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Inhibition of HERG potassium channels by domiphen bromide and didecyl dimethylammonium bromide

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Abstract

Domiphen bromide and didecyl dimethylammonium bromide were widely used environmental chemicals with potent activity on blockade of HERG channels. But the mechanism of their action is not clear. The kinetics of block of HERG channels by domiphen bromide and didecyl dimethylammonium bromide was studied in order to characterize the inhibition of HERG currents by these quaternary ammonium compounds (QACs). Domiphen bromide and didecyl dimethylammonium bromide inhibited HERG channel currents in a dose-dependent manner with IC₅₀ values of 9 nM and 5 nM, respectively. Block of HERG channel by domiphen bromide and didecyl dimethylammonium bromide was found to be voltage-dependent and use-dependent. Domiphen bromide and didecyl dimethylammonium bromide caused substantial negative shift of the activation curves, accelerated activated process, but had no effects on the deactivation and reactivation processes. The docking models implied that these two compounds bound to PAS domain of HERG channels and inhibited its function. Our data demonstrated that domiphen bromide and didecyl dimethylammonium bromide blocked the HERG channel with a preference for the activated channel state.

Keywords

Quaternary ammonium compounds; domiphen bromide; didecyl dimethylammonium bromide; patch clamp

1. Introduction

Domiphen bromide (DB) and didecyl dimethylammonium bromide (DDB), two members of quaternary ammonium compounds (QACs), are widely used in clinical and industrial fields. Domiphen bromide is used in the treatment of acute infectious oral diseases (Scaglione et

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al., 1983). Didecyl dimethylammonium bromide is being used in various industrial fields including bio-chemical industries (Kuo and Yu, 2011a, b). The chloride form of DDAB is authorized for use in food industries (Mechin et al., 1999). These two compounds, similar to the well-known voltage-gated potassium channel blocker tetraethylammonium (TEA), have four ethyl groups attached to a central nitrogen atom. Previous electrophysiological studies demonstrated that QA's binding site was located inside the channel pore, and it accessed this binding site through open potassium channel pore (Armstrong, 1969, 1971). Moreover, findings have been verified that TEA could be trapped inside the channel pore by closure of the activation gate. On the other hand, large QA compounds were reported to block K^+ channels by a foot in the door mechanism (Armstrong, 1969, 1971). These two mechanisms may reflect that different compounds cause various alterations on HERG channels kinetics.

The human ether-a-go-go related gene (HERG) potassium channel, a member of voltage-gated potassium channels, plays a pivotal role in cardiac rhythm regulation, especially in the repolarization of the cardiac action potential. Drugs selectively inhibiting HERG channels may reduce the repolarizing cardiac potassium currents, causing the prolonged cardiac action potential and producing long QT syndromes. Thus, the HERG channel has been subjected to a routine test for compound cardiac toxicity in the drug development process. Recently, several QACs including benzethonium chloride, domiphen bromide, and tetra-n-octylammonium bromide have been found to block the HERG channel (Long et al., 2013; Xia et al., 2011). To further investigate the potential mechanisms for the efficacy of HERG inhibition of domiphen bromide and didecyl dimethylammonium bromide, two QACs, we performed detailed studies to explore the effects of domiphen bromide and didecyl dimethylammonium bromide on the use-dependence, voltage-dependence and state-dependence of HERG channels expressed in Chinese hamster ovary (CHO) cells.

2. Materials and methods

2.1 Materials

The two quaternary ammonium compounds domiphen bromide and didecyl dimethylammonium bromide as well as other chemicals were purchased from Sigma (St. Louis, MO, USA).

2.2 Cell culture

HERG K^+ channels stably transfected CHO cell line was purchased from ChanTest (Cleveland, OH, USA). The cells were cultured in 35 mm plastic dishes with culture medium of HAMS F-12 (Invitrogen, Carlsbad, CA, USA), supplemented with 1 mM l-glutamine and 10% fetal bovine serum (Hyclone, Logan, UT, USA) in a humidified, 5% CO_2 incubator at 37°C.

2.3 General electrophysiologic recordings

HERG potassium current was recorded with the method published previously (Long et al., 2013). Briefly, whole-cell patch clamp technique was conducted at room temperature (22°C). The extracellular solution contained (mM): NaCl 137; KCl 4; $CaCl_2$ 1.8; $MgCl_2$ 1.0; glucose 10; HEPES 10; pH was adjusted to 7.4. An Axopatch 200B patch clamp amplifier in

conjunction with a Digidata 1400 interface (Axon Instruments) was used for recording. Using a Flaming/Brown micropipette puller (P-97; Sutter Instruments, Co.), patch pipettes were pulled and had resistances of 2–4 M Ω when filled with the internal pipette solution, which contained (mM): KCl 130; MgCl₂ 1; EGTA 5; Mg-ATP 5; HEPES 10; pH was adjusted to 7.2. Cell and pipette capacitances were nulled and series resistance was compensated (85–95%) before recording. Data were acquired using pCLAMP programs (10.0; Axon Instruments).

2.4 Recording HERG tail currents

To determine the concentration-response of domiphen bromide and didecyl dimethylammonium bromide on HERG tail currents (I_{HERG}), cells were depolarized from a holding potential of –80 mV to 20 mV for a period of 4 s, followed by 2 s repolarization to –40 mV, this cycle was repeated for 5 min in the absence of drugs (control) until the HERG current became stable. Thereafter, the test compound was added to the bath solution from low concentrations to high concentrations cumulatively and each concentration recorded for 5 min.

2.5 Recording voltage-dependent inhibition of HERG channels

To determine the current-voltage (I–V) relations of domiphen bromide and didecyl dimethylammonium bromide on I_{HERG} , cells were assessed with a standard I–V protocol. With the standard I–V protocol, the activating currents were elicited by 4 s depolarizing pulses ranging from –70 to +50 mV and the tail currents by 2 s repolarizing pulses to –40 mV. The voltage steps were delivered from a holding potential (HP) of –80 mV at an inter-pulse interval of 10 s.

2.6 Recording HERG channel kinetics

The activation curves were constructed with the standard I–V protocols. Activation time courses were obtained by fitting the currents elicited by a step to 0 mV from a holding potential of –80 mV to the double exponential function (fast and slow time course). To construct the inactivation curves, the following voltage protocols were employed: a 2 s depolarizing pulse to +40 mV to inactivate the HERG channels followed by varying repolarizing pulses to potentials between –140 to +30 mV for a short period to allow full recovery of channels from inactivation at more negative potentials and rapid inactivation at less negative potentials. Deactivation time course was measured using a double pulse protocol consisting of a depolarization to +20 mV followed by a test pulse of –40 mV. The decaying phase of the tail current was fitted to the double exponential function and calculated fast and slow components. Recovery from inactivation was measured using a double pulse protocol consisting of a depolarization to +50 mV followed by a test pulse of –140 mV. The time constants derived from the fits for the first phase (descending phase or increasing inward current) with the single exponential function.

2.7 Recording use-dependent inhibition

The use-dependent inhibition was recorded as follow: firstly, 4 s duration voltage steps to +20 mV were applied from the holding potential of –80 mV, then repolarized to –40 mV with

an interval of 30 s in the absence of compounds (Control run). Then, 3 nM domiphen bromide or didecyl dimethylammonium bromide was added to the bath solution. Every ten minutes later, the same protocol was repeated with the same cycle lengths (the second, third and fourth run).

2.8 Data analysis

All data were presented as means and standard error of the mean (S.E.M.). The statistical significance of data was analyzed with Student's t-test or one-way analysis of variance followed by the Dunnett's test using SPSS software. A probability of $P < 0.05$ was considered as indicating a statistically significant difference.

3. Results

3.1 Concentration-dependent effects on HERG tail currents

The voltage clamp protocol and the representative traces were shown in Figure 1, fitting of the data with the Hill equation indicated that the calculated half-maximal inhibitory concentration (IC_{50}) value for domiphen bromide block of peak tail I_{HERG} was 9 nM with a Hill coefficient of 0.24 (Figure 1A and 1C). The calculated IC_{50} value for didecyl dimethylammonium bromide block of peak tail I_{HERG} was 5 nM with a Hill coefficient of 0.45 (Figure 1B and 1C). In addition, the inhibitory effects of these two compounds did not recover after ten minutes of washing.

3.2 Voltage-dependent inhibition of HERG channels

The I–V relationship in the absence and presence of about IC_{50} concentration of domiphen bromide and didecyl dimethylammonium bromide (3 nM) on I_{HERG} were shown in Figure 2. In the absence of drug, the standard I–V curves at the pulse end demonstrated a strong inward rectification at potentials positive to 0 mV, whereas domiphen bromide and didecyl dimethylammonium bromide caused a significant shift of the curves to the negative direction (Fig 2C and 2G). Domiphen bromide at concentration of 3 nM substantially diminished the currents at varying test potentials: the blockade was significant increased between -70 mV and $+20$ mV (0–39.2%), then slightly declined at more positive potentials up to $+50$ mV (39.2–27.4%), whereas the blockade of tail current by 3 nM domiphen bromide was persistently raised between -70 mV and $+20$ mV (0–40.4%), then maintained in a stable level (Fig 2D).

Compared with domiphen bromide, didecyl dimethylammonium bromide at concentration of 3 nM caused a weaker inhibition on the currents elicited at the pulse end, with a blockade of 0–22.4% between -70 mV and $+10$ mV, then slightly declined at more positive potentials up to $+50$ mV (22.4–7.0%), whereas the blockade of tail current by 3 nM didecyl dimethylammonium bromide was persistently increased between -70 mV and $+20$ mV (0–38.9%) and maintained in a constant level (Fig 2H).

3.3 Effects on the kinetics of activation, deactivation, inactivation and reactivation of HERG currents

State-dependent channel blockers often demonstrate characteristic effects on channel kinetics. Once the HERG channel is activated, it undergoes a complex process of state changes from closed state to open/activation, inactivation, and then recovers from inactivation followed by back to closed state again. Therefore the effects of quaternary ammonium compounds on the kinetics of the HERG channels were then analyzed.

Figure 3 showed the effects of domiphen bromide and didecyl dimethylammonium bromide on the activation and inactivation curves. The activation curves were constructed by normalizing the tail currents recorded with the standard I–V protocols. The normalized data were plotted against the prepulse potentials and fitted to the Boltzmann distribution. As shown in Figure 3, both activation curves of domiphen bromide and didecyl dimethylammonium bromide were dramatically shifted to the negative direction, indicating both quaternary ammonium compounds could alter the activation properties of HERG channel. The $V_{1/2}$ value (the half-maximal activation voltage) and slope factor (κ) without domiphen bromide (3 nM) were -7.1 ± 0.3 mV and 8.3 ± 0.2 mV, and -19.9 ± 0.5 mV ($p < 0.05$, $n = 9$) and 8.3 ± 0.4 mV ($p > 0.05$, $n = 9$) respectively after domiphen bromide (3 nM) exposure. The $V_{1/2}$ value was changed from -7.5 ± 0.6 mV under control condition to -20.8 ± 0.6 mV with didecyl dimethylammonium bromide ($p < 0.01$, $n = 6$). The slope factor was not significantly altered (9.4 ± 0.6 mV for control vs 10.0 ± 0.5 mV for didecyl dimethylammonium bromide, $p > 0.05$, $n = 6$).

As shown in Figure 3, both domiphen bromide and didecyl dimethylammonium bromide (3 nM) did not alter the inactivation properties of HERG channel. The $V_{1/2}$ value (the half-maximal inactivation voltage) and slope factor (κ) without domiphen bromide (3 nM) were -88.2 ± 1.0 mV and -24.3 ± 0.8 mV, -91.1 ± 1.6 mV ($p > 0.05$, $n = 8$) and -27.4 ± 1.2 mV ($p > 0.05$, $n = 8$) respectively after domiphen bromide exposure. The $V_{1/2}$ value was changed from -89.0 ± 1.2 mV under control condition to -92.5 ± 0.8 mV with didecyl dimethylammonium bromide ($p > 0.05$, $n = 5$). The slope factor was not significantly altered (-26.5 ± 0.9 mV for control vs -23.2 ± 0.6 mV for didecyl dimethylammonium bromide, $p > 0.05$, $n = 5$).

In addition, the Figure 4 illustrated that domiphen bromide and didecyl dimethylammonium bromide had different effects on different states of HERG channels. As showed in Figure 4A, the activation time course was fitted to a double exponential function on the currents evoked at a test potential of 0 mV from a holding potential of -80 mV. Both domiphen bromide and didecyl dimethylammonium bromide significantly accelerated the channel activation time. The fast and slow time constants were 425 ± 37 and 2635 ± 392 ms in the control group, respectively. After 3 nM domiphen bromide treatment, the fast and slow time constants were reduced to 268 ± 32 and 1129 ± 117 ms ($p < 0.01$, $n = 8$), respectively. The fast and slow time constants decreased from 435 ± 41 and 2836 ± 414 ms to 250 ± 31 and 1165 ± 193 ms ($p < 0.01$, $n = 8$) after the treatment with 3 nM didecyl dimethylammonium bromide. The results together indicated that these two compounds may bind to its binding site on the open channel state because they significantly affected the channel activation kinetics.

The deactivation tail currents were elicited by a test pulse to -40 mV which were preceded by a 4 s prepulse to $+20$ mV (Fig 4B). The decay of the tail current was biexponential with fast and slow time constants of 133 ± 6 and 957 ± 64 ms in the control group, respectively. After application of 3 nM domiphen bromide, the tail current was well fitted by two components with time constants of 103 ± 8 and 903 ± 64 ms (paired t-tests, $p>0.05$, $n=8$). In addition, the fast and slow time constants in the presence of 3 nM didecyl dimethylammonium bromide were 157 ± 10 and 1078 ± 66 ms that were not significantly different from 114 ± 6 and 853 ± 47 ms ($n=9$) in the control group ($p>0.05$, $n=9$).

The effects of domiphen bromide and didecyl dimethylammonium bromide on recovery from inactivation were measured using a standard double-pulse protocol consisting of a depolarization to $+50$ mV for 1 s followed by a test pulse of -140 mV. The initial descending phase of the tail current upon hyperpolarization to -140 mV was taken as an indication of reactivation process. The currents were fitted with the single exponential function. Both compounds did not cause any significant changes in the recovery time course (Fig. 4C), which indicated that domiphen bromide and didecyl dimethylammonium bromide may not associate with the reactivated channels.

3.4 Use-dependent effects on HERG tail currents

Use-dependent inhibition usually shows that the compound inhibitory activity increases when the ion channels are repetitively activated and inactivated. The use dependency of HERG channel blockade by 3 nM domiphen bromide and didecyl dimethylammonium bromide were examined with the voltage pulses shown in Figure 5A. The typical traces and the normalized tail currents were recorded in 40 min after the treatment with 3 nM domiphen bromide (Fig 5B and 5D). During the first voltage step pulse after the application of domiphen bromide, the initial peak current was modest suppressed with a 32.7% inhibition (Fig. 5D). The inhibition increased to 48.9% in the second voltage step pulse. A stronger inhibition of 63.7% was observed in the third voltage step pulse followed by a 75.5% of inhibition at the fourth cycle. The results indicate that domiphen bromide blocks HERG channels in a use-dependent manner.

Compared to domiphen bromide, didecyl dimethylammonium bromide showed a similar effect on the use-dependency of HERG channel inhibition (Fig. 5C and 5E). The inhibition on HERG channel currents was 19.8% after the first step pulse that progressed slightly from 40.9% inhibition in the second cycle to 49.9% inhibition in the third cycle. A strong inhibition (58.6 %) was reached at the forth cycle.

3.5 The docking models of two QACs on the surface of the PAS domain of HERG

As shown in Figure 6, domiphen bromide and didecyl dimethylammonium bromide were docked into the PAS domain of HERG channels. The X-ray crystal structure of PAS (PDB ID: 4HQA) was minimized using protein modeling software MOE (Chemical Computing Group, Montreal, CANADA). The force field of Amber12:EHT was selected for both energy minimization and molecular docking. The molecular docking was carried out using the Induced Fit method provided in MOE.

The interaction potential energies, as evaluated using Amber 12 EHT force field embedded in MOE, were -68.63 kcal/mol (-31.49 kcal/mol contributed from van de Waals and -37.14 kcal/mol from electrostatic energies) for domiphen bromide and -60.44 kcal/mol (-27.13 kcal/mol from van de Waals and -33.31 kcal/mol from electrostatic energies) for didecyl dimethylammonium bromide. In addition, the results also demonstrated that the positively charged tertiary amine in both molecules were adjacent to the negatively charged residue GLU37, while the rest of the molecules fit very well in the hydrophobic cleft formed by residues ALA34, VAL36, PRO63, ALA83, LEU86, LEU87, ALA89, VAL122, and PHE125, indicating the binding sites of two compounds located on the PAS domain at the HERG N-terminus.

4. Discussion

Quaternary ammonium compounds were a large family including numerous widely used environmental chemicals such as tetraethylammonium (TEA), tetra-n-octylammonium bromide, domiphen bromide and benzethonium chloride etc. Tetra-n-octylammonium bromide and benzethonium chloride have been demonstrated to be highly effective blockers of HERG channels, these two compounds were reported as open HERG channel blockers (Long et al., 2013; Xia et al., 2011). In this study, we compared the kinetics of block of HERG channels by domiphen bromide and didecyl dimethylammonium bromide.

Our findings demonstrated that there were some similarities in the mechanism of HERG blockade by domiphen bromide and didecyl dimethylammonium bromide. First, both domiphen bromide and didecyl dimethylammonium bromide blocked HERG channel currents expressed in CHO cells in a voltage-dependent and use-dependent manner. Second, domiphen bromide and didecyl dimethylammonium bromide shifted the activation curve to the hyperpolarized direction, accelerated activation process. Besides, these two compounds had no effects on the deactivation and reactivation time constants of HERG channels. Third, both domiphen bromide and didecyl dimethylammonium bromide did not cause any shift in HERG inactivation I-V curve in transfected CHO cells. Overall, we suspected that the HERG blockade by domiphen bromide and didecyl dimethylammonium bromide were mainly associated with the binding of the compounds to open channels.

Domiphen bromide and didecyl dimethylammonium bromide preferentially binds to open channels was supported by two evidences. First, these two compounds displayed significant use dependency, indicating that it was so-called open channel blockers. These results agree with the previously results (Snyders and Chaudhary, 1996; Weerapura et al., 2002). Second, both domiphen bromide and didecyl dimethylammonium bromide altered the activation properties of HERG channels. External application of domiphen bromide and didecyl dimethylammonium bromide induced an acceleration of activation of HERG channels, demonstrated with a negative shift of 12.8 mV and 13.3 mV in $V_{1/2}$ of the activation I-V curves (Fig. 3) and the considerably decreased activation time course (Fig. 4). A similar shift in the gating of cardiac I_{Kr} current has been observed with antiarrhythmic drugs suggesting that this may be a common mechanism of many different HERG channel inhibitors (Carmeliet, 1992, 1993). In addition, the domiphen bromide and didecyl dimethylammonium bromide inhibition of heterologously expressed HERG channels was

voltage sensitive, increasing for steps more depolarized than -40 mV and reaching a maximum at approximately 0 mV (Fig. 2). This increase in compounds inhibition parallels the voltage-dependent activation of the channels suggesting that activation or opening may play an important role in domiphen bromide and didecyl dimethylammonium bromide binding.

Compared with the reported two compounds tetra-n-octylammonium bromide and benzethonium chloride (Long et al., 2013), the HERG channel blockade features of domiphen bromide and didecyl dimethylammonium showed high similarities to benzethonium chloride. They both exhibited a progressive concentration-dependent inhibition rather than a rapid inhibitory effect. In addition, the voltage-dependency and state-dependency including activation, inactivation, deactivation, and reactivation characteristics of HERG channel blockade are similar to benzethonium chloride. However, domiphen bromide and didecyl dimethylammonium also showed significant use-dependent inhibitory effects on HERG channels, which are similar to tetra-n-octylammonium bromide. With the help of in silico modeling approach, we further verified that these QACs shared a similar pharmacophore, the positively charged tertiary amine, which was adjacent to the negatively charged residue GLU37, while the hydrophobic cleft formed by residues ALA34, VAL36, PRO63, ALA83, LEU86, LEU87, ALA89, VAL122, and PHE125 on HERG channels helped the binding of QACs. This positively charged center plus one or two hydrophobic sites on QACs has been reported as a common pharmacophore for a broad spectrum of structurally diverse HERG channel inhibitors (Aronov, 2005; Sun et al., 2013).

Our understanding is that the interactions between the PAS domain and the HERG channel are required for the channel to function normally, including activation and deactivation (Morais Cabral et al., 1998; Sanguinetti and Tristani-Firouzi, 2006). Although the four quaternary ammonium compounds contain the similar NR_4^+ scarf, they have different substituted alkyl group or aryl group that may have different effects on their channel inhibition properties. Domiphen bromide, benzethonium chloride, and didecyl dimethylammonium bromide may bind to the PAS domain tightly to interrupt the interactions between PAS and HERG, which may further influence the deactivation behavior of the channel.

In conclusion, our data demonstrated that domiphen bromide and didecyl dimethylammonium bromide inhibited HERG channel in similar voltage-dependent, use-dependent, and state-dependent way. The in silico modeling approach further verified that both compounds shared an identical pharmacophore of positively charged tertiary amine. However, there were some limitations in our study. First, our experiments did not exclude the possibility of close state-dependent block by the compounds. Second, extensive site-directed mutagenesis was required to confirm the precise molecular determinants. Nevertheless, these topics were beyond the scope of the present study, it was of great interest and importance to make further investigation in order to have better understand of mechanisms of quaternary ammonium compounds actions.

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Fig. 1.

Concentration-dependence of HERG blockade by domiphen bromide and didecyl dimethylammonium bromide in transfected CHO cells. (A, B) Typical traces of the HERG currents recorded before and after different concentrations of domiphen bromide and didecyl dimethylammonium bromide. (C) Dose-response curves of the HERG channel blockade by quaternary ammonium compounds ($n=5-12$). Shown were averaged data and fitted to the Hill equation: $B(\%) = 100 / [1 + (IC_{50}/D)^n]$, where $B(\%)$ is the percent changes of HERG tail current at a drug concentration D , IC_{50} is the concentration of tested compounds that produces 50% effects, and n is the Hill coefficient.

Fig. 2.

Voltage-dependence of HERG channel blockade by domiphen bromide and didecyl dimethylammonium bromide in transfected CHO cells. (A, B and E, F) Representative traces of the HERG currents recorded before and after domiphen bromide and didecyl dimethylammonium bromide with a standard protocol. I-V relationship (C and G) and block percentage (D and H) of the HERG channels before and after administration of domiphen bromide and didecyl dimethylammonium bromide determined by the standard I-V protocol.

Fig. 3.

Effects of domiphen bromide and didecyl dimethylammonium bromide on the activation and inactivation curves of the HERG channels expressed in CHO cells. (A and B) Negative shifts of the activation curves produced by domiphen bromide and didecyl dimethylammonium bromide. Currents were elicited with the standard I-V protocols and the amplitude of the HERG tail currents at a repolarized test potential of -40 mV was normalized to the maximum values and plotted against the prepulse potentials. (C and D) Effects of domiphen bromide (3 nM) and didecyl dimethylammonium bromide (3 nM) on the inactivation conductance curve. To construct the inactivation curves, the voltage protocols employed were: a 2 s depolarizing pulse to $+40$ mV to inactivate the HERG channels, followed by varying repolarizing pulses to potentials from -140 mV to $+30$ mV for 5 ms followed by a test pulse to $+20$ mV. The amplitude of the HERG currents at the test potential was normalized and plotted against the prepulse potentials. The curves represent the best fits to the Boltzmann distribution: $I/I_{\max} = 1 / \{1 + \exp[(V_{1/2} - V)/\kappa]\}$, where I is the HERG tail current amplitude at a prepulse potential V , $V_{1/2}$ is the voltage for half-maximal activation/inactivation, and κ is a slope factor. ($n=6-9$).

Fig. 4. Effects of domiphen bromide (DB) and didecyl dimethylammonium bromide (DDB) on the kinetics of the HERG channels. (A) Both domiphen bromide and didecyl dimethylammonium bromide accelerated HERG activation. After the treatments with these two compounds, the activation time courses of HERG channels were significantly decreased. Both domiphen bromide and didecyl dimethylammonium bromide had no effects on the HERG deactivation (B) and recovery from inactivation (C). * $p < 0.05$, ** $p < 0.01$ (n=5–8).

Fig. 5. Use-dependence of HERG channel blockade by domiphen bromide and didecyl dimethylammonium bromide in HERG transfected CHO cells. (A) The voltage clamp protocol to evoke HERG currents expressed in CHO cells (Methods 2.7). Typical HERG currents measured in the absence and presence of 3 nM domiphen bromide (B) and didecyl dimethylammonium bromide (C). Use-dependent blockade of domiphen bromide (D) and didecyl dimethylammonium bromide (E) on HERG channels ($n = 5$). The peak tail current in the second, third and fourth run were normalized to their corresponding amplitude in the first run and plotted against time after application of the compounds.

Fig. 6.

The docking models of domiphen bromide and didecyl dimethylammonium bromide on the surface of the PAS domain of HERG (PDB ID: 4HQA). (A and B) Domiphen bromide was docked into the PAS domain of HERG. The ligand was colored in green (stick), and the protein in cyan (wire in (a) and surface in (b)). (C and D) Didecyl dimethylammonium bromide was docked into the PAS domain of HERG. The ligand was colored in green (stick), and the protein in blue (wire in (a) and surface in (b)).