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Original Paper

Downregulation of Long Non-Coding RNA Kcnq1ot1: An Important Mechanism of Arsenic Trioxide-Induced Long QT Syndrome

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Key Words
Arsenic trioxide • Long QT syndrome • LncRNA Kcnq1ot1 • Kcnq1

Abstract
Background/Aims: Arsenic trioxide (ATO) is a known anti-acute promyelocytic leukemia (APL) reagent, whose clinical applications are limited by its serious cardiac toxicity and fatal adverse effects, such as sudden cardiac death resulting from long QT syndrome (LQTS). The mechanisms of cardiac arrhythmia due to ATO exposure still need to be elucidated. Long non-coding RNAs (lncRNAs) are emerging as major regulators of various pathophysiological processes. This study aimed to explore the involvement of lncRNAs in ATO-induced LQTS *in vivo* and *in vitro*. ***Methods:*** For *in vivo* experiments, mice were administered ATO through the tail vein. For *in vitro* experiments, ATO was added to the culture medium of primary cultured neonatal mouse cardiomyocytes. To evaluate the effect of lncRNA *Kcnq1ot1*, siRNA and lentivirus-shRNA were synthesized to knockdown lncRNA *Kcnq1ot1*. ***Results:*** After ATO treatment, the *Kcnq1ot1* and *Kcnq1* expression levels were down regulated. lncRNA *Kcnq1ot1* knockdown prolonged the action potential duration (APD) *in vitro* and exerted LQTS *in vivo*. Correspondingly, *Kcnq1* expression was decreased after silencing lncRNA *Kcnq1ot1*. However, the knockdown of Kcnq1 exerted no effect on lncRNA *Kcnq1ot1* expression. ***Conclusions:*** To our knowledge, this report is the first to demonstrate that lncRNA *Kcnq1ot1* downregulation is responsible for QT interval prolongation induced by ATO at least partially by repressing *Kcnq1* expression. lncRNA *Kcnq1ot1* has important pathophysiological functions in the heart and could become a novel antiarrhythmic target.

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Introduction

Arsenic trioxide (ATO) is a well-known traditional medicine with the chemical formula As_2O_3. In the 1970s, Dr. Zhang Tingdong, a Chinese researcher, uncovered the extraordinary therapeutic potential of ATO against acute promyelocytic leukemia (APL). Since then, ATO (Trisenox™) has become a choice for the treatment of relapsed or refractory APL worldwide. Thus far, it is the most active and effective single agent in the treatment of APL. ATO-based salvage therapy offers long-lasting remissions and cures up to 90% of patients with relapsed APL [1-3].

However, the high frequency and severity of complex arrhythmias as a consequence of the compound's cardiotoxicity limited the clinical application of ATO. Long QT syndrome (LQTS) is the most common cardiac rhythm abnormality observed during ATO therapy [3, 4]. Approximately, two thirds of patients receiving ATO develop significant but rapidly reversible QTc prolongation, which can predispose patients to temporary discontinuation, torsade de pointes and even sudden cardiac death unless stringent precautions are taken [5]. Due to these adverse cardiac effects, some patients have to be precluded from ATO therapy for APL. Therefore, clarifying the molecular toxicity mechanisms of ATO and minimizing the risk of LQTS are very important.

LQTS is characterized by delayed cardiac repolarization and increased risk of developing potentially fatal ventricular arrhythmias. It can be divided into congenital and acquired LQTS. Congenital LQTS is identified as LQT1 to LQT13 according to mutations in different genes [6, 7]. *KCNQ1/KCNE1*-encoded slow delayed rectifying K^+ channel (I_{Ks}) are expressed in diverse tissues and serve a variety of functions [8-10]. Mutation of *KCNQ1* is a cause of LQT1, making up approximately 40% to 55% of all LQTS cases [11]. In addition, *Kcnq1*-deficient mice show an LQTS phenotype [12]. I_{Ks} blockade significantly contributes to acquired LQTS, especially when repolarization reserve is reduced [13]. Most studies have shown that the QT prolongation induced by ATO can be attributed to cardiac repolarization abnormalities [14]. The chaos of ion channel stability has been well characterized in ATO-related cardiotoxicity. The alteration of *Kcnq1* is involved in ATO-induced QT interval prolongation [14]. However, the regulatory mechanisms for this involvement are still not fully understood.

Long non-coding RNAs (lncRNAs) are functional RNA molecules larger than 200 nucleotides without protein-coding potential. Recently, increasing evidence has suggested that lncRNAs play important roles in cardiac development and diseases, including acute myocardial infarction, hypertrophy and heart failure [15-18]. *KCNQ1* overlapping transcript1 (*Kcnq1ot1*) is an lncRNA transcribed from the *KCNQ1* locus. It plays pivotal regulatory role in the expression of both ubiquitously and tissue-specific imprinted genes within the *Kcnq1* domain and is thus involved in the pathogenesis of cancer, acute myocardial infarction and Beckwith-Wiedemann syndrome [19-22]. Therefore, it is likely that alteration of the lncRNA *Kcnq1ot1* may affect cardiac rhythm through the regulation of *Kcnq1*.

In preliminary experiments, we found that ATO inhibited the expression of both *Kcnq1ot1* and *Kcnq1* in primary cultured neonatal mouse cardiomyocytes. In addition, ATO-induced LQTS was associated with the blockage of I_{Ks} [14]. Based on these findings, we hypothesized that the alteration of lncRNA *Kcnq1ot1* may be an important mechanism of ATO-induced LQTS.

Materials and Methods

Animals

Mice were obtained from the Experimental Animal Center of Harbin Medical University. The methods were performed in accordance with the National Guidelines for Experimental Animal Welfare (the Ministry of Science and Technology, People's Republic of China, 2006) and the Guide for the Care and Use of Laboratory Animals published by the US National Institutes of Health (NIH Publication No. 85-23, revised 1996). All experimental protocols were pre-approved by the Experimental Animal Ethic Committee of Harbin Medical University, China (No. HMUIRB 20150034).

Isolation and culture of neonatal mouse cardiomyocytes

One- to three-day-old neonatal mice were rinsed quickly in 75% ethanol solution for surface sterilization. Hearts were extracted from the body and were transferred immediately into the bacterial dish containing cold DMEM (HyClone, UT, USA). The ventricles were washed and minced into small pieces. The cells were dissociated at 37°C with trypsin-EDTA solution (Beyotime Institute of Biotechnology, Jiangsu, China). Cells from subsequent digestion were added to an equal volume of DMEM containing 10% fetal bovine serum (BI, Kibbutz Beit Haemek, Israel) and were kept at 4°C. After final collection, the resulting mixture was filtered and centrifuged for 3 min at 1500 rpm to obtain pellet cells. The cells were resuspended in culture medium and incubated for 1.5 h under a water-saturated atmosphere of 5% CO_2-95% air to allow the attachment of non-cardiomyocytes. The suspended cells were then collected, plated into a new culture plate and incubated under the same conditions as mentioned above.

In vitro and *in vivo* model of ATO-induced cardiotoxicity

Neonatal mouse cardiomyocytes were incubated with or without ATO (2.5 or 5 μM) for 48 h. Subsequently, the total RNA and protein were extracted for the following experiments. Male adult Kunming mice weighting 25-30 g were used in this experiment. ATO (0.5, 1.5 or 4.5 $mg \cdot kg^{-1}$) or an equivalent volume of saline was administered to mice through the tail vein on alternate days. The electrocardiograms (ECGs) were monitored before and 7 days after treatment. The cardiac tissues were collected and frozen at -80°C until further analysis.

siRNA transfection

Neonatal mouse cardiomyocytes were transfected with siRNA targeting *Kcnq1ot1* or scrambled negative control siRNA (GenePharma, Shanghai, China) using X-treme GENE siRNA transfection reagent (Roche Applied Science, Prague, Czech Republic) according to the manufacturer's instructions. The siRNA sequences were as follows: si-Kcnq1ot1-1, GGGAAUCUGGUCUAAUGAATT, UUCAUUAGACCAGAUUCCCTT; si-Kcnq1ot1-2, CCUGGUGAAGGUACUAAAUTT, AUUUAGUACCUUCACCAGGTT.

Lentivirus-shRNA transfection

Lentivirus-shRNAs were designed and synthesized by GenePharma (GenePharma, Shanghai, China). Initially, the effects of lentivirus-shRNA were verified *in vitro*. Lentivirus-shRNAs were added to culture medium to yield a final multiplicity of infection (MOI) of 1. For *in vivo* experiments, lentivirus-shRNA (1×10^9 TU) diluted in 50 μL of saline was administered to mice through the tail vein.

Whole-cell patch clamp recording

Action potential duration (APD) was measured by the whole-cell patch-clamp method using a MultiClamp 700B amplifier (Axon Instruments, CA, USA). All experiments were performed using pCLAMP 10.2 software (Axon Instruments, Foster City, CA, USA). Boltzman's fits were performed.

Electrocardiograms

The standard limb lead II ECG was continuously recorded using a BL420s multichannel recorder (TME Technology, Chengdu, China). Arrythmias in mouse hearts were measured by ECG for a continuous period of 10 min. The rate-corrected QT interval (QTc) was calculated using Bazett's formula: $QTc = QT/(RR/100)^{1/2}$ [23].

Real-time reverse transcriptase polymerase chain reaction (RT-PCR)

Total RNA was extracted using TRIzol reagent (Invitrogen, CA, USA) from neonatal mouse cardiomyocytes or mouse myocardium. First-strand cDNA was synthesized using a reverse transcriptase kit (TaKaRa, Shiga, Japan) according to the manufacturer's instructions. Real-time RT-PCR analysis was performed on an ABI 7500 fast Real Time PCR system (Applied Biosystems, CA, USA) using SYBR Green I (Toyobo, Osaka, Japan). The thermal cycling conditions consisted of one cycle for 60s at 95°C and 40 cycles of 15 s at 95°C, 15 s at 60°C and 45 s at 72°C. GAPDH served as an internal control. Relative expression levels of target genes were calculated using the $2^{-\Delta\Delta CT}$ method. The primers sequences were as follows: *Gapdh*, 5'-AAGAAGGTG-GTGAAGCAGGC-3' (forward), 5'-TCCACCACCCTGTTGCTGTA-3' (reverse); *Kcnq1ot1*, 5'-GCACTCTGGGTCCT-GTTCTC-3' (forward) 5'-CACTTCCCTGCCTCCTACAC-3' (reverse); *Kcnq1*, 5'-CAAAGACCGTGGCAGTAAC-3' (forward), 5'-CCTTCATTGCTGGCTACAAC-3' (reverse); *Kcnh2*, 5'-TGGGGAGAAAAGTGACACCTG -3' (forward),

5'-CAGGGCTAGACAAGGGGATG -3' (reverse); *Kcne2*, 5'-GGTCTCAAGCTGAAAGTGCC -3' (forward), 5'-TGCT-GTGTGGTATGTGAGCA -3' (reverse).

Western blot analysis

Total protein extractions were obtained from neonatal mouse cardiomyocytes or cardiac tissues. The protein samples were separated by electrophoresis in 10% sodium dodecyl sulfate polyacrylamide gels, and the protein was transferred to polyvinylidene difluoride membranes (Millipore, MA, USA). Membranes were blocked with 5% non-fat dry milk in phosphate-buffered saline (PBS) for 2 h. The blots were incubated overnight at 4°C with primary antibodies. The next day, membranes were washed three times for 10 min each time with PBS containing 0.5% Tween 20 (PBS-T) and were then incubated with secondary antibodies for 1 h at room temperature. Finally, membranes were rinsed with PBS before scanning. Images were captured on the GelDox XR System (Bio-Rad, CA, USA). Western blotting bands were quantified using Quantity one software. Gapdh served as an internal control.

Data analysis

Data are expressed as the means ± SEM and were analyzed with SPSS 13.0 software. Statistical comparisons between two groups were performed using Student's t-test. Statistical comparisons among multiple groups were examined using analysis of variance (ANOVA). A two-tailed $P < 0.05$ was considered statistically significant. Graphs were generated using GraphPad Prism 5.0.

Results

QTc interval is prolonged in mice treated with ATO

To evaluate the effect of ATO on QTc interval, mice were administered different doses of ATO. No significant difference was observed between the 0.5 mg·kg^{-1} group and the control group after 7 days of treatment. However, the QTc interval was prolonged in the 1.5 and 4.5 mg·kg^{-1} groups compared with the control group (Fig. 1).

ATO downregulates Kcnq1 and lncRNA Kcnq1ot1 expression both in vivo and in vitro

The cardiac tissues were harvested after ECG recording in mice. The expression levels of lncRNA *Kcnq1ot1* and *Kcnq1* were determined using real-time PCR and western blot analysis. ATO at the doses of 1.5 and 4.5 mg·kg^{-1} significantly decreased *Kcnq1ot1* expression (Fig. 2A). Meanwhile, the *Kcnq1* mRNA and protein expression levels were both downregulated (Fig. 2B & C). Similarly, *Kcnq1ot1* and *Kcnq1* expression levels were also downregulated in neonatal mouse cardiomyocytes incubated with ATO (Fig. 2D-F).

siRNA targeting lncRNA Kcnq1ot1 downregulates Kcnq1 expression in cardiomyocytes

To assess the effect of lncRNA *Kcnq1ot1* knockdown on *Kcnq1* expression, two different siRNAs were used. lncRNA *Kcnq1ot1* expression was significantly downregulated after transfection in neonatal mouse cardiomyocytes. Both of the siRNAs targeting lncRNA

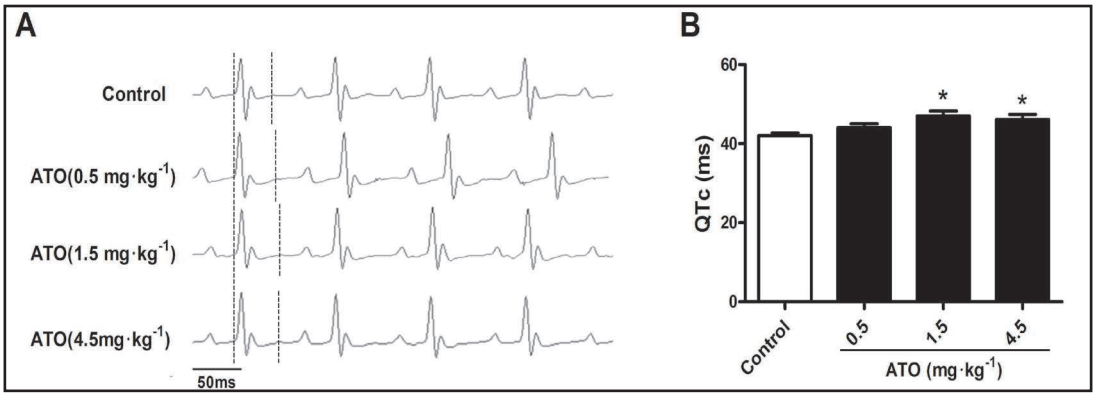

Fig. 1. Prolongation of corrected QT (QTc) in mice treated with arsenic trioxide (ATO). (A) Representative electrocardiogram. (B) ATO treatment prolonged QTc in mice. *P<0.05 versus Control; n = 6.

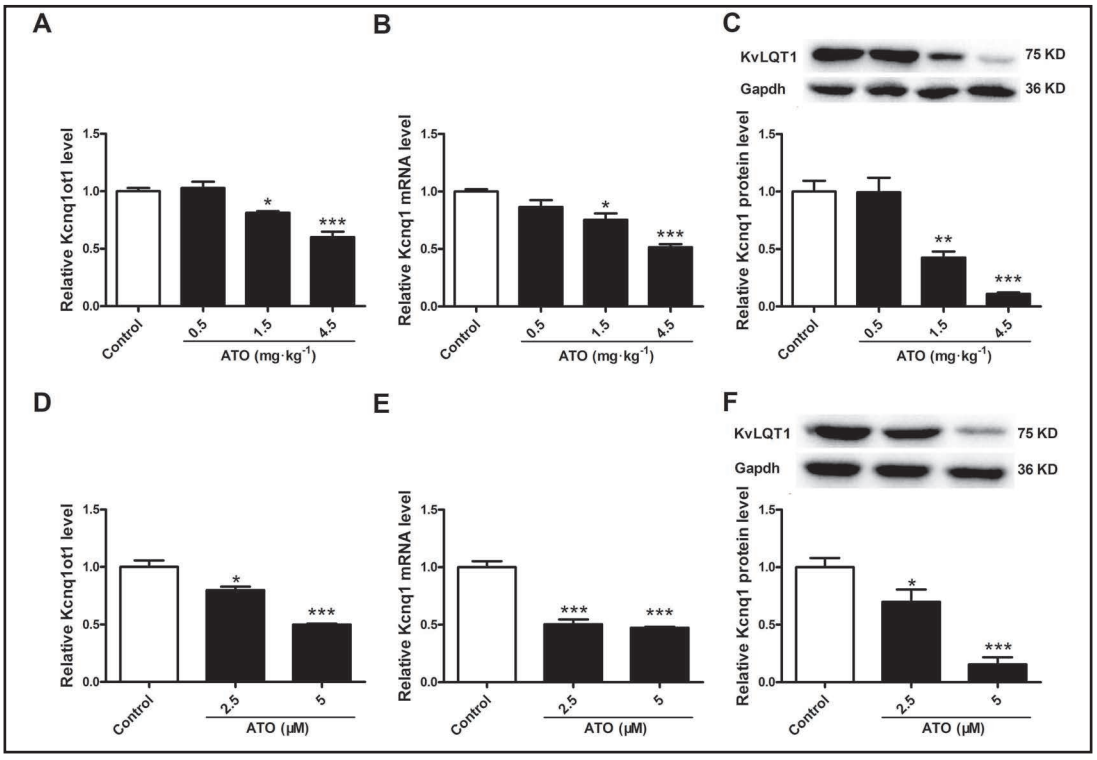

Fig. 2. Expression downregulation of lncRNA Kcnq1ot1 and Kcnq1 by arsenic trioxide (ATO) in vivo and in vitro. lncRNA Kcnq1ot1 (A), Kcnq1 mRNA (B) and KvLQT1 (C) expression in the myocardium of ATO-treated mice. lncRNA Kcnq1ot1 (D), Kcnq1 mRNA (E) and KvLQT1 (F) expression in ATO-treated neonatal mouse cardiomyocytes. *P<0.05 versus Control, **P<0.01 versus Control, ***P<0.001 versus Control; n = 3.

Kcnq1ot1 exerted similar effects on *Kcnq1* expression. The mRNA and protein expression levels of *Kcnq1* were both downregulated after the silencing of *Kcnq1ot1* (Fig. 3). The siRNA with better efficiency was selected for the subsequent experiments.

lncRNA Kcnq1ot1 knockdown lengthens APD in cardiomyocytes

APD in cardiac cells is the major determinant of the length of the QT interval. APD is governed by the balance between membrane depolarization and repolarization. The delayed rectifier K^+ current carried by the I_{Ks} channel is responsible for the terminal phase of cardiac repolarization, and the latter is encoded by *Kcnq1*, which is regulated by *Kcnq1ot1*. Our results showed that after *Kcnq1ot1* knockdown, the APD was significantly prolonged in primary cultured neonatal mouse cardiomyocytes (Fig. 4A & B). However, *Kcnh2* and *Kcne2* mRNA expression levels were not changed (Fig. 4C & D).

Lentivirus-shRNA targeting lncRNA Kcnq1ot1 decreases Kcnq1 expression in cardiomyocytes

The lentivirus carrying shRNA targeting lncRNA *Kcnq1ot1* was constructed based on the siRNA sequence. After incubation with the lentivirus-shRNA, *Kcnq1ot1* expression was significantly downregulated in neonatal mouse cardiomyocytes. Meanwhile, *Kcnq1* mRNA and protein expression levels were also inhibited (Fig. 5).

lncRNA Kcnq1ot1 knockdown exhibits a long QT phenotype in mice

To determine whether lncRNA *Kcnq1ot1* plays a role in cardiac repolarization, ECG recordings from *Kcnq1ot1* knockdown and negative control mice were evaluated. The lentivirus-shRNA or the negative control shRNA was administered to mice through the tail vein. The QTc interval was prolonged from day 7 after *Kcnq1ot1* knockdown (Fig. 6).

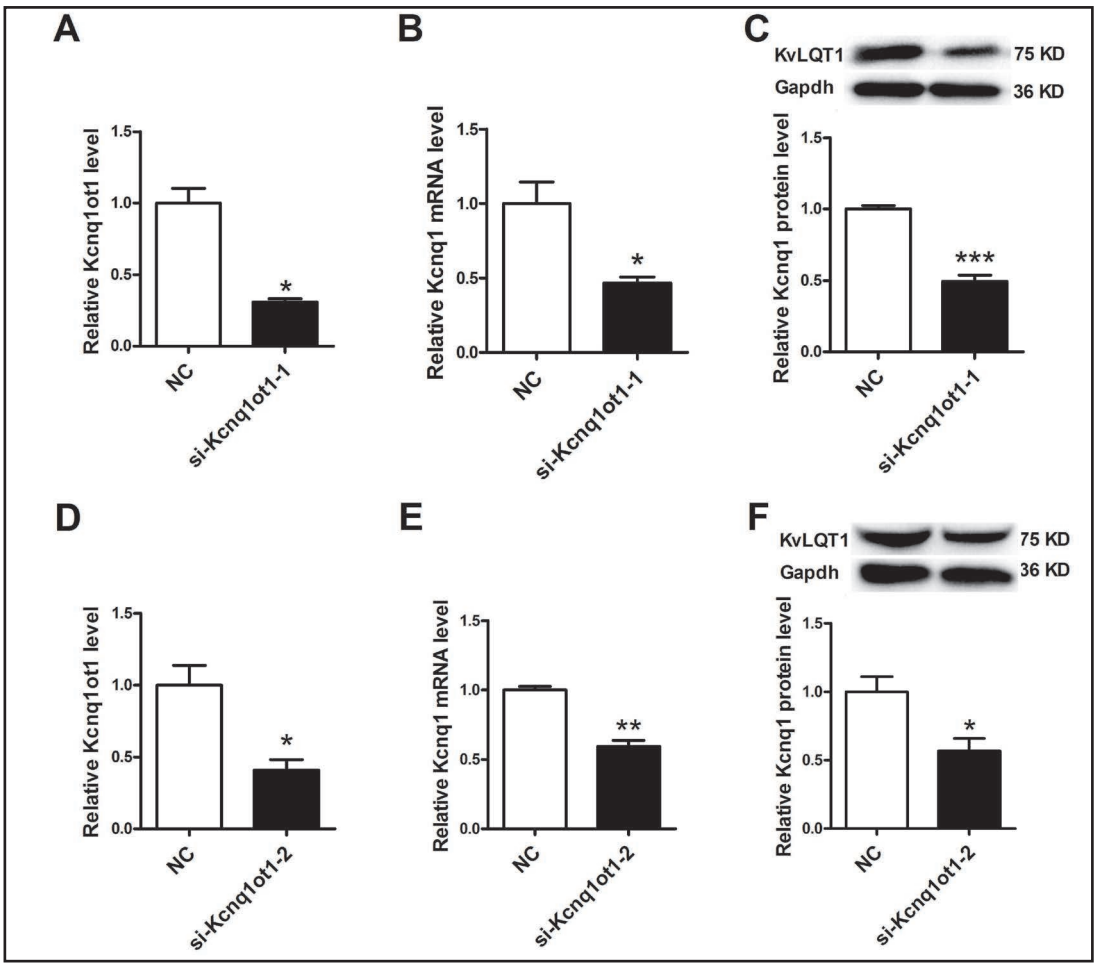

Fig. 3. Downregulation of Kcnq1 upon knockdown of endogenous Kcnq1ot1 by siRNA in neonatal mouse cardiomyocytes. si-Kcnq1ot1-1 and si-Kcnq1ot1-2 were used to knockdown lncRNA Kcnq1ot1. lncRNA Kcnq1ot1 (A & D) and Kcnq1 mRNA (B & E) expression levels were decreased. The expression of KvLQT1 (C & F) expression was also decreased. Gapdh served as an internal control. *P<0.05 versus Control, **P<0.01 versus Control, ***P<0.001 versus Control; n = 3.

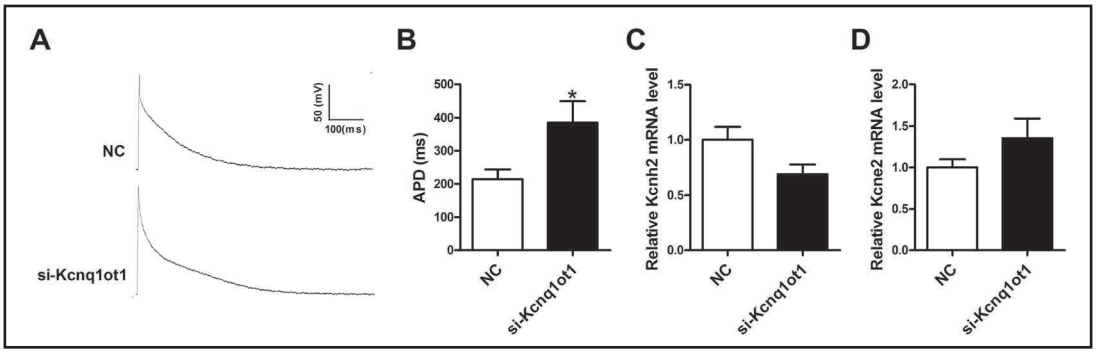

Fig. 4. Prolongation of action potential duration (APD) upon knockdown of endogenous Kcnq1ot1 by siRNA in neonatal mouse cardiomyocytes. (A) Representative cardiomyocyte action potential waveforms. (B) Action potential duration was prolonged after Kcnq1ot1 knockdown. (C) Kcnh2 mRNA expression was not changed after Kcnq1ot1 knockdown. (D) Kcne2 mRNA expression was not changed after Kcnq1ot1 knockdown. Gapdh served as an internal control. *P<0.05 versus NC. n = 4-5.

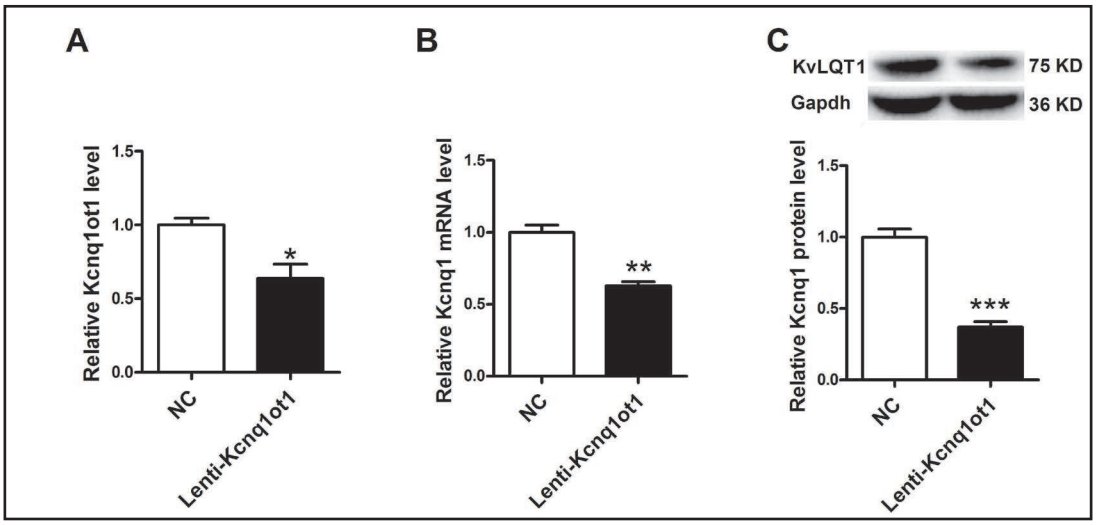

Fig. 5. Downregulation of Kcnq1 upon knockdown of endogenous Kcnq1ot1 by a lentivirus carrying shRNA in neonatal mouse cardiomyocytes. lncRNA Kcnq1ot1(A) and Kcnq1 mRNA (B) expression levels were decreased. KvLQT1 expression was also decreased. Gapdh served as an internal control. *P<0.05 versus NC, **P<0.01 versus NC, ***P<0.001 versus Control; n = 3.

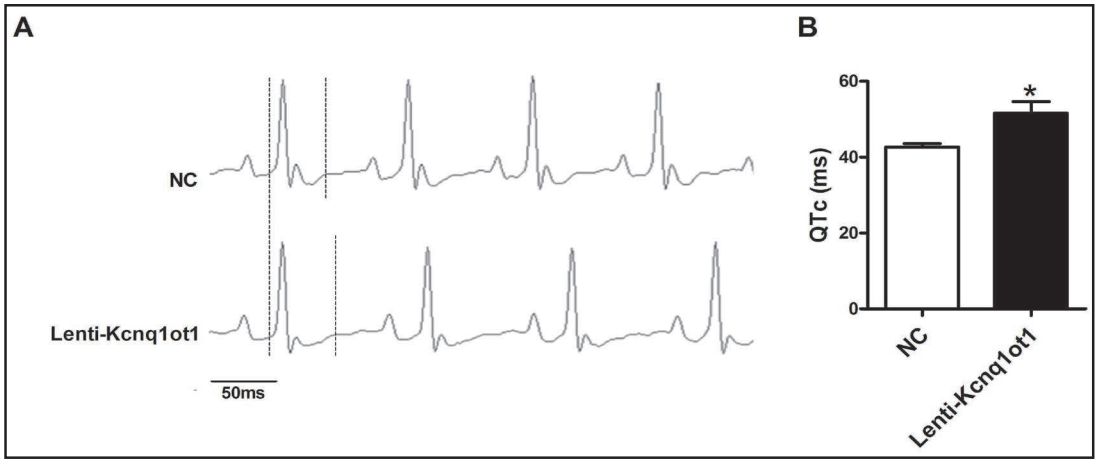

Fig. 6. Prolongation of corrected QT (QTc) upon knockdown of Kcnq1ot1 by lentivirus-shRNA in mice. (A) Representative electrocardiogram. (B) lncRNA Kcnq1ot1 knockdown prolonged QTc in mice. *P<0.05 versus NC; n = 5.

Kcnq1ot1 knockdown decreases Kcnq1 expression in mice

To determine the effects of lncRNA *Kcnq1ot1* on *Kcnq1* expression *in vivo*, cardiac tissues were harvested 7 days after administration of the lentivirus-shRNA. In accordance with *in vitro* experiments, lncRNA *Kcnq1ot1* expression was downregulated. Meanwhile, *Kcnq1* mRNA and protein expression levels were also decreased (Fig. 7A-C). However, downregulation of *Kcnq1* exerted no effect on lncRNA *Kcnq1ot1* expression (Fig. 7D-F).

Discussion

ATO is a highly effective treatment for APL. However, it can cause sudden cardiac death due to LQTS. Although dozens of studies have proposed several possible mechanisms, including disturbance of ion channel balance, apoptosis stimulation and promotion of oxidative injury, the exact regulatory network of ATO-induced cardiac toxicity has not been fully clarified [24-26].

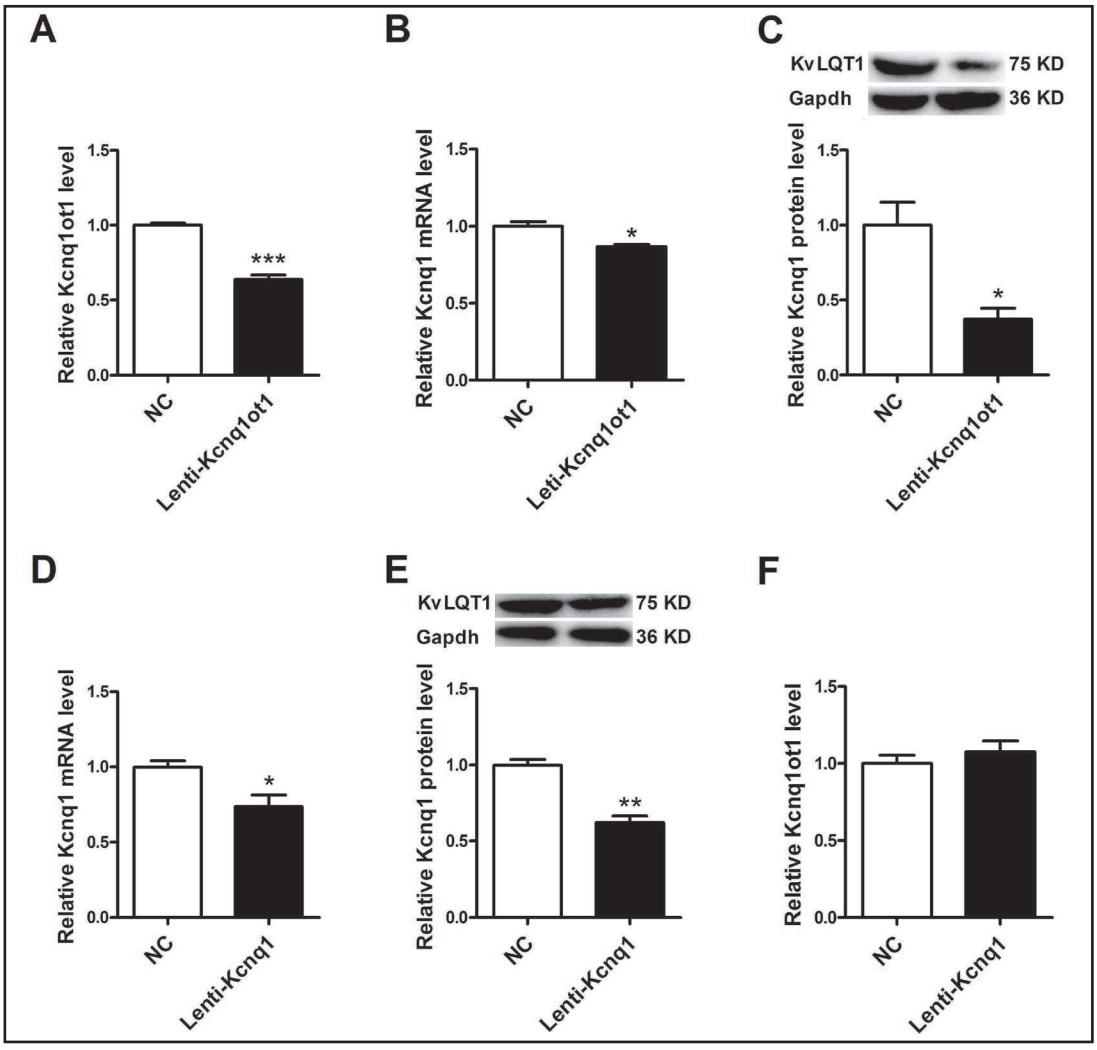

Fig. 7. Expression downregulation of Kcnq1 upon knockdown of Kcnq1ot1 by lentivirus-shRNA in mice. lncRNA Kcnq1ot1 (A), Kcnq1 mRNA (B) and KvLQT1 (C) expression levels were decreased in the myocardium of lenti-Kcnq1ot1-treated mice. Kcnq1 mRNA (D) and KvLQT1 (E) expression levels were decreased with no significant alteration of lncRNA Kcnq1ot1 (F) in the myocardium of lenti-Kcnq1ot1-treated mice. Gapdh served as an internal control. *P<0.05 versus NC, **P<0.01 versus NC, ***P<0.001 versus NC; n = 3-8.

ATO at a median dose of 0.15 mg·kg^{-1}·d^{-1}, ranging from 0.06 to 0.2 mg·kg^{-1}·d^{-1}, is recommended for the clinical treatment of APL. The QT changes that occur are usually observed within 7 days after treatment [5]. To mimic clinical drug administration, mice were treated with ATO at dose of 0.5, 1.5 or 4.5 mg·kg^{-1}, with the median dose of ATO in mice being equal to that in humans. The ECGs of mice were then monitored during the experiment. Seven days after treatment, the QTc was prolonged in mice treated with median and high dose of ATO (Fig. 1). This result was in accordance with the research exploring the effect of ATO on QTc interval prolongation in mice [27]. Based on this model, we explored the molecular mechanisms of ATO-induced LQTS.

lncRNAs constitute a novel class of non-coding RNAs that regulate gene expression. Although there is a growing interest in lncRNA-mediated biological and pathophysiological functions, little is known about the involvement of lncRNAs in the regulation of cardiac rhythm. *Kcnq1ot1* is a ubiquitously expressed lncRNA that is considered to have regulatory effects on cardiac development and is aberrantly expressed in the setting of myocardial ischemia [28, 29]. To explore the molecular mechanisms of ATO-induced LQTS, we focused

on *Kcnq1ot1*. Our results showed that ATO inhibited the expression of *Kcnq1ot1* both *in vitro* and *in vivo* (Fig. 2A & D). Moreover, *Kcnq1* expression was also downregulated (Fig. 2B, C, E & F).

To investigate the effects of lncRNA *Kcnq1ot1* on *Kcnq1*, siRNAs targeting lncRNA *Kcnq1ot1* were synthesized to knock down *Kcnq1ot1* in neonatal mouse cardiomyocytes. Both of the siRNAs could efficiently and specifically down regulate *Kcnq1ot1* expression, resulting in significant inhibition of *Kcnq1*(Fig. 3), and the APD was prolonged due to lncRNA *Kcnq1ot1* knockdown (Fig. 4A & B). Although I_{Kr} is also considered as an important regulator in LQTS pathogenesis, lncRNA *Kcnq1ot1* knockdown exerts no significant effects on the mRNA expression of *Kcnh2* and *Kcne2*, which encode I_{Kr} (Fig. 4C & D).

To further detect the effect of lncRNA *Kcnq1ot1* knockdown *in vivo*, lentivirus-shRNA targeting lncRNA *Kcnq1ot1* was constructed. The effects of lentivirus-shRNA were verified *in vitro*. After transfection, *Kcnq1ot1* expression was inhibited in neonatal mouse cardiomyocytes (Fig. 5A). Meanwhile, *Kcnq1* mRNA and protein expression levels were also decreased (Fig. 5B & C). We then performed an *in vivo* experiment. The mice treated with lentivirus-shRNA targeting lncRNA *Kcnq1ot1* exhibited a long QT phenotype in 7 days after lentivirus-shRNA administration (Fig. 6). In addition, *Kcnq1* expression was also inhibited. These observations indicate that lncRNA *Kcnq1ot1* knockdown could inhibit *Kcnq1* expression (Fig. 7A-C). These results disagreed with the research conducted by Korostowski et al., who purposed that the absence of *Kcnq1ot1* leads to *Kcnq1* overexpression [29]. This discrepancy could be explained by the variability of lncRNA effects depending on development stage and pathologic status. We also investigated whether a positive feedback loop existed between lncRNA *Kcnq1ot1* and *Kcnq1* by knocking down *Kcnq1* to observe the changes in lncRNA *Kcnq1ot1 in vivo*. Silencing of *Kcnq1* exerted no significant effect on lncRNA *Kcnq1ot1* (Fig. 7D-F). These findings indicated that ATO-induced LQTS was at least partially mediated by the regulation of the lncRNA *Kcnq1ot1*/*Kcnq1* axis.

Theoretically, a rescue experiment aiming to observe whether *Kcnq1ot1* upregulation could attenuate ATO-induced LQTS is indispensable. Unfortunately, due to technical difficulties, lncRNA *Kcnq1ot1* overexpression could not be achieved. Therefore, RNA interference was performed to mimic the effect of ATO on *Kcnq1ot1* in cardiomyocytes. Considering the characteristics of lncRNAs, it was difficult to proceed from mouse to human. Compared with protein-coding sequences, most lncRNAs are poorly conserved among vertebrates [30]. A recent study identified only 18 conserved lncRNAs in humans from 3133 mouse lncRNAs, confirming the fact that only a small minority of lncRNAs in mice or humans have transcribed homologous sequences across different species [31, 32]. In contrast, highly conserved lncRNAs could play critical and conserved roles across different species [33]. Fortunately, *Kcnq1ot1* is a relatively conserved lncRNA and likely exerts similar effects in different species. For instance, the dysfunction of *Kcnq1ot1* is responsible to Beckwith-Wiedemann syndrome in humans, similar to what has been observed for large offspring syndrome in bovines [34, 35]. In the present study, we found that the aberrant expression of lncRNA *Kcnq1ot1* was involved in ATO-induced LQTS in mice and could likely have a similar effect in humans. However, it remains to be further investigated whether homologous lncRNA *KCNQ1OT1* is aberrantly expressed in APL patients treated with ATO or merely in those with LQTS.

Overall, this study is the first to demonstrate that lncRNAs have regulatory potential for cardiac rhythm. The discovery of lncRNA *Kcnq1ot1*-mediated effects on cardiac rhythm in ATO-induced LQTS not only proposes a therapeutic target for ameliorating the cardiac toxicity of ATO but also provides a novel strategy for the treatment of arrhythmias and other related diseases.

Disclosure Statement

The authors declare no conflicts of interest.

Acknowledgements

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