricular fibrillation (VF), remains the leading cause of death in the USA. Recent experimental and theoretical studies suggest that VF could be caused by spiral wave re-entry. The initiation and subsequent break-up of spiral waves has been linked to electrical alternans, a phenomenon produced in cardiac tissue that has a steeply sloped restitution relation. Agents that reduce the slope of the restitution relation have been shown to suppress alternans and, presumably by that mechanism, terminate VF. These results suggest that electrical restitution could be a promising new target for antiarrhythmic therapies.

Robert F. Gilmour, Jr
Dept of Biomedical Sciences
T7 012C VRT
Cornell University
Ithaca, NY 14853-6401, USA
e-mail: rfg2@cornell.edu

Catastrophic rhythm disturbances of the heart, such as ventricular fibrillation (VF), are the major cause of death in the United States [1,2]. Treatment of these rhythm disorders remains largely empirical, in part because of an incomplete understanding of underlying cellular electrophysiological mechanisms. Recent studies, building on earlier theoretical work by Krinsky, Winfree and colleagues [3,4] and experiments by Allessie [5], have suggested that spiral wave re-entry could be the 'engine' that drives VF [6–11]. Although there is substantial evidence that spiral wave re-entry contributes significantly to the induction and maintenance of VF, the exact mechanism by which spiral waves sustain VF is currently being debated. Correctly identifying the mechanism (or, more likely, mechanisms) by which spiral waves cause VF is likely to be a crucial step in the development of pharmacological approaches to VF prevention.

Restitution hypothesis for VF

One of the hypotheses to account for the apparent link between spiral waves and VF is the restitution hypothesis, which proposes that VF is caused by the break up of a single spiral wave into multiple self-perpetuating wavelets [8,10,12–17]. This mechanism is similar to that proposed decades ago by Moe [18], but with one important difference: it can occur in intrinsically homogeneous cardiac tissue (for reasons to be described below). The transition from normal planar wave excitation to a single spiral wave and, ultimately, to multiple wavelets is thought to underlie the transition from normal sinus rhythm to ventricular tachycardia and VF characteristic of patients who succumb to sudden death (Fig. 1).

The exact mechanism for the break up of spiral waves is unknown. However, there is considerable evidence that break up is closely related to action potential duration (APD) restitution, which is the relationship between APD and diastolic interval (DI, the time interval between action potentials), in which APD is determined by the preceding DI (Fig. 2). During pacing of cardiac tissue at progressively shorter cycle lengths, APD decreases, until at sufficiently short cycle lengths a period-doubling bifurcation occurs and APD begins to alternate between a long duration and a short duration, a phenomenon known as APD alternans (Fig. 2) [19–24]. The development of APD alternans requires that the slope of the restitution relation exceeds 1, whereas if the slope of the restitution is less than 1, alternans will not occur.

The slope of the restitution relation, and the corresponding presence or absence of APD alternans, has been linked to spiral wave stability. If the slope of the APD restitution relation is <1, a spiral wave tends to stabilize and produce a periodic rhythm, the manifestation of which might be monomorphic ventricular tachycardia. If, however, the slope of the APD restitution relation is ≥1, a single spiral wave might disintegrate into many spiral waves, manifested as VF. These transitions are illustrated in Fig. 3, using results generated by a computer model. Once initiated, a single stable spiral wave can be destabilized by increasing the slope of the restitution relation from <1 to ≥1. Similarly, multiple wavelets can be induced to coalesce into a single spiral wave

PII: S1359-6446(02)02567-9

by reducing the slope of the restitution relation from ≥1 to <1. Thus, a reduction of the slope of the restitution relation from ≥1 to <1 is expected to prevent the induction of VF and to convert existing VF into a periodic rhythm.

Several recent studies have provided experimental and theoretical support for a causal relationship between APD restitution and VF [12,13,25–29]. If the APD restitution relation contains a region of slope ≥1, APD alternans and VF are induced by pacing at short cycle lengths. If the slope of the APD restitution relation is reduced to less than one, APD alternans is suppressed and VF is not induced. Furthermore, if a fibrillating ventricle is exposed to a drug that reduces the slope of the restitution to less than 1, VF is converted to a periodic rhythm sustained by a single stable spiral wave [12,13,26].

Unfortunately, interventions that suppress VF experimentally (verapamil [13], bretylium [12] and hyperkalemia [26]) have unwanted actions that severely limit their clinical use. Nevertheless, the effects of these interventions on restitution and VF have provided valuable insights regarding new, potentially more clinically relevant, drug targets. In particular, attempts to understand the mechanism by which calcium channel blockers alter APD restitution have led to the realization that increasing selected outward repolarizing currents might also flatten restitution, as discussed in the following sections.

Calcium channel antagonists and VF

The suppression of VF by verapamil, in association with a reduction in the slope of the APD restitution relation (Fig. 4), suggests that the L-type calcium current (I_{Ca}) has a key role in restitution and in the development of VF [13,30]. To better define these roles, we recently developed an ionic model for the cardiac ventricular myocyte (CVM) based on the Winslow [31] and Luo–Rudy models [32] that generates physiologically realistic

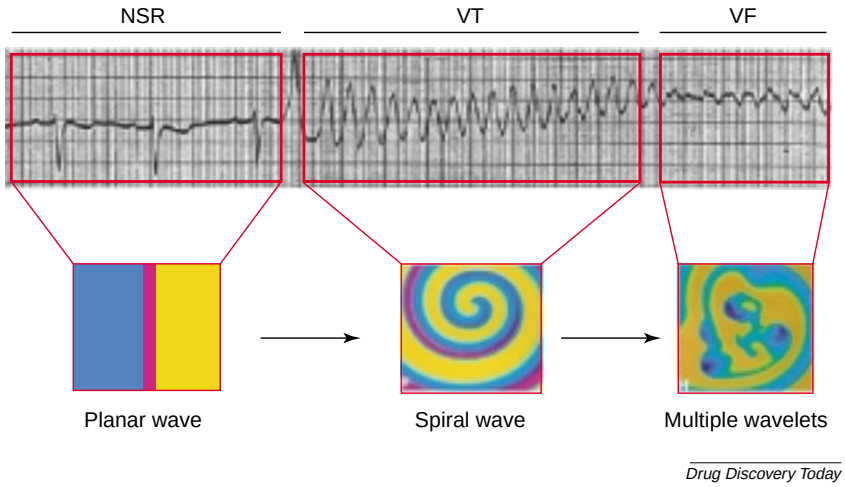

Figure 1. Proposed evolution of cardiac wave propagation patterns underlying the transition from a normal cardiac rhythm to ventricular fibrillation. Transition from normal sinus rhythm (NSR) to ventricular tachycardia (VT) and ventricular fibrillation (VF), as recorded on the surface electrocardiogram. Corresponding transitions from a planar wave to a single spiral wave to multiple spiral waves.

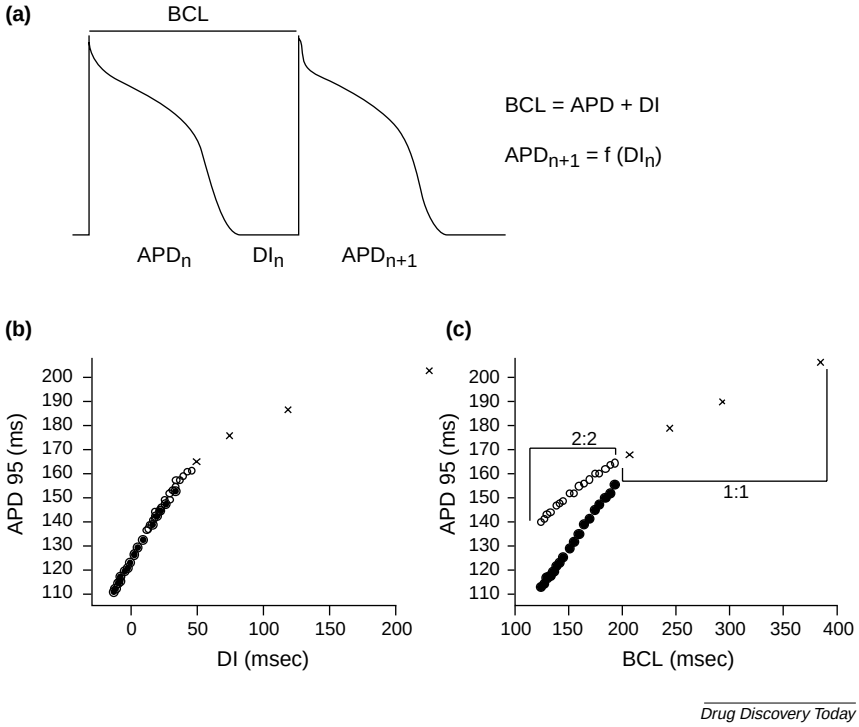

Figure 2. Restitution of action potential duration (APD). **(a)** Definition of APD, diastolic interval (DI) and cycle length (CL). The duration of the n+1th action potential (APD_{n+1}) is a function of the preceding DI (DI_n). **(b)** Plot of APD versus preceding DI. **(c)** Plot of APD versus CL. Crosses are APDs during the CLs in which no alternans occurs. During APD alternans, unfilled circles are APDs during the long action potential, and filled circles are APDs during the short action potential. Modified, with permission, from Koller *et al.* [25].

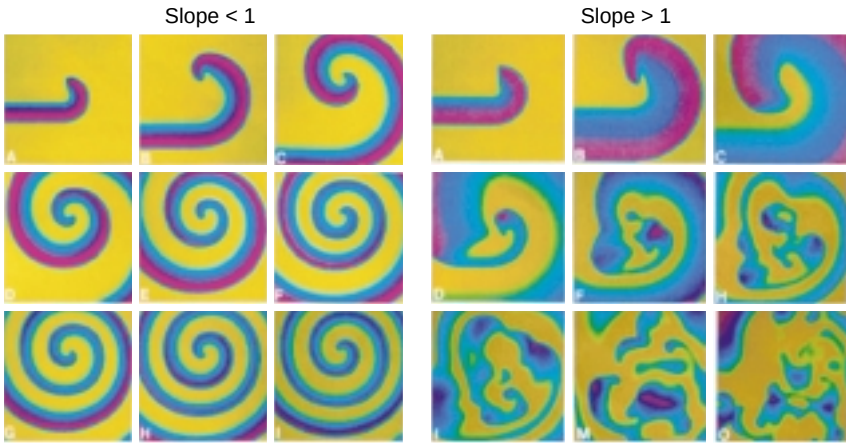

Figure 3. Wave patterns in a computer model of cardiac activation. Sequential snapshots (A–I) of activation during the development of a stable single spiral wave under conditions in which the slope of the restitution relation for action potential duration is <1 (left panel). The transition from a single spiral wave into many spiral waves for a restitution slope >1 (A–O, with some intervening steps deleted: right panel). Results courtesy of Dante R. Chialvo (Department of Physiology, University of California, Los Angeles: http://www.ucla.edu).

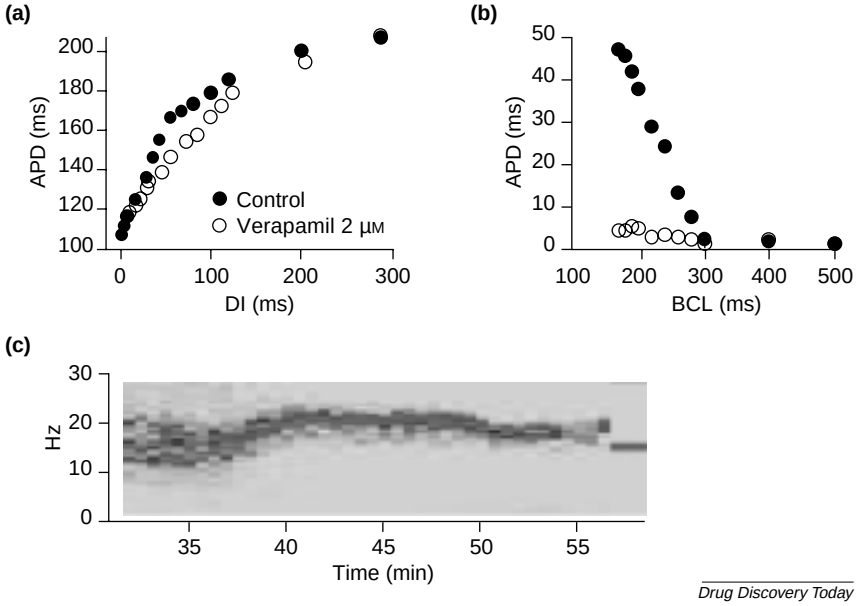

Figure 4. Effects of verapamil on action potential duration (APD) restitution and ventricular fibrillation (VF). **(a)** Example of the reduction in the slope of the APD restitution relation after exposure to verapamil. **(b)** Corresponding reduction in the magnitude of APD alternans (AM) during pacing at different basic cycle lengths (BCL) after exposure to verapamil. **(c)** Effects of verapamil on the composite Fast Fourier Transform (FFT) of monophasic action potential (MAP) recordings during VF in arterially perfused canine ventricle. Verapamil, added after 35 min of VF had elapsed, converted the broad FFT spectra during VF to a single frequency. Modified, with permission, from Riccio *et al.* [13].

APD alternans over a wide range of pacing basic cycle lengths (BCL) [33]. Using this model, the L-type Ca^{2+} current was implicated as an important determinant of APD alternans, according to the mechanism shown in Fig. 5. On initiation of pacing at a short BCL, I_{Ca} is fully recovered before the first action potential and activates fully during the action potential, which results in a long APD. The long action potential is subsequently followed by a short diastolic interval, during which I_{Ca} fails to recover completely from Ca^{2+}-induced inactivation. Because of decreased availability of I_{Ca}, the duration of the subsequent action potential is shorter, which results in a longer succeeding diastolic interval, more complete recovery of I_{Ca} and a long action potential duration. This cycle then repeats, eventually establishing a steady-state alternans of I_{Ca} and APD.

Potassium channel agonists and VF

From the mechanism for APD alternans outlined in Fig. 5, we anticipate that shortening of APD during rapid pacing will prolong DI, which, in turn, might provide adequate time for complete recovery of I_{Ca}. If so, reducing the magnitude of APD alternans could be accomplished by increasing outward repolarizing currents, rather than by decreasing I_{Ca}. This idea has been supported by additional computer modeling studies in which APD alternans was suppressed by increasing any one of several outward repolarizing currents [e.g. the rapid (I_{Kr}) and slow (I_{Ks}) components of the delayed rectifier and the inward rectifier (I_{K1})] [33]. Although agonists for these currents are generally not available, it might be possible to modify currents such as I_{Kr} and I_{Ks} by increasing phosphatidyl inositol bisphosphate (PIP2) levels [34] or by altering the phosphorylation state of the channels [35–37], provided the channels can be phosphorylated without concomitant phosphorylation of calcium channels or can be upregulated

in the presence of a calcium channel antagonist, to offset increased I_{Ca} secondary to phosphorylation [38].

Recently, we have focused on increasing I_{Kr} as an approach to suppressing APD alternans. I_{Kr} has an important role in cardiac repolarization, increasing to a maximum during phase three of the action potential, as the channel recovers from inactivation, and then decreasing as the electrical driving force decreases and as deactivation of the channel increases [39–43]. Because I_{Kr} contributes minimally to the action potential plateau, increasing I_{Kr} can have little or no effect on the Ca^{2+} transient. Consequently, by increasing I_{Kr} it can be possible to suppress APD alternans without adversely affecting contractility.

At present, however, there are no I_{Kr} agonists available to test this hypothesis. To circumvent this problem, we infected isolated ventricular myocytes with an adenovirus expressing HERG, the gene that encodes the pore-forming domain of I_{Kr}, to increase HERG protein expression level and the corresponding I_{Kr} current [44] [Hua, F. *et al.*, unpublished data]. After verifying that I_{Kr} had increased post-infection, the myocytes were paced at rapid rates to determine whether increasing I_{Kr} by overexpressing HERG suppressed APD alternans. As shown in Fig. 6, overexpression of HERG markedly increased I_{Kr}, as recorded during action potential clamp, and suppressed APD alternans during rapid pacing. Overexpression of HERG did not, however, significantly reduce I_{Ca}, suggesting that this approach to suppression of alternans need not be accompanied by a reduction in contractility.

The current paradigm regarding I_{Kr} and VF is that blocking I_{Kr} is expected to be anti-arrhythmic, secondary to prolongation of APD and refractoriness [45,46]. However, this approach has not been successful in preventing VF and, moreover, is associated with pro-arrhythmia [45]. Prolongation of APD, as reflected by an increased duration of the QT interval on the ECG, and an increase in the incidence of ventricular arrhythmias are also associated with the administration of numerous non-cardiac drugs that block I_{Kr} [47–50]. Similarly, inherited loss-of-function mutations in I_{Kr} are accompanied by a prolongation of the QT interval and by an increased risk of lethal ventricular tachyarrhythmias, such as *torsade de pointes* [51–54].

The cardiac arrhythmias associated with inherited or drug-induced abnormalities of I_{Kr} are thought to be precipitated primarily by bradycardia-induced prolongation of repolarization [51,52]. The potential mechanisms by which I_{Kr} might facilitate the induction of arrhythmias at slow heart rates have been studied extensively [46,50,53]. However, the contribution of I_{Kr} to repolarization during tachycardia, which might also be important for the development of cardiac arrhythmias, has not been well

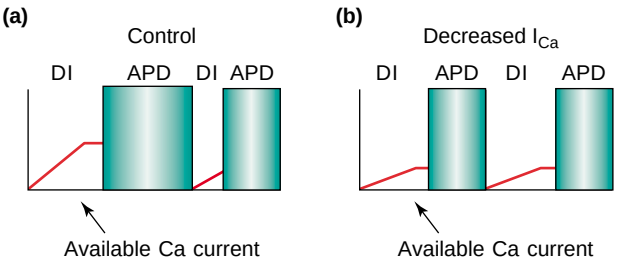

Figure 5. Putative mechanism for action potential duration (APD) alternans and the suppression of alternans by calcium channel blockade. **(a)** Under control conditions, alternation of the length of the diastolic intervals (DI) during APD alternans causes alternation of the magnitude of I_{Ca}, secondary to incomplete recovery of I_{Ca} from inactivation during the short DI. **(b)** Reduction of I_{Ca} decreases APD, which prolongs DI, so that DI following each APD is sufficiently long to enable complete recovery of I_{Ca}. In the absence of alternations of I_{Ca}, APD alternans is suppressed.

characterized. The observation that reducing I_{Kr} increases the magnitude of APD alternans could provide an additional mechanism to account for the proarrhythmic effects of I_{Kr} blockers, in that increased alternans magnitude would be expected to destabilize ventricular tachyarrhythmias, leading to the development of VF. Conversely, the observation that increasing I_{Kr} reduces the magnitude of APD alternans and the slope of the APD restitution relation provides a rationale for the development of a new class of compounds, I_{Kr} agonists, with the expectation that such compounds might have anti-fibrillatory effects.

The latter hypothesis relies, however, on the expectation that I_{Kr} can be increased sufficiently to reduce the slope of the restitution relation without shortening action potential duration to such an extent that contractility is impaired, secondary to a shortening of the plateau duration and attenuation of I_{Ca}. In addition, shortening of APD could lead to a reduction in the wavelength of re-entry circuits (in which wavelength = refractory period × conduction velocity). Reduction of the wavelength could, in turn, precipitate wavebreak by other mechanisms (e.g. so-called 'head–tail' interactions, in which a wavefront encounters a waveback and fragments) [55]. These and other potential drawbacks to the use of K-channel agonists to suppress VF remain to be evaluated critically.

Potential implications for drug development and evaluation

Until recently, therapy for the prevention of sudden cardiac death had been based on the presumption that frequent ventricular ectopy, in particular ventricular tachycardia, is a prelude to ventricular fibrillation [2]. Accordingly, drugs

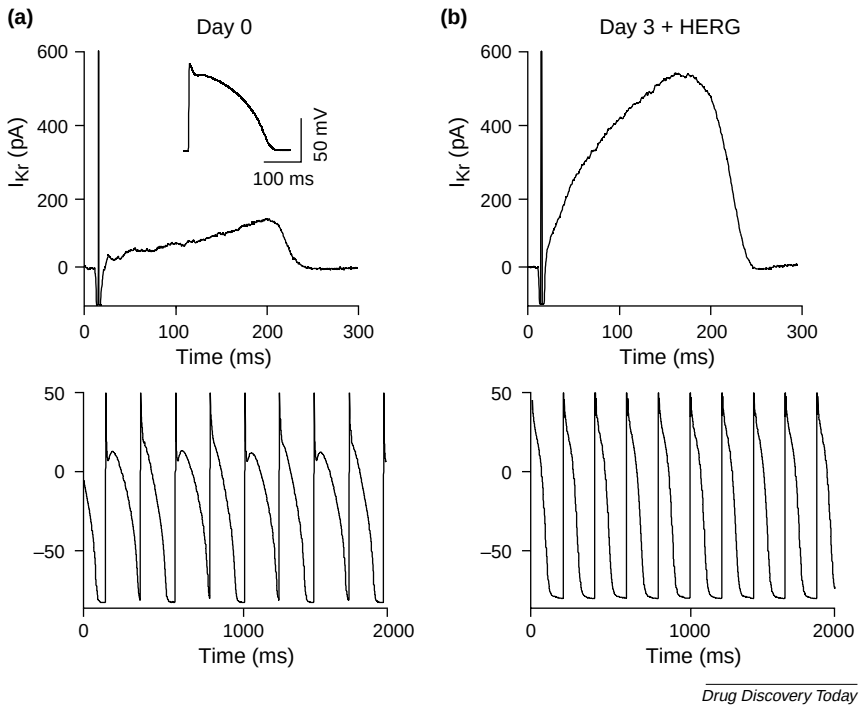

Figure 6. Effects of HERG overexpression on I_{Kr} and action potential duration (APD) alternans in isolated canine cardiac myocytes. **(a)** Current traces of I_{Kr} (upper panel) and action potentials during pacing at CL = 200 msec (lower panel) obtained from myocytes in the day of isolation (Day 0). **(b)** Current traces of I_{Kr} (upper panel) and action potentials during pacing at cycle length (CL) = 200 msec (lower panel) obtained from myocytes after three days in cell culture and infection with adenovirus (Day three + HERG). HERG overexpression increased I_{Kr} and suppressed APD alternans.

that suppress inducible or spontaneously occurring ventricular tachycardia are expected to prevent sudden death. However, recent large-scale clinical trials have indicated that classes of drugs that are effective for the suppression of ventricular tachycardia, for example, Class I and Class III antiarrhythmic drugs, do not prevent sudden death and, in fact, could be proarrhythmic [45,56]. In contrast, other classes of drugs that are not particularly effective for the suppression of most forms of ventricular tachycardia, such as β-adrenergic receptor antagonists [57] and calcium channel antagonists [58], could reduce mortality from sudden death.

If a causal relationship between the slope of the APD restitution relation and VF is confirmed, such a result could have significant implications for the pharmacological therapy of sudden cardiac death. Drugs that reduce the slope of the restitution relation would be expected to prevent the development of VF, but would not be expected to suppress ventricular tachycardia, if ventricular tachycardia is caused by some variant of spiral wave re-entry. Conversely, drugs that do not reduce the slope of the restitution relation would not be expected to prevent VF, although they might suppress ventricular tachycardia, perhaps via a mechanism that does not involve alteration of restitution kinetics (e.g. slowing of conduction or prolongation of refractoriness).

Given these expectations, the value of existing agents for the prevention of VF could be re-evaluated in the light of their effects on the restitution relation and new drugs targeted against restitution could be developed. Naturally, such an effort would need to recognize that factors other than APD restitution significantly contribute to the induction and maintenance of VF. Furthermore, drugs intended to alter restitution selectively might have additional effects that could offset their intended effect. Nevertheless, judicious alteration of the APD restitution slope appears to be a promising approach to the treatment of VF and, as such, represents a potentially fruitful opportunity for drug development.

Conclusions

Electrical restitution is a recent addition to factors that play a key role in the development of ventricular tachyarrhythmias. Experimental interventions that reduce the slope of the restitution relation have been effective in suppressing VF. These results encourage further investigation of altering restitution as a means of preventing sudden death, with the realization that clinically useful interventions that flatten restitution without having untoward effects have yet to be developed.

Acknowledgements

I thank Fei Hua, Jeffrey Fox and Mark Riccio for helpful comments and criticisms. This work was supported by grants from the National Institutes of Health, the National Science Foundation and the American Heart Association, New York State Affiliate.

References

1 Zipes, D.P. and Wellens, H.J. (1998) Sudden cardiac death. *Circulation* 98, 2334–2351
2 Myerburg, R.J. *et al.* (1995) Life-threatening ventricular arrythmias: the link between epidemiology and pathophysiology. In *Cardiac Electrophysiology: From Cell to Bedside* (Zipes, D.P. and Jalife, J. eds), pp. 723–731, WB Saunders Co., Philadelphia
3 Krinsky, V.I. and Efimov, I.R. (1992) Votices with linear cores in mathematical models of excitable media. *Physica A* 188, 55–60
4 Winfree, A.T. (1998) Evolving perspectives during 12 years of electrical turbulence. *Chaos* 8, 1–20

5 Allessie, M.A. *et al.* (1977) Circus movement in rabbit atrial muscle as a mechanism of tachycardia. III. The 'leading circle' concept: a new model of circus movement in cardiac tissue without the involvement of an anatomical obstacle. *Circ. Res.* 41, 9–18

6 Frazier, D.W. *et al.* (1989) Stimulus-induced critical point. Mechanism for electrical initiation of reentry in normal canine myocardium. *J. Clin. Invest.* 83, 1039–1052

7 Witkowski, F.X. *et al.* (1998) Spatiotemporal evolution of ventricular fibrillation. *Nature* 392, 78–82

8 Weiss, J.N. *et al.* (1999) Chaos and the transition to ventricular fibrillation: a new approach to antiarrhythmic drug evaluation. *Circulation* 99, 2819–2826

9 Chen, P.S. *et al.* (1988) Mechanism of ventricular vulnerability to single premature stimuli in open-chest dogs. *Circ. Res.* 62, 1191–1209

10 Gilmour, R.F., Jr and Chialvo, D.R. (1999) Electrical restitution, critical mass, and the riddle of fibrillation. *J. Cardiovasc. Electrophysiol.* 10, 1087–1089

11 Pertsov, A.M. *et al.* (1993) Spiral waves of excitation underlie reentrant activity in isolated cardiac muscle. *Circ. Res.* 72, 631–650

12 Garfinkel, A. *et al.* (2000) Preventing ventricular fibrillation by flattening cardiac restitution. *Proc. Natl. Acad. Sci. U. S. A.* 97, 6061–6066

13 Riccio, M.L. *et al.* (1999) Electrical restitution and spatiotemporal organization during ventricular fibrillation. *Circ. Res.* 84, 955–963

14 Panfilov, A. and Pertsov, A. (2001) Ventricular fibrillation: evolution of the multiple wave hypothesis. *Philos. Trans. R. Soc. Lond. Ser. A.* 359, 1315–1325

15 Fenton, F.H. *et al.* (2002) Multiple mechanisms of spiral wave breakup in a model of cardiac electrical activity. *Chaos* 12, 852–892

16 Karma, A. (1993) Spiral breakup in model equations of action potential propagation in cardiac tissue. *Phys. Rev. Lett.* 71, 1103–1106

17 Karma, A. (1994) Electrical alternans and spiral wave breakup in cardiac tissue. *Chaos* 4, 461–472

18 Moe, G.K. (1964) A computer model of atrial fibrillation. *Am. Heart J.* 67, 200–220

19 Guevara, M.R. *et al.* (1984) Electrical alternans and period doubling bifurcations. *IEEE Comp. Cardiol.* 562, 167–170

20 Chialvo, D.R. *et al.* (1990) Low dimensional chaos in cardiac tissue. *Nature* 343, 653–657

21 Karagueuzian, H.S. *et al.* (1993) Action potential alternans and irregular dynamics in quinidine-intoxicated ventricular muscle cells. Implications for ventricular proarrhythmia. *Circulation* 87, 1661–1672

22 Nolasco, J.B. and Dahlen, R.W. (1968) A graphic method for the study of alternation in cardiac action potentials. *J. Appl. Physiol.* 25, 191–196

23 Rosenbaum, D.S. *et al.* (1994) Electrical alternans and vulnerability to ventricular arrhythmias. *New Engl. J. Med.* 330, 235–241

24 Pastore, J.M. *et al.* (1999) Mechanism linking T-wave alternans to the genesis of cardiac fibrillation. *Circ. Res.* 99, 1385–1394

25 Koller, M.L. *et al.* (1998) Dynamic restitution of action potential duration during electrical alternans and ventricular fibrillation. *Am. J. Physiol.* 275, H1635–H1642

26 Koller, M.L. *et al.* (2000) Effects of $[K^+]_o$ on electrical restitution and activation dynamics during ventricular fibrillation. *Am. J. Physiol. Heart Circ. Physiol.* 279, H2665–H2672

27 Fox, J.J. *et al.* (2002) Spatiotemporal transition to conduction block in canine ventricle. *Circ. Res.* 90, 289–296

28 Chen, P.S. *et al.* (1997) Spirals, chaos, and new mechanisms of wave propagation. *Pacing Clin. Electrophysiol.* (*PACE*) 20, 414–421

29 Panfilov, A. (1998) Spiral breakup as a model of ventricular fibrillation. *Chaos* 8, 57–64

30 Chudin, E. *et al.* (1999) Intracellular Ca^{2+} dynamics and the stability of ventricular tachycardia. *Biophys. J.* 77, 2930–2941

31 Winslow, R.L. *et al.* (1999) Mechanisms of altered excitation-contraction coupling in canine tachycardia-induced heart failure, II. *Circ. Res.* 84, 571–586

32 Luo, C.H. and Rudy, Y. (1994) A dynamic model of the cardiac ventricular action potential, I: simulation of ionic currents and concentration changes. *Circ. Res.* 74, 1071–1096

33 Fox, J.J. *et al.* (2002) Ionic mechanism of cardiac alternans. *Am. J. Physiol.* 282, H516–H530

34 Bian, J. *et al.* (2001) HERG K^+ channel activity is regulated by changes in phosphatidyl inositol 4,5-bisphosphate. *Circ. Res.* 89, 1168–1176

35 Kiehn, J. (2001) Regulation of the cardiac repolarizing HERG potassium channel by protein kinase A. *Trends Cardiovasc. Med.* 10, 205–209

36 Heath, B.M. and Terrar, D.A. (2000) Protein kinase C enhances the rapidly activated delayed rectifier potassium current, I_{Kr}, through a reduction in C-type inactivation in guinea pig ventricular myocytes. *J. Physiol.* 522, 391–402

37 Marx, S.O. *et al.* (2002) Requirement of a macromolecular signaling complex for β adrenergic receptor modulation of the KCNQ1-KCNE1 potassium channel. *Science* 295, 496–499

38 Reuter, H. and Scholz, H. (1977) The regulation of calcium conductance of cardiac muscle by adrenaline. *J. Physiol.* 246, 49–62

39 Sanguinetti, M.C. and Jurkiewicz, N.K. (1990) Two components of cardiac delayed rectifier K^+ current. Differential sensitivity to block by class III antiarrhythmic agents. *J. Gen. Physiol.* 96, 195–215

40 Rocchetti, M. *et al.* (2001) Rate dependency of delayed rectifier currents during the guinea pig ventricular action potential. *J. Physiol.* 534, 721–732

41 Gintant, G.A. (2000) Characterization and functional consequences of delayed rectifier current transient in ventricular repolarization. *Am. J. Physiol.* 278, H806–H817

42 Gintant, G.A. (1995) Regional differences in I_K density in canine left ventricle: role of I_{Ks} in electrical heterogeneity. *Am. J. Physiol.* 268, H604–H613

43 Clay, J.R. *et al.* (1995) A quantitative description of the E-4031-sensitive repolarization current in rabbit ventricular myocytes. *Biophys. J.* 69, 1830–1837

44 Nuss, H.B. *et al.* (1999) Overexpression of a human potassium channel suppresses cardiac hyperexcitability in rabbit ventricular myocytes. *J. Clin. Invest.* 103, 889–896

45 Waldo, A.L. *et al.* (1996) Effect of d-sotalol on mortality in patients with left ventricular dysfunction after recent and remote myocardial infarction. The SWORD Investigators. Survival with ORal D-sotalol. *Lancet* 348, 7–12

46 Hondeghem, L.M. and Snyders, D.J. (1990) Class III antiarrhythmic agents have a lot of potential but a long way to go. Reduced effectiveness and dangers of reverse use dependence. *Circulation* 81, 686–690

47 Mohammad, S. *et al.* (1997) Blockage of the HERG human cardiac K^+ channel by the gastrointestinal prokinetic agent cisapride. *Am. J. Physiol.* 273, H2534–H2538

48 Roy, M. *et al.* (1996) HERG, a primary human ventricular target of the nonsedating antihistamine terfenadine. *Circulation* 94, 817–823

49 Mitcheson, J.S. *et al.* (2000) A structural basis for drug-induced long QT syndrome. *Proc. Natl. Acad. Sci. U. S. A.* 97, 12329–12333

50 Mitcheson, J.S. *et al.* (2000) A structural basis for drug-induced long QT syndrome. *Proc. Natl. Acad. Sci. U. S. A.* 97, 12329–12333

51 Keating, M.T. and Sanguinetti, M.C. (1996) Molecular genetic insights into cardiovascular disease. *Science* 272, 681–685

52 Curran, M.E. *et al.* (1995) A molecular basis for cardiac arrhythmia: HERG mutations cause long QT syndrome. *Cell* 80, 795–803

53 Roden, D.M. and Balser, J.R. (1999) A plethora of mechanisms in the HERG-related long QT syndrome. Genetics meets electrophysiology. *Cardiovasc. Res.* 44, 242–246

54 Sanguinetti, M.C. *et al.* (1995) A mechanistic link between an inherited and an acquired cardiac arrhythmia: HERG encodes the I_{Kr} potassium channel. *Cell* 81, 299–307

55 Hund, T.J. *et al.* (2000) Dynamics of action potential head–tail interaction during reentry in cardiac tissue: ionic mechanisms. *Am. J. Physiol.* 279, H1869–H1879

56 Echt, D.S. *et al.* (1991) Mortality and morbidity in patients receiving encainide, flecainide or placebo: the cardiac arrhythmia suppression trial. *New Engl. J. Med.* 324, 781–788

57 Yusuf, S. *et al.* (1988) Overview of results of randomized clinical trials in heart disease: I. Treatments following myocardial infarction. *J. Am. Med. Assoc.* 260, 2088–2095

58 Held, P.H. and Yusuf, S. (1994) Impact of calcium channel blockers on mortality. In *Cardiovascular Pharmacology and Therapeutics* (Singh, B.N. *et al.*, eds), pp. 525–533, Churchill Livingston