Increased Delayed Afterdepolarisation Susceptibility in Hypertrophic Cardiomyopathy Cardiomyocytes Under Exercise Conditions

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Abstract

Electrophysiological mechanisms underlying exercise as a potential arrhythmic trigger in hypertrophic cardiomyopathy (HCM) remain incompletely understood, while clinical exercise guidelines continue to evolve. This study aimed to analyse delayed afterdepolarisation (DAD) formation in cells affected by HCM ionic remodelling.

Control and HCM action potential model populations were constructed by varying ionic conductance multipliers and calibrating models to experimentally reported ranges. Both populations underwent a DAD induction protocol consisting of fast pacing and beta-adrenergic stimulation, followed by a 10 second quiescent period.

HCM models showed more than double the DAD incidence of controls and longer trains of DADs, with longer DAD trains being associated with upregulated I_{Kr} conductance in the HCM population. Targeting the downregulated Na^+/K^+ pump in HCM notably reduced DAD incidence and the length of DAD trains.

In conclusion, this study demonstrates a higher risk of DADs in human HCM cardiomyocytes under exercise conditions, compared to non-diseased cardiomyocytes.

1. Introduction

Clinical exercise guidance in hypertrophic cardiomyopathy (HCM) is continuously evolving, with recent studies challenging traditional exercise restrictions [1]. As a potential trigger of arrhythmias during exercise, *in vitro* experiments have shown that beta-adrenergic receptor stimulation (β-ARS) can increase delayed afterdepolarisation (DAD) incidence in HCM cardiomyocytes [2]. A sufficiently large DAD can produce a premature action potential (AP), which can initiate re-entries and clinically significant arrhythmias.

In this study, we aim to evaluate if the risk of DADs after exercise is greater in human HCM ventricular cardiomyocytes, and to investigate the ionic mechanisms that drive DAD formation. We used computational modelling and simulation to analyse DAD inducibility in human HCM-remodelled cardiomyocytes under exercise conditions, to elucidate the disease-specific ionic determinants of DAD risk.

2. Methods

2.1. Control and HCM Populations

Cellular electrophysiology was simulated using the endocardial T-world ventricular AP model, which is the first to reproduce all key cellular arrhythmic mechanisms including DADs [3].

A population of 1,500 candidate control models was first constructed following [4,5] with variability in conductances of 10 main ionic currents and fluxes (I_{CaL}, I_{Kr}, I_{Ks}, I_{NaCa}, I_{NaK}, I_{Cab}, I_{pCa}, I_{ClCa}, J_{Rel}, J_{Up}), sampled within ±50% of their baseline values using Latin Hypercube Sampling.

The experimental Ca^{2+} biomarker ranges at 0.2 Hz and 1 Hz pacing documented by [6] and the experimental AP biomarker ranges at 1 Hz detailed in [7,8] were used to calibrate this initial control population. Models showing alternans at 1 Hz pacing were also filtered out of the control population.

Models were paced for 250 beats to reach steady state before performing calibration.

A corresponding HCM population was created by applying ionic remodelling to each model, based on the experimental data in [4,6] and the HCM phenotype modifications to the ToR-Ord model and β-ARS cascade detailed in [9].

2.2. Selective Reversal of HCM Ionic Remodelling

To analyse the effects of individual components of HCM ionic remodelling, further disease AP models were generated by selectively reversing specific aspects of the remodelling.

In disease model M1, I_{NaK} maximal conductance was reset. In M2, I_{CaL} maximal conductance was reset. In M3, the I_{CaL} slow and fast time constants were reset. In M4, the affinity of troponin for Ca^{2+} was reset. In M5, I_{CaL} maximal conductance and the affinity of troponin for Ca^{2+} were reset. In M6, J_{rel} maximal flux was reset.

2.3. DAD Pacing Protocol

To evaluate DAD inducibility under an exercise-like protocol, the models were paced for 200 beats at 2 Hz pacing, followed by a prolonged quiescent period of 10 s, all under full β-ARS.

Like the detection of afterdepolarisations in [7], we used a positive gradient greater than 0.02 mV/ms at a membrane potential less than or equal to -65 mV to detect DADs. The number of DADs detected was calculated as one less than the number of such depolarisations. The detection of DADs was performed on the membrane potential trace during the 10 s quiescent period, following the final paced beat. We used a DAD exceeding -60 mV in membrane potential to detect triggered APs [10].

3. Results

Calibration of the control population to human physiological ranges of AP and Ca^{2+} transient biomarkers yielded 229 accepted models forming the population of healthy cardiomyocytes. The HCM population comprised 229 paired models with ionic remodelling.

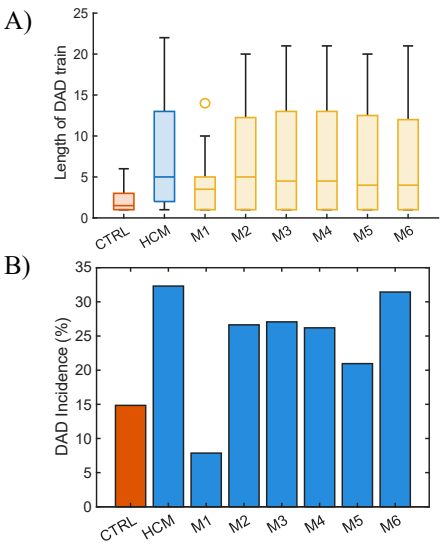

Figure 1. DAD inducibility quantified as (A) the percentage of models that developed DADs, and (B) the number of DADs during the 10 s quiescent period, across the control (CTRL), HCM and M1-6 populations.

Figure 1A shows that the incidence of DADs in the HCM population (32.3%) was more than double that of the control population (14.8%) after 2 Hz pacing under β-ARS. We also observed that, for all the control and HCM models that developed DADs, full APs were triggered. Moreover, the HCM models that developed DADs had significantly longer trains of DADs than the control models that developed DADs (Figure 1B).

The M1-6 populations with partial ionic remodelling reversal had lower DAD incidence than the original HCM population (Figure 1A). Most notably, only 7.9% of the M1 models (with I_{NaK} remodelling reversal) developed DADs, significantly lower than the 32.3% incidence with full ionic remodelling. Similarly, the number of DADs in a train was lower for the M1 models that developed DADs (median: 3.5, upper quartile: 5) than for the HCM models that developed DADs (median: 5, upper quartile: 13) (Figure 1B and 2A).

During the 10 s quiescent period, HCM and M1 models that did not develop DADs had a higher propensity to early afterdepolarisation (EAD) formation than the control models that did not develop DADs (Figure 2B). We also observed that, for both HCM and M1, none of the models that developed DADs (74 and 18 models developed DADs, respectively) had exhibited EADs during the quiescent period. Similarly, only 1 control model out of 34 that developed DADs exhibited EADs.

Ionic conductance scaling factors were compared between models, stratified by DAD inducibility (Figure 3). Figure 3A shows that, in HCM models with DADs, I_{Kr} conductance was upregulated, with I_{NCX}, I_{NaK}, I_{Cab} and J_{rel} downregulated. The HCM models that developed larger numbers of DADs in a train had higher I_{Kr} conductance (Figure 3B). In both control and HCM populations, action potential durations at baseline (1 Hz pacing, no β-ARS) were smaller in models with DADs during the DAD pacing protocol than those without DADs (Figure 3C).

4. Discussion and Conclusion

This study aimed to assess if human HCM ventricular cardiomyocytes have greater risk of DADs after exercise compared to non-diseased cardiomyocytes and to gain insights into the ionic mechanisms underlying DAD formation. The key findings are that: (i) the exercise DAD incidence in HCM was more than twice that of controls; (ii) reversal of the I_{NaK} remodelling in HCM significantly reduced DAD incidence; and (iii) almost all HCM models that developed DADs did not develop EADs.

This study found that the HCM population had notably greater DAD incidence than the control population and greater intensity of DAD trains. These findings complement the in-vitro studies in [6] showing the higher prevalence of DADs in HCM cardiomyocytes compared to controls. Future in-silico studies focusing on the propagation of exercise-induced DAD trains through human cardiac ventricular tissue would be a valuable next step to understand if the higher incidence and intensity of DADs in HCM ventricular cardiomyocytes causes a higher incidence of localised arrhythmias in HCM patients.

Targeting the downregulation of I_{NaK} in the HCM phenotype reduced the DAD incidence to almost half that of the control population. The targeting of I_{NaK} downregulation also reduced the severity of the DAD

trains. While other investigations into DAD occurrence under β-ARS in HCM have focused on the importance of I_{NaL} remodelling in the HCM phenotype [2], our findings highlight the contribution of downregulated I_{NaK} within the HCM phenotype to DAD formation. The I_{NaK} downregulation used in the HCM modelling relates to impaired energetics in HCM [4], which suggests that targeting impaired energetics may be a way to decrease arrhythmic risk in HCM, deserving further analysis.

While [11] shows that DADs and EADs occur together under Ca^{2+} overload in cardiomyocytes of heart failure patients, our findings show the rareness of HCM models that form DADs and EADs. This may suggest another impact of the HCM on the mechanisms underlying EADs and DADs.

As limitations, the present study did not also test the mid-myocardial and epicardial cell types. However, this is in part mitigated by the wide range of parameter variability used in the population of models approach.

In conclusion, the present study used a population of models approach to evaluate DAD inducibility in control and HCM-remodelled cells under exercise conditions. The incidence and length of DAD trains was elevated in HCM, with greater I_{Kr} conductance associated with longer DAD trains. Finally, we identified that the downregulated Na^+/K^+ pump can be targeted in HCM to decrease the intensity and propensity of DAD formation after exercise.

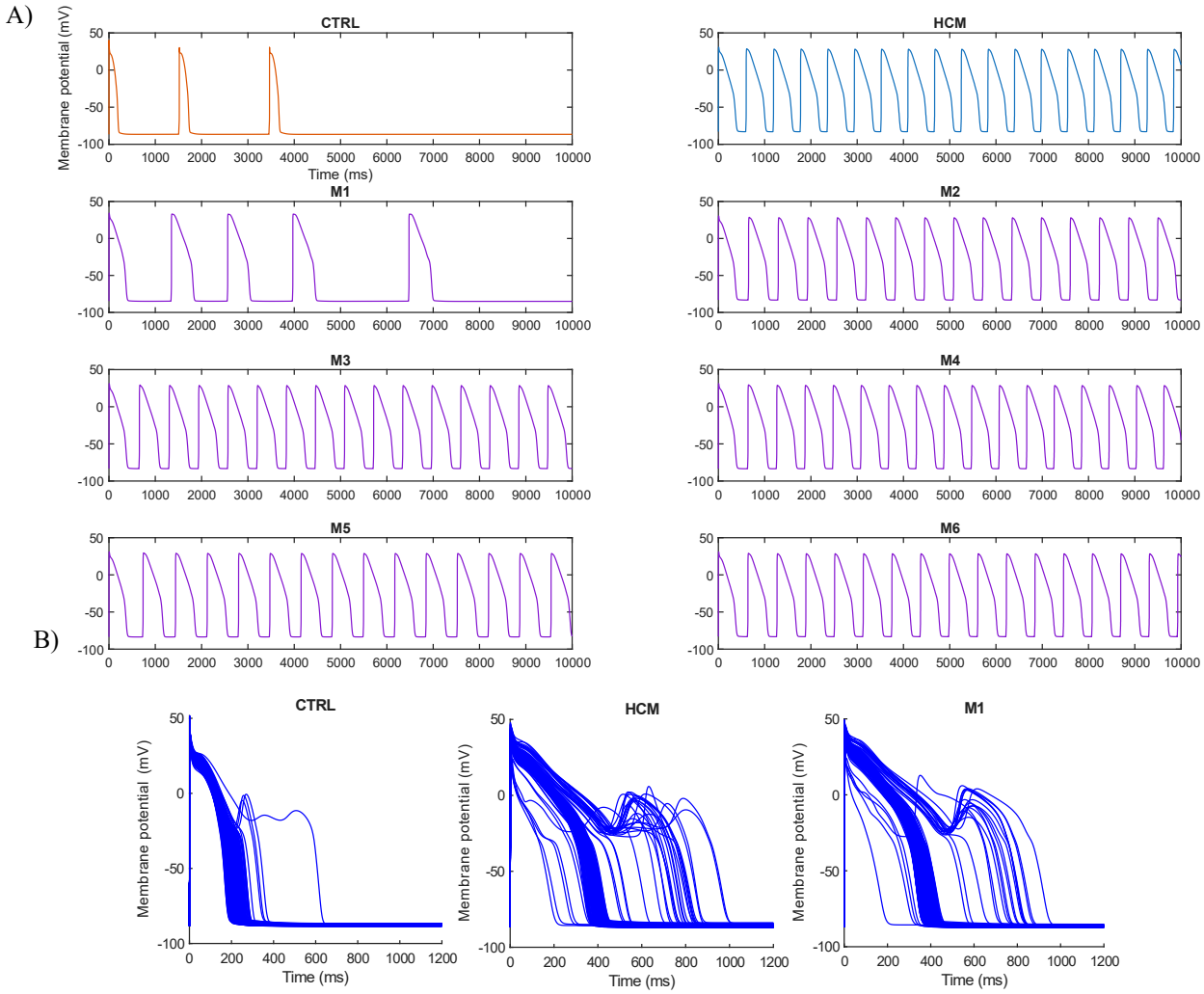

Figure 2. (A) Representative membrane potential traces during the 10 s quiescent period from a control model that developed DADs, alongside its corresponding HCM and M1-6 model traces. (B) AP traces of the 10 s quiescent period from control, HCM, and M1 models that do not develop DADs.

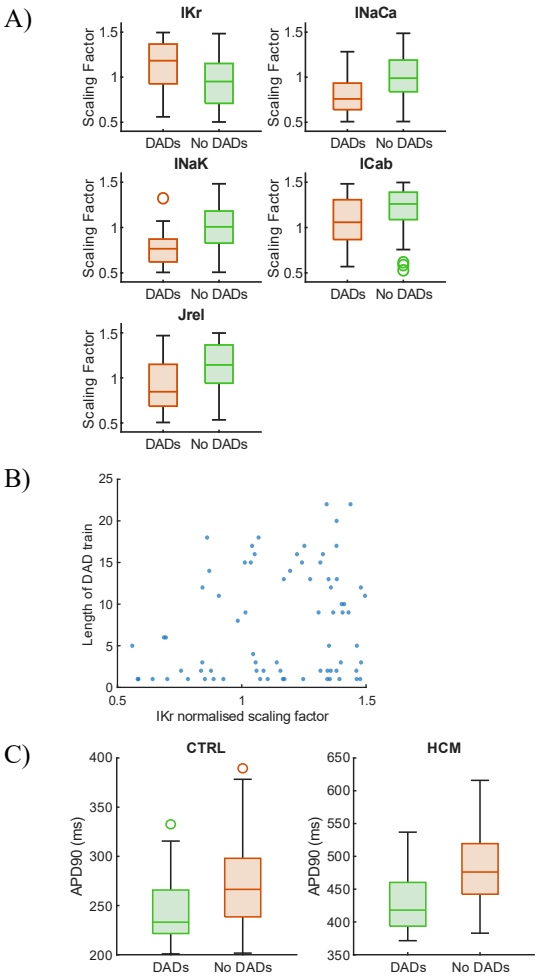

Figure 3. Ionic determinants of DAD inducibility. (A) Boxplots of ionic conductance scaling factors in HCM models, comparing models with vs. without DADs inducible. (B) Scatterplot of number of DADs during the quiescent period vs. I_{Kr} scaling factor for HCM models with DADs inducible. (C) Action potential duration at 90% repolarisation (APD90) at baseline (1 Hz pacing, no β-ARS) in control and HCM models, comparing models with vs. without DADs inducible.

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