HESI - ILSI Meeting , November 2-3, 2005, Cristal City, Virginia, USA.

Break-out Session on " Dynamics of Periodicity"

Some Suggestions for Future Approaches

De Clerck, Fred M.Sc., Ph. D., Pharm.D.

Professor, University of Utrecht , the Netherlands

Former Senior Director, Center of Excellence for Cardiovascular Safety Research & Benefit-Risk Management
Johnson & Johnson Pharmaceutical Research & Development
A division of Janssen Pharmaceutica NV
Turnhoutseweg 30, B-2340 Beerse, Belgium

Private address: Laakstraat 7, B-2340 Beerse , Belgium
Phone & fax 0032(0)14614835
É-mail : fred.de.clerck@pandora.be

October 13th, 2005

Content

A. Background

A.1. Several pre-clinical tests on isolated cardiac tissues as well as in experimental animals indicate that the arrhythmogenic potential of drugs is determined not only by their effect on the duration of ventricular repolarization (prolongation or shortening) but also by the way in which they affect the heterogeneity of ventricular repolarization and/or autonomic tone (see, for example: ref 1-8).

A.2. Recently, several new data on this topic became available. For instance, results with a large set of compounds in isolated rabbit hearts suggest that either drug-induced prolongation (induction of torsades de pointes) or shortening (induction of ventricular fibrillation) of the duration of the action potential (AP) is arrhythmogenic, provided it is accompanied by changes in AP characteristics. Such changes include triangulation of the shape, reverse use dependency, beat-to-beat instability of the AP duration and dispersion of AP duration between epicardial and endocardial layers (TRIaD; ref 2-4). Also, in canine experiments *in vivo*, torsadogenic drugs eliciting early afterdepolarizations such as dofetilide, sotalol, erythromycin (see, for example: ref 5, 6) not only prolong QTc but also induce beat-to-beat instability in QT prolongation (time-dependent QT dispersion) as well as increases in the difference in the duration of QT peak to QT end of the surface ECG (transmural QT dispersion as defined in ref 1; see ref 5, 8). By contrast, induction of QT time-dependent and/or transmural dispersion without QTc prolongation as with isoproterenol, or QTc prolongation without induction of QT heterogeneity as with moxifloxacin at certain doses do not elicit pro-arrhythmogenic early afterdepolarizations *in vivo* in dogs (ref 8). Such *in vitro* and *in vivo* experimental data strongly suggest that the arrhythmogenic potential of compounds (induction of torsades de pointes or/ and of ventricular fibrillation) depends on their overall effect on the electrophysiological characteristics of ventricular repolarization, rather than a "simple" effect of QTc duration as such, despite intrinsic differences (ref 1) in ion channel density/activity between several transmural layers of the myocardium .

A.3. The mechanisms which may explain such differential drug effects on the dynamics of periodicity in ventricular repolarization are multiple, but still remain elusive. They may include :

A.3.1. Pharmacodynamic factors: drug effects on multiple cardiac ion channels and/or adrenergic receptors.

Examples of such mechanisms are provided compounds blocking the hERG channel (IKr) in isolated cells (verapamil, sertindole, pimozide..), yet not producing a high level of arrhythmogenesis *in vitro* or

in vivo, possibly via their additional effect on other cardiac ion channels such as ICa, INa or on cardiac adrenergic receptors (amiodarone) (see, for example: ref 5, 9) and by compounds attenuating cardiac sodium and/or calcium channels (lidocaine, nisoldipine, n-3 polyunsaturated fatty acids), thus reducing QT heterogeneity and pro-arrhythmias induced by Class III agents such as almokalant or dofetilide in isolated rabbit ventricular muscles or hearts (ref 10,11).

A.3.2.Pharmacokinetic factors: drug distrubution/ effect throughout myocardial layers

Despite an intrinsic difference (ref 1) in ion channel density/ activity between several transmural layers of the myocardium, the arrhythmogenic- torsadogenic potential of a given compound can depend on the rate of its biological exposure. A classic example is that of almokalant which is highly torsadogenic in anaesthetized rabbits challenged with methoxamine when delivered at a fast infusion rate, but not when administered at a prolonged infusion rate , despite similar plasma levels and effects on QTc duration as the former delivery rate (ref 12).
This raises the possibility that a *time-dependent or uniform* drug distribution/effect on ion channels or/and receptors throughout the different layers of myocardium can avoid arrhythmogenicity. This may depend on the specific physicochemical characteristics of each individual compound at hand.

A.4. Drug-induced effects on cardiac electrophysiology are bound to be far more complex in patients than those in normal or "diseased" animal preparations. Therefore, interactions between clinical and pre-clinical investigators, as well as with regulatory authorities, are needed to determine the nature and validity of future pre-clinical approaches for a refined detection of drug-induced pro-arrhythmias.

B. Purpose of the break- out session on "Dynamics of Periodicity"

The purposes of this break-out session can include :

1. identify mechanisms in terms of cellular biology/pharmacology by which such differences in arrhythmogenic potential of drugs operate
    - ✓ see detailed section below (C. Some Suggestions for Future Approaches)

2. mobilize resources for a set of targeted tests identifying a next approach to characterize such drug effects:
    - ✓ identify and validate suitable research organizations & obtain prices/timelines
    - ✓ mobilize sponsors in private and governmental institutions
    - ✓ produce detailed protocols for the projects

3. select/activate a set of tests, capable of convincing regulatory agencies.
    - ✓ obtain agreement on intensions, protocols, timelines with regulatory agencies

- ✓ obtain agreement with a major peer-reviewed journal to eventually publish results
- ✓ initiate the projects and report the results in peer-reviewed journal and to regulatory agencies

C. Some Suggestions for Future Approaches

C.1. Item 1. Identify mechanisms in terms of cellular biology/pharmacology by which such differences in arrhythmogenic potential of drugs operate

C.1.1. In vitro tests

C.1.1.1. Approaches via tests on isolated cardiac tissues.

The pioneering work of Dr Antzelevitch C and his group revealed the role of temporal and transmural dispersion of ventricular repolarization in the eventual torsadogenic effect of QT prolonging drugs (see, for example: ref 1). Several recent experiments (ref 2-4, 13) support the importance of changes in the characteristics of ventricular repolarization (triangulation, reverse use dependency, instability in time between consecutive beats, dispersion/ heterogeneity between epi-and endocardial AP duration: TRIaD; see ref 2) and of ventricular conduction time as prerequisites for the induction of cardiac arrhythmias (torsades de pointes = TdP or ventricular fibrillation= VF) by compounds prolonging (TdP induction) or shortening the duration of ventricular repolarization (VF induction).

In principle, additional analyses/tests can be performed in any appropriate *in vitro* set-up, but an approach in the isolated female rabbit hearts (ref 2-4) would have the advantage of a high throughput & resolution and a solid validation via a large data base on cardiovascular and non-cardiovascular compounds (see: Hondeghem LM, presentation at the ILSI meeting, november 2, 2005).

Future approaches in this model (or another equivalent one) to be considered include …

- A search on the available data base to identify & select interesting compounds with differential effects on ventricular repolarization and conduction time, in particular those..
    1. producing APD prolongation, plus TRIaD and eliciting TdP or other AR
    2. producing APD prolongation , without TRIaD , not eliciting TdP or other AR
    3. producing APD shortening, plus TRIaD and eliciting TdP or other AR
    4. producing APD shortening, without TRIaD, not eliciting TdP or other AR

    (obviously, the identity of compounds can/will only be disclosed when the sponsor of the tests agrees in an appropriate statement).

- A search on the available data base to identify (if possible) a *dominant* parameter (if any..) in the TRIaD series, leading to an eventual AR effect. This is importance to determine whether measurements of beat- to -beat differences in QT duration (time-dependent " QT instability") is sufficient or whether other ECG parameters than beat-to-beat variability in QT duration (for example, differences in the QT Peak to QT end..) should be quantified as well in drug-treated patients.

- A contact with regulatory agencies to obtain a (strictly confidential) list of compounds of special clinical importance in terms of their electrophysiological effects and test them in the *in vitro* set-ups (and , if possible, in the in vivo set-ups), if practically feasible.

- Additional tests with a number of salient compounds, which are already available in the public domain. Such compounds should already be documented for the fact that they can produce QTc prolongation but have a low torsadogenic effect in man. Compounds of particular interest in this respect could be, for example, sertindole, pimozide, moxifloxacin, ebastine, fenoxifenadine.

- A check for potential pharmacodynamic effects: organize tests in which some selected compounds are tested 1. via a short exposure at increasing concentrations versus 2. a long exposure to the same preparation at a constant concentration during a protracted time period , allowing drug-saturation/homogeneous distribution throughout different layers of the myocardium. The observation by Di Diego JM *et al* in the group of Dr Antzelevitch C in their canine wedge preparation that cisapride, at higher concentrations, but also ***longer contact times*** further prolongs QT vis -a- vis lower concentrations/shorter contact times without increasing transmural dispersion or inducing TdP, leaves open an important research quest. Indeed, that research quest is whether or not such a " biphasic" effect of cisapride (QT prolongation + prolonged Tpeak-Tend +TdP at low concentrations/short exposure times versus QT prolongation without increased Tpeak-Tend and without TdP at higher concentrations/ longer exposure times) is due to an additional drug effect on "correcting" channels at higher concentrations (INa, ICa..), or to a saturation/ homogeneous effect on IKr channels throughout different myocardial layers at longer exposition time *in vitro* (ref 14).

C.1.1.2. Additional approaches via tests on ion channels/cardiac receptors activity.

- The potential torsadogenic effect of a drug-induced IKr blockade can be modulated by an effect on other ion channels (INa, ICa..) or on cardiac autonomic receptors (see, for example: ref 9, 11). However, drug effects on the latter ion channels can shorten action potential duration and favour other types of arrhythmias.

- Therefore, we suggest to test the compounds, scrutinized in isolated rabbit hearts, in depth for effects on several cardiac ion channels and receptors. (concentration dependency, incubation times, kinetics of effect at different cycle lengths and stimulation rates..).

C.1.2. In vivo tests

- So far, only limited data seem to be available from tests in experimental animals such as anaesthetized dogs in which detailed analyses of ECG parameters - such as beat-to-beat variability of QT duration or differences between QT peak to QT end as a parameter for transmural dispersion – have been applied systematically (sotalol, dofetilide, isoproterenol, flecainide, sertindole; see, for example; ref 5-8, 16).
- To ensure their validity for clinical practise, the feasibility/ relevance of such detailed ECG analyses in man should be checked on beforehand.
- Additional compounds of interest should be tested in this model using ECG readings & analyses using appropriate soft ware programmes, together with monophasic action potential recordings in the ventricle to check on incidences of pro-arrhythmogenic early afterdepolarizations. Again, the same set of compounds, tested for effects on action potential characteristics in isolated cardiac tissues and on cardiac ion channels/ receptors, should be considered for testing here.
- To check the potential importance of pharmacokinetic factors (see, for example: ref 12), tests with the same compound delivered via bolus versus slow infusion are to be considered.
- Measurements of drug concentrations in plasma and in heart tissues seems imperative for all such experiments.

D. References

1. Antzelevitch C. Cardiac repolarisation. The long and short of it. Eurospace 2005 sep;7. Supp 2: 3-9.

2. Hondeghem LM. TRIad: foundation for proarrhythmia (triangulation, reverse use dependence and instability). Novartis Found Symp. 2005;266:235-44; discussion 244-50.

3.Shah RR, Hondeghem LM. Refining detection of drug-induced proarrhythmia: QT interval and TRIaD. Heart Rhythm. 2005 Jul;2(7):758-72.

4. Hondeghem LM, Lu HR, van Rossem K, De Clerck F. Detection of proarrhythmia in the female rabbit heart: blinded validation. J Cardiovasc Electrophysiol 2003; 14: 287-294.

5. van der Linde H, Van de Water A, Loots W, De Clerck F. Effects on dispersion and heterogeneity of ventricular repolarization beyond its duration in anaesthetized dogs. J Pharmacol Toxicol Methods 49, 225, 2004.

6 .Thomsen MB, Verduyn SC, Stengl M, Beekman JD, de Pater G, van Opstal J, Volders PG, Vos MA. Increased short-term variability of repolarization predicts d-sotalol-induced torsades de pointes in dogs. Circulation 2004 Oct 19; 110 (16): 2453-2459).
7. Fossa AA, Wisialowski T, Magnano A, Wolfgang E, Winslow R, Gorczyca W, Crimin K, Raunig DI. Dynamic beat-to-beat modelling of the QT-RR interval relationship: analysis of QT prolongation during alterations of autonomic state versus human ether a-go-go- related gene inhibition. J Pharmacol Exp Ther 2005 Jan; 312 (1): 1-11.
8. De Clerck F, Gallacher D. Effects on dispersion and heterogeneity of ventricular repolarization beyond its duration in anaesthetized dogs. Scientific Meeting Maastricht, the Netherlands, April 14-15 , 2005. " Beat-to-beat variability of repolarization to tests for the pro-arrhythmic potential of drugs. Challenging normal and diseased hearts" Organizers: Volders P, Vos, M. In press, 2005.
9. Haverkamp W, Eckhardt L, Matz J, Frederiksen K. Sertindole: cardiac electrophysiological profile. J Pschychiatry in Clinical Practice 2002, 6 (supp 1), 511-520.
10. Hondeghem LM , Dujardin K. Dietary spplementation with n-3 polyunsaturated fatty acids: a far less toxic alternative and perhaps superior antiarrhythmic therapy to amiodarone? . Abstract Number 12298, accepted for presentation at the American Heart Association Scientific Sessions, 2005.
11. Abrahamsson C, Carlsson L, Duker G. Lidocaine and nisoldipine attenuate almokalant-induced dispersion of repolarization and early afterdepolarizations in vitro. J Cardiovasc. Electrophysiol 1996 Nov; 7 (11): 1074-1081.
12. Carlsson L, Abrahamsson C, Andersson B, Duker G, Schiller-Linhardt G. Proarrhythmic effects of the class III agent almokalant: importance of infusion rate, QT dispersion, and early afterdepolarisations. Cardiovasc Res 1993 Dec; 27 (12): 2186-2193.
13. Lu HR, Vlaminckx E, Van Ammel K, De Clerck F. Drug-induced long QT in isolated rabbit Purkinje fibers: importance of action potential duration, triangulation and early depolarizations. Eur J Pharmacol 2002, 452: 183-192.
14. Di Diego JM, Bernardillelli L, Antzelevitch C. Cisapride-induced transmural dispersion of repolarization and torsades de pointes in the canine left ventricular wedge preparation during epicardial stimulation. Circulation. 2003; 108: 1027- 1033.
15. Sanguinetti MC, Chen J, Fernandez D, Kammiya K, Mitcheson J, Sanchez-Chapulala JA. Physicochemical basis for binding and voltage-dependent block of hERG channels by structurally diverse drugs. Novartis Found Symp 2005; 266: 159-1566; discussion.
16. Thomsen MB. Beat-to-beat variability of repolarization and drug-induced torsades de pointes in the canine heart. Doctoral Thesis. Promotor: Prof. Dr. Vos MA. Maastricht, 2005.