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Inhibition of the Activity of Human Lymphocyte Kv1.3 Potassium Channels by Resveratrol

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Abstract. The whole-cell patch-clamp technique was applied to study the modulatory effect of resveratrol on voltage-gated potassium channel Kv1.3 expressed in human lymphocytes. Results demonstrate that application of resveratrol in the concentration range 1–200 μM inhibited the channel activity in a concentration-dependent manner to about 18% of the control value. The half-blocking concentration of resveratrol was 40.9 μM, whereas the Hill coefficient was 1.05. The inhibition was time-dependent and slowly reversible. The inhibitory effect of resveratrol was correlated in time with a significant slowing of the current activation, whereas the inactivation rate remained unaffected upon application of resveratrol. The inhibition of Kv1.3 channels was voltage-independent. The steady-state activation of the currents remained unchanged upon resveratrol application. The magnitude of the inhibitory effect of resveratrol was not altered when resveratrol was coapplied with genistein. The possible mechanism of the inhibitory effect and its significance for biological activity of resveratrol are discussed.

Key words: Patch clamp — Potassium channel — T lymphocyte — Resveratrol — Cancer cell proliferation — Lymphocyte proliferation

Introduction

It has been known for some years that ion channels, and particularly potassium channels, participate in cancer development and apoptosis (Pardo, 2004; Wang, 2004, Felipe et al., 2006). Potassium channel inhibitors may inhibit cell proliferation in different cancer cell lines (Abdul & Hoosein, 2002; Rouzaire-Dubois & Dubois, 1990; Rouzaire-Dubois, Gerard & Dubois, 1993). It is also known that some inhibitors of multidrug resistance of cancer cells, e.g., verapamil and cyclosporin A, inhibit the activity of voltage-gated potassium channels (Panyi et al., 1996; Rybalchenko et al., 2001). Therefore, potassium channel inhibitors may be potentially useful drugs in cancer chemotherapy (Conti, 2004).

To the group of voltage-gated potassium channels, which participate in development and apoptosis of cancer cells in prostate, breast and colon cancer, belong also Kv1.3 channels (Abdul & Hoosein, 2002; Abdul, Santo & Hoosein, 2003). These channels are expressed in many different tissues, including human T lymphocytes (TLs) (Cahalan, Wulff & Chandy, 2001; Chandy et al., 2004). Activity of Kv1.3 channels is required for TL mitogenesis, and blockade of these channels inhibits cell mitogenesis in its early stage (Cahalan et al., 2001; Chandy et al., 2004). Therefore, Kv1.3 channels are considered molecular targets for immunosuppression (Chandy et al., 2004).

Resveratrol (3,4′,5– trihydroxystilbene) is a natural anticancer agent, which is present at the highest concentration in red grapes and wine (Fremont, 2000). Resveratrol is also involved in inhibition of lipid peroxidation, chelation of copper ions, scavenging of free radicals, alteration of eicosanoid synthesis, inhibition of platelet aggregation, anti-inflammation, vasorelaxation, estrogenic activity and cardioprotection (Fremont, 2000; Delmas et al., 2006). Therapeutic potential of resveratrol in these areas was confirmed by data obtained *in vivo* (Baur & Sinclair, 2006).

One of resveratrol's recently discovered areas of biological activity is the ability to modulate various types of potassium channels. There is evidence that application of resveratrol at micromolar concentrations increased the activity of large- and intermediate-conductance calcium-activated potassium channels in vascular endothelial cells (Li, Chen & Wu, 2000). Activation of these channels is associated with the peripheral antinociceptive effect of resveratrol (Grandos-Soto, Arguelles & Ortiz, 2002).

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The activating effect of resveratrol applied at micromolar concentrations was also observed in case of slowly activating, delayed rectifier, voltage-gated potassium channels expressed in guinea pig ventricular myocytes (Zhang et al., 2006). Stimulation of these channels combined with inhibition of L-type potassium channels by resveratrol is correlated with shortening of cardiac action potentials and antiarrhythmic efficacy (Zhang et al., 2006). In contrast, adenosine triphosphate-sensitive potassium channels (Grandos-Soto et al., 2002) and rapidly activating, delayed rectifier, voltage-gated potassium channels in guinea pig ventricular myocytes (Zhang et al., 2006) were both unaffected by resveratrol.

Moreover, resveratrol may also exert inhibitory effects on potassium channels. It was shown that *trans*-resveratrol applied at the concentration of 90 μM inhibited 28.9% of the rapidly inactivating, inward-rectifying potassium currents (I_{ERG}) and 42.9% of the delayed rectifier potassium currents (I_{DR}) in the F-11 neuroblastoma cell line (Orsini et al., 2004). Recently obtained data showed that application of *trans*-resveratrol at micromolar concentrations inhibited both the delayed rectifier (I_K) and fast-transient (I_A) currents in rat dissociated hippocampal neurons (Hu & Gao, 2005). The authors concluded that the resveratrol-induced inhibition of both I_K and I_A currents might be correlated with the neuroprotective effect exerted by resveratrol (Hu & Gao, 2005).

The aim of the present study was to investigate the influence of resveratrol on the activity of Kv1.3 channels. No data in this matter are yet available. Since Kv1.3 channels are expressed endogenously and predominantly in human TLs, these cells were used as a model system to study the effect of resveratrol on channel activity. The experiments showed that resveratrol inhibited the activity of Kv1.3 channels in a concentration- and time-dependent manner. The inhibitory effect of resveratrol on Kv1.3 channels was similar to the effect exerted by genistein. However, it was shown that the inhibitory effects of genistein and resveratrol occurred independently of each other.

Preliminary results of our study were published as an abstract (Michalak, Teisseyre & łania-Pietrzak, 2005).

Materials and Methods

Cell Separation, Solutions and Pipettes

Human TLs were separated from peripheral blood samples from 10 healthy donors using a standard method described elsewhere (Hirano et al., 1977). After separation, cells were cultured for at least 24 h in the standard RPMI-1640 medium (Sigma, St. Louis, MO) supplemented with 5% vol/vol horse serum (Sigma).

The external solution used in the experiments contained (in mM) 150 NaCl, 4.5 KCl, 1 $CaCl_2$, 1 $MgCl_2$ and 10 4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid (HEPES) (pH 7.35), adjusted with NaOH, 300 mOsm. The pipette solution contained (in mM) 150 KCl, 1 $CaCl_2$, 1 $MgCl_2$, 10 HEPES and 10 ethyleneglycoltetraacetic acid (EGTA) (pH 7.2), adjusted with KOH and osmomolarity, 300 mOsm. The concentration of free calcium in the internal solution was below 100 nM, assuming the dissociation constant for EGTA at pH 7.2 of 10^{-7} M (Grissmer, Nguyen & Cahalan, 1993). Such a low calcium concentration was applied in order to prevent the activation of calcium-activated IKCa1 channels (Grissmer et al., 1993). The reagents were provided by the Polish Chemical Company (Gliwiee, Poland), except for HEPES and EGTA, which were purchased from Sigma (St. Louis, MO). Resveratrol in the form of the *trans* isomer was also purchased from Sigma. Dishes with cells were placed under an inverted Olympus (Tokyo, Japan) IMT-2 microscope. External solutions containing resveratrol were applied using a perfusion system developed in our laboratory. Pipettes were pulled from borosilicate glass (Hilgenberg GmbH, Malsfeld, Germany) and fire-polished before the experiment. The pipette resistance was in the range 3–5 MΩ.

Electrophysiological Recordings

Whole-cell potassium currents in TLs were recorded, applying the patch-clamp technique (Hamill et al., 1981). The currents were recorded using an EPC-7 amplifier (Heka, Lambrecht, Germany), low pass-filtered at 3 kHz and digitized using the CED Micro 1401 (Cambridge Electronic Design, Cambridge, UK) analogue-to-digital converter with a sampling rate of 10 kHz. A standard depolarizing sequence contained 10 voltage ramps applied every 20 s, depolarizing the cell membrane from –100 to +40 m*V*, ramp duration 340 ms, holding potential –90 m*V*. The linear (ohmic) component of the current was subtracted off-line from the final record. For measurements of the steady-state activation and activation kinetics of the channels, another protocol containing seven voltage steps in the range–60 to +60 m*V* (20-m*V* increment) was applied every 10 s; pulse duration was 40 ms and holding potential –90 m*V*. For measurements of the time dependence of resveratrol-induced inhibition of the current amplitude and slowing of the activation kinetics, a different sequence was applied: seven depolarizing pulses of +60 m*V* each were applied every 30 s, pulse duration was 20 ms and holding potential was –90 m*V*. The influence of resveratrol application on the inactivation kinetics was investigated, applying a modified sequence containing seven depolarizing pulses of +60 m*V* each applied every 30 s, pulse duration was 1,000 ms and holding potential was –90 m*V*. The linear (ohmic) component of the current was subtracted on-line from the final records. The data were analyzed using the WCP J. Dempster program (J. Demster, Strathclyde, UK).

The data are given as the mean ± standard error. All experiments were carried out at room temperature (22–24°C).

Data Analysis

Since the values of current amplitudes varied significantly among the cell population, the magnitude of the inhibitory effect of resveratrol was presented in terms of the relative peak current recorded at +40 m*V* applying the voltage ramp protocol. The dose-response curve for resveratrol application was fitted applying the Hill equation: $y = a + (b - a)/(1 + [x/c]^d)$, where y is the fraction of blocked current, a is the minimal value of y, b is the maximal value of y, x is the logarithm of resveratrol concentration, c is the logarithm of the 50% inhibitory concentration (IC_{50}) value and d is the Hill coefficient. The steady-state activation of the channels was presented in terms of a normalized relative chord conductance

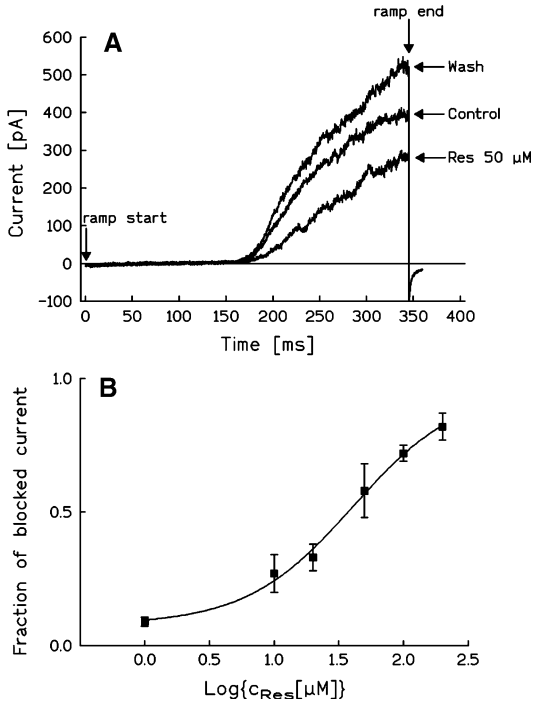

Fig. 1. (*A*) Effects of application of resveratrol on whole-cell potassium currents recorded, applying the ramp current protocol (see Materials and Methods) under control conditions (*middle trace*), upon application of 50 μM resveratrol for 2 min (*lower trace*) and after washout of the drug for 1 min (*upper trace*). The start and end of application of each voltage ramp are indicated by *vertical arrows*. (*B*) The fraction of blocked current as a function of the logarithm of resveratrol concentration. The data were fitted applying the Hill equation (*see* Materials and Methods).

(gK_{norm}) defined by the equation $gK_{norm} = gK/gK_{60}$, where gK is chord conductance and gK_{60} is chord conductance at the membrane potential of +60 mV. Chord conductance was calculated according to the definition $gK = I_p/(V - V_{rev})$, where I_p is the amplitude of the current, V is the membrane potential and V_{rev} is the reversal potential of the current, which is –75 mV. The voltage dependence of steady-state activation was fitted by a Boltzmann function given by the equation $gK_{norm}(V) = 1/[1 + \exp\text{-}(V - V_{1/2})/k_n]$, where V_n is the activation midpoint and k_n is the steepness of the voltage dependence. The activation kinetics was fitted by applying a power function, given by the equation $I(t) = I_p[1 - \exp(-t/\tau_n)]^2$, where τ_n is the time constant of activation.

Results

Whole-cell potassium currents recorded in TLs by application of the voltage ramp protocol are depicted in Figure 1A. The middle trace presents the currents recorded under control conditions, the lower trace shows the current recorded upon application of 50 μM resveratrol and the upper trace depicts the current recorded after the drug had been washed out. In each case, application of a voltage ramp (ramp start and ramp end are marked by the vertical arrows) evoked a current, which contained two components: a linear nonspecific leak current subtracted during the off-line analysis followed by a nonlinear current activated upon membrane depolarization. Results of our experiments performed previously demonstrated that the nonlinear component was predominantly due to activation of Kv1.3 channels (Teisseyre & Mozrzymas, 2002). Apparently, the Kv1.3 component of the currents was significantly reduced in the presence of resveratrol (Fig. 1A). Washout of resveratrol caused a recovery of the Kv1.3 component, and the value of the peak current was even higher than before application of resveratrol (Fig. 1A). In fact, the inhibitory effect of resveratrol on the channels was time-dependent and slowly reversible (*see below*).

The dose-response curve for resveratrol-induced inhibition of Kv1.3 currents is presented in the form of a semilogarithmic plot in Figure 1B. The magnitude of the blocking effect was strongly concentration-dependent in the range 1–100 μM. Application of 1 μM resveratrol blocked only about 5% of the currents, whereas the currents were inhibited by about 70% at 100 μM concentration. The calculated IC_{50} value was 40.9 ± 5.0 μM, whereas the Hill coefficient was 1.05 ± 0.27. Raising the resveratrol concentration from 100 to 200 μM decreased the current amplitude to about 18% of the control value (Fig. 1B). Thus, the inhibitory effect of resveratrol on Kv1.3 channels was not complete at concentrations below 200 μM.

The steady-state activation curve of Kv1.3 currents recorded under control conditions and in the presence of 20 μM resveratrol is plotted in Figure 2A. The activation midpoint ($V_{1/2}$) parameter values calculated for currents recorded under control conditions and in the presence of 20 μM resveratrol were –22.6 ± 1.2 mV and –24.1 ± 1.3 mV (n = 8), respectively. These values were not statistically different from each other ($P > 0.05$, Student's t-test). The steepness of the voltage dependence (k_n) parameter values were 4.4 ± 1.8 mV and 7.0 ± 1.2mV (n = 8), respectively. Also, these values were not statistically different from each other ($P > 0.05$, Student's t-test). Lack of influence on the steady-state activation of the currents was also observed in case of application of 50 μM resveratrol (*not shown*). Figure 2B depicts the relationship between the relative peak current (I_{Res}/I_{Ctrl}) and the membrane potential for resveratrol applied at concentrations of 20 and 50 μM. The value of the relative peak current did not depend markedly on the membrane potential for any concentration tested. This may suggest that the inhibitory effect of resveratrol on the currents was voltage-independent.

Figure 3A depicts the time course of normalized potassium currents recorded, applying a voltage step from the holding potential of –90 to +60 mV (see Materials and Methods). It can be observed that the currents recorded in the presence of resveratrol exhibited much slower activation than the currents

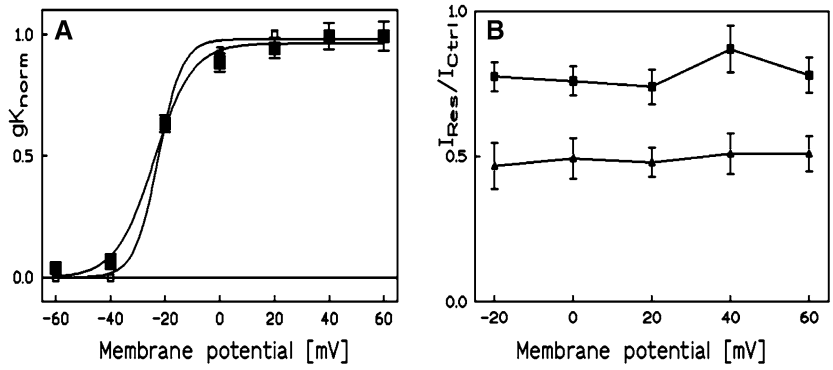

Fig. 2. (*A*) Steady-state activation in terms of a normalized chord conductance (gK_{norm}) as a function of membrane potential: *open squares*, control conditions ($n = 8$), *filled squares*, application of 20 μM resveratrol ($n = 8$). Data points were fitted by the Boltzmann function (*see* Materials and Methods). (*B*) Current amplitude ratio (I_{Res}/I_{Ctrl}) as a function of membrane potential upon application of 20 μM ($n = 8$) and 50 μM of resveratrol ($n = 11$). Data points in *B* were connected by a point-to-point line.

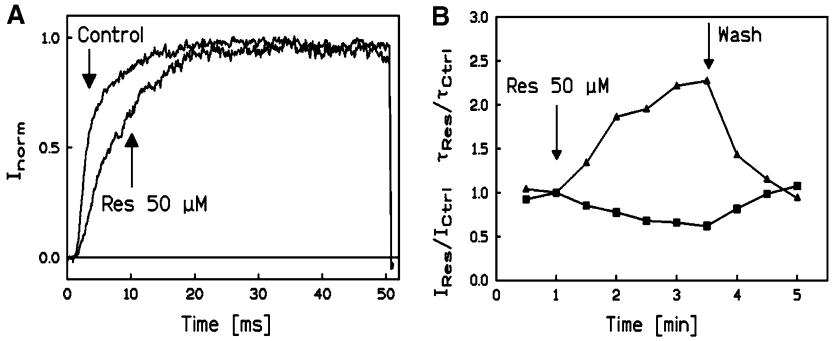

Fig. 3. (*A*) Normalized currents recorded, applying a voltage step at +60 m*V* (*see* Materials and Methods) under control conditions and in the presence of 50 μM resveratrol. (*B*) Current amplitude ratio (I_{Res}/I_{Ctrl}) at +60 m*V* (*filled squares*) and time constant ratio (τ_{Res}/τ_{Ctrl}) at +60 m*V* (*filled triangles*) as a function of time. After 1 min, 50 μM resveratrol was applied; after 3.5 min, resveratrol was washed out.

recorded under control conditions. Therefore, it was of interest to study whether the slowing of activation was correlated in time with the resveratrol-induced inhibition of the currents. For this purpose, currents evoked by voltage steps from the holding potential of –90 to +60 m*V* were recorded upon application and washout of resveratrol. Then, the relative peak currents (I_{Res}/I_{Ctrl}) and the activation time constant ratios (τ_{Res}/τ_{Ctrl}) were calculated. Figure 3B presents the relative peak currents and the activation time constant ratios as a function of time. The current inhibition by resveratrol was clearly correlated in time with the slowing of activation. Moreover, both the current inhibition upon application of resveratrol and the recovery after washout of the drug were time-dependent. The inhibition of the currents was, in fact, much slower than the recovery from the inhibition (Fig. 3B).

Since the activation kinetics of Kv1.3 channels is strongly voltage-dependent (Teisseyre & Michalak, 2005), activation time constants (τ_n) were calculated at different membrane potentials for currents recorded under control conditions and in the presence of resveratrol applied at the concentrations of 20 and 50 μM. The results shown in Figure 4A demonstrate that the time constants at all the applied potentials were significantly higher in the presence of resveratrol than under control conditions. The effect of resveratrol on the activation kinetics was clearly concentration-dependent. Figure 4B shows the time constant ratios (τ_{Res}/τ_{Ctrl}) as a function of the membrane potential. The value of the time constant ratios increased slightly with the membrane potential for both concentrations applied; however, this increase was statistically insignificant ($P > 0.05$, one-way analysis of variance). Thus, the slowing of activation upon resveratrol application was voltage-independent.

The influence of application of resveratrol on the inactivation kinetics of Kv1.3 currents was also investigated, applying the sequence containing long depolarizing voltage pulses (see Materials and Methods). Our results provide evidence that the inactivation kinetics remained unaffected upon application of resveratrol (*data not shown*).

The inhibitory effect of resveratrol on Kv1.3 channels reported in this study is similar to the inhibitory effect of genistein published recently (Teisseyre & Michalak, 2005). It might therefore be possible that resveratrol and genistein affect the channels by the same mechanism or that both substances share a common binding site on the channels. In such a case, the inhibitory effect of resveratrol should have been diminished or been abolished in the presence of genistein. To verify this hypothesis, experiments with resveratrol applied in the presence of 50 μM genistein were performed. The degree of current inhibition by 50 μM resveratrol in the presence of 50 μM genistein was calculated by dividing the relative peak current re-

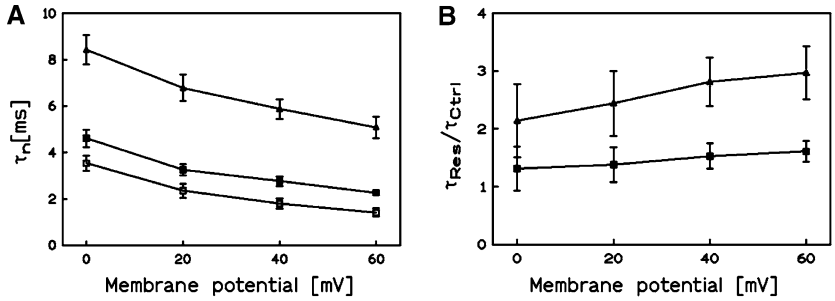

Fig. 4. (*A*) Activation time constants as a function of membrane potential: *open squares*, control conditions ($n = 11$); *filled squares*, application of 20 μM resveratrol ($n = 8$); *filled triangles*, application of 50 μM resveratrol ($n = 11$). (*B*) Time constant ratio (τ_{Res}/τ_{Ctrl}) as a function of membrane potential upon application of 20 μM (*filled squares*, $n = 8$) and 50 μM (*filled triangles*, $n = 11$) of resveratrol. Data points in *A* and *B* were connected by a point-to-point line.

corded upon coapplication of genistein and resveratrol by the relative peak current obtained in the presence of genistein only. The relative peak current recorded upon application of 50 μM resveratrol in the presence of the same concentration of genistein was 0.44 ± 0.05 ($n = 5$) of the control value (*not shown*). This value was almost the same as the relative peak current recorded when 50 μM of resveratrol was applied without genistein (0.42 ± 0.05, $n = 5$, *not shown*).Thus, the inhibitory effect of resveratrol on Kv1.3 channels was genistein-independent. This suggests that genistein and resveratrol probably inhibit the channels by different mechanisms and act on different binding sites.

Discussion

Results of this study demonstrated that resveratrol inhibited the activity of Kv1.3 channels in human lymphocytes in a concentration- and time-dependent manner. The inhibitory effect was correlated in time with a significant slowing of the activation of the currents. The channel inhibition was voltage-independent.

As mentioned above, the inhibitory effect of resveratrol on Kv1.3 channels was similar to the effect exerted by genistein (Teisseyre & Michalak, 2005). According to the data, genistein inhibited the activity of lymphocyte Kv1.3 channels in a concentration- and time-dependent manner. The inhibitory effect of genistein was voltage-independent. It was correlated in time with a significant slowing of the channel activation, whereas the inactivation kinetics remained unaffected (Teisseyre & Michalak, 2005). However, the inhibitory effects of resveratrol remained unchanged in the presence of genistein. This may suggest that the inhibitory effects of both substances occurred independently of each other and that the two compounds interacted with different binding sites.

Little is known about the mechanisms of the inhibitory effects of resveratrol on voltage-gated potassium channels. It was shown that resveratrol inhibited both delayed rectifier (I_K) and fast-transient (I_A) potassium currents in rat dissociated hippocampal neurons (Hu & Gao, 2005). In I_K currents, the IC_{50} value was 13.6 ± 1.0 μM; this was much lower than the 40.9 ± 5.0 μM reported in this study for Kv1.3 channels. In I_A currents, the IC_{50} was 45.7 ± 7.5 μM; this was not markedly different from the value obtained in our experiments for Kv1.3 channels. However, the inhibition of I_A currents by resveratrol was clearly voltage-dependent and correlated with a shift of the steady-state activation curve by about 16 m*V* toward positive membrane potentials (Hu & Gao, 2005). In contrast, our results provide evidence that the inhibitory effect of resveratrol on Kv1.3 channels was voltage-independent and that there was no shift of the steady-state activation curve toward positive membrane potentials. Moreover, the value of the Hill coefficient in I_A currents was 1.50 ± 0.40. This may suggest that two molecules of resveratrol had to bind to the channel protein to promote inhibition (Hu & Gao, 2005). In our study, the Hill coefficient value was 1.05 ± 0.27, and this might indicate that only one resveratrol molecule was needed to inhibit Kv1.3 channels. Finally, there was no evidence that application of resveratrol slowed the activation of voltage-gated potassium channels in rat hippocampal neurons (Hu & Gao, 2005). Thus, the mechanism of resveratrol-induced inhibition of voltage-gated potassium channels expressed in lymphocytes and in rat hippocampal neurons was probably different.

The time dependence of the effect of resveratrol on Kv1.3 channels may suggest that signal-transduction pathways were involved in the inhibition. Results obtained in rat primary hippocampal neurons showed that application of 20 μM of resveratrol caused activation of protein kinase C (PKC) (Han et al., 2004). It is known that PKC-induced

phosphorylation influences the activity of Kv1.3 channels in TLs (Chung & Schlichter, 1997). It might, therefore, be possible that the inhibitory effect of resveratrol on Kv1.3 channels was due to activation of PKC and channel phosphorylation. However, this is unlikely for several reasons. First, it was shown that activation of PKC enhanced the activity of Kv1.3 channels (Chung & Schlichter, 1997). In contrast, our study demonstrated that channel activity was inhibited upon application of resveratrol. Moreover, the PKC-induced activation of channels occurred after 30 min of cell preincubation with the PKC activator phorbol ester (Chung & Schlichter, 1997). The experiments performed with resveratrol applied in rat hippocampal neurons demonstrated that the PKC-induced phosphorylation occurred 30 min after the application (Han et al., 2004). Our experiments showed that the inhibitory effect of resveratrol on Kv1.3 channels reached its maximum 2–3 min after application. It was one order of magnitude faster than resveratrol-induced phosphorylation by PKC. Thus, the inhibition of the activity of Kv1.3 channels by resveratrol observed in our study was not due to activation of PKC. Nevertheless, the time dependence seems to be a characteristic feature of the inhibition of Kv1.3 channels by resveratrol. A similar time dependence was observed in resveratrol-induced inhibition of voltage-gated potassium channels in rat hippocampal neurons (Hu & Gao, 2005). In both cases, the mechanism of the inhibitory effect of resveratrol on the channels requires more study to be elucidated.

The inhibitory effect of resveratrol on Kv1.3 channels might be involved in the biological activities of resveratrol observed *in vivo*. It is known that the peak serum and plasma concentrations of resveratrol and its metabolic derivatives after oral dosing in rat and mouse may reach 100 or even 300 μM (Baur & Sinclair, 2006). Taking into account the data presented in this study, such a concentration is high enough to inhibit most Kv1.3 channels. It is known that activity of Kv1.3 channels is required for TL mitogenesis and that blockade of these channels inhibits cell mitogenesis in its early stage (Cahalan et al., 2001; Chandy et al., 2004). Therefore, it can be suggested that resveratrol might inhibit TL mitogenesis. More studies are necessary to verify this hypothesis. It is also known that Kv1.3 channels are present in many different tissues including cancer cells and that they participate in cancer development as was shown for prostate, colon and breast cancers (Abdul & Hoosein, 2002; Abdul et al., 2003). Moreover, it is known that inhibition of Kv1.3 channels may inhibit the proliferation of cancer cell lines (Abdul & Hoosein, 2002). Thus, inhibition of Kv1.3 channels may also contribute to the anticancer activity of resveratrol in prostate, breast and colon cancers. In order to verify such a hypothesis, more studies with Kv1.3 channels in cancer cell lines will have to be done.

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References

Abdul, M., Hoosein, N. 2002. Expression and activity of potassium ion channels in human prostate cancer. *Cancer Lett*. **186:**99–105

Abdul, M., Santo, A., Hoosein, N. 2003. Activity of potassium channel-blockers in breast cancer. *Anticancer Res*. **23:**3347–3351

Baur, J.A., Sinclair, D.A. 2006. Therapeutic potential of resveratrol: The *in vivo* evidence. *Nat. Rev. Drug Discov*. **5:**493–506

Cahalan, M., Wulff, H., Chandy, K.G. 2001. Molecular properties and physiological roles of ion channels in the immune system. *J. Clin. Immunol*. **21:**235–252

Chandy, K.G., Wulff, H., Beeton, C., Pennington, M., Gutman, G., Cahalan, M. 2004. K^+ channel as targets for specific immunomodulation. *Trends Pharmacol. Sci*. **25:**280–289

Chung, I., Schlichter, L. 1997. Native Kv1.3 channels are upregulated by protein kinase C. *J. Membr. Biol*. **156:**73–85

Conti, M.J. 2004. Targeting K^+ channels for cancer therapy. *Exp. Ther. Oncol*. **4:**161–166

Delmas, D., Lancon, A., Colin, D., Jannin, B., Latruffe, N. 2006. Resveratrol as a chemopreventive agent: A promising molecule for fighting cancer. *Curr. Drug Targets* **7:**423–442

Felipe, A., Vincente, R., Villalonga, N., Roura-Ferrer, M., Martinez-Marmol, R., Sole, L., Ferres, J., Condom, E. 2006. Potassium channels: New targets in cancer therapy. *Cancer Detect. Prev*. **30:**375–385

Fremont, L. 2000. Minireview. Biological effects of resveratrol. *Life Sci*. **66:**663–673

Grandos-Soto, V., Arguelles, C.F., Ortiz, M.I. 2002. The peripheral antinociceptive effect of resveratrol is associated with activation of potassium channels. *Neuropharmacology* **43:**917–923

Grissmer, S., Nguyen, A., Cahalan, M. 1993. Calcium-activated potassium channels in resting and activated human T lymphocytes. *J. Gen. Physiol*. **102:**601–630

Hamill, O.P., Marty, A., Neher, E., Sakmann, B., Sigworth, F. 1981. Improved patch-clamp techniques for high-resolution current recording from cells and cell-free membrane patches. *Pfluegers Arch*. **39:**85–100

Han, Y., Zheng, W., Bastianetto, S., Chabot, J., Quirion, R. 2004. Neuroprotective effects of resveratrol against β-amyloid-induced neurotoxicity in rat hippocampal neurons: Involvement of protein kinase C. *Br. J. Pharmacol*. **141:**997–1005

Hirano, T., Kuritani, T., Kishimoto, Y., Yamamura, Y. 1977. T cell dependency of PWM-induced Ig production by B cells. *J. Immunol*. **119:**1235

Hu, G., Gao, Z. 2005. *Trans*-resveratrol, a red wine ingredient, inhibits voltage-activated potassium currents in rat hippocampal neurons. *Brain Res*. **1056:**68–75

Li, H., Chen, S., Wu, S. 2000. Evidence for the stimulatory effect of resveratrol on Ca^{2+} activated K^+ current in vascular endothelial cells. *Cardiovasc. Res*. **45:**1035–1045

Michalak, K., Teisseyre, A., Łania-Pietrzak, B. 2005. Inhibition of multidrug resistance-associated protein MRP and Kv channels by natural polyphenols. *Eur. Biophys. J./Biophys. Lett*. **34:**805 abstract P-875

Orsini, F., Verotta, L., Lecchi, M., Restano, R., Curia, G., Redaelli, E., Wanke, E. 2004. Resveratrol derivatives and their role as potassium channel modulators. *J. Nat. Prod*. **67:**421–426

Panyi, G., Gaspar, R., Krasznai, J., Ameloot, M., Aszalos, A., Steels, P., Damjanovich, S. 1996. Immunosuppressors inhibit voltage-gated potassium channels in human peripheral blood lymphocytes. *Biochem. Biophys. Res. Commun.* **221:**254–258

Pardo, LA. 2004. Voltage-gated potassium channels in cell proliferation. *Physiology (Bethesda)* **19:**285–292

Rouzaire-Dubois, B., Dubois, JM. 1990. Tamoxifen blocks both proliferation and voltage-dependent K channels of neuroblastoma cells. *Cell. Signal.* **2:**387–393

Rouzaire-Dubois, B., Gerard, V., Dubois, J.M. 1993. Involvement of K channels in quercetin-induced inhibition of neuroblastoma cell growth. *Pfluegers Arch.* **423:**202–205

Rybalchenko, V., Prevarskaya, N., van Coppenolle, F., Legrand, G., Lemonnier, L., Bourhis, X., Skryma, R. 2001. Verapamil inhibits proliferation of LNCaP human prostate cancer cells influencing K^+ channel gating. *Mol. Pharmacol.* **59:**1376–1387

Teisseyre, A., Michalak, K. 2005. Genistein inhibits the activity of Kv1.3 potassium channels in human T lymphocytes. *J. Membr. Biol.* **205:**71–79

Teisseyre, A., Mozrzymas, J.W. 2002. Inhibition of the activity of T lymphocyte Kv1.3 channels by extracellular zinc. *Biochem. Pharmacol.* **64:**595–607

Wang, Z. 2004. Roles of K^+ channels in regulating tumour cell proliferation and apoptosis. *Pfluegers Arch.* **448:**274–286

Zhang, Y., Liu, Y., Wang, T., Li, B., Li, H., Wang, Z., Baofeng, Y. 2006. Resveratrol, a natural ingredient of grape skin: Antiarrythmic efficacy and ionic mechanisms. *Biochem. Biophys. Res. Commun.* **340:**1192–1199