Supporting Information for

Short Communication

Inhibition of temperature-sensitive TRPV3 channel by two natural isochlorogenic acid isomers for alleviation of dermatitis and chronic pruritus

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Table S1 The docking scores of IAA and IAB in mTRPV3.

Pocket	Pocket description	Glide score of IAA	Glide score of IAB
A	CBD binding to TRPV2 pocket and A967079 binding to TRPA1 pocket	–8.9	–9.1
B	Capsaicin binding to TRPV1 pocket	–7.3	–6.5
C	2-APB binding to TRPV3 pocket	–7.2	–5.2

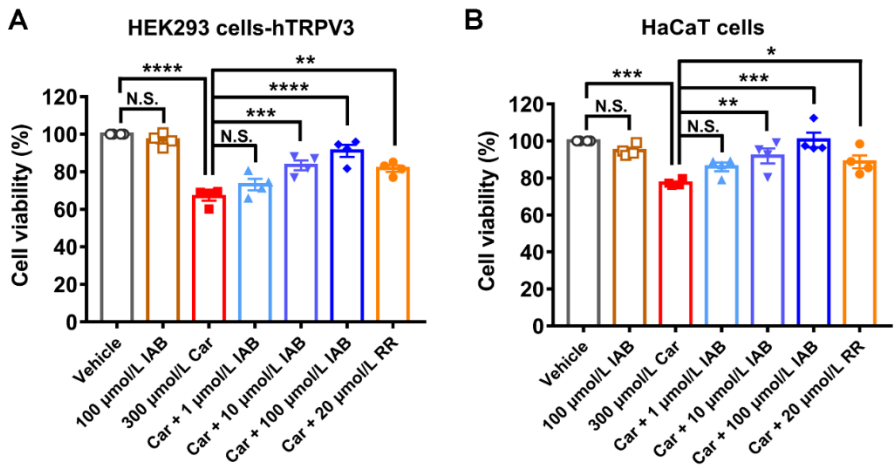

Figure S1 IAB reversed the proliferation inhibition of HEK293 cells expressing TRPV3 or HaCaT cells induced by overactiving TRPV3. Cell viability of HEK293 cells expressing hTRPV3 (A) or HaCaT cells (B) after treatment with different TRPV3 modulators for 24 h in MTT assay ($n = 4$; *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$, ****$P < 0.0001$; N.S., no significance; by one-way ANOVA, followed by the Dunnet's test). Data are presented as the mean ± SEM.

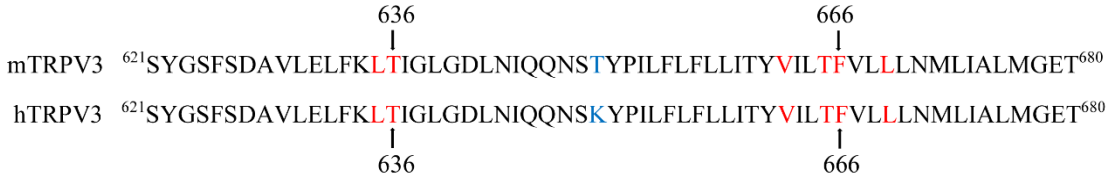

Figure S2 Residue sequence alignment of pore helix and upper portion of S6 between mouse TRPV3 and human TRPV3.

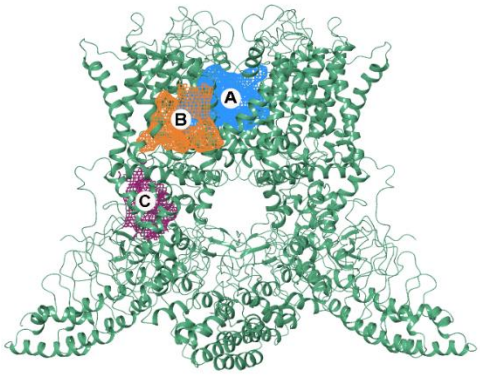

Figure S3 Illustration of three docking pockets in meshes in three different colors in mTRPV3. Pocket A in blue for the isomers IAA and IAB binding, correlating to the binding site for TRPV2 agonist CBD and TRPA1 inhibitor A967079. Pocket B in orange consisted of S4 and S4–S5 linker, corresponding to the capsaicin binding site in TRPV1. Pocket C in purple showing the 2-APB binding site in mTRPV3.

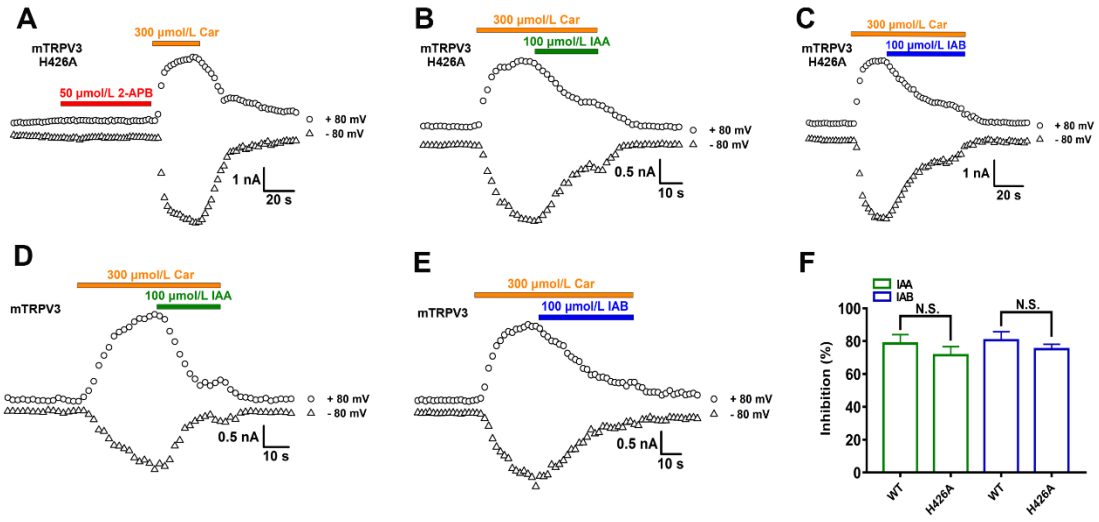

Figure S4 The residue H426 in mTRPV3 is critical for the binding of 2-APB, but not for IAA and IAB. (A) A representative current trace of mTRPV3 H426A mutant in response to carvacrol (300 μmol/L) or 2-APB (50 μmol/L). (B and C) Representative current traces for inhibition of carvacrol-activated mTRPV3 H426A mutant channels by IAA or IAB. (D and E) Representative current traces for inhibition of WT mTRPV3 channels by IAA or IAB. (F) Summary for current inhibition of WT mTRPV3 or H426A mutant channel by 100 μmol/L IAA and IAB (n = 4). Data are expressed as the mean ± SEM. N.S., no significance, by unpaired t test.