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The antipsychotic drug chlorpromazine inhibits HERG potassium channels

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1 Acquired long QT syndrome (aLQTS) is caused by prolongation of the cardiac action potential because of blockade of cardiac ion channels and delayed repolarization of the heart. Patients with aLQTS carry an increased risk for *torsade de pointes* arrhythmias and sudden cardiac death. Several antipsychotic drugs may cause aLQTS. Recently, cases of QTc prolongation and *torsade de pointes* associated with chlorpromazine treatment have been reported. Blockade of human ether-a-go-go-related gene (HERG) potassium channels, which plays a central role in arrhythmogenesis, has previously been reported to occur with chlorpromazine, but information on the mechanism of block is currently not available. We investigated the effects of chlorpromazine on cloned HERG potassium channels to determine the biophysical mechanism of block.

2 HERG channels were heterologously expressed in *Xenopus laevis* oocytes, and ion currents were measured using the two-microelectrode voltage-clamp technique.

3 Chlorpromazine blocked HERG potassium channels with an IC_{50} value of 21.6 μM and a Hill coefficient of 1.11.

4 Analysis of the voltage dependence of block revealed a reduction of inhibition at positive membrane potentials.

5 Inhibition of HERG channels by chlorpromazine displayed reverse frequency dependence, that is, the amount of block was lower at higher stimulation rates. No marked changes in electrophysiological parameters such as voltage dependence of activation or inactivation, or changes of the inactivation time constant were observed.

6 In conclusion, HERG channels were blocked in the closed and activated states, and unblocking occurred very slowly.

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Abbreviations: HERG, human ether-a-go-go-related gene; I_{Kr}, rapidly activating component of I_K; LQTS, long QT syndrome; aLQTS, acquired long QT syndrome

Introduction

Higher than average total cardiovascular mortality and cases of sudden cardiac death in psychiatric patients have raised the concern that antipsychotic drugs may cause life-threatening arrhythmias. Recent studies have revealed the potential of several antipsychotic drugs to induce acquired long QT syndrome (aLQTS), characterized by prolonged rate-corrected QT intervals (QTc), and a high risk for both *torsade de pointes* tachyarrhythmias and sudden cardiac death (Reilly *et al*., 2000; Glassman & Bigger, 2001; Haddad & Anderson, 2002). Chlorpromazine is a conventional antipsychotic drug that has been used for the management of psychotic disorders since its FDA approval in 1954. Although there have been reports on chlorpromazine-induced QTc lengthening and *torsade de pointes* (Hoehns *et al*., 2001), Reilly *et al*. (2000) did not find a statistically significant association of QTc prolongation with chlorpromazine use.

Acquired and hereditary long QT syndromes are based on disorders of cardiac repolarization. Repolarization of ventricular myocytes is mainly driven by outward potassium currents, with the rapid component of the delayed rectifier potassium current, I_{Kr}, as one of their most important members. The human ether-a-go-go-related gene (HERG) (Sanguinetti *et al*., 1995) encodes the major protein underlying I_{Kr}, and mutations in HERG account for chromosome 7-linked inherited long QT syndrome (LQT-2) (Viskin, 1999; Ficker *et al*., 2000). Pharmacological inhibition of HERG potassium channels is a property of the class III antiarrhythmic drugs dofetilide (Kiehn *et al*., 1996), amiodarone (Kiehn *et al*., 1999), BRL-32872 (Thomas *et al*., 2001), or bertosamil (Zitron *et al*., 2002). In addition, several other nonantiarrhythmic compounds block HERG currents, such as the β-adrenoceptor antagonist carvedilol (Karle *et al*., 2001), the tricyclic antidepressants imipramine and amitriptyline (Teschemacher *et al*., 1999), the selective serotonin reuptake inhibitor fluoxetine (Thomas *et al*., 2002), the histamine receptor antagonists terfenadine and astemizole (Suessbrich *et al*., 1996), and fluoroquinolone-based antibacterial drugs (Kang *et al*., 2001). Finally, the antipsychotic drugs haloperidol (Suessbrich *et al*., 1997), thioridazine (Drolet *et al*., 1999), and

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sertindole (Lacerda *et al*., 2001) have been shown to inhibit HERG channels. Blockade of I_{Kr} causes lengthening of the cardiac action potential, which may produce a beneficial class III antiarrhythmic effect. On the other hand, excessive prolongation of the cardiac action potential may lead to aLQTS and life-threatening '*torsade de pointes*' arrhythmias (Napolitano *et al*., 1994).

HERG potassium-channel inhibition by chlorpromazine has been reported by Tie *et al*. (2000). The aim of the present study was to investigate the biophysical mechanism of this action, using the *Xenopus laevis* oocyte expression system and the two-microelectrode voltage-clamp technique.

Methods

Molecular biology

Details on procedures for *in vitro* transcription and oocyte injection have been published previously (Kiehn *et al*., 1999). Briefly, HERG wild-type (GenBank accession number: hs04270) cRNA (Warmke & Ganetzky, 1994) was prepared with the mMESSAGE mMACHINE kit (Ambion, Austin, U.S.A.) using SP6 RNA polymerase after linearization with *Eco*RI (Roche Diagnostics, Mannheim, Germany). Stage V–VI defolliculated *Xenopus* oocytes were injected with 46 nl of cRNA per oocyte. The investigation conforms to the *Guide for the Care and Use of Laboratory Animals* published by the U.S. National Institute of Health (NIH Publication No. 85-23, revised 1996).

Electrophysiology and statistics

Two-microelectrode voltage-clamp recordings from *Xenopus laevis* oocytes were carried out as published previously (Thomas *et al*., 1999). In brief, recordings were performed using a Warner OC-725A amplifier (Warner Instruments, Hamden, U.S.A.) and pClamp software (Axon Instruments, Foster City, U.S.A.) for data acquisition and analysis. Microelectrodes had tip resistances ranging from 1 to 5 MΩ. The recording chamber was continually perfused. All experiments were carried out at room temperature (20–22°C), and no leak subtraction was carried out during the experiments.

Concentration–response relations for chlorpromazine block were fit with a Hill equation of the form $I_{chlorpromazine}/I_{control} = 1/[1 + (C/IC_{50})^n]$, where I indicates the current, C is the chlorpromazine concentration, n is the Hill coefficient, and IC_{50} is the concentration necessary for 50% block. Activation curves were fit with a single-power Boltzmann distribution of the form $I_{tail} = I_{tail-max}/[1 + e^{(V_{1/2}-V)/k}]$, where V is the test pulse potential, $V_{1/2}$ is the half-maximal activation/inactivation potential, and k is the slope factor. Inactivation curves were fit to the following single-power Boltzmann equation: $I = I_{max}/[1 + e^{(V-V_{1/2})/k}]$. All data are expressed as mean ± standard deviation. We used the paired and unpaired Student's t-test (two-tail test) to compare the statistical significance of the results: $P < 0.05$ was considered statistically significant.

Solutions and drug administration

Voltage-clamp measurements of *Xenopus* oocytes were performed in a solution containing (in mM): 5 KCl, 100 NaCl, 1.5 $CaCl_2$, 2 $MgCl_2$, and 10 HEPES (pH 7.4 with NaOH). Current and voltage electrodes were filled with 3 M KCl solution. Chlorpromazine (Sigma, St Louis, U.S.A.) was prepared as a 100 mM stock solution in water and stored at −20°C. On the day of experiments, aliquots of the stock solution were diluted to the desired concentration with the bath solution.

Results

HERG potassium currents are inhibited by chlorpromazine

Chlorpromazine blocked HERG potassium channels expressed in *Xenopus laevis* oocytes in a concentration-dependent manner, as displayed in Figure 1. HERG currents were elicited by a 2 s depolarizing step to +20 mV followed by a repolarizating step to −40 mV for 1.6 s to produce large, slowly decaying outward tail currents that are a characteristic of HERG potassium currents (Sanguinetti *et al*., 1995). The holding potential was −80 mV. This voltage protocol was repeated every 10 s during superfusion with the drug solution for 25 min. When 1 mM chlorpromazine was applied, the incubation time had to be reduced to 10 min because of the death of the cells after longer perfusion periods. After the monitoring period, test pulses were applied to determine the amount of block. The recorded HERG tail currents were blocked by chlorpromazine (Figure 1a). To study the concentration dependence of HERG current block by chlorpromazine, HERG peak tail currents were normalized to the respective control values and plotted as relative current amplitudes in Figure 1b ($n = 3–5$ oocytes were investigated at each concentration). The half-maximal inhibition concentration (IC_{50}) for block of tail currents was 21.6 ± 6.9 μM with a Hill coefficient n_H of 1.11 ± 0.18. Chlorpromazine (100 μM) blocked HERG peak tail currents by 86.8 ± 4.8%, whereas currents at the end of the depolarizing pulses were blocked by 82.8 ± 5.7% ($n = 5$).

The time course of block is shown in Figure 1c. The onset of block was fast. After a control period of 21 min, HERG channel block by 100 μM chlorpromazine occurred rapidly within approximately 10 min. During the following 21 min of drug application, a slight increase in the degree of block (less than 7%) could be observed. Upon washout of chlorpromazine, the blocking effects on HERG were at this high concentration to a low degree reversible within 30 min.

Chlorpromazine blocks HERG potassium channels in the activated state

To investigate whether the channel is blocked in the closed or activated (open and/or inactivated) state, we activated currents using a protocol with a single depolarizing step to 0 mV for 7.5 s. After having obtained the control measurement, we allowed 100 μM of the drug to wash in for 25 min, while holding all channels in the closed state at −80 mV membrane potential. Then measurements with chlorpromazine were performed (Figure 2a). The degree of inhibition (i.e. current after chlorpromazine/control current × 100) after the incubation period is displayed with linear and logarithmic timescale in Figures 2b and c, respectively. Analysis of the test pulse after chlorpromazine application revealed a time-dependent

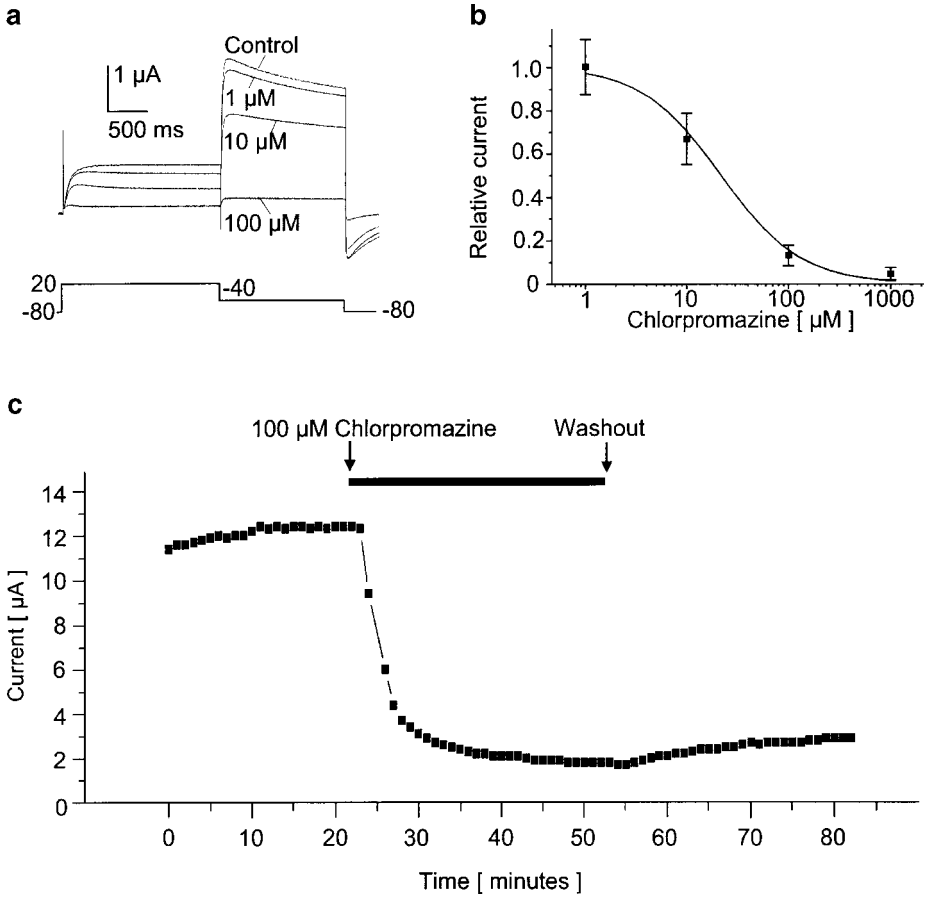

Figure 1 Inhibition of HERG channels by chlorpromazine. Currents were evoked by a depolarizing pulse to +20 mV (2 s), and tail currents were recorded during a step to −40 mV (1.6 s). Current amplitudes were monitored during control periods and after 25 min of drug application with the same voltage protocol (0.1 Hz pulsing frequency). Representative HERG current traces recorded under control conditions and after perfusion with chlorpromazine (1, 10, and 100 μM) are displayed in panel (a). (b) Dose–response curve for the effect of chlorpromazine on HERG peak tail currents. Error bars denote s.d. ($n = 3-5$ oocytes). The IC_{50} yielded 21.6 μM. (c) Time course of HERG tail current inhibition by 100 μM chlorpromazine. Currents were measured as described above. For clarity, not all current measurements are displayed. After a control period of 21 min, currents decreased rapidly upon perfusion with the drug solution within 10 min. Subsequent 21 min of drug application leads to only a slight further increase in the degree of block (<7%).

increase of block to about 78% at 400 ms (Figure 2c), which is consistent with a fast block of open HERG potassium channels. Some degree of inactivated or closed state block cannot be ruled out by this protocol. In this series of experiments, chlorpromazine reduced HERG outward currents at the end of the 0 mV pulse by 72.0±5.1% ($n = 5$).

To determine the voltage dependence of HERG channel block by chlorpromazine, we applied the following methodical approach. Since unblocking was very slow, only one experiment at each potential could be carried out with one individual oocyte. Currents were elicited by 35 s depolarizing pulses ranging from −40 to 40 mV from a holding potential of −80 mV, and peak inward tail currents were recorded during a second step to −120 mV (400 ms). First, control currents were recorded. Then the oocyte was superfused with the drug solution (50 μM chlorpromazine), while holding the cell at constant −80 mV for 25 min, where HERG channels are in the closed state. After this, measurements at the test pulse potential were performed. Relative inhibition of the outward currents (measured at the end of the 35 s test pulse) and block of peak tail currents were plotted as functions of the preceding test pulse potential in Figure 3a and b ($n = 3-5$ cells studied at each potential). Chlorpromazine reduced outward currents and peak tail currents between −40 and 20 mV in a voltage-dependent manner, with the block being more pronounced at negative membrane potentials. In contrast, at 40 mV membrane potential, there was virtually no effect on HERG tail currents, and even a slight increase of outward currents could be observed. This is because of the chlorpromazine-induced activation of endogenous chloride currents in *Xenopus* oocytes at potentials greater than 20 mV, as reported by Quamme (1997). The degree of block (outward currents and tail currents) was not significantly different between the values measured at −40, −20, or 0 mV membrane potential, respectively. In contrast, the reduction of block at positive potentials (20, 40 mV) was significantly different from the latter values.

Chlorpromazine has no marked effect on HERG channel activation

The effect of chlorpromazine on the HERG current voltage ($I-V$) relation was investigated under isochronal recording conditions using the following protocol. Oocytes were clamped

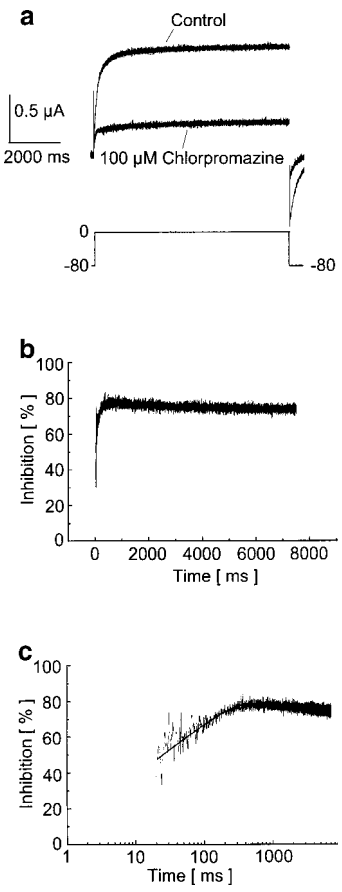

Figure 2 Chlorpromazine primarily blocks activated HERG channels. HERG currents were activated by a depolarizing voltage step to 0 mV (7500 ms) from a holding potential of −80 mV (a). After having recorded the control measurements, the channels were held at −80 mV in the closed state for 25 min during perfusion with the drug solution. The control recording and the first pulse measured after incubation with 100 μM chlorpromazine are displayed. (b, c) show the degree of inhibition in per cent, starting 20 ms after the beginning of the test pulse ((b), linear timescale; (c), logarithmic timescale). Inhibition of current increased time dependently to approximately 78% at 400 ms, indicating that open channels were blocked.

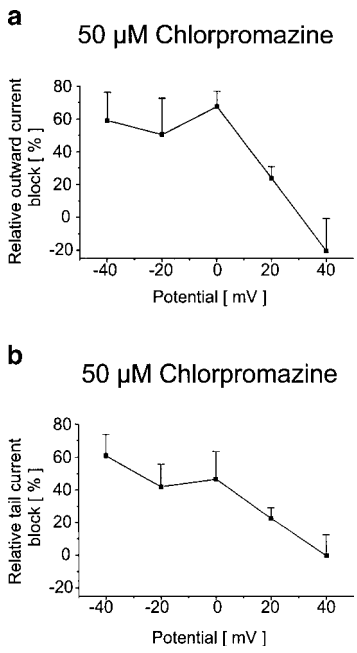

Figure 3 Voltage dependence of chlorpromazine block. Displayed are fractions of blocked outward currents measured at the end of the long depolarizing pulse (a) and peak tail currents (b) as a function of various test pulse potentials. HERG channel block was voltage dependent at potentials <40 mV, whereas at 40 mV HERG channel block by chlorpromazine was completely abolished (see text for details). Data are expressed as mean ± s.d., and $n = 3-5$ cells were studied at each potential. Voltage protocol: peak tail currents were measured during a repolarizing step to −120 mV (400 ms), following a test pulse to potentials ranging from −40 to 40 mV (35 s) to measure outward currents. Holding potential: −80 mV.

at a holding potential of −80 mV. Depolarizing pulses were applied for 2 s to voltages between −80 and +70 mV in 10 mV increments, and tail currents were recorded during a constant repolarizing step to −60 mV for 1.6 s. Families of current traces from one cell are shown for control conditions and after exposure to 25 μM chlorpromazine (25 min) in Figure 4a and b. The currents activated at potentials greater than −50 mV, reached a peak at −10 mV and then decreased at more positive potentials because of inactivation (Sanguinetti *et al*., 1995; Smith *et al*., 1996), giving the $I-V$ relation its typical bell-shaped appearance (Figure 4c). HERG currents at the end of the test pulse to 0 mV were reduced by 46.7 ± 7.4%. Figure 4d displays peak tail currents as a function of the preceding test pulse potential, resulting in activation curves. The peak tail current, measured during the second step of the voltage protocol, increased with voltage steps from −40 to +20 mV and then plateaued with test pulse potentials positive to +20 mV. HERG peak tail currents were reduced by 25 μM chlorpromazine by 44.2 ± 8.7% ($n = 5$). Chlorpromazine caused no significant change in the half-maximal activation voltage $V_{1/2}$ (from −15.7 ± 3.3 to −17.1 ± 4.3 mV; $n = 4$). The mean slope factor of the activation curve yielded 7.7 ± 1.1 under control conditions and 7.1 ± 1.1 after drug application ($n = 4$). This difference was not statistically significant.

Effects of chlorpromazine on HERG current inactivation

The effects of chlorpromazine on HERG current inactivation were investigated using two different approaches. First, it was tested whether the rate of inactivation was affected by the drug. Pulses were applied to 40 mV for 900 ms where channels are partially open but predominantly inactivated. A brief repolarization to −100 mV for 16 ms caused rapid recovery from inactivation without marked deactivation. During a second depolarizing pulse (150 ms) to different voltages ranging from −60 to 40 mV (increment 20 mV), large, rapidly inactivating currents were produced. The holding potential was −80 mV. Inactivating currents were recorded before (Figure 5a) and after equilibration of the block with 25 μM chlorpromazine (Figure 5b) by current monitoring after 25 min. Single-exponential fits to the large inactivating currents yielded the time constants of inactivation at different voltages. In these experiments, no pronounced changes in the time constants for HERG channel inactivation were observed (Figure 5c; $n = 6$).

In a second approach, we measured steady-state inactivation relations. Channels were inactivated at a holding potential of 20 mV, before being recovered from inactivation at various potentials from −120 to 30 mV (increment 10 mV) for 20 ms. Finally, the resulting peak outward currents at constant 20 mV

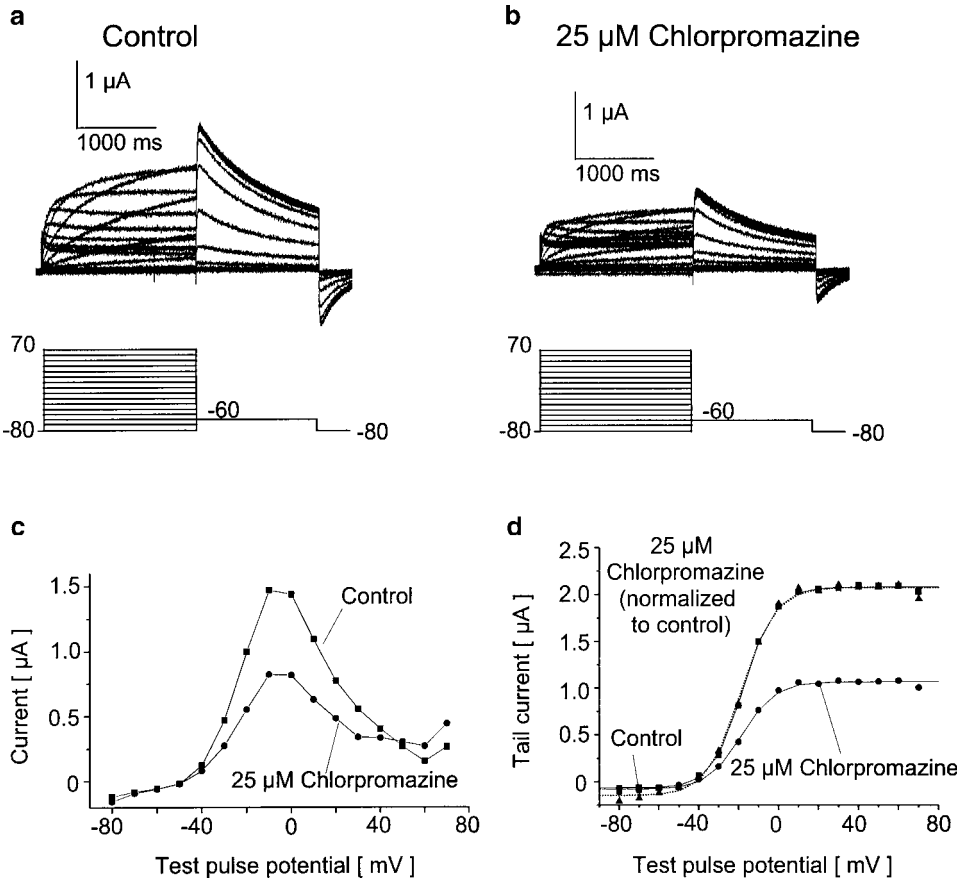

Figure 4 Chlorpromazine has no marked effect on HERG channel activation kinetics. Control measurement (a) and the inhibitory effects of 25 μM chlorpromazine (25 min; (b)) in one representative oocyte. (c) shows the resulting current amplitude at the end of the test pulse as a function of the preceding test pulse potential under control conditions and after incubation with 25 μM chlorpromazine. The maximum current amplitude at 0 mV is reduced in this measurement by 45.2%. The peak tail current amplitudes as a function of the preceding test pulse potentials during the first step of the voltage protocol, recorded under isochronal conditions, give the HERG activation curves (d). In addition, the activation curve in the presence of the drug normalized to control values is displayed (dashed line). Peak tail currents were reduced by 47.6%. No pronounced changes in the half-maximal activation potential $V_{1/2}$ were apparent ($\Delta V_{1/2} = -1.0$ mV). Voltage protocol in (a, b): holding potential −80 mV, test pulse −80 to 70 mV (2 s) in 10 mV increments, return pulse −60 mV (1.6 s).

as a measure of steady-state inactivation were recorded (Smith *et al*., 1996). After having obtained the control measurements (Figure 5d), 25 μM chlorpromazine were applied to the oocytes. The holding potential was −80 mV during the incubation period of 25 min to avoid destruction of the cell, as it would occur when holding the cell at 20 mV, and no pulsing was carried out. One typical recording in the presence of the drug is displayed in Figure 5e. The inactivating outward current amplitudes measured at 20 mV were normalized and plotted against the test pulse potentials, giving the steady-state inactivation curves (Figure 5f). Mean values for the half-maximal inactivation voltage yielded -72.7 ± 4.7 mV for control and -74.7 ± 6.2 mV for chlorpromazine measurements ($n = 4$), displaying only a small difference of -2.0 ± 6.2 mV. The mean slope factor of the inactivation curve was not significantly altered (-20.8 ± 3.8 under control conditions *versus* -23.1 ± 5.9 after drug application; $n = 4$).

Chlorpromazine block is reverse frequency dependent

The frequency dependence of chlorpromazine block was investigated after 25 μM chlorpromazine was allowed to wash into the bath for 5 min at −120 mV without pulsing. After this wash-in period, pulses were applied at intervals of 1 or 10 s for 20 min, with each cell studied only at one stimulation rate. HERG channels were rapidly activated by a depolarizing step to 20 mV for 300 ms followed by a repolarizing step to −40 mV (300 ms) to elicit outward tail currents, before returning to the holding potential of −80 mV. Four oocytes were used at each stimulation rate. The development of current reduction was plotted *versus* time (Figure 6). The resulting level of steady-state block is a measure for the frequency dependence of block. The amount of block after 25 min was significantly higher at lower stimulation rates. HERG current inhibition yielded $26.2 \pm 7.2\%$ at a rate of 1 Hz and $45.8 \pm 10.1\%$ at 0.1 Hz stimulation rate, respectively. Therefore, block was reverse frequency dependent.

Discussion

This study confirms that chlorpromazine is an inhibitor of HERG potassium channels, a finding in line with QTc prolongation observed among psychiatric patients treated with

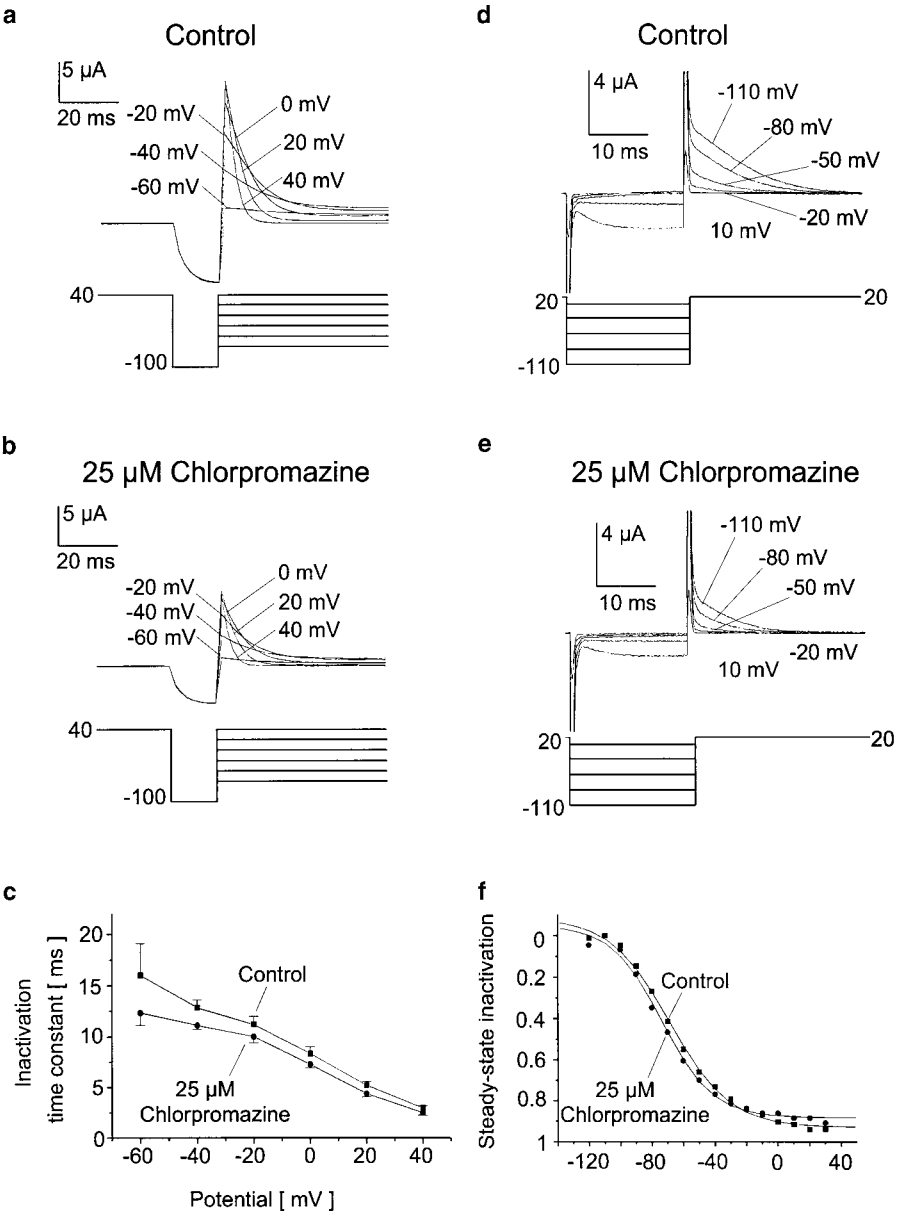

Figure 5 Effects of chlorpromazine on HERG current inactivation. The inactivation time constant was assessed from the third step of the following voltage protocol: currents were activated by 900-ms pulses to 40 mV, followed by a brief repolarization to −100 mV (16 ms). Variable voltage steps ranging from −60 to 40 mV (150 ms; increment 20 mV) were consecutively applied to evoke inactivating currents (a). The holding potential was −80 mV. Current measurements recorded before (a) and after incubation with 25 μM chlorpromazine (b) are displayed. (c) shows the corresponding inactivation time constants obtained from single-exponential fits to the inactivating current traces ($n=5$). (d, e) show measurements of the steady-state inactivation at constant 20 mV after various potentials from −120 to 30 mV (increment 10 mV). Note that, for clarity, not all current traces are displayed. The normalized inactivating current amplitude at 20 mV is shown in panel (f), giving the steady-state inactivation curve. There was a small shift of $V_{1/2}$ from −65.9 to −71.3 mV in this experiment. Error bars denote s.d.

this agent. Blockade of HERG channels heterologously expressed in *Xenopus laevis* oocytes displayed an IC_{50} value of 21.6 μM with a Hill coefficient of 1.11. Therapeutic plasma concentrations in humans have been reported to be 61.4–260 nM (Tokunaga *et al*., 1997), reaching up to 4.32 μM in cases of renal insufficiency (Dorson & Crismon, 1988). The IC_{50} values found in our oocyte experiments differ to some degree from the physiological IC_{50} values. Tie *et al*. (2000) reported that HERG channels expressed in Chinese Hamster Ovary cells were blocked by chlorpromazine with an IC_{50} value of 1.47 μM. Owing to the specific properties of the *Xenopus* oocyte expression system, higher concentrations of drugs are necessary when applied to the extracellular surface of whole oocytes under *in vitro* conditions. For example, the antiarrhythmic drug BRL-32872 blocked HERG channels expressed in *Xenopus* oocytes with an IC_{50} of 241 nM, approximately 12-fold higher than the IC_{50} value for mammalian HEK 293 cells (19.8 nM) (Thomas *et al*., 2001). One explanation for this observation is that the vitelline membrane and the yolk reduce the concentration of drugs at the cell membrane. Thus, it is reasonable to assume that HERG current inhibition by chlorpromazine should be of physiological relevance, since

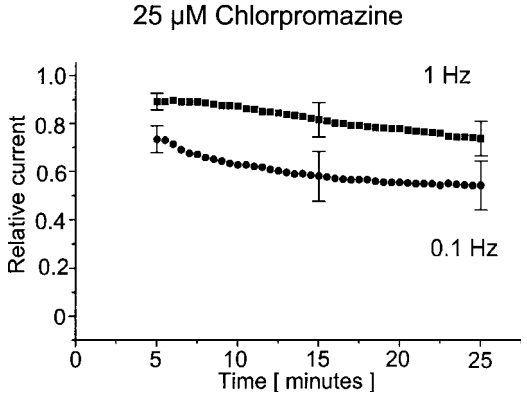

Figure 6 Chlorpromazine block is reverse frequency dependent. Trains of pulses were applied at intervals of 1 and 10 s under control conditions and in the presence of 25 μM chlorpromazine after an incubation period of 5 min at −120 mV without pulsing, until steady-state block was achieved. HERG channels were activated with a test pulse to 20 mV (300 ms) from a holding potential of −80 mV, and outward tail currents were recorded during a repolarizing step to −40 mV (300 ms), before returning to the holding potential. The resulting mean relative tail current amplitudes are plotted *versus* time ($n=4$ oocytes were studied at each rate). For the purpose of clear presentation, only six error bars were drawn in this figure. The amount of steady-state block was higher when pulses were applied at lower stimulation rates compared to higher rates, indicating that blockade by chlorpromazine is reverse frequency dependent.

the physiological IC_{50} value for chlorpromazine block of HERG in human cells is likely to be markedly lower than 21.6 μM.

The biophysical analysis of HERG channel block by chlorpromazine revealed that HERG currents are inhibited by the drug molecule in the closed and open states, whereas inactivated state block was less pronounced. Unblocking upon repolarization or washout occurred very slowly, and a complete washout could not be achieved. This may be because of a trapping mechanism of the drug at its binding site (Mitcheson *et al*., 2000). Chlorpromazine block of HERG is reverse frequency dependent, that is, the degree of inhibition is lower at higher stimulation rates. This is a common property of many inhibitors of I_{Kr} and the underlying HERG potassium channel. One possible explanation for the reverse frequency dependence is a different amount of closed-channel block that occurs during the time between the pulses (i.e., the time during which the channels are in the closed state). In addition, the pronounced block at negative membrane potentials (particularly at −40 mV; Figure 3) further supports the hypothesis that chlorpromazine binding to closed channels occurs. Under clinical conditions, reverse frequency dependence causes less block at higher heart rates than at lower heart rates. In contrast, only few drugs, such as amiodarone or BRL-32872, display frequency dependent blocking characteristics (Kiehn *et al*., 1999; Thomas *et al*., 2001), with the degree of inhibition being higher at higher stimulation rates. The action of these drugs is more pronounced at higher heart rates, which may account for the positive outcome of clinical trials with amiodarone (Naccarelli *et al*., 2000).

Channel inactivation at positive membrane potentials reduced HERG channel block by chlorpromazine. This observation could be explained by the following mechanism. The drug binding site is more likely to be accessible for chlorpromazine when the channel is in the open (in contrast to inactivated) state, similar to the block of HERG potassium channels by dofetilide (Kiehn *et al*., 1996; Ficker *et al*., 1998), BRL-32872 (Thomas *et al*., 2001) and fluoxetine (Thomas *et al*., 2002). This further supports the hypothesis that primarily open channels are blocked by chlorpromazine, although the voltage protocols do not clearly distinguish between open and inactivated states. Moreover, the following results illustrate that chlorpromazine preferentially binds to open channels. The degree of peak tail current block by 100 μM chlorpromazine after recovery from inactivation (which promotes the open state) is higher compared with the percentage of mean outward current inhibition at the end of the test pulse (which promotes the inactivated state) (86.8 *versus* 82.8%; Figure 1). This difference is even more pronounced when the degree of mean current inhibition at the end of the long depolarizing pulse in Figure 2 (72.0%) is compared with the reduction of peak tail currents in Figure 1.

The block of HERG currents by chlorpromazine probably underlies the QTc interval prolongation associated with chlorpromazine treatment. Compared with other antipsychotic drugs such as thioridazine, droperidol, haloperidol, or sertindole (Reilly *et al*., 2000; Haddad & Anderson, 2002), chlorpromazine seems to have less proarrhythmic potential. This may be attributed to the additional inhibition of L-type calcium channels by chlorpromazine (Lee *et al*., 1999), as suggested earlier for other HERG channel antagonists (Thomas *et al*., 2001; 2002). However, HERG channel blockade might be particularly important when chlorpromazine is prescribed in combination with other drugs that inhibit HERG potassium channels. In these cases, the additional inhibitory effects of chlorpromazine on HERG channels might lead to severe proarrhythmic events.

In conclusion, the present results highlight the significance of chlorpromazine block of HERG potassium channels for the proarrhythmic potential of this drug.

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