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Aristolochic acid, a plant extract used in the treatment of pain, is a regulator of K2P channels.

Aristolochic acid (AristA) is found in plants used in traditional medicines to treat pain(1). A role for K2P channels, in particular TREK and TRESK channels, as therapeutic targets for the treatment of pain has been proposed(2). The aim of this studywas to investigate the action of AristA on these channels to determine whether this may contribute to its apparent therapeutic usefulness in the treatment of pain.

Currents through wild-type and mutated human K2P channels expressed in tsA201 cells were measured using whole-cell patch-clamp recordings in the presence and absence of AristA.Application of AristA resulted in an enhancement of current through TREK1 and TREK2 channels of 26 ± 6 % (mean ± SEM, n = 6) and 44 ± 11 % (n = 6), respectively. Enhancement of both currents was rapid and easily reversible. There was some voltage-dependence to the enhancement for both currents. By contrast, AristA (100 μM) had little effect on current through TRAAK channels with an enhancement of 8 ± 5 % (n = 5), or on the short form of TREK1 channels (TREK1_ΔN), with an enhancement of -4 ± 5 % (n = 5).

In contrast to the enhancement seen at high concentrations for TREK1 and TREK2 channels, we observed that AristA was an*inhibitor* of TRESK channelswith a calculated 50% effective concentration of 13 μM for AristA on TRESK (95% confidence intervals 11 – 18 μM) and a Hill slope of 0.56 (95% confidence intervals 0.43 – 0.65). As for enhancement of TREK channels, the effect of AristA was voltage-dependent. Altering the internal calcium buffer concentration had nosignificant effect on inhibition of TRESK by AristA (100 μM) with 73 ± 4 % (n = 6)inhibition in 0.1 mM EGTA and 79 ± 2 % (n = 6) inhibition in5 mM EGTA.Furthermore, inhibition of TRESK did not depend on the phosphorylation of key intracellular serine residues(3).S252A_S264A mutated TRESK channels had a significantly larger current density than WT TRESK channels (48 ± 3 pA/pF, n = 31 versus 27 ± 2 pA/pF, n = 28, $p < 0.05$, Student t-test), however, AristA (100 μM) still inhibited channel current by 80 ± 2% (n = 6). For S252E_S264E channels, current density was not significantly different to wild-type channels (33 ± 3 pA/pF, n = 39). AristA (100 μM) was, again, able to inhibit current through these channels by 73 ± 5 % (n = 5). On the other hand, mutations of bulky residuesin the M2 and M4 inner pore regions (F145A_F352A) completely abolished AristA inhibition of TRESK (0 ± 5 %, n = 7).

Enhancement of both TREK1 and TREK2 channel activity by AristAmay contribute to a therapeutically useful effect of this compound in pain and may help to explain the persistent use of plant extracts containing this compound in herbal remedies for pain (1). Hydroxy-α-sanshool, a primary active ingredient of Szechuan peppers, has been shown to block TRESK channels and this action has been proposed to underlie the distinctive numbing effect induced by this natural, widely-used analgesic(4).Since AristA is a potent inhibitor of TRESK channels, it might be predicted to act in a similar manner to hydroxy-α-sanshool to produce analgesia.

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