

Ponissery Saidu et al., <http://www.jgp.org/cgi/content/full/jgp.201210937/DC1>

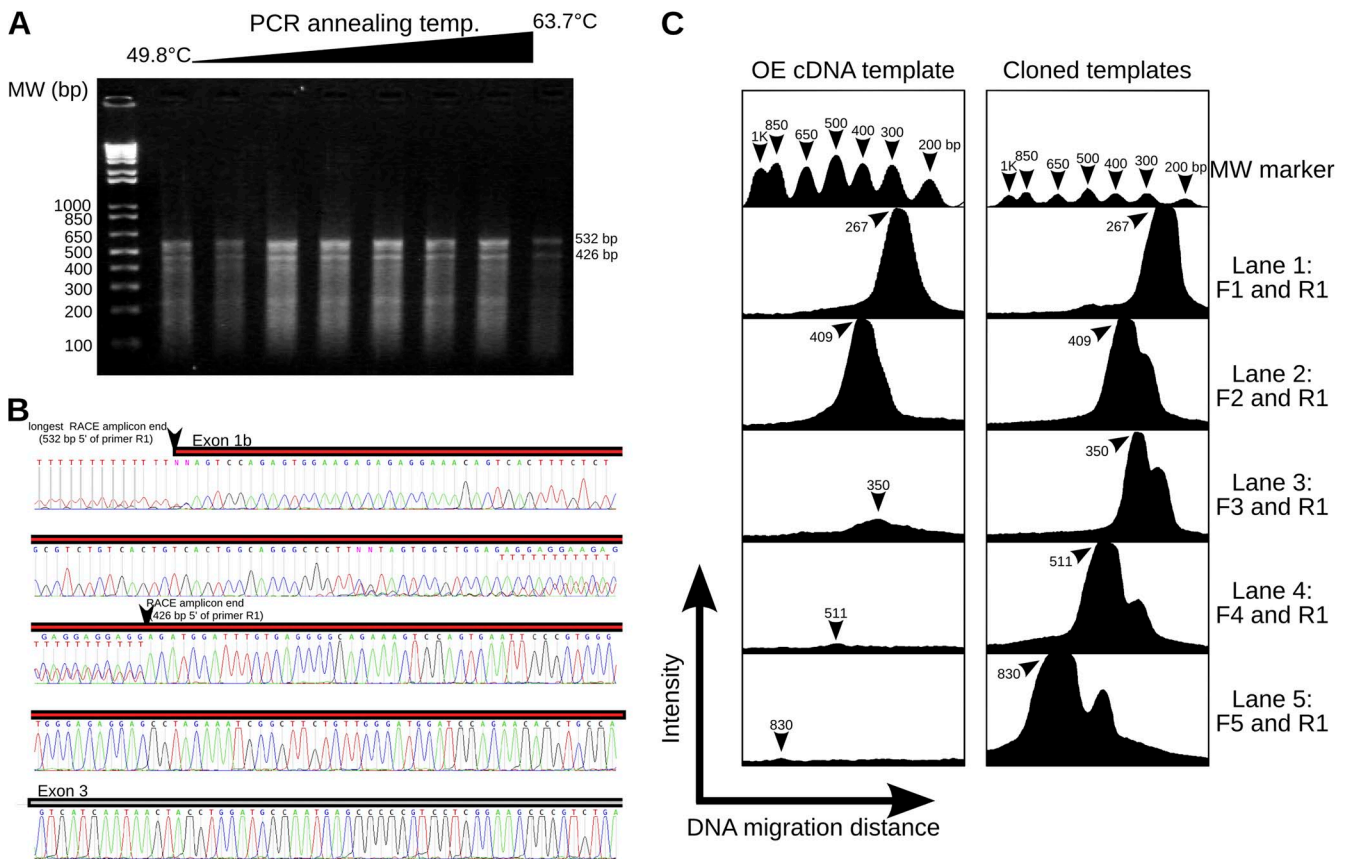


Figure S1. 5' RACE analysis to determine the 5' end of olfactory *Ano2* transcripts. (A) Agarose gel showing 5' RACE PCR products for *Ano2*. The predominant bands are of the sizes 532 and 426 bp. These PCR products were sequenced directly (B), and were also cloned and sequenced (see Table S1). (B) Reverse complement sequencing chromatogram showing that the predominant RACE PCR products include a new exon, named exon 1b (red), which is joined to exon 3 (gray). (C) Densitometry quantification of band from Fig. 1 (B and C). Arrowheads indicate peak positions.

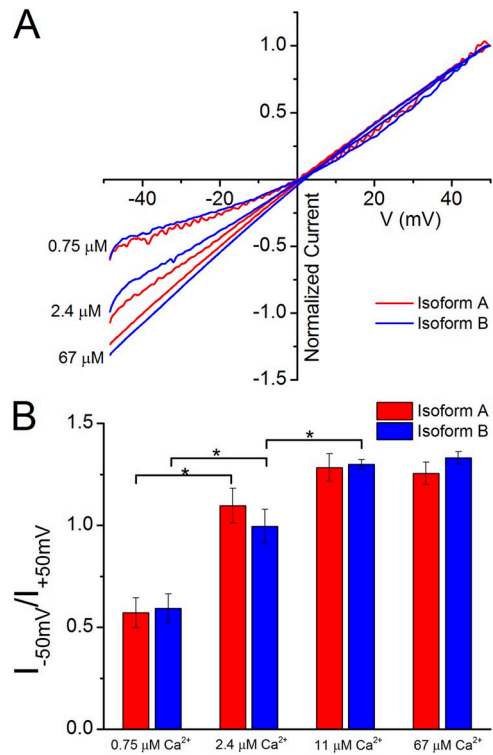


Figure S2. Isoforms A and B exhibit similar rectification. (A) I-V relationships for Isoforms A and B at three different Ca²⁺ concentrations. All currents were normalized to their current value at +50 mV to allow comparison. (B) Rectification was calculated as the current ratio at -50 mV divided by the current at +50 mV as a function of Ca²⁺ concentration. *, statistical significance at P < 0.01; values are averages of five patches.

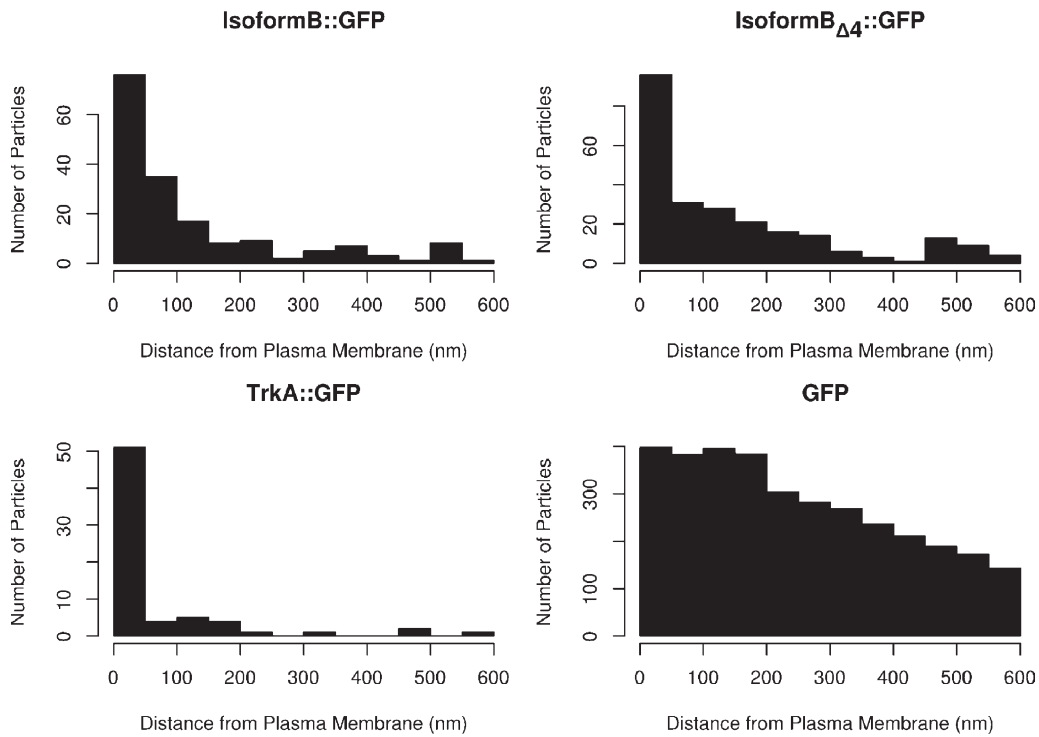


Figure S3. Cellular distribution of ANO2 splicing isoforms by immunogold electron microscopy. The distance of each immunogold particle to the cell membrane was measured for the four proteins shown in Fig. 5 B. Each plot is a histogram showing the frequency of distance measurements in 50-nm bins. The first bin represents particles that are considered to be localized to the cell membrane. For Isoform B, 173 gold particles were measured from 47 images. For Isoform B_{Δ4}, 249 particles were measured from 32 images. For TrkA::GFP, 69 particles were measured from 21 images. For GFP, 3,726 particles were measured from 30 images.

Table S1

Summary of olfactory Ano2 5' RACE PCR and clone sequences

Clone	Length	Exons included
	<i>bp</i>	
PCR product	426/532	1b, 3
1-11	87	3
1-2	89	3
4-6	104	3
1-12	201	3
3-2	245	3
1-8	321	3
2-5	416	1b, 3
1-7	426	1b, 3
2-10	426	1b, 3
4-2	426	1b, 3
4-5	426	1b, 3
4-9	426	1b, 3
1-9	530	1b, 3
2-9	531	1b, 3
2-4	533	1b, 3

5' RACE PCR products were sequenced directly and were also cloned. 14 of the clones were then sequenced. Two predominant species, of lengths 426 and 532 bp, were identified by gel analysis (Fig. S1 A) and sequencing. Both of these species contained a novel exon, named exon 1b.