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A mutation of TASK2 K2P channels (T108P), found in certain patients predisposed to Balkan Endemic Nephropathy, reduces TASK2 current density and alters ion selectivity.

Balkan Endemic Nephropathy (BEN) is a renal disease restricted to rural areas of the Balkans and characterised by insidious onset, chronic renal failure and strong association with urothelial carcinoma of the upper urinary tract(1). It is widely believed that BEN is caused by low-level aristolochic acid (AristA) exposure, probably from contamination of wheat flour seeds by *Aristolochiaclematitis* (1). A number of patients predisposed to BEN have been shown to carry a mutation of the physiological important renal(2) two pore domain potassium (K2P) channel, TASK2(3). In this study, we describe how the mutation of TASK2 channels seen in patients susceptible to BEN (T108P), influenced the functional properties of TASK2 channels. We also determined whether AristA altered the activity of TASK2 channels and thus whether this may contribute to the putative action of AristAin BEN.

Currents through wild-type (WT) and mutated human TASK2 channels expressed in tsA201 cells were measured using whole-cell patch-clamp recordings. In control conditions, current density was 52 ± 4 pA/pF (mean ± SEM, n = 27) for WT TASK2, 9 ± 1 pA/pF (n = 15) for the conservative mutant, TASK2_T108C and just 2 ± 1 pA/pF (n = 26) for TASK2_T108P channels. Furthermore, unlike WT TASK2 channels, the reversal potentials of TASK2_T108P channels measured in different external [K] were not consistent with the presence of a K-selective current. For example, in 2.5 mM external K, reversal of current for WT TASK2 was -86 ± 1 mV (n = 27) but for TASK2_T108P this was -43 ± 3 mV (n = 26). It is of interest that the TASK2_T108C channel also showed a reduced K selectivity with a reversal potential in 2.5 mM external K of -70 ± 3 mV (n = 15). Altering external pH to 8.4 (from 7.4) resulted in a 120 ± 17 % (n = 9) enhancement for WT TASK2 and a 143 ± 22 % (n = 9) enhancement for TASK2_T108C channels. In contrast, pH 8.4 had little effect on TASK2_T108P channels with 10 ± 12 % (n = 7) enhancement of current. Similarly, application of flufenamic acid (FFA,100 μM), enhanced WT TASK2 current by 80 ± 31 % (n = 7) and produced a 97 ± 25 % (n = 6) enhancement of TASK2_T108C channels. However, FFA (100 μM) produced no enhancement of TASK2_T108P channels with 15 ± 14 % (n = 9) *inhibition* of current. At a concentration of 300 μM, AristA was able to induce a very modest enhancement of WT TASK2 channels (21 ± 13%, n = 5). At this high concentration, AristA also produced a very small enhancement of TASK2_T108P channels (12 ± 6 %, n = 5). AristA had no detectable effect on these channels at lower concentrations.

TASK2 channels have an important role in renal ion transport (2). Poorly or non-functioning TASK2 channels in patients carrying the T108P mutation would be anticipated to result in a reduced capacity for HCO_3^- transport and compromised renal function. AristA has little effect on either WT TASK2 channels or on TASK2_T108P channels except to induce a modest enhancement of current at a high concentration (300 μM). This suggests that AristA is unlikely to interact directly with TASK2 channels in contributing to its action in BEN. Rather, loss of functional TASK2 channels in BENmay indirectly increase susceptibility to AristA toxicity (and thus to BEN) acting through independent molecular pathways.

1. Grollman AP *et al.* (2007). *Proc Natl AcadSci U S A.***104:** 12129-12134.

2. Sepúlveda FV *et al.* (2015).*Physiol Rev.***95:** 179-217.

3. Toncheva D *et al.* (2014). *Biomed Res Int.* **2014:** 920723.