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Supplementary Material

Supplementary Table 1. Primers for PCR

Name	Sequence
GPR68-1	aagcttccaccATGAGGAGTGTGGCCCT
GPR68-2	gaattcCTAGGCCAACCTGCCCGTGGGA
GPR68-2HA	gaattcCTAAGCGTAATCTGGAACATCGTATGGTAGGCCAACCTG CCCGTGGGA
pcDNA3-T7	TAATACGACTCACTATAAGGG
pcDNA3-BGH	TAGAAGGCACAGTCGAGGCT
GAPDH-1	TGATGACATCAAGAAGGTGGTGAAG
GAPDH-2	TCCTTGGAGGCCATGTAGGCCAT

Supplementary Fig. 1. Prediction of secondary structure for human OGR1 by PSIPRED V4.0 (The PSIPRED Protein Sequence Analysis Workbench at <http://bioinf.cs.ucl.ac.uk/psipred/>). Abbreviations used were: Conf, configuration values (0-9); Pred, prediction of secondary structure (H, helix; E, strand; C, coil) as described in The PSIPRED Protein Sequence Analysis Workbench (<http://bioinf.cs.ucl.ac.uk/psipred/>). The transmembrane domains (TMI-TMVII) are boxed in purple, and the histidine residues that have been shown to be involved in proton-sensing in OGR1 (H17, H20, H84, H169, and H269) are shown in green letter as reported previously [2, 8]. A notable leucine (L74) in TMII is shown in red letter. A putative amino acid sequence (10 residues) are underlined in the N-terminal of the OGR1 (GenBank: NM_003485).

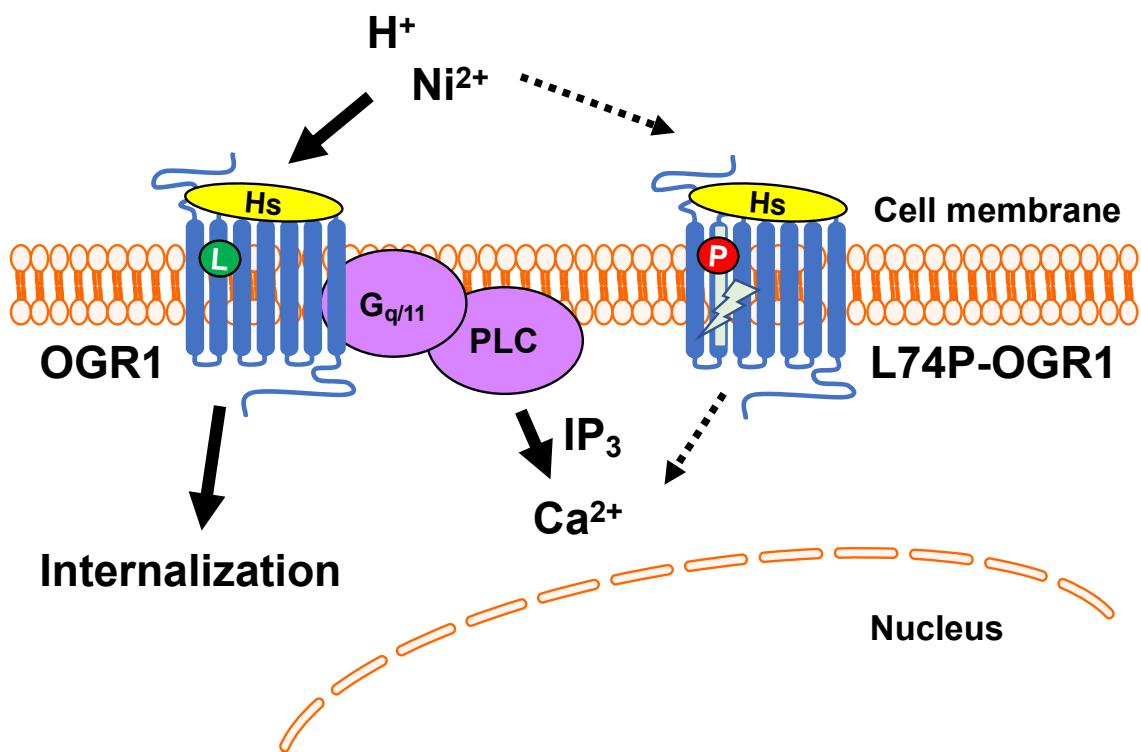
OGR1

Conf: 23998899999999999999992899999997599985
Pred: CCCCCHHHHHHHHHHHHHHHHHHHHHHHHHHCCCCCCC
AA: KARNELGVYLCNLTVADLFYICS **I**PFWLQYVL**Q**HDNWSHG
51 60 70 80 90

L74P-
OGR1

Conf: 34998899899999999999728989999971999997
Pred: CCCCCHHHHHHHHHHHHHHHHHHHHHHHHCCCCCCC
AA: KARNELGVYLCNLTVADLFYICS **P**PFWLQYVL**Q**HDNWSHG
51 60 70 80 90

Supplementary Fig. 2. Prediction of secondary structure around second transmembrane domain in human OGR1 by PSIPRED V4.0. The positions of L74P and H84 are shown in red and green letters, respectively. The predicted secondary structure around L74 and TMII (boxed in purple) for WT and L74P are affected by the substitutions as shown in blue letters. Thus, the mutations in OGR1 may become damaged in its receptor functions.



Supplementary Fig. 3. The postulated Ca^{2+} signaling by OGR1 and different modes of WT and L74P receptors. Extracellular protons or Ni^{2+} induce OGR1 activation through histidine residues and subsequent activation of G protein/ Ca^{2+} signaling pathways. Similar to other GPCRs, OGR1 is internalized in response to the extracellular stimuli. The L74P receptor is distributed in the plasma membranes without sensing extracellular stimuli, and dose not influence anything on the proton/intrinsic OGR1-receptor signaling. See text more detail.