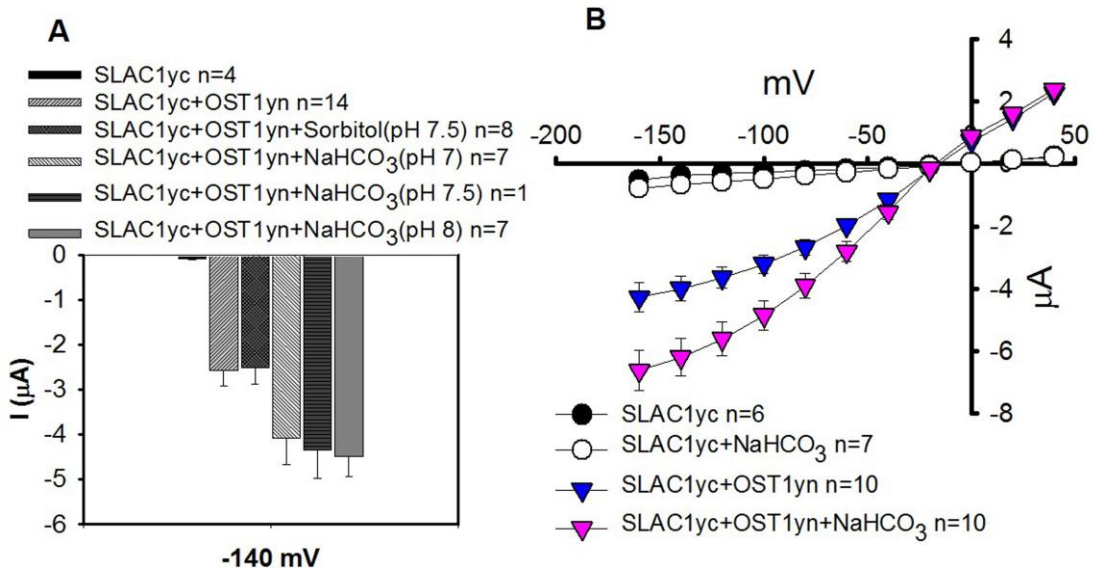
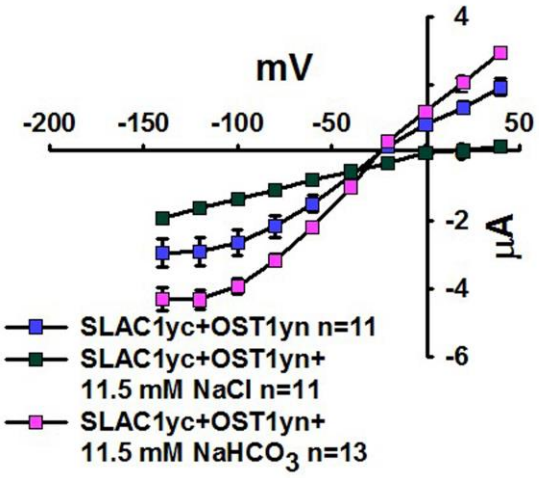
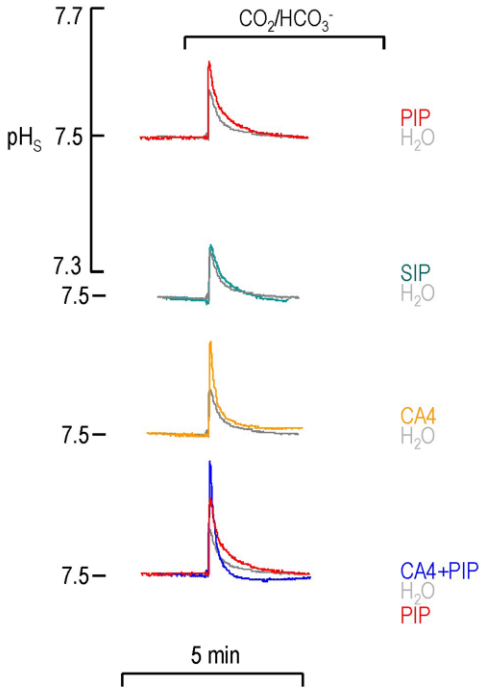


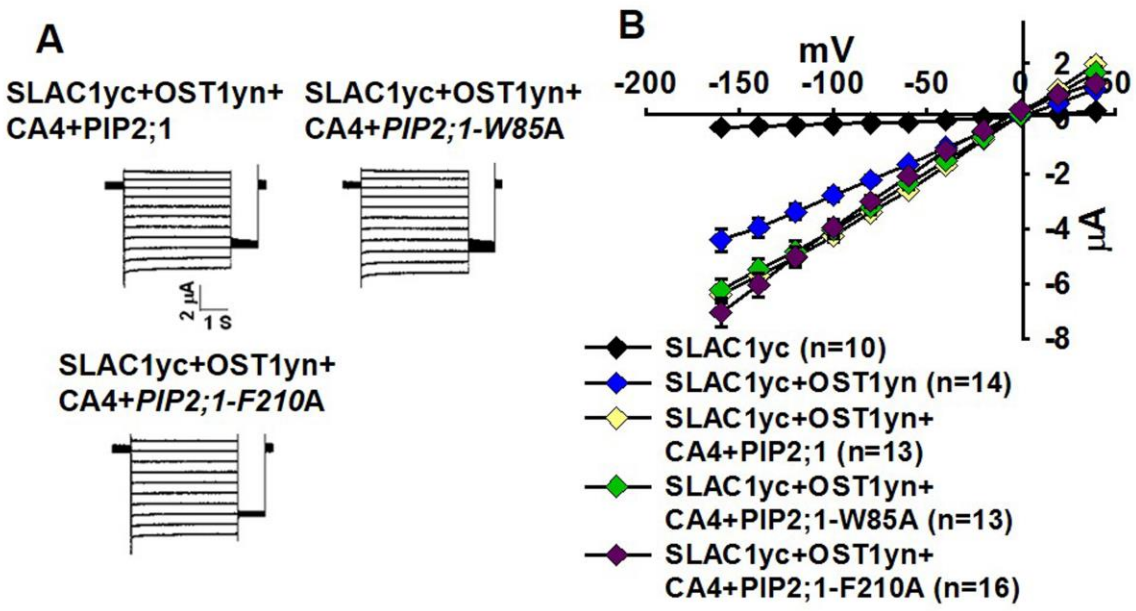
Supplemental Figure 1. (A, B) Injection of 11.5 mM $NaHCO_3$ **(A)** at pH 7 or pH 8 into oocytes and **(B)** recordings in 96 mM NaCl extracellular buffer also cause enhancement of SLAC1-mediated anion channel currents, whereas **(A)** 23 mM sorbitol injection has no effect on SLAC1 activity. **(B)** Note that the reversal potential of SLAC1yc + OST1yn-mediated currents in (B) was close to the Cl- equilibrium potential. Data are mean ± s.e.m.



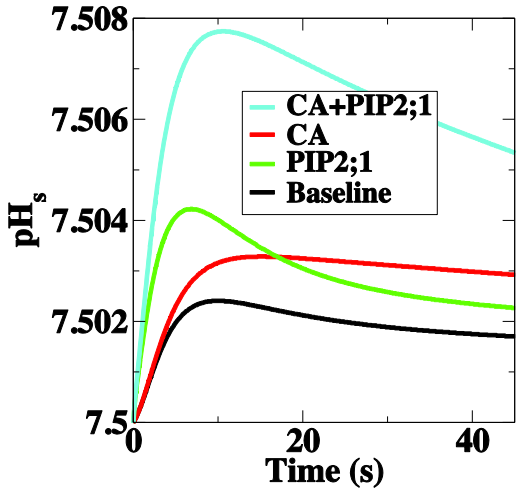
Supplemental Figure 2. Steady-state current-voltage relationships show the average magnitude of SLAC1yc/OST1yn-mediated anion channel currents recorded from oocytes injected with 11.5 mM NaCl were reduced rather than enhanced. Data are mean ± s.e.m. Data are representative of experiments performed on three independent oocyte batches.



Supplemental Figure 3. Surface pH (pH_s) measurements from oocytes exposed to CO_2/HCO_3^-. Oocytes were exposed to 5% CO_2/33 mM HCO_3^- long enough for the pH_s to rise and then decay exponentially to a stable value. Traces from oocytes recorded in the same batch are shown.



Supplemental Figure 4. The *PIP2;1-W85A and PIP2;1-F210A* mutation isoforms do not impair the PIP2;1-CA4 enhancement of SLAC1/OST1-mediated anion channel currents in oocytes by extracellular CO_2/HCO_3^-. **(A)** Whole-cell currents were recorded from oocytes expressing the indicated cRNAs with 11.5 mM $NaHCO_3$ in the bath solution. The voltage protocol was the same as in Figure 1. **(B)** Steady-state current-voltage relationships from oocytes recorded as in (A). Data are mean ± s.e.m. Results from three independent batches of oocytes showed similar results.



Supplemental Figure 5. Simulated membrane surface pH (pH_s) as a function of time for baseline parameter values (black curve), in the presence of intracellular carbonic anhydrase (CA) activity (red curve), for simulated increased membrane CO_2 permeability (green curve), and in the presence of both intracellular CA activity and increased membrane CO_2 permeability (blue curve). An extracellular CO_2 increase from 200 ppm to 800 ppm (Hanstein et al., 2001) was simulated (see Methods), resulting in smaller modeled membrane surface pH (pH_s) changes than when larger CO_2 concentration steps were applied experimentally in oocytes.

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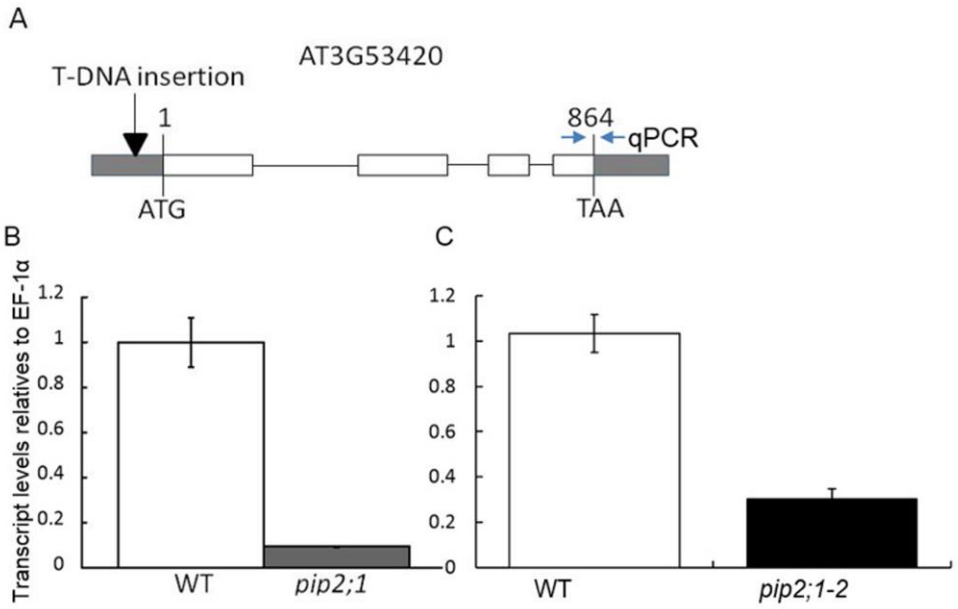

Supplemental Figure 6. (A) Structure of *PIP2;1* gene and T-DNA insertion. *PIP2;1 c*onsists of four exons (open boxes); black boxes highlight the 5' and 3' untranslated regions, respectively. Mutant line *pip2;1* (ABRC stock name CS320492) has a T-DNA insertion in 5'-UTR region. The location of qPCR primers is shown by opposing arrows. **(B)** qPCR analyses suggested that *pip2;1* is a knockdown mutant. Expression level was compared to *EF-1a.* **(C)** qPCR analyses suggest that *pip2;1-2* (Grondin et al., 2015) is a knockdown mutant. Expression level was compared to *EF-1a*. Data were presented by means ±s.e.m, n=3.

Name	sequence (5'-3')
CA4NBF	GGCTTAAUAATGGCTCCTGCATTCGG
CA4NBR	GGTTTAAUTTCCGGTAGCTTTCTTTC
CA4GBKF	CAGAATTCGACGAAATGGCAACGGAATC
CA4GBKR	CAGGATCCTTCCGGTAGCTTTCTTTC
PIP2;1-pNB1F	GGCTTAAUTAACTATGGCAAAGGATG
PIP2;1-pNB1R	GGTTTAAUGACTGATTTAGATTTGTACAGAGAG
SIP1A-pNB1F	GGCTTAAUTACCACCCACCTAACCAC
SIP1A-pNB1R	GGTTTAAUCTTTCTACAGCCCAAACC
CA4F	GGGGACAAGTTTGTACAAAAAAGCAGGCTAATGGCTCCTGCATTCGG
CA4R	GGGGACCACTTTGTACAAGAAAGCTGGGTAGGCAAAAGCAGGAGTG
PIP2;1F	GGGGACAAGTTTGTACAAAAAAGCAGGCTTCTCACTAACCACTCCAACA
PIP2;1R	GGGGACCACTTTGTACAAGAAAGCTGGGTCACTTCTGAATGATCCAAGA
RHC1-pNB1F	GGCTTAAUATGTCAGAAACATCAAAGTC
RHC1-pNB1R	GGTTTAAUCTATGAGTGGCTATCTTGTCC
qEF-1aF	TGAGCACGCTCTTCTTGCTTTCA
qEF-1aR	GGTGGTGGCATCCATCTTGTTACA

Supplemental Table 1. Primers Used for Construct and Expression Studies.