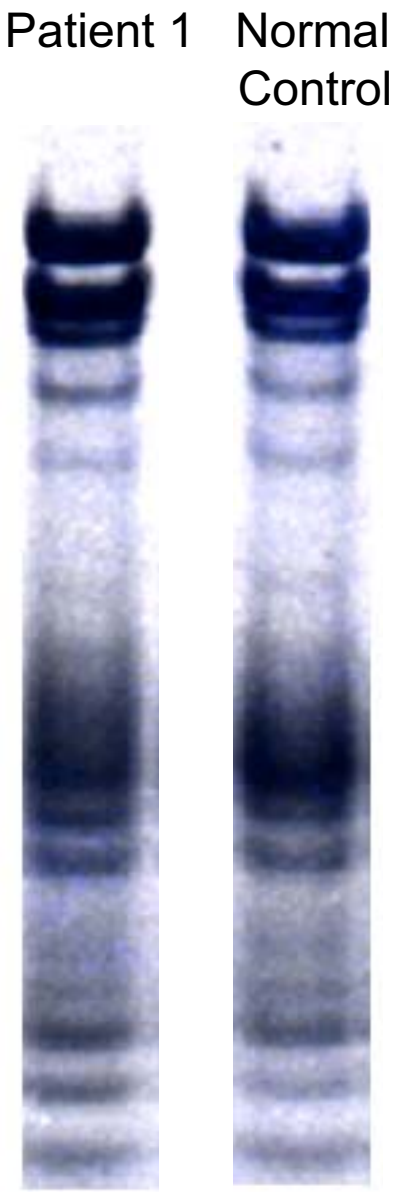
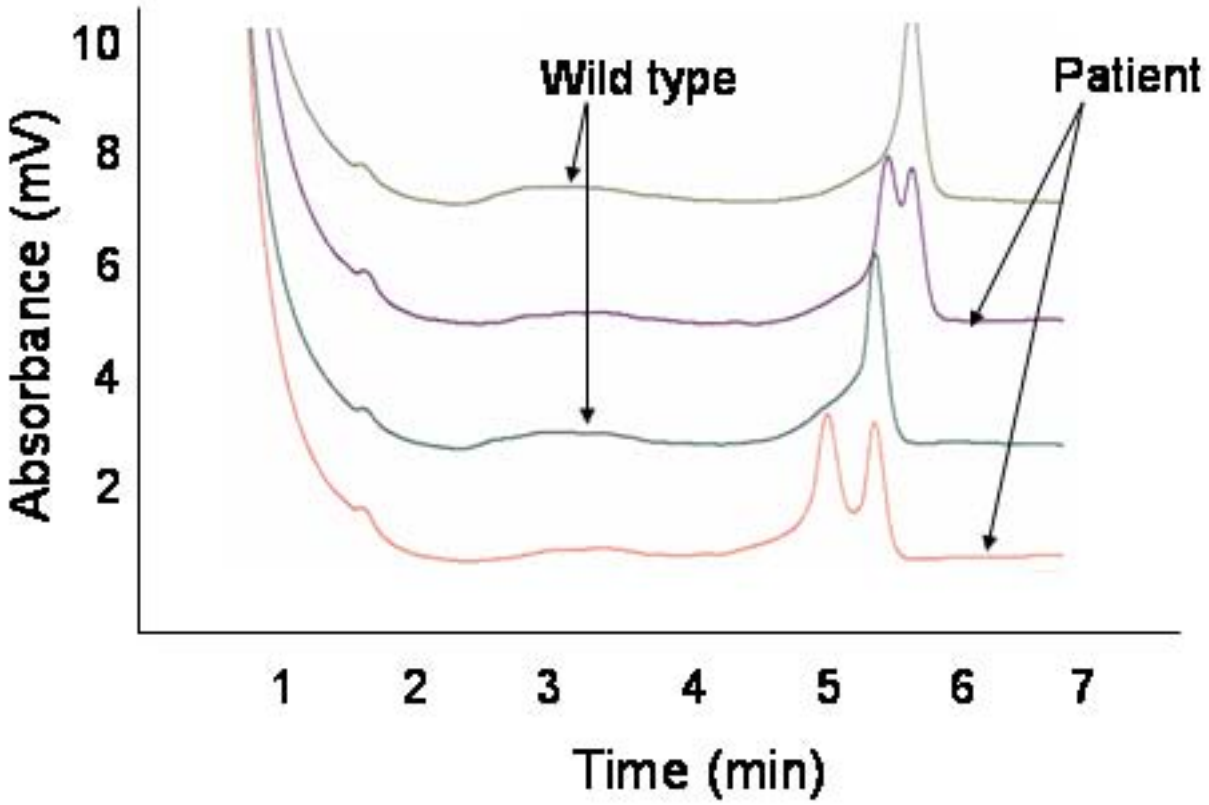
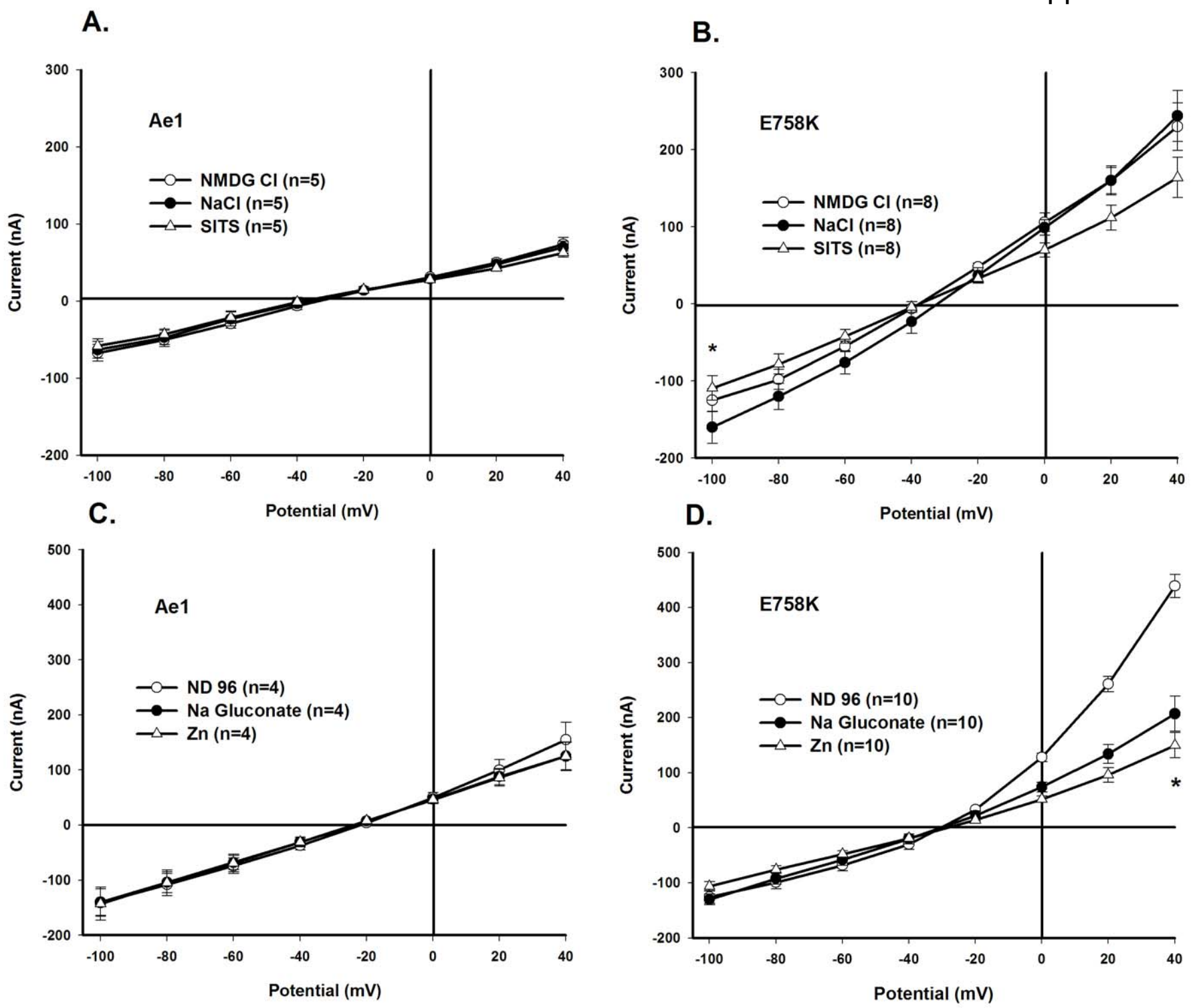
Supplemental Figure 1

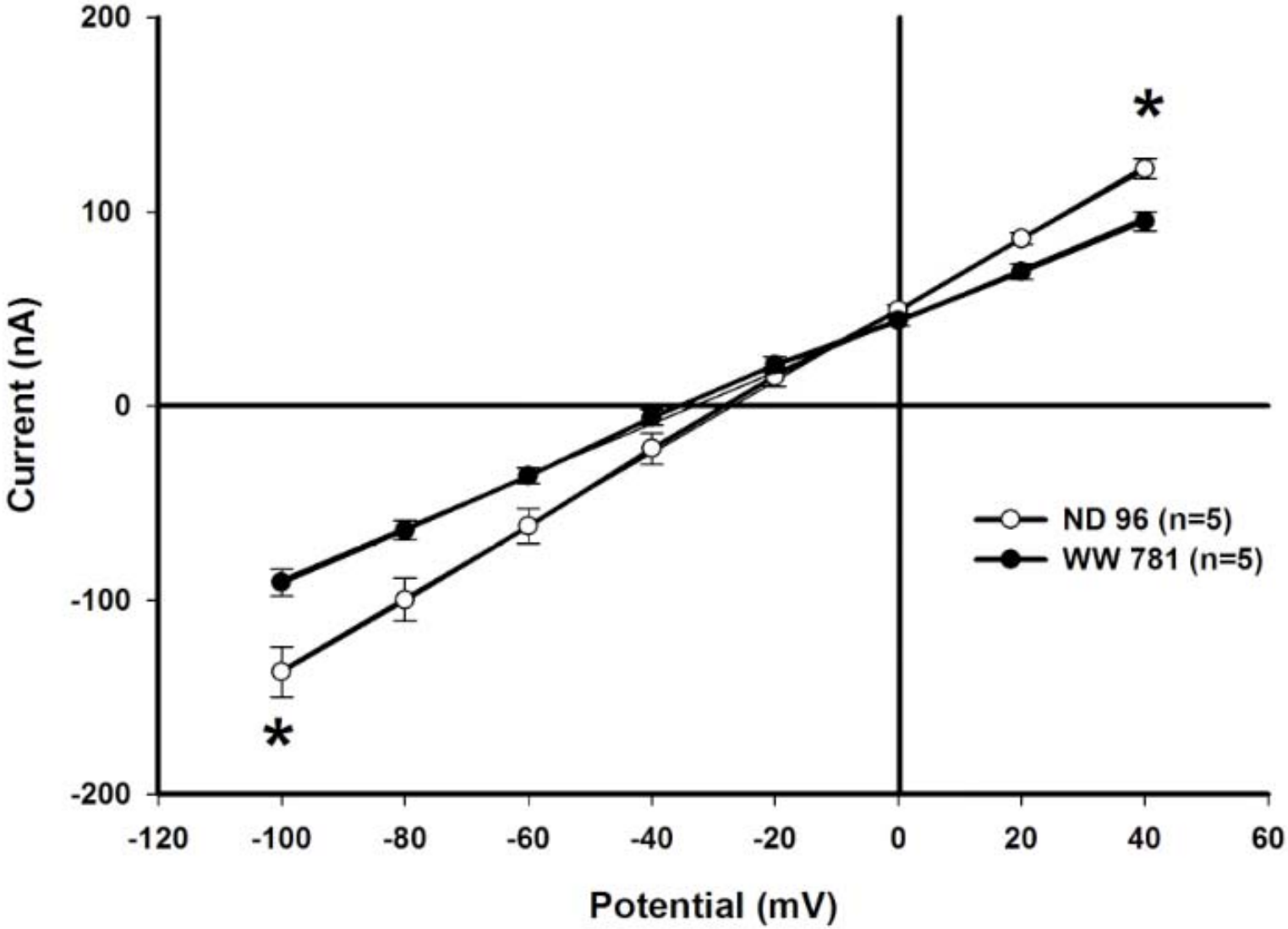


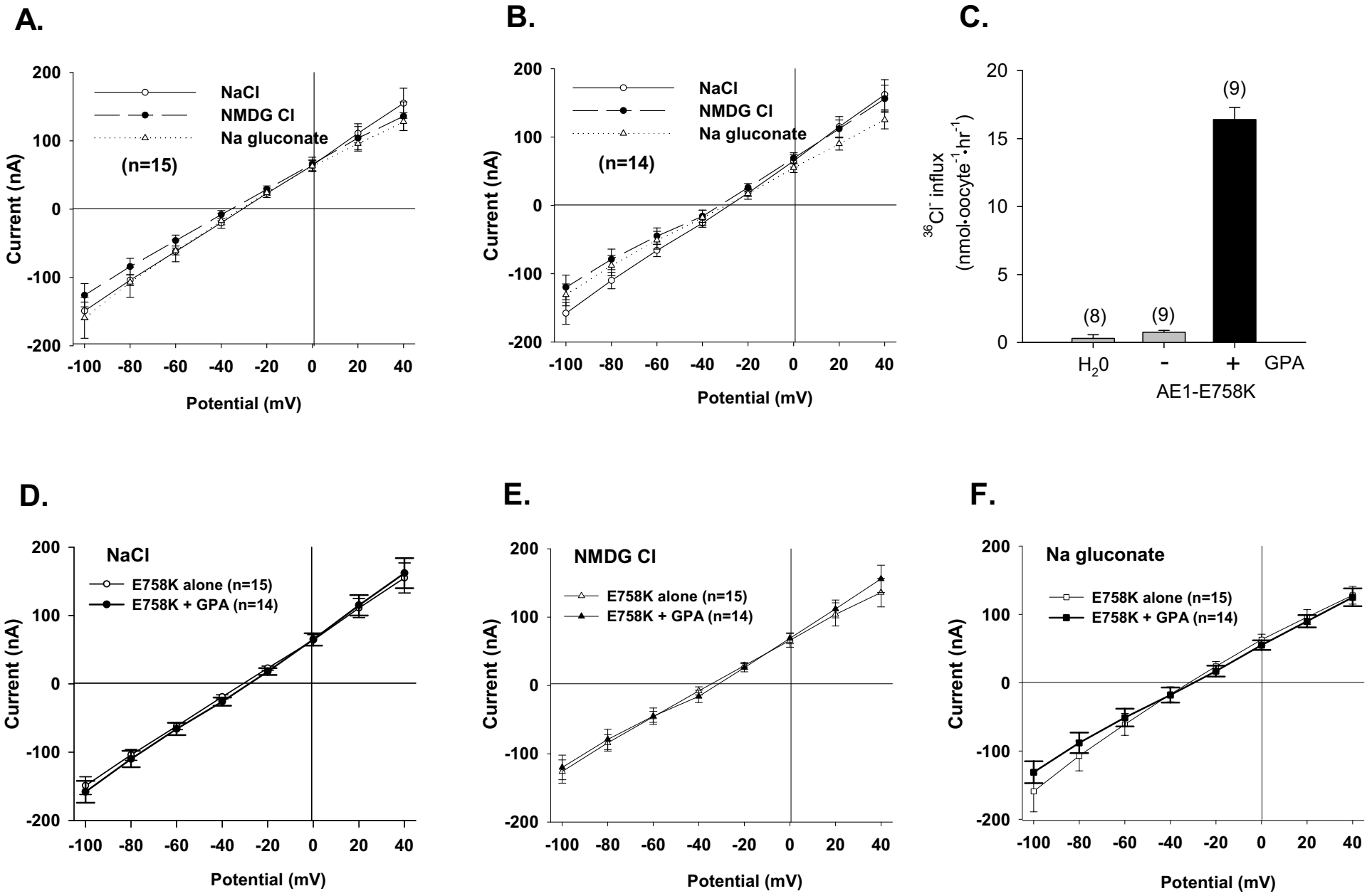
Supplemental Figure 2




A.
Ae1
NMDG Cl (n=5)
NaCl (n=5)
SITS (n=5)
Current (nA)
Potential (mV)
B.
E758K
NMDG Cl (n=8)
NaCl (n=8)
SITS (n=8)
*
Current (nA)
Potential (mV)
C.
Ae1
ND 96 (n=4)
Na Gluconate (n=4)
Zn (n=4)
Current (nA)
Potential (mV)
D.
E758K
ND 96 (n=10)
Na Gluconate (n=10)
Zn (n=10)
*
Current (nA)
Potential (mV)

Supplemental Figure 4




A.
B.
C.
D.
E.
F.
NaCl
NMDG Cl
Na gluconate
(n=15)
(n=14)
Current (nA)
Potential (mV)
(8)
(9)
H20
-
+
GPA
AE1-E758K
E758K alone (n=15)
E758K + GPA (n=14)

Supplemental Figure Legends

Supplemental Figure 1. Coomassie Blue profile of erythrocyte membrane proteins from patient 1 and a normal control.

Supplemental Figure 2. DHPLC profile of amplified exon 17 from patient 2 and from a normal control.

Supplemental Figure 3. Distinct patterns of partial inhibition by SITS and Zn^{2+} of currents in *Xenopus* oocytes expressing AE1 E758K. A. Oocytes expressing wildtype AE1 display basal currents insensitive to SITS applied in NMDG chloride. **B.** Inward current in AE1 E758K-expressing oocytes is partially inhibited by SITS applied in NMDG chloride. *, $p < 0.001$ comparing absence and presence of inhibitor at -100 mV ($p=0.07$ at +40 mV). **C.** Oocytes expressing wildtype AE1 display basal currents insensitive to SITS applied in Na gluconate. **D.** Oocytes expressing AE1 E758K exhibit outwardly rectifying current reduced after transition from chloride to gluconate, and inhibited by Zn^{2+}. All oocytes coexpressed GPA (2 ng cRNA) with wildtype or mutant AE1 (10 ng cRNA). Values are means $\pm$ s.e.m. for (n) oocytes. *, $p < 0.001$ comparing absence and presence of inhibitor at +40 mV.

Supplemental Figure 4. Inhibition by WW-781 of current in *Xenopus* oocytes expressing AE1 E758K. Oocytes previously injected with AE1 E758K (10 ng cRNA) and GPA (2 ng cRNA) were subjected to two-electrode voltage clamp measurement of currents in ND=96 prior to (open circles) and following exposure to 100 μM WW-781 (filled circles). Values are means $\pm$ s.e.m. for (n) oocytes. *, $p<0.01$ comparing absence and presence of inhibitor at -100 mV and +40 mV.

Supplemental Figure 5. Coexpression of GPA does not increase currents in *Xenopus* oocytes expressing AE1 E758K. A. I-V relationship of AE1 E758K-expressing oocytes (n=15) exposed sequentially to bath solutions of NaCl (open circles), NMDG Cl (filled circles), and Na gluconate (open triangles). **B.** I-V relationship of oocytes coexpressing AE1 E758K (10 ng) and GPA (2 ng; n=14) exposed sequentially to bath solutions of NaCl (open circles), NMDG Cl

(filled circles), and Na gluconate (open triangles). **C.** GPA-dependence of $^{36}Cl^-$ influx in oocytes from one of the two frogs used in the experiments of panels A and B. **D.** Comparison of I-V relationships of oocytes in NaCl bath expressing AE1 E758K without coexpressed GPA (open circles, n=15; from panel A) with that from different oocytes (from the same two frogs) coexpressing AE1 E758K and GPA (filled circles, n=14; from panel B). **E.** Comparison of I-V relationships of oocytes in NMDG Cl bath expressing AE1 E758K without coexpressed GPA (open circles, n=15; from panel A) with that from different oocytes (from the same two frogs) coexpressing AE1 E758K and GPA (filled circles, n=14; from panel B). **F.** Comparison of I-V relationships of oocytes in Na glucoante bath expressing AE1 E758K without coexpressed GPA (open circles, n=15; from panel A) with that from different oocytes (from the same two frogs) coexpressing AE1 E758K and GPA (filled circles, n=14; from panel B). In panels D-F, the GPA-dependent current (the difference between the two plotted currents) is statistically indistinguishable from zero in all bath solutions tested.