Torsades de Pointes in the Guinea-Pig Heart

Editorial to: “Dofetilide Promotes Repolarization Abnormalities in Perfused Guinea-Pig Heart” by O.E. Osadchii

Ewa Soltysinska · Morten B. Thomsen

Published online: 26 September 2012
© Springer Science+Business Media New York 2012

It is increasingly recognized that certain cardiovascular and non-cardiovascular medicines may induce a potentially life threatening form of ventricular tachyarrhythmia known as torsades de pointes (TdP; [1, 2]). This realization led to the withdrawal of a number of drugs from the market, and proarrhythmic testing of a drug candidate is routinely performed today using a battery of pre-clinical tests. Yet, as development of TdP is multifactorial with female gender, bradycardia, hypokalemia and structural heart disease as the major risk factors, it is not surprising that the incidence of arrhythmia in healthy pre-clinical animal models is negligible. Since TdP is frequently not a direct read-out from the screening models, the determination of proarrhythmic risks is based on surrogate markers. Many such biomarkers for TdP have been proposed, including QT and APD prolongation, reverse-use dependence of drugs, and temporal and spatial dispersion of repolarization duration [3–9]. Undoubtedly, evaluation of biomarkers for proarrhythmia advanced our knowledge of TdP mechanisms; however, utility of these biomarkers in drug development is still limited as both their specificity and sensitivity is far from optimal. Moreover, regulatory authorities approving novel drugs for use in humans are not recognizing proarrhythmic animal models as informative of human risk. Instead, human safety is determined based on perhaps the most imperfect biomarker, the QT interval, recorded from healthy volunteers. The potential consequences are two-fold: approval of a drug that turns out to be proarrhythmic and discontinuation of the

development of a promising therapy for an unmet medical need.

In the present issue of *Cardiovascular Drugs and Therapy*, the proarrhythmic mechanism of dofetilide is investigated and various biomarkers in isolated, perfused guinea-pig heart preparations are reported [10]. Dofetilide is a potent I_{Kr} blocker and class-III antiarrhythmic drug with experimentally and clinically documented TdP risk. The author shows that addition of dofetilide (10 nM) to the perfusion fluid of isolated hearts is associated with electrophysiological changes that we would normally consider harbingers of TdP: reverse-use-dependent prolongation of repolarization, augmented spatial dispersion of repolarization, and increased steepness of action-potential rate adaptation. In spite of these proarrhythmic changes and experimental conditions favoring TdP induction (female gender, bradycardia), dofetilide only infrequently induced ectopic beats and short-lasting (2–5s) runs of monomorphic ventricular tachycardia; TdP never developed. Was the dofetilide concentration simply too low to trigger TdP? Complete block of I_{Kr} in guinea-pig cardiomyocytes required 1000 nM dofetilide in early reports [11], yet the data presented by Dr Osadchii in this issue confirms the findings of two previous studies showing that neither 10 nor 1000 nM dofetilide is able to reproducibly induce TdP in perfused, guinea-pig hearts [12, 13]. In addition, Guo et al. who used the guinea-pig model to analyze cardiovascular risk of 15 reference compounds, including dofetilide, reported arrhythmias for only 3 drugs, namely aconitine, ouabain and pinacidil [13]. Interestingly, none of these drugs are associated with considerable I_{Kr} block.

The low incidence of arrhythmias in guinea-pig hearts appears to be at odds with data obtained from rabbit hearts, where a much higher sensitivity for proarrhythmic drugs is

E. Soltysinska · M. B. Thomsen (✉)
Danish National Research Foundation Centre for Cardiac
Arrhythmia, University of Copenhagen,
Blegdamsvej 3B,
DK-2200 Copenhagen, Denmark
e-mail: mbthom@sund.ku.dk

reported [3, 13]. What is the underlying mechanism? Primarily, the guinea pig has a prominent participation of I_{Ks} in cardiac repolarization [14], indicating that there is a large repolarizing current remaining even during circumstances of dofetilide administration. Secondly, the smaller heart size may reduce the chance of sustaining the perpetuation of a triggered event [15], although this notion has been challenged by many recent reports of ventricular arrhythmias and fibrillation in mice and rats. Thirdly, guinea-pig cardiomyocytes are renowned for lacking the transient outward current [16], which by adjusting the plateau voltage may impact on the frequency of early afterdepolarizations secondary to reactivation of the L-type calcium channel.

How do we sensitize the guinea-pig heart to TdP arrhythmias?

In the continued absence of reliable or verified biomarkers, despite the effort put into the area, the goal for drug testing should be triggering the arrhythmia. Based on the finding that drug-induced TdP in the clinical setting is a product of complex interplay of predisposing factors, it is apparent that useful proarrhythmia models should be primed or sensitized according to our knowledge on risk factors and contributors [17]. Repolarization reserve has previously been introduced [18], and we have suggested to define it as the *ability of the heart to withstand a challenge on repolarization* [4]. For example, repolarization reserve was reduced in patients with a history of drug-induced TdP [19] and they showed excessive QT prolongation upon challenge with sotalol, an I_{Kr} blocking drug, when compared to control patients [20]. Hence, the sensitivity of guinea pigs to proarrhythmia might be improved by introducing experimental conditions known to compromise repolarization reserve. These conditions potentially include: (i) lowering K^+ concentration in tissue perfusate; (ii) co-application of agents blocking repolarizing currents or enhancing depolarizing currents; and (iii) performing experiments on remodeled hearts.

Hypokalemia is a well-recognized risk factor of development of TdP. Hypokalemic conditions increase the driving force for potassium increasing the outward currents e.g. I_{Ks} ; however, the kinetics of ion channel governing I_{Kr} is sensitive to extracellular potassium in such a way that decreased potassium in the perfusion fluid actually causes a decrease in I_{Kr} amplitude [21]. Moreover, hypokalemia inhibits I_{K1} , and the combined cellular electrophysiological effects of low external K^+ is action potential prolongation and hyperpolarization-induced slowing of impulse conduction [21, 22]. The guinea pig with predominant I_{Ks} and limited I_{Kr} may thus, theoretically, be more resistant to arrhythmias in periods of relative hypokalemia. This may explain why in guinea-pig heart, lowering K^+ concentration to 2.1 mM

failed to significantly sensitize the model towards development of TdP associated with dofetilide infusion and only rare incidence of drug-induced TdP is noted (e.g. 1/24 hearts in [12]). Conversely, in isolated rabbit hearts with repolarization governed predominantly by I_{Kr} , hypokalemic solution (3 mM) greatly exacerbated torsadogenic action of dofetilide [23]. Dr. Osadchii and colleagues have previously shown that guinea pig action-potential duration and effective refractory periods during physiological pacing rates are only modestly changed during hypokalemia, although increased heterogeneity and facilitated action potential alternans and ventricular fibrillation were observed [24]. Thus, presently it is unclear whether hypokalemia will increase sensitivity to drug-induced proarrhythmia in the guinea-pig heart.

Accumulating evidence suggests that reducing repolarization reserve by pharmacological I_{Ks} blockade greatly enhances occurrence of TdP associated with I_{Kr} inhibition. In this regard, it has been reported that inhibition of I_{Ks} with either chromanol-293B or HMR1556 increased torsadogenic propensity of I_{Kr} blockers in rabbits and dogs. The combined blockade of I_{Ks} and I_{Kr} in anesthetized guinea pigs was not proarrhythmic, but a concomitant administration of adrenaline triggered TdP [25]. Moreover, pharmacological gain of I_{Na} function has been used successfully to reduce repolarization reserve and induce TdP in ex-vivo studies [12, 26, 27]. Thus, co-administration of drugs attenuating repolarization strength or augmenting depolarizing currents increase sensitivity to TdP; however, it may also lower specificity of the model considerably, and additional studies are prompted to optimize experimental conditions of pharmacologically-primed proarrhythmia in the guinea pig [28].

Structural heart disease predisposes to TdP. Patients with a history of heart failure have enhanced susceptibility towards TdP associated with I_{Kr} inhibition using d, l-sotalol or dofetilide [29, 30]. Experimental data from dogs and rabbits confirm clinical findings, and testing drugs on remodeled guinea-pig hearts may substantially increase the susceptibility towards development of arrhythmia. To our knowledge, no studies were performed to evaluate drug-induced proarrhythmia in hypertrophied or failing guinea pig hearts. This may be because, up to now, there was a paucity of clinically relevant models with well-described significant structural, mechanical and electrophysiological cardiac remodeling in guinea pigs. Recently, a novel model of heart failure induced by chronic β -adrenergic stimulation has been developed [31, 32]; however additional follow-up studies are required to determine whether this model has an increased risk of drug-induced TdP.

In conclusion, the study by Dr. Osadchii contributes importantly to our understanding of the guinea-pig electrophysiology and to its potential as proarrhythmic animal

model. Presently, TdP has not been induced in large frequency, highlighting the requirement for additional fine tuning of the model before the species can be adopted for drug-screening. In the era of imperfect biomarkers, pre-clinical assessment of proarrhythmic risk should be based on the incidence of TdP in susceptible animal models.

References

1. Fenichel RR, Malik M, Antzelevitch C, Sanguinetti MC, Roden DM, Priori SG, et al. Drug-induced torsades de pointes and implications for drug development. *J Cardiovasc Electrophysiol*. 2004;15:475–95.
2. Roden DM. Drug-induced prolongation of the QT interval. *N Engl J Med*. 2004;350:1013–22.
3. Hondeghem LM, Carlsson L, Duker G. Instability and triangulation of the action potential predict serious proarrhythmia, but action potential duration prolongation is antiarrhythmic. *Circulation*. 2001;103:2004–13.
4. Thomsen MB, Verduyn SC, Stengl M, Beekman JD, de Pater G, van Opstal J, et al. Increased short-term variability of repolarization predicts d-sotalol-induced torsades de pointes in dogs. *Circulation*. 2004;110:2453–9.
5. Graff C, Andersen MP, Xue JQ, Hardahl TB, Kanters JK, Toft E, et al. Identifying drug-induced repolarization abnormalities from distinct ECG patterns in congenital long QT syndrome: a study of sotalol effects on T-wave morphology. *Drug Saf*. 2009;32:599–611.
6. Fossa AA, DePasquale MJ, Raunig DL, Avery MJ, Leishman DJ. The relationship of clinical QT prolongation to outcome in the conscious dog using a beat-to-beat QT-RR interval assessment. *J Pharmacol Exp Ther*. 2002;302:828–33.
7. Said TH, Wilson LD, Jeyaraj D, Fossa AA, Rosenbaum DS. Transmural dispersion of repolarization as a preclinical marker of drug-induced proarrhythmia. *J Cardiovasc Pharmacol*. 2012;60:165–71.
8. Thomsen MB, Louch WE. Special Interest Group on Cardiac Physiology established within the Scandinavian Physiological Society. *Acta Physiol (Oxf)*. 2012;204:464.
9. Wang Q, Curran ME, Splawski I, Burn TC, Millholland JM, VanRaay TJ, et al. Positional cloning of a novel potassium channel gene: KVLQT1 mutations cause cardiac arrhythmias. *Nat Genet*. 1996;12:17–23.
10. Osadchii OE. Dofetilide promotes repolarization abnormalities in perfused guinea-pig heart. *Cardiovasc Drugs Ther*. 2012;26: this issue.
11. Jurkiewicz NK, Sanguinetti MC. Rate-dependent prolongation of cardiac action potentials by a methanesulfonanilide class III antiarrhythmic agent. Specific block of rapidly activating delayed rectifier K⁺ current by dofetilide. *Circ Res*. 1993;72:75–83.
12. Cheng HC, Incardona J. Models of torsades de pointes: effects of FPL64176, DPI201106, dofetilide, and chromanol 293B in isolated rabbit and guinea pig hearts. *J Pharmacol Toxicol Methods*. 2009;60:174–84.
13. Guo L, Dong Z, Guthrie H. Validation of a guinea pig Langendorff heart model for assessing potential cardiovascular liability of drug candidates. *J Pharmacol Toxicol Methods*. 2009;60:130–51.
14. Lu Z, Kamiya K, Ophthof T, Yasui K, Kodama I. Density and kinetics of I(Kr) and I(Ks) in guinea pig and rabbit ventricular myocytes explain different efficacy of I(Ks) blockade at high heart rate in guinea pig and rabbit: implications for arrhythmogenesis in humans. *Circulation*. 2001;104:951–6.
15. Garrey WE. Auricular fibrillation. *Physiol Rev*. 1924;4:215–50.
16. Varro A, Lathrop DA, Hester SB, Nanasi PP, Papp JG. Ionic currents and action potentials in rabbit, rat, and guinea pig ventricular myocytes. *Basic Res Cardiol*. 1993;88:93–102.
17. Farkas AS, Nattel S. Minimizing repolarization-related proarrhythmic risk in drug development and clinical practice. *Drugs*. 2010;70:573–603.
18. Roden DM. Taking the “idio” out of “idiosyncratic”: predicting torsades de pointes. *Pacing Clin Electrophysiol*. 1998;21:1029–34.
19. Hinterseer M, Thomsen MB, Beckmann BM, Pfeufer A, Schimpf R, Wichmann HE, et al. Beat-to-beat variability of QT intervals is increased in patients with drug-induced long-QT syndrome: a case control pilot study. *Eur Heart J*. 2008;29:185–90.
20. Kaab S, Hinterseer M, Nabauer M, Steinbeck G. Sotalol testing unmasks altered repolarization in patients with suspected acquired long-QT-syndrome—a case-control pilot study using i.v. sotalol. *Eur Heart J*. 2003;24:649–57.
21. Sanguinetti MC, Jurkiewicz NK. Role of external Ca²⁺ and K⁺ in gating of cardiac delayed rectifier K⁺ currents. *Pflugers Arch*. 1992;420:180–6.
22. Osadchii OE. Mechanisms of hypokalemia-induced ventricular arrhythmogenicity. *Fundam Clin Pharmacol*. 2010;24:547–59.
23. Barrett TD, Hennan JK, Fischbach PS, O'Neill BP, Driscoll Jr EM, Lucchesi BR. Tedisamil and dofetilide-induced torsades de pointes, rate and potassium dependence. *Br J Pharmacol*. 2001;132:1493–500.
24. Osadchii OE, Larsen AP, Olesen SP. Predictive value of electrical restitution in hypokalemia-induced ventricular arrhythmogenicity. *Am J Physiol Heart Circ Physiol*. 2010;298:H210–20.
25. Michael G, Kane KA, Coker SJ. Adrenaline reveals the torsadogenic effect of combined blockade of potassium channels in anaesthetized guinea pigs. *Br J Pharmacol*. 2008;154:1414–26.
26. Wu L, Shryock JC, Song Y, Li Y, Antzelevitch C, Belardinelli L. Antiarrhythmic effects of ranolazine in a guinea pig in vitro model of long-QT syndrome. *J Pharmacol Exp Ther*. 2004;310:599–605.
27. Gerhardt A, Scholtysik G, Schaad A, Haltiner R, Hess T. Generating and influencing Torsades de Pointes-like polymorphic ventricular tachycardia in isolated guinea pig hearts. *Basic Res Cardiol*. 1998;93:285–94.
28. Thomsen MB. Double pharmacological challenge on repolarization opens new avenues for drug safety research. *Br J Pharmacol*. 2007;151:909–11.
29. Lehmann MH, Hardy S, Archibald D, Quart B, MacNeil DJ. Sex difference in risk of torsade de pointes with d, l-sotalol. *Circulation*. 1996;94:2535–41.
30. Pedersen HS, Elming H, Seiback M, Burchardt H, Brendorp B, Torp-Pedersen C, et al. Risk factors and predictors of Torsade de pointes ventricular tachycardia in patients with left ventricular systolic dysfunction receiving Dofetilide. *Am J Cardiol*. 2007;100:876–80.
31. Soltysinska E, Thiele S, Olesen SP, Osadchii OE. Chronic sympathetic activation promotes downregulation of beta-adrenoceptor-mediated effects in the guinea pig heart independently of structural remodeling and systolic dysfunction. *Pflugers Arch*. 2011;462:529–43.
32. Soltysinska E, Olesen SP, Osadchii OE. Myocardial structural, contractile and electrophysiological changes in the guinea-pig heart failure model induced by chronic sympathetic activation. *Exp Physiol*. 2011;96:647–63.