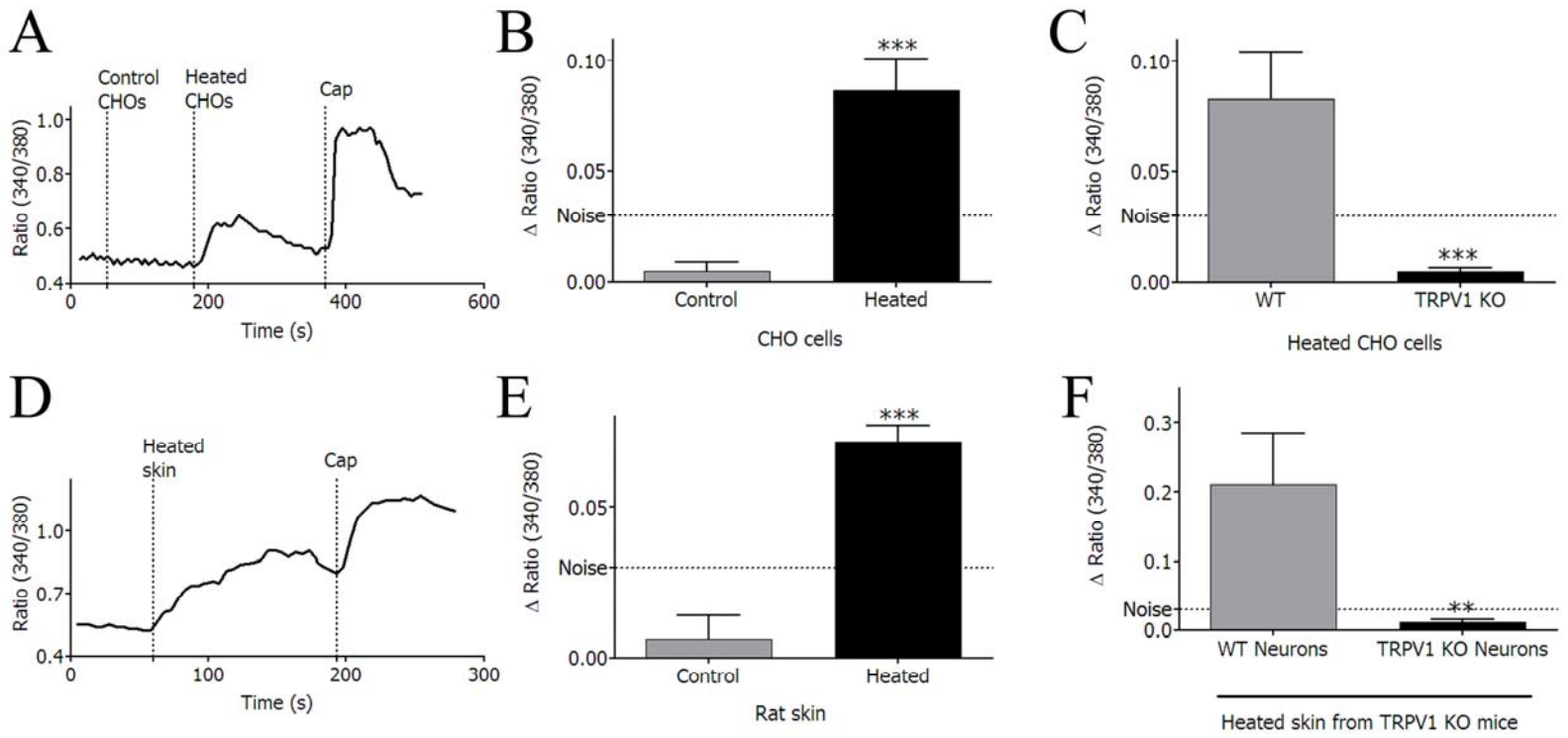


Supplementary Figure 1: Heat evokes release of endogenous TRPV1 ligand(s) in other cells/tissues

A: Effect of application of Hanks buffer, collected after exposure of CHO cells to noxious heat (48°C, for 20min) or control (37°C for 20 min) temperatures, on intracellular calcium levels in TRPV1-expressing CHO cells that also response to capsaicin (CAP 100nM). Both control and heated buffer were applied after cooling to room temperature.

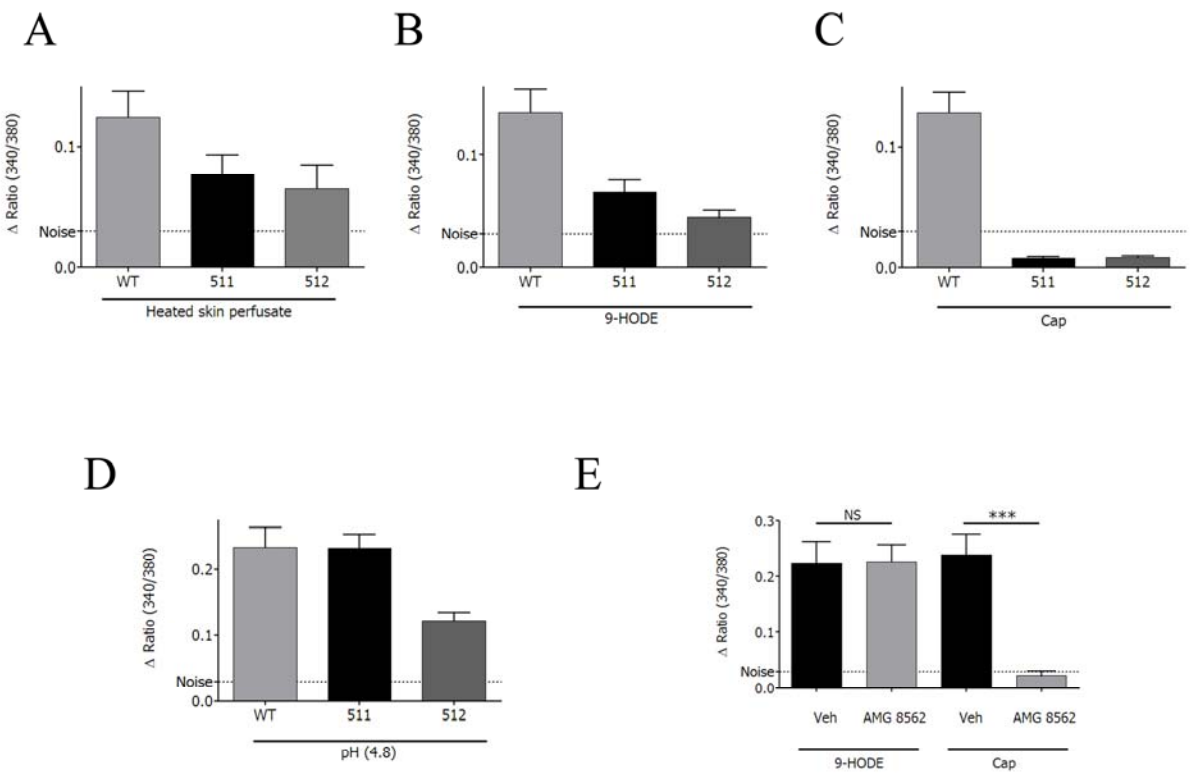
B: Summary presentation of the effect of Hanks buffer, collected from heated (48°C for 20 min) or non-heated (37°C) CHO cells, on [Ca]i after application to separate cultures of control or TRPV1-transfected CHO cells ($p<0.001$). Control CHO cells were transfected with GFP.

C: Summary of the effect of superfusates collected from heated (48°C for 20 min) or non-heated (37°C for 20 min) control CHO cells on [Ca]i after application to cultured trigeminal ganglia (TG) neurons from WT or TRPV1 KO mice ($p<0.001$).

D: Effect of applying a superfusate, collected from a rat skin biopsy (4 X 4 cm) after exposure to noxious heat (48°C, for 20min), to a TRPV1-expressing CHO cell that also responded to capsaicin (CAP 100 nM).

E: Summary of the effect of applying superfusates, collected from rat skin biopsies after exposure to noxious heat (48°C, for 20min) or control temperatures (37°C for 20 min), to
CHO cells expressing TRPV1 (n=45 and 61 cells for non-heated and heated skin respectively, $p=0.0001$).

F: Summary of the effect of applying superfusates, collected from skin biopsies from TRPV1 KO mice (1.5 X 1.5 cm. 6 skins) after exposure to noxious heat (48°C, for 20min), to TG neurons cultured from WT (n=22) or TRPV1 KO (n=18) mice ($p<0.01$).



Supplementary Fig 2: Heated skin superfusate and 9-HODE activate TRPV1 by a mechanism different than capsaicin.

A: Effect of applying a superfusate, collected from mouse skin biopsies (1.5 X 1.5 cm, 6 mice) after exposure to noxious heat (48°C, for 20min), to CHO cells expressing WT TRPV1, or the TRPV1 511 mutant or the 512 mutant (n=8-18) as measured by calcium imaging.

B: Effect of applying synthetic 9-HODE (100 μM) to CHO cells expressing WT TRPV1, or the TRPV1 511 mutant or 512 mutant (n=66-77).

C: Effect of applying capsaicin (100 nM) to CHO cells expressing WT TRPV1, 511 mutant and 512 mutant (n=25-61)

D: Comparison of calcium accumulation evoked by pH (4.8) in CHO cells expressing WT TRPV1, 511 mutant and 512 mutant (n=66-88)

E: Effect of pre and co-treatment with AMG 8562 (1 μM) on 9-HODE (100 μM) or capsaicin (100 nM)-evoked calcium accumulation in rat TG neurons.

Supplementary Fig 3: Effect of intracellular NDGA and Anti-HODE Antibodies on Inward Currents generated from Extracellular Application of synthetic 9-HODE (100 uM)

A: Representative tracers demonstrating the effect of preincubation with either NDGA (30uM, 5min) or the combination of anti-9-HODE and anti-13-HODE antibodies dialyzed intracellularly via the patch pipette (5min) on extracellular bath application of 9-HODE (100 uM) in rat TG neurons.

B: Summary of the NDGA and antibody effect on 9-HODE (n=6-10, p>0.05 in all comparisons)