Conclusions and future developments

The main findings of this work are: 1) heterocellular electrotonic interactions between myofibroblasts and cardiomyocytes precipitate spontaneous ectopic activity in the heart; 2) dynamic electrical restitution properties of the rat ventricle and their pharmacological modulation can be measured at the cellular level.

In the first part of the work, in particular, the main finding is that, for monolayer cultures of cardiomyocytes, spontaneous activity is not due to a culture-dependent dedifferentiation of cardiomyocyte toward a spontaneously active immature phenotype, but is the specific result of electrotonic interactions with a sufficient number of myofibroblasts. In regard to intact cardiac tissue, the findings of the first part of the thesis open the perspective that contact regions between cardiomyocytes and sufficiently large numbers of myofibroblasts as occurring in the borderzone of healing infarcts (Sun et al., 2002) a few days after the acute event or in the fibrotic working myocardium (Clement et al., 1999) might give rise to arrhythmogenic ectopic activity

In the second part of the work the main findings are 1) intrinsic beat-to-beat dispersion of repolarization increases with intrinsic APD and, when normalized to it, is inversely proportional to pacing frequency, increased by nifedipine and decreased by 4-AP, 2) both random and linear changes in pacing CL tend to correlate positively with APD changes, and 3) nifedipine abolishes this correlation, uncoupling APD from the preceding CL, whereas 4-AP increases correlation, coupling the time course of the two parameters over consecutive beats. A direct effect of these blockers on intrinsic beat-to-beat variability of APD has not been shown previously and is of interest, given that one of the beneficial effects of anti-arrhythmic drugs is thought to be their ability to decrease temporal dispersion of repolarization in the heart (Kuo *et al.* 1983; Fynn *et al.* 2003). In this work we did not consider the effect on repolarization dispersion of homo and hetero-cellular coupling. It is well known the importance of intercellular coupling in the synchronization of repolarization between neighboring myocytes and hence in the dispersion of repolarazation. Both local membrane currents and electrotonic current flow between neighboring cells govern transmembrane potential. Computer modeling studies have shown that a progressive reduction of cell-to-cell coupling can reduce electrotonic current flow and reveal

intrinsic transmural heterogeneity of membrane repolarization currents (Viswanathan et al., 1999). It was also found, using experimental studies combined with computer simulation (Laurita et al., 1997), that electrotonic current flow between neighboring cells can attenuate the expected APD shortening associated with restitution at short diastolic intervals and hence can reduce dispersion of repolarization.

In the present study, it is demonstrated that the appearance of interstitial myofibroblasts contribute to establish higher propensity to arrhythmis inducing depolarization of cardiomyocytes, but the effects of heterocellular electrotonic interactions between myofibroblasts and cardiomyocytes on repolarization and electrical restitution properties of the cardiac tissue (properties characterized in the study showed in the second chapter) are still to be measured. In this context, a possible future development of this research could be, not only the study of the influence of homo-cellular coupling, but also of hetero-cellular coupling on APD rate-dependency and restitution properties of ventricular cardiomyocytes.

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