

2 Fig. S2 Electrophysiological analyses of AtALMT1.

a, Representative I-V curves of empty vector (left) or AtALMT1 (middle) transfected HEK293T 1 cells at various concentrations of extracellular  $Al^{3+}$ . The "Bath" and "0" *I–V* curves indicate the 2 first and the last sweep of ramp recording model before  $Al^{3+}$  was perfused established by whole-3 cell configuration.  $Al^{3+}$  was perfused with different concentrations (in  $\mu M$ ) when getting the 4 steady-state currents. The membrane potential was held at 0 mV, and a voltage ramp of 500 ms 5 duration from -200 mV to +80 mV was applied every 5 s. The right panel shows the concentration-6 dependent extracellular Al<sup>3+</sup> activation of wild-type (WT) AtALMT1. Data are reported as mean 7  $\pm$  s.e.m. of at least five independent biological replicates. Curves are least-square fits to a Hill 8 equation with half-maximum activation concentration (EC<sub>50</sub>) values of 98.17  $\pm$  3.53  $\mu$ M. **b**. The 9 same current recording as that in **a** except that the bath solution pH was changed to 5.  $EC_{50} = 120.7$ 10  $\pm$  11.93 µM. c-e, Representative *I–V* curves of mutant AtALMT1-transfected HEK293T cells at 11 various concentrations of extracellular  $Al^{3+}$ . f, Representative fluorescent (upper) and bright field 12 (lower) images of cells expressing green fluorescent protein (GFP)-strep-tagged WT and mutant 13 AtALMT1. GFP fluorescence was readily detected in WT and mutant AtALMT1 cells with similar 14 patterns. g, Western Blots with anti-strep antibody to probe the membrane extract of HEK293 cells 15 overexpressing GFP-strep-tagged WT and mutant AtALMT1 shows that the WT and mutant 16 AtALMT1 display comparable expression levels on the membrane. 17

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