The role of the M1-P1 loop in acid sensitive two-pore domain potassium (TASK) channel regulation

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Background potassium channels control the resting membrane potential of many cells and regulate their excitability. Two-pore-domain potassium (2PK) channels have been shown to underlie a number of such background currents. Although often classed as "leak" channels, currents through 2PK channels are tightly regulated. For example, the acid sensitive 2PK (TASK) channels are inhibited by extracellular acidification with pKa's for TASK-1, TASK-2 and TASK-3 ranging from 6.5 to 8.5. TASK channels show relatively high homology in their transmembrane domains, but very little homology in extracellular domains, most notably in the ~50 residue linker between the first transmembrane domain and the first pore domain (the M1P1 loop). The M1P1 loop has previously been shown to be involved in the differential sensitivity of TASK channels to block by zinc (Clarke *et al.*, 2004). The aim of the present study was to test the hypothesis that the M1P1 loop contributes to the differential pH sensitivity of TASK channels. Chimaeric TASK channels were constructed by swapping M1P1 loops between family members and expressed in Chinese Hamster Ovary cells. Channels were characterised using standard whole cell patch clamp techniques. The chimaera formed from TASK-3 with the TASK-1 M1-P1 loop (T3/T1-M1P1) had a pK_a for inhibition that was much more similar to that of TASK-1 than TASK-3 (T1/T3 M1P1: $pK_a = 7.0 \pm 0.05$, TASK-1: $pK_a = 7.1 \pm 0.04$; TASK-3, $pK_a = 6.7 \pm 0.07$). This indicates that the M1P1 loop contributes to the differential pH sensitivity of TASK-1 and TASK-3 channels. All other chimaeras, however, were non-functional. This further suggests that the M1P1 loops are important for function and/or structure. However, further work needs to be done to assess whether these non-functional chimaeras are reaching the membrane and whether function can be restored through re-addition of further regions of the channel, such as the P-loop.

Clarke, C.E., Veale, E.L., Green, P.J., Meadows, H.J. & Mathie, A. (2004) *Journal of Physiology* **560**, 51-62.