Effect of Blocking I_{Kur} on the Genesis of Repolarisation Alternans in Canine Atrium

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Abstract

The Kv1.5 channel-mediated, ultra-rapid delayed rectifier K^+ current (I_{Kur}) is a promising target for anti-arrhythmic drugs due to its 'atrial-selective' property. However, whether blocking I_{Kur} induced action potential (AP) prolongation in normal atria can increase the alternans susceptibility has not yet been investigated.

This study aimed to investigate the effects of blocking I_{Kur} on the genesis of repolarisation alternans which is associated with arrhythmogenesis. The updated canine atrial cell model developed by Ramirez et al. was used to implement the dynamic pacing protocol and the standard S1-S2 protocol to obtain rate dependent curve and restitution curve to analyse the alternans.

Simulation results showed the prolonged AP and increased Ca^{2+} transient by blocking I_{Kur}. I_{Kur} block from 40% to 80% produced long-short-long-short AP and Ca^{2+} transient alternans at pacing rate of ~ 2.3 Hz to 5Hz. Further analysis demonstrated that I_{Kur} block promoted the genesis of AP and Ca^{2+} transient alternans, via a mechanism of increased AP duration that led to changes of L-type Ca^{2+} current dynamic, which was a major determinant coupling AP and Ca^{2+} transient. Our findings suggest that blocking I_{Kur} may promote the genesis of AP alternans, promote the genesis of AP alternans, indicating a latent pro-arrhythmic effect in normal atria.

1. Introduction

The Kv1.5 channel-mediated, ultra-rapid delayed rectifier K^+ current (I_{Kur}) contributes to the early and late repolarization of the action potential (AP), of which function is prominent in atria and negligible in ventricle [1]. Therefore, it is a promising target for class III anti-arrhythmic drugs [2,3], due to the 'atrial-selective' property of the channel. Blocking I_{Kur} is thought to be a treatment for atrial fibrillation (AF) via reducing repolarization reserve leading to prolonging action potential duration (APD) or effective refractory period (ERP) with reduced re-entrant tendency [4,5].

In the constant pacing rate, APD prolongation leads to shortening of diastolic interval (DI), resulting in increasing the gradient of APD restitution curves. Some studies have demonstrated that the steep slope of APD restitution is related to the development of APD alternans underlying the voltage-driven mechanism [6-8], even though short-term cardiac memory and calcium cycling dynamics limit predicting APD alternans by the APD restitution slope >1[9]. However, whether blocking I_{Kur} induced APD prolongation in normal atria can increase the alternans susceptibility has not yet been investigated. Therefore, this study aimed to investigate the effects of blocking I_{Kur} on the genesis of repolarization alternans which was associated with arrhythmogenesis.

2. Methods

In this study, the canine atrial cell model (the RNC model) developed by Ramirez *et al*. [10] was implemented to examine the effects of blocking I_{Kur} on genesis of the action potential (AP) alternans. For simulation of I_{Kur} block, the conductance of I_{Kur}, g_{Kur}, was multiplied by 0.9 to 0.2 responding to blocking I_{Kur} from 10% to 80%. The RNC model was modified, including reducing the conductance of I_{to}, I_{Kur}, I_{Kr}, I_{Ks} and I_{K1} by 10% and increasing the I_{NaK} by 30%, to maintain the APD relatively stable during the 100 beats and the APD of the 100^{th} AP was 184.4 ms at 1Hz in good agreement with experiments [11].

The action potential duration at 90% repolarization (APD_{90}) rate dependent curve and restitutions curve were calculated using the dynamic pacing protocol and the standard S1-S2 protocol respectively [12] to analyze the genesis of repolarization alternans under control and I_{Kur} block conditions. Firstly, the cell model was paced at 1000ms for 100 beats to reach relatively steady state as

initial values for next simulation under control and different I_{Kur} block conditions. Then, the two protocols were executed. In the dynamic pacing protocol, the cell model was pacing at a fixed cycle length (CL) for 100 beats to reach steady state from 1000 ms to which the APD alternans disappeared. The last two APs were recorded to observe whether there was an alternation with the longer AP and shorter AP. The APDs against different CLs constituted the APD rate dependent curve. In the standard S1-S2 protocol, the cell model was pacing at 1000 ms for 100 beats to reach steady state and then one S2 stimulus was applied after a certain diastolic interval (DI). The resulting APD of S2 stimulus was depending on DI, so that the APD against various DIs constituted the APD restitutions curve.

In addition, the changes in the intracellular Ca^{2+} transient (CaT) amplitude and decay time (calculated as the duration from the peak of the CaT to the time when the intracellular Ca^{2+} level had been reduced by a factor of e) were evaluated using the two protocols under control and I_{Kur} block conditions.

3. Results

3.1. Effect of blocking I_{Kur} on AP and CaT

Figure 1 shows the effect of blocking I_{Kur} on the AP morphology, intracellular calcium transient, APD_{90} and CaT amplitude. The canine atrial APD_{90} at a pacing rate of 1Hz was prolonged from 184.4 ms in the control condition to 200.4 ms, 205.6 ms, 211.7 ms and 222.4 ms in the 20%, 40%, 60% and 80% I_{Kur} block condition, and the plateau phase of AP was elevated due to blocking I_{Kur}. The CaT amplitude was increased from 450.6 nM to 503.7 nM, 545.5 nM, 579.9 nM and 644.8 nM.

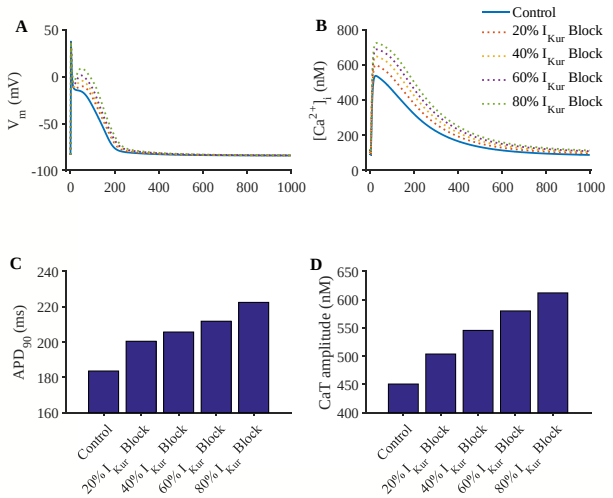

Figure 1. (A) AP morphology, (B) intracellular calcium transient, (C) APD_{90} and (D) CaT amplitude under control and I_{Kur} block conditions.

Table 1. The longest CL for the genesis of alternans in APD_{90}, CaT amplitude (CaT_{amp}) and CaT decay time (CaT_{DT}).

Bloking I_{Kur}	CL (APD_{90})	CL (CaT_{amp})	CL (CaT_{DT})
40%	440 ms	-	440 ms
50%	430 ms	425 ms	435 ms
60%	400 ms	395 ms	405 ms
70%	365 ms	370 ms	375 ms
80%	330 ms	340 ms	340 ms

3.2. APD and CaT alternans under I_{Kur} block conditions

Using the dynamic pacing protocol, the APD_{90}, CaT amplitude and CaT decay time rate dependent curves were presented in Figure 2 under control and I_{Kur} block conditions (only shown 20%, 40% and 60% I_{Kur} block). Genesis of alternans was considered when the beat-to-beat variation > 10 ms in APD_{90}, > 10 nM in CaT amplitude or > 10 ms in CaT decay time at the longest CL (details in Table 1). The longest CL for the genesis of alternans was 440 ms in the APD_{90} and in the CaT decay time when blocking I_{Kur} by 40%, and the one was 425 ms in the CaT amplitude when blocking I_{Kur} by 50%. There was no alternans in control condition and in blocking I_{Kur} by 10% to 30% conditions. APD_{90} and CaT decay time alternans occurred when blocking I_{Kur} by 40% to 80%, and CaT amplitude alternans occurred when blocking I_{Kur} by 50% to 80% (beat-to-beat variation < 10nM when blocking I_{Kur} by 40%).

Using the standard S1-S2 protocol, the APD_{90} restitutions curve were presented in Figure 3 under control and I_{Kur} block conditions (only shown 20%, 40% and 60% I_{Kur} block). The maximum slope in the control and the 20% I_{Kur} block conditions were 0.48 and 0.8 which were < 1 and corresponding to no alternans of rate dependent curves in Figure 2. The maximum slope in the 40% and 60% I_{Kur} block conditions were 1.46 and 2.54 which were > 1 and corresponding to alternans of rate dependent curves in Figure 2 and Table 1. When blocking I_{Kur} by 40%, the DI with maximum slope was 320 ms with APD_{90} of 154 ms so that the CL was 474 ms. When blocking I_{Kur} by 60%, the DI with maximum slope was 234 ms with APD_{90} of 167ms so that the CL was 401 ms, which was shorter than the one in 40% I_{Kur} block.

3.3. Mechanism of alternans under I_{Kur} block conditions

Further analysis for the mechanism of alternans under I_{Kur} block condition was demonstrated in Figure 4 which showed the alternans of I_{Kur}, AP, intracellular CaT, L-type

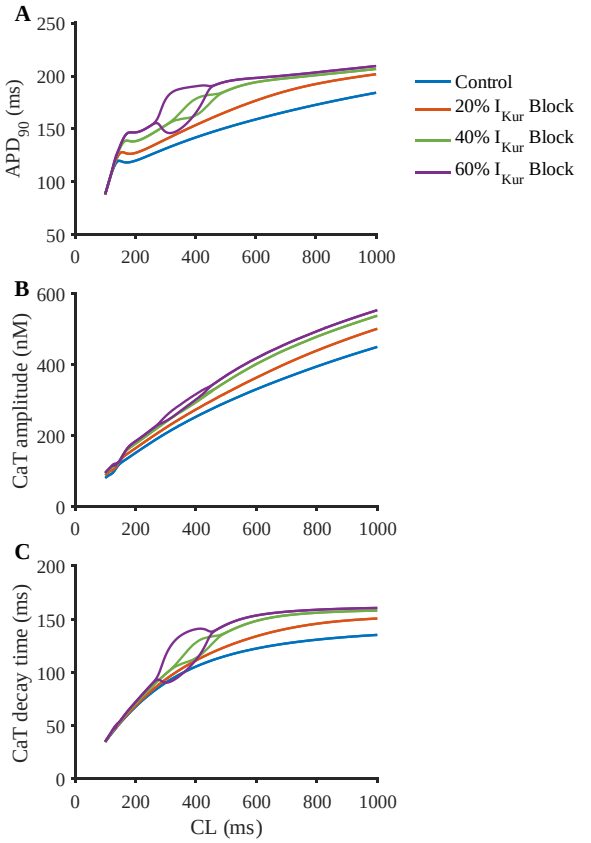

Figure 2. (A) APD_{90} and (B) CaT amplitude rate dependent curves under control and I_{Kur} block conditions.

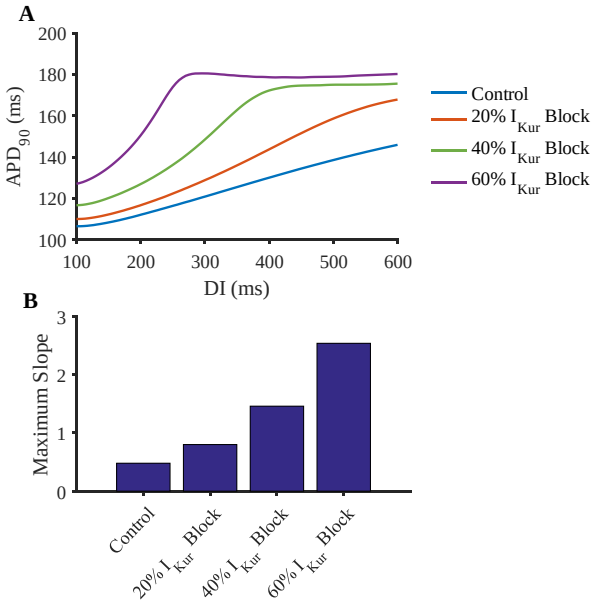

Figure 3. (A) APD_{90} restitutions curves under control and I_{Kur} block conditions. (B) The maximum slopes of the APD_{90} restitution curves in (A).

calcium current (I_{CaL}) and its voltage-dependent activation gate (d), voltage-dependent inactivation gate (f) and calcium-dependent inactivation gate (f_{Ca}), and Na-Ca exchange current (I_{NCX}) at pacing CL of 400 ms in control and 50% I_{Kur} block condition. The alternans was in-phase electromechanically representing the long AP with large CaT amplitude and slow rate of decay and the short AP with small CaT amplitude and fast rate of decay (Figure 4B and 4C).

In the long AP, the plateau phase of AP was elevated by blocking I_{Kur}, which led to extended open time of the I_{CaL} voltage-dependent activation gate (Figure 4E) resulting in increased transmembrane Ca^{2+} influx through I_{CaL}. More Ca^{2+} influx increased the CaT amplitude and slowed the decay time of Ca^{2+} transient, so that before the next AP intracellular Ca^{2+} did not completely recover to diastolic level. Therefore, calcium-dependent inactivation gate of I_{CaL} was smaller (Figure 4G) before the next AP resulting smaller I_{CaL} of the next AP (Figure 4D). In addition, due to the long AP, voltage-dependent inactivation gate of I_{CaL} did not completely recover to the level before this AP (Figure 4F) resulting smaller I_{CaL} of the next AP. The consequent smaller I_{CaL} shortened the AP and the plateau phase resulting reduced open time of the I_{CaL} voltage-dependent activation gate and decreased transmembrane Ca^{2+} influx that led to the short AP and small Ca^{2+} transient. Moreover, the alternans of Ca^{2+} transient caused alternans of I_{NCX} which played a significant role in AP repolarisation (Figure 4H).

4. Discussion and conclusion

In this study, the updated canine atrial cell model was used to investigate the effect of blocking I_{Kur} on the genesis of repolarisation alternans. In this simulation, blocking I_{Kur} from 40% to 80% produced noticeable long-short-long-short AP and CaT alternans at pacing CL of ~ 200ms to 440 ms. During these CL, the slope of DI-dependent APD_{90} restitutions curve was steeper which was related to the genesis of AP alternans. Further analysis for the the mechanism of alternans was demonstrated that the effect of I_{Kur} block on the plateau phase of AP played a significant role in I_{CaL} which was a major determinant of AP and CaT, consistent with rabbit atrial experimental results that alternation in AP morphology contributed to atrial alternans via I_{CaL} [13].

In conclusion, this study demonstrates that although prolonging the APD, blocking I_{Kur} may promote the genesis of AP alternans, indicating a latent pro-arrhythmic effects in normal atria.

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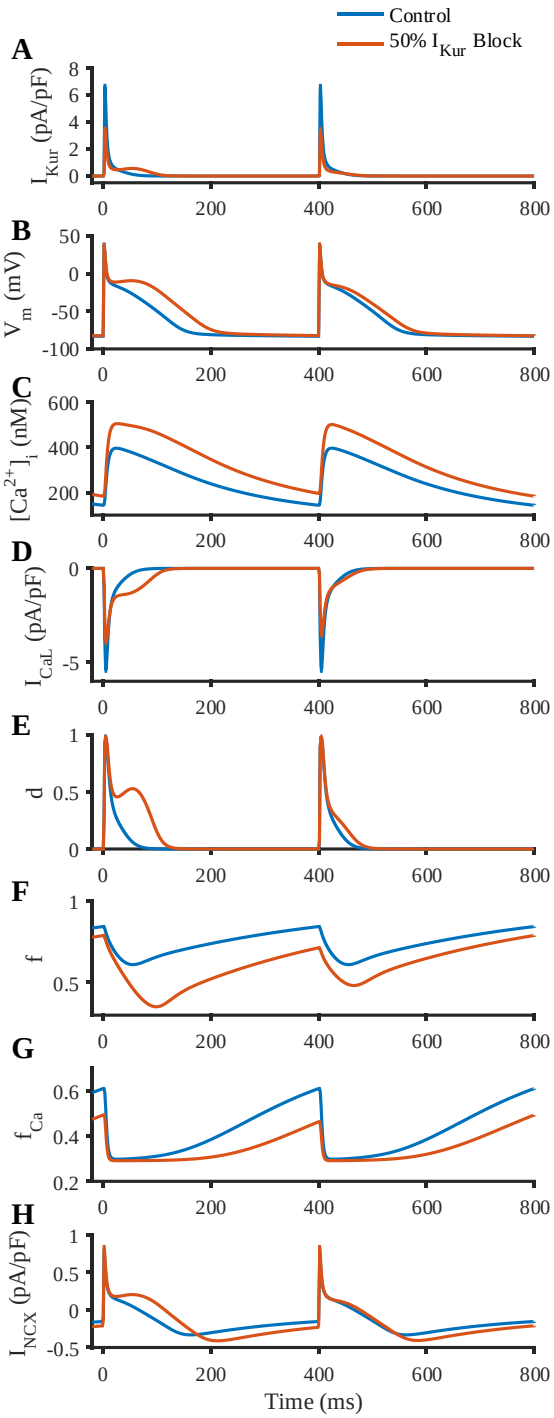

Figure 4. Alternans of (A) I_{Kur}, (B) AP, (C) intracellular CaT, (D) I_{CaL}, (E) d, (F) f , (G) f_{Ca}, and (H) I_{NCX} at pacing CL of 400 ms in control and 50% I_{Kur} block condition.

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