

Supplementary Figure legends

Supplementary Figure 1. Effects of a TREK-1 agonist ML335 in HEK293 cells or TREK-1-expressing HEK293 cells. (A) HEK293 cells were treated with vehicle or ML335 (10^{-5} M) and intensity of fluorescent were measured using a FluxOR™ potassium ion channel assay. Representative tracing of the fluorescence intensity in randomly selected HEK293 cells treated with vehicle ($n = 15$, the right figure) or ML-335 ($n = 15$, the center figure). The activity of TREK-1 was estimated using the time to peak fluorescence following vehicle ($n = 63$) or ML335 ($n = 39$) treatment (the left figure). (B) Representative tracing of the fluorescence intensity in randomly selected TREK-1-expressing HEK293 cells treated with vehicle ($n = 15$, the right figure) or ML-335 ($n = 15$, the center figure). The activity of TREK-1 was estimated using the time to peak fluorescence following vehicle ($n = 102$) or ML335 ($n = 105$) treatment (the left figure). The data were expressed as the mean \pm SEM. *** indicated $p < 0.001$, compared with vehicle; Mann-Whitney test.

Supplementary Figure 2. Effects of a TREK-1 agonist ML335 in TREK-1-expressing HEK293 cells using a FluxOR™ potassium ion channel assay. The cells were pretreated with vehicle for 2 min and subsequently treated with vehicle or ML335 (10^{-5} M). Representative tracing of the fluorescence intensity in randomly selected HEK293 cells treated with vehicle ($n = 15$, A) or ML-335 ($n = 15$, B). The activity of TREK-1 was estimated using the time to peak fluorescence following 2nd vehicle ($n = 98$) or 2nd ML335 ($n = 119$) treatment (C). The data were expressed as the mean \pm SEM. *** indicated $p < 0.001$, compared with vehicle; Mann-Whitney test.