The “M” CELLS OF VENTRICULAR MIDMYOCARDIUM

Dispersion of repolarization across the ventricular wall has been suggested to underlie the inscription of the normal electrocardiographic T wave and when amplified to contribute prominently to the development of cardiac arrhythmias.[1–3]

The magnitude of transmural dispersion of repolarization (TDR) is due to intrinsic differences in the action potential duration (APD) of the three principal cell types that comprise the ventricular myocardium and the extent to which these repolarization differences are damped by electrotonic forces. An increase in tissue resistivity in the deep subepicardium,[4] due to a sharp transition in cell orientation,[3] reduced expression of connexin 43 (Cx43)[5, 6] and increased density of collagen[7] in this region, contributes to the expression of repolarization heterogeneities across the ventricular wall by limiting the degree of electrotonic interaction between the myocardial layers. Thus, the degree of electrotonic coupling, together with the intrinsic differences in APD, determine the extent to which TDR is expressed, its impact on arrhythmogenesis as well as on the morphology of the T wave. It is noteworthy that even in the absence of any difference in final repolarization time, electrotonic forces generated by transmural differences in the shape of the action potential can inscribe an upright T wave in the electrocardiogram (ECG). Theoretical studies have also been helpful in our understanding of the role of electrical coupling in the expression of TDR.[8]

TDR is in large part attributable to the presence of M cells between the endocardial and epicardial layers of the heart. The M cell, discovered in the early 1990’s, and named in memory of Gordon K. Moe,[9, 10] has as its hallmark the ability to prolong its action potential more than that of normal epicardium or endocardium in response to a slowing of rate or exposure to agents that prolong APD.[9, 11–13]

Histologically, M cells are similar to epicardial and endocardial cells. Electrophysiologically and pharmacologically, they appear to be a hybrid between Purkinje and ventricular cells.[14] Like Purkinje fibers, M cells show a prominent APD prolongation and develop early afterdepolarizations (EAD) in response to rapidly activating delayed rectifier potassium current (I_{Kr}) blockers, whereas epicardium and endocardium generally are less likely to do so. Like Purkinje fibers, M cells develop delayed afterdepolarizations (DAD) more readily in response to agents that calcium load or overload the cardiac cell. Thus, they may contribute to the development of both substrate and triggers associated with arrhythmogenesis.

In the dog, the ionic basis for these features of the M cell include the presence of a smaller slowly activating delayed rectifier current (I_{Ks}),[15] a larger late sodium current (late I_{Na})[16] and a larger Na-Ca exchange current ($I_{Na\text{-}Ca}$).[17]

Distribution of M cells within the ventricular wall has been investigated in greatest detail in the left ventricle of the canine heart. Although transitional cells are found throughout the wall in the canine left ventricle, M cells displaying the longest action potentials (at BCLs ≥ 2000 msec) are often localized in the deep subendocardium to midmyocardium in the anterior wall [4] and throughout the wall in the region of the right ventricular outflow tract (RVOT).[10] M cells are also present in the deep cell layers of endocardial structures, including papillary muscles, local APD gradient between the islands of delayed repolarization (M cells) and the neighboring myocardium.

trabeculae and the inter-ventricular septum [18, 19]. Sicouri et al.,[19] reported a greater TDR across the canine inter-ventricular septum than across the left ventricular (LV) free wall, a finding that is at odds with that of Morita and co-workers.[20] Cells with characteristics of M cells have been described in a wide variety of animal species including canine, guinea pig, rabbit, and pig ventricles.[1, 4, 9, 12, 15, 18, 21–38]

There is a paucity of data dealing with the presence of M cells in the human heart. Drouin and co-workers were the first to report the presence of M cells in humans based on microelectrode recordings from tissue slices isolated from normal human hearts.[39] Li and co-workers[28] studied the characteristics of cells enzymatically dissociated from normal and failing human hearts and thus identified the presence of cells with the characteristics of M cells. Taggart and co-workers[40] attempted to map transmural distinctions of repolarization in the human heart *in vivo* using unipolar plunge electrodes, but failed to detect the presence of M cells or any type of TDR.

The extent to which transmural dispersion of repolarization (TDR) exists within the normal heart of animals *in vivo* has been a matter of considerable debate.[10, 13, 26, 41–44] The controversy derives in large part from the fact that quantitation of TDR in animals *in vivo* is hampered by 1) the inability to record local repolarization accurately, 2) the unavoidable use of anesthesia, which reduces TDR and 3) less than optimal recording conditions. These same constraints apply to the measurement of TDR in the human heart [40, 45]. Indirect evidence for the presence of a prominent TDR in the human heart in the absence of general anesthesia was provided in a recent study.[46]

Detailed maps of repolarization characteristics in the human heart similar to those available for the dog heart were not available until recently. In this issue of *Circulation Research*, Glukhov et al. [47] present the results of optical recording of action potentials from coronary-perfused left ventricular wedge preparations isolated from normal and failing human hearts.

In nominally normal hearts, they recorded an average TDR of 104±17 ms, which increased in some regions to values as high as 147 ms. M cells were found to cluster in islands located in the deep subendocardium with action potential duration (APD) averaging 527±43ms. Outside of the islands, subepicardial, midmyocardial, and subendocardial APD measured at 80% repolarization (APD_{80}) values were 380±17ms, 450±26ms, and 479±23ms. Although these TDR values are much greater than those reported in canine LV wedge preparations, the clustering of M cells in islands was observed in the canine heart by Akar et al. using optical recording techniques.[48] Interestingly, in failing hearts, TDR was reduced to 47±6 ms and the authors report the absence of M cells. In failing hearts APD_{80} increased to 492±38 ms, 512±29 ms and 470±28ms in subendocardial, midmyocardial and subepicardial regions, respectively, approximating the APD values of the M cells in the normal heart, raising the interesting possibility that the ion channel remodeling associated with heart failure (reduced outward and augmented inward ion channel currents)[49,50] transforms epicardial and endocardial cells into those with properties similar to those of M cells. The hallmark of the M cell is its ability to prolong more than the other normal ventricular cell types in response to a slowing of heart rate and/or in response to APD prolonging drugs.[11] This has been the traditional definition of M cell for the past 20 years. Glukhov et al [47] propose a new definition, which is based on the Based on their definition there is no evidence of M cells in failing hearts. Based on the traditional definition of the M cell, which distinguishes a M cell based on the ability of its APD to prolong prominently at slower rates, all of the cells in all three layers of the failing hearts are M cells.

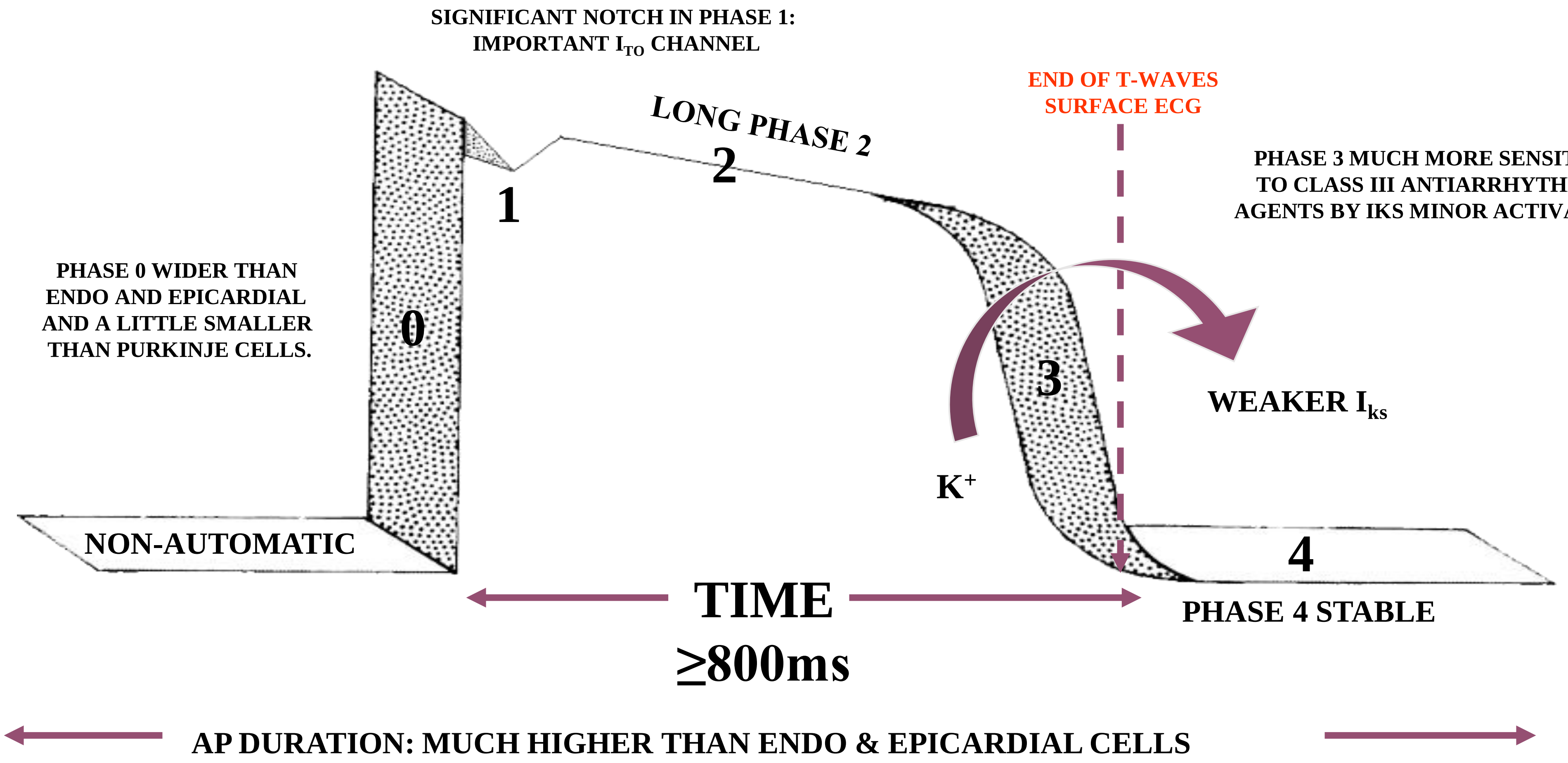
The authors are to be applauded for making the effort to record a transmural ECG. This is the most important parameter to monitor the quality of the preparation, which is often circumvented by other groups claiming "high quality" wedge preparations. The normal convention for recording a transmural pseudo-ECG is to place the + electrode of the ECG leads facing the epicardial side of the preparation and the – terminal facing the endocardial surface, following the convention of precordial leads on the body surface. This appears to be reversed in their experiments, resulting an inverted pseudo-ECG. Thus the apparent ST segment elevation shown in Figure 1 of their paper is actually a depression and the negative T wave is actually a concordant positive T wave, which is expected based on the repolarization sequence. The ST segment depression suggests the presence of mild ischemia on the endocardial side of the preparation. It is difficult to determine the extent to which the ischemia/hypoxia may have been heterogeneous and it is possible that this non-homogeneity may be responsible for creating islands of abbreviated responses, which could give rise to the appearance of islands of M cells.

Congruent with the studies of Poelzing et al.[5] in the dog and Yamada et al. in the rat [6] showing reduced Cx43 in the epicardium, Glukhov et al., using immunostaining techniques, showed reduced expression of Cx43 in epicardium in normal, but particularly in failing, human hearts.

Also of interest are the results of Glukhov et al. showing reduced TDR in human failing hearts. This result is opposite to the that obtained in animal models, in which TDR has been shown to be markedly augmented with the development of congestive heart failure secondary to tachypacing.[51]

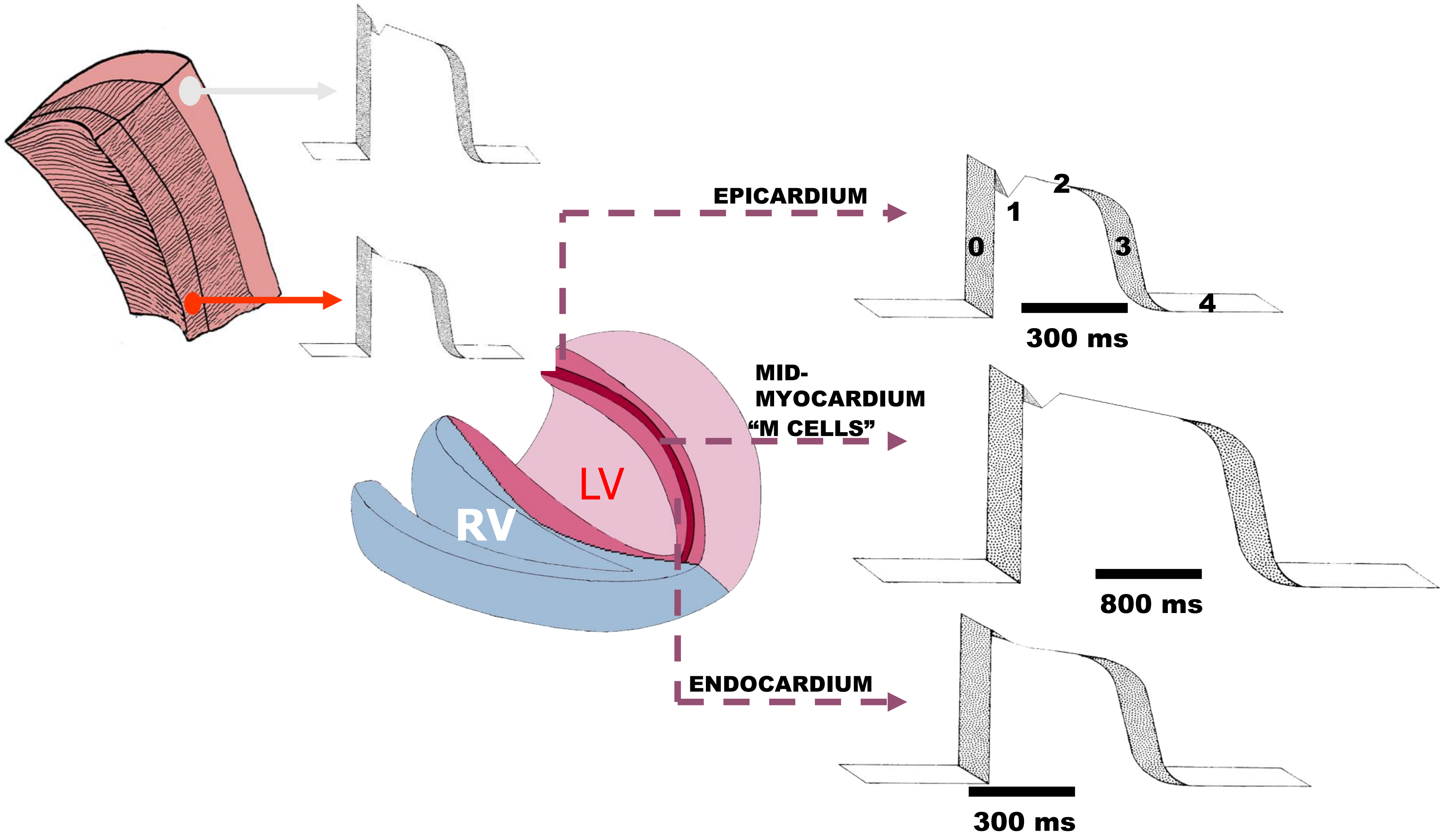
As the authors point out, these conclusions are based on limited data derived from relatively few (5) non-failing hearts that were not entirely normal and few (5) failing hearts of various etiologies. Accordingly, additional studies are needed to confirm and expand these findings, but the authors are to be congratulated on this significant advance in our understanding of human electrophysiology.

CHARACTERISTICS OF AP OF "M" CELLS OF VENTRICULAR MIDMYOCARDIUM



Characteristics of action potential of M cells. It is a fast and non-automatic fiber; therefore, we could say that it is a mixture between Purkinje cells and contractile ventricular myocardium. It is very sensitive to bradycardia and class III antiarrhythmic drugs, such as amiodarone and sotalol.

AP OF VENTRICULAR CONTRACTILE CELLS IN WALL THICKNESS: EPI, MESO & ENDOCARDIUM: HETEROGENEITY IN VENTRICULAR WALL THICKNESS



The thickness of the ventricular wall is formed by three functional layers, with different action potential. In the depth of the middle layer we find M cells, which have a subpopulation of cells with a great conduction velocity and electrophysiological properties of their own, which are very relevant in the pathophysiology of long and short QT syndromes.

ELECTROPHYSIOLOGICAL CHARACTERISTICS OF VENTRICULAR MYOCARDIAL CELLS

	EPICARDIUM.	**MID-MYOCARDIUM: TRANSITIONAL CELLS.**	**ENDOCARDIUM.**
AP DURATION	300ms.	800ms.	300ms.
PHASE 1: NOTCH	I_{to} channel very abundant, responsible for prominent notch in phase 1.	Abundant I_{to} channel	I_{to} channel absent. There is no phase 1 with notch.

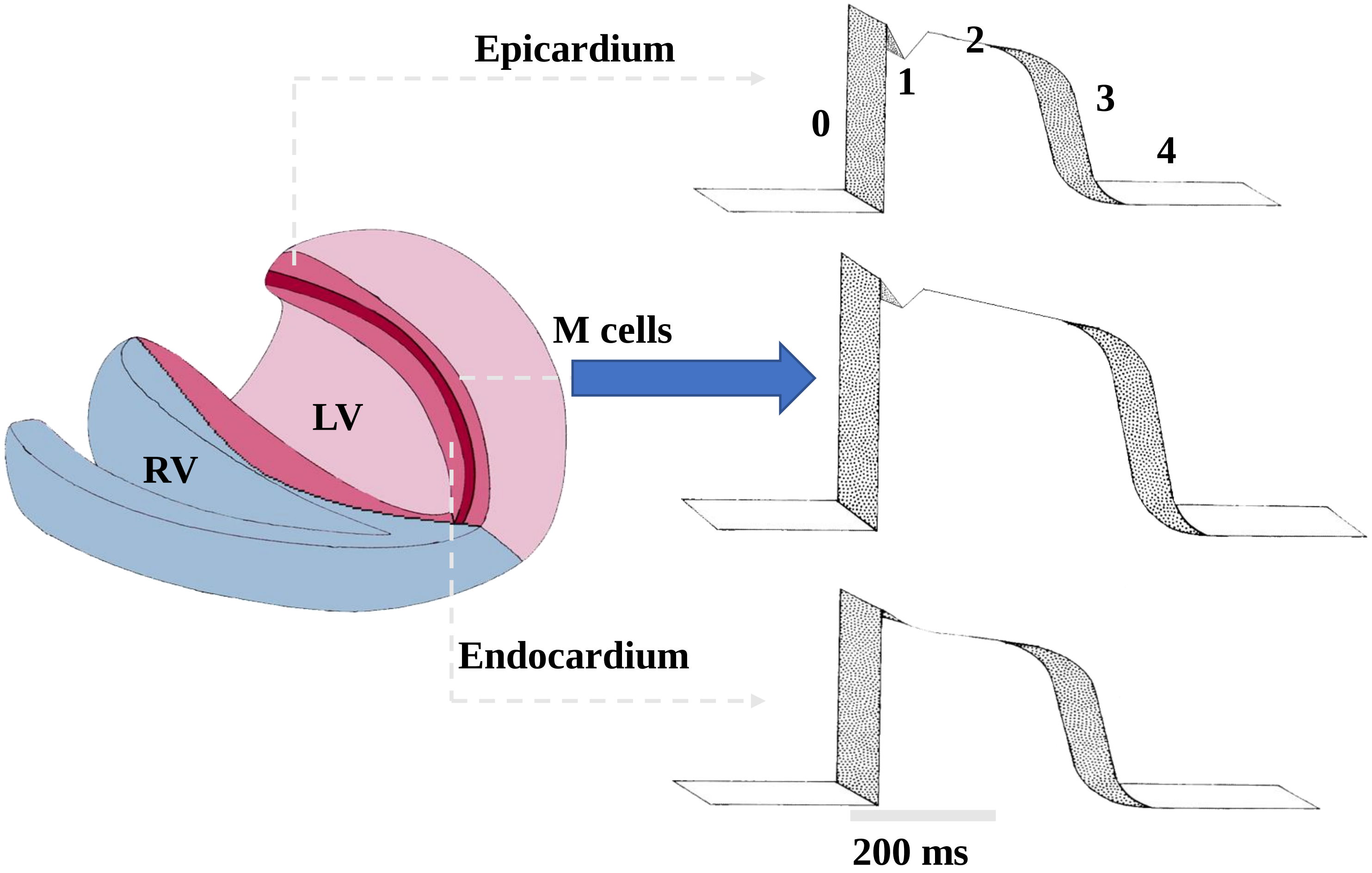
Electrophysiological characteristics of the three cellular types in ventricular wall thickness.

THE SOURCE OF U WAVE - THEORIES

1) Repolarization of Purkinje fibers.
2) Delayed repolarization of papillary muscles.
3) Residual late potentials of the septum.
4) Electro-mechanic coupling.
5) Theory of origin in “M” cells.
6) Post-potentials of triggered activity.

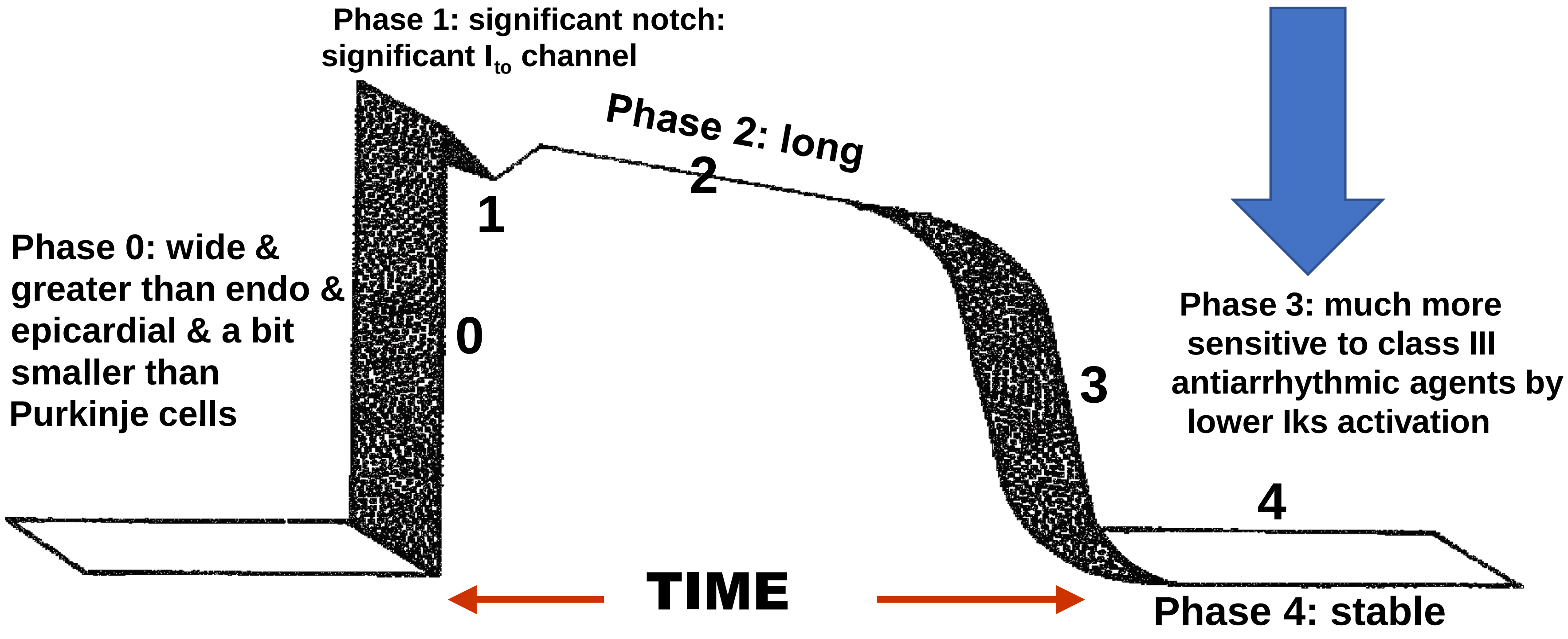
Theories existing on U wave origin.

THEORY OF ORIGIN IN “M” CELLS



It shows the theory of U wave origin in M cells and it explains the electrophysiological characteristics of these cells.

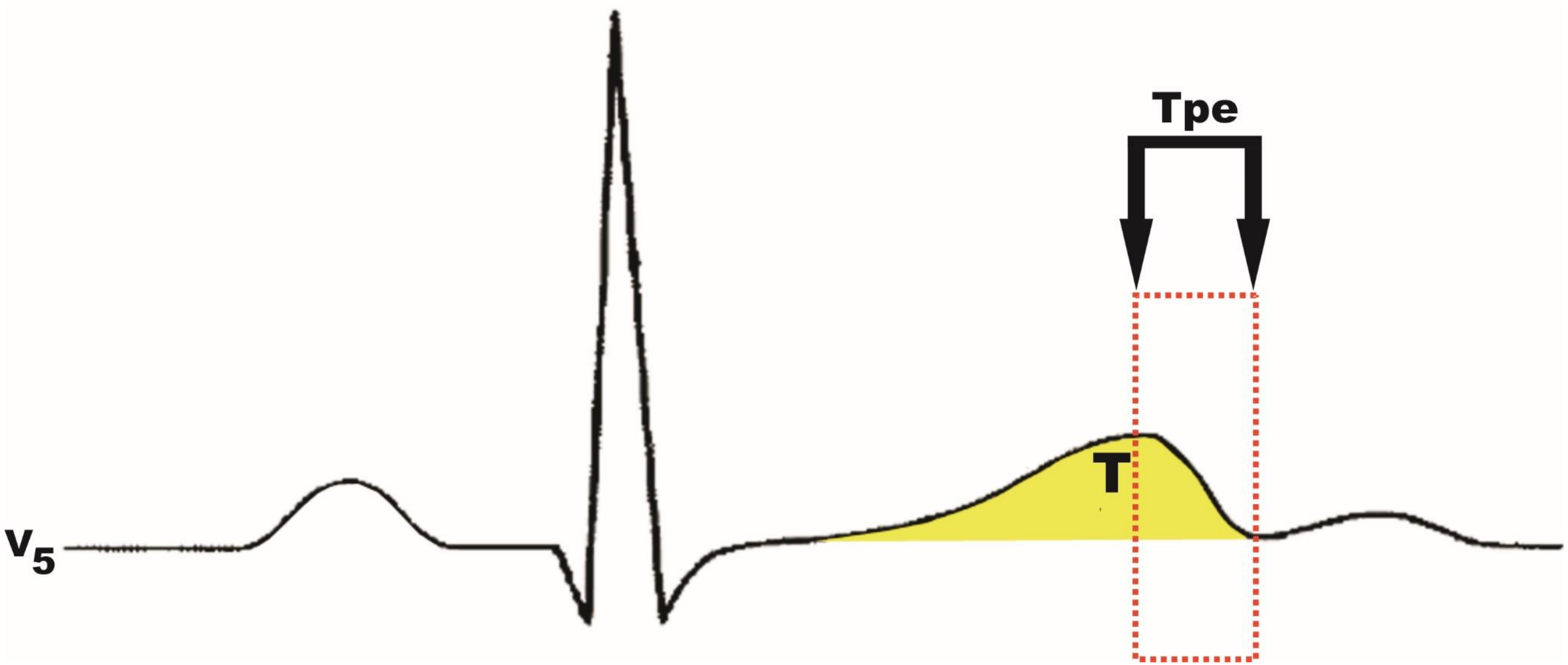
ACTION POTENTIAL OF “M” CELLS



It shows the theory of U wave origin in M cells and it explains the electrophysiological characteristics of these cells.

$T_{\mathrm{peak}} - T_{\mathrm{end}}$ prolongation and $T_{\mathrm{peak}} - T_{\mathrm{end}}$ dispersion is explained in channelopaties by M cells presence

The normal value of Tpeak/Tend interval (Tpe) is 94 ms in men and 92 in women when measured in the V_5 lead. Tpe prolongation to values ≥120 ms is associated to a greater number of events in patients carriers of BrS



Interval elapsed from the apex to the end of T wave (Tpeak-Tend interval or Tpe). Tpe may correspond to transmural dispersion of repolarization and consequently, the amplification of this interval is associated to malignant ventricular arrhythmias.

U wave origin in “M” cells

The authors from the Masonic Medical Research Laboratory of Utica, NY, suggest that “M” cells, more abundant in mass and having a prolonged repolarization time comparable to Purkinje cells, may be responsible for the pathophysiologic recording of the U wave in the presence of long QT interval, acquired or congenital. Thus, bimodal T waves with hump-like morphology represent different levels of interruption of the descending slope of the T wave, called T2 instead of U wave. Besides the three basic types of cells in the ventricular myocardium: epicardial, mesocardial and endocardial, there is a cellular subpopulation called “M cells”, located in the midmyocardium with very differentiated electrophysiological and pharmacological features. Studies have established the presence of 3 distinct cell types in the ventricular myocardium: epicardial, M and endocardial cells. Epicardial and M cell APs differ from endocardial cells with respect to the phase 1shape. These cells possess a prominent Ito-mediated notch responsible for the 'spike and dome' morphology of the epicardial and M cell response. M cells are distinguished from the other cell types in that they display a smaller slowly activating delayed rectifier current (I(Ks)), but a larger late sodium current (late I(Na)) and sodium-calcium exchange current (I(Na-Ca)). These ionic distinctions underlie the longer APD and steeper APD-rate relationship of the M cell, which is more pronounced in the presence of antiarrhythmic agents with class III actions. The preferential prolongation of the M cell action potential results in the development of a TDR, which can be estimated from the ECG as the interval between the peak and the QTpeak-QTend interval.

Using the canine arterially perfused ventricular wedge model, TAPs of the various cardiac cell types can be correlated to the waveforms of the ECG, providing insight into the cellular etiology of ECG abnormalities. Two congenital syndromes of sudden cardiac death that have been modeled using this technique are the long QT and Brugada syndromes.

The long QT syndrome has been linked to 5 gene mutations on chromosomes 3, 7, 11, and 21. Mutations in the cardiac sodium channel SCN5A have been linked to families with a history of the Brugada syndrome. Although the etiologies of these two syndromes are different, lethal arrhythmias in both are thought to arise due to amplification of intrinsic electrical heterogeneities. Similar mechanisms are likely responsible for life-threatening arrhythmias in a variety of other cardiomyopathies ranging from heart failure and hypertrophy, which involve mechanisms similar to those operative in LQTS, to ischemia and infarction, which may involve mechanisms more closely resembling those responsible for the Brugada syndrome.

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