ABSTRACT

The research field of my PhD concerns mathematical modeling and numerical simulation, applied to the cardiac electrophysiology analysis at a single cell level. This is possible thanks to the development of mathematical descriptions of single cellular components, ionic channels, pumps, exchangers and subcellular compartments. Due to the difficulties of vivo experiments on human cells, most of the measurements are acquired in vitro using animal models (e.g. guinea pig, dog, rabbit). Moreover, to study the cardiac action potential (AP) and all its features, it is necessary to acquire more specific knowledge about single ionic currents that contribute to the cardiac activity. Electrophysiological models of the heart have become very accurate in recent years giving rise to extremely complicated systems of differential equations. Although describing the behavior of cardiac cells quite well, the models are computationally demanding for numerical simulations and are very difficult to analyze from a mathematical (dynamical-systems) viewpoint.

In addition to the traditional methods for the cardiac electrophysiology, the thesis is focused on the development of an innovative approach to the modelling of cardiac action potentials. My dissertation uses the dynamic clamp technique (DC), as the central technique, applying to different mathematical modelling of cardiac AP with the aim of gaining a clearer insight into the origin of the dynamics of electrophysiological models. In order to implement the dynamic clamp we have used the open source Real-Time eXperiment Interface (RTXI), which is a real-time biological experimentation system based on Real-Time Linux. To increase my knowledge on the RTXI, I have spent five months at the Weill Cornell Medical College of New York, in David Christini’s laboratory, which, together with his team, has developed the RTXI software and used it in several biologic applications. The implementation of the system features and the custom user code, as modules, written in C++ for running RTXI was one of my work aspect.

The second chapter of the thesis has concerned a collaboration with the Instituto de Investigaciones Biomédicas Alberto Sols, CSIC-UAM, of Madrid where we have studied a novel mutation to its short QT phenotype through multiscale computational modelling. My contribution has been the simulation of a novel mutation (responsible for short QT syndrome) using two human AP models, the ten Tusscher -Panfilov and the O’Hara Rudy. The simulation results predicted a similar shortening of the AP than observed in the patient, and were useful to strengthen the experimental results.

All the other works used DC in different applications. The first application was aimed to: 1) optimize the Luo-Rudy mathematical formulation of the guinea-pig rapid delayed rectifier K+ current (IKr) and 2) validate the new model, analyzing the effects of the current block (using the
specific blocker E4031) and of the modelled current injection on the AP duration. We were able to provide an optimized $I_{Kr}$ model able to reproduce AP- and Dynamic- clamp experimental data. A positive message conveyed by the results is that, once a reliable numerical $I_{Kr}$ model is developed, it can be expected to accurately reproduce the native $I_{Kr}$ behaviour in the context of myocyte complexity. This is particularly relevant in consideration of the peculiar role of $I_{Kr}$ in repolarization and of the high cost of biological tests currently used to assess the drug arrhythmogenic potential due to $I_{Kr}$ alterations. The optimized model has been integrated into action potential models to explore the effects of changes in individual kinetic parameters on cardiac repolarization.

The second study investigated the $I_{Kr}$ impact on repolarization and its variability using the optimized model (obtained as described in chapter 3). Identification of the relative impact of $I_{Kr}$ gating properties may provide a new framework for predicting the consequences of $I_{Kr}$ mutations on APD and its variability, two quantities strongly associated with arrhythmogenic risk. If validated by correlation with clinical observations, such a framework would be of obvious practical use.

In line with the work presented by Ahrens-Nicklas and Christini (2009) where they tried to anthropomorphize the mouse cardiac AP, we have used DC to compare different species. In particular, the third study regarded the comparison between different species (dog and guinea-pig) for what concerns the transient outward K$^+$ current ($I_{to}$) presence. We investigated whether a spike – and – dome profile (SaD) is the main factor determining the direction of APD response to β – adrenergic stimulation. The possibility to inject in a guinea pig a “synthetic” $I_{to}$ current (which is absent) has allowed to examine 1) the ISO effects on APD (in guinea-pig and canine myocyte); 2) the presence of AP spike – and – dome profile.

The last DC application regards the “funny” current $I_f$ present in the sinoatrial node pacemaker cells. This work has shown that the $I_f$ formulation by Severi et al. 2012 allows to restore the basal condition when used to calculate the DC-based re-injection of $I_f$ blocked by Ivabradine whereas the Maltsev-Lakatta 2009 model can only minimally compensate for the $I_f$ block. In addition the study of the physiological mechanisms of rate modulation has revealed that the known isoprenaline-induced $I_f$ activation shift accounts for most of the ISO-induced pacemaking rate increase.

In conclusion, our results can assert that dynamic clamp is a promising approach as it allows to test how the properties of a numerical model affect the AP generated by a real myocyte and they show how mathematical approach is suitable to provide new insights into the cardiac AP mechanisms in physiological but also arrhythmogenic mechanisms in a pathological condition, especially when fully integrated with experimental data.