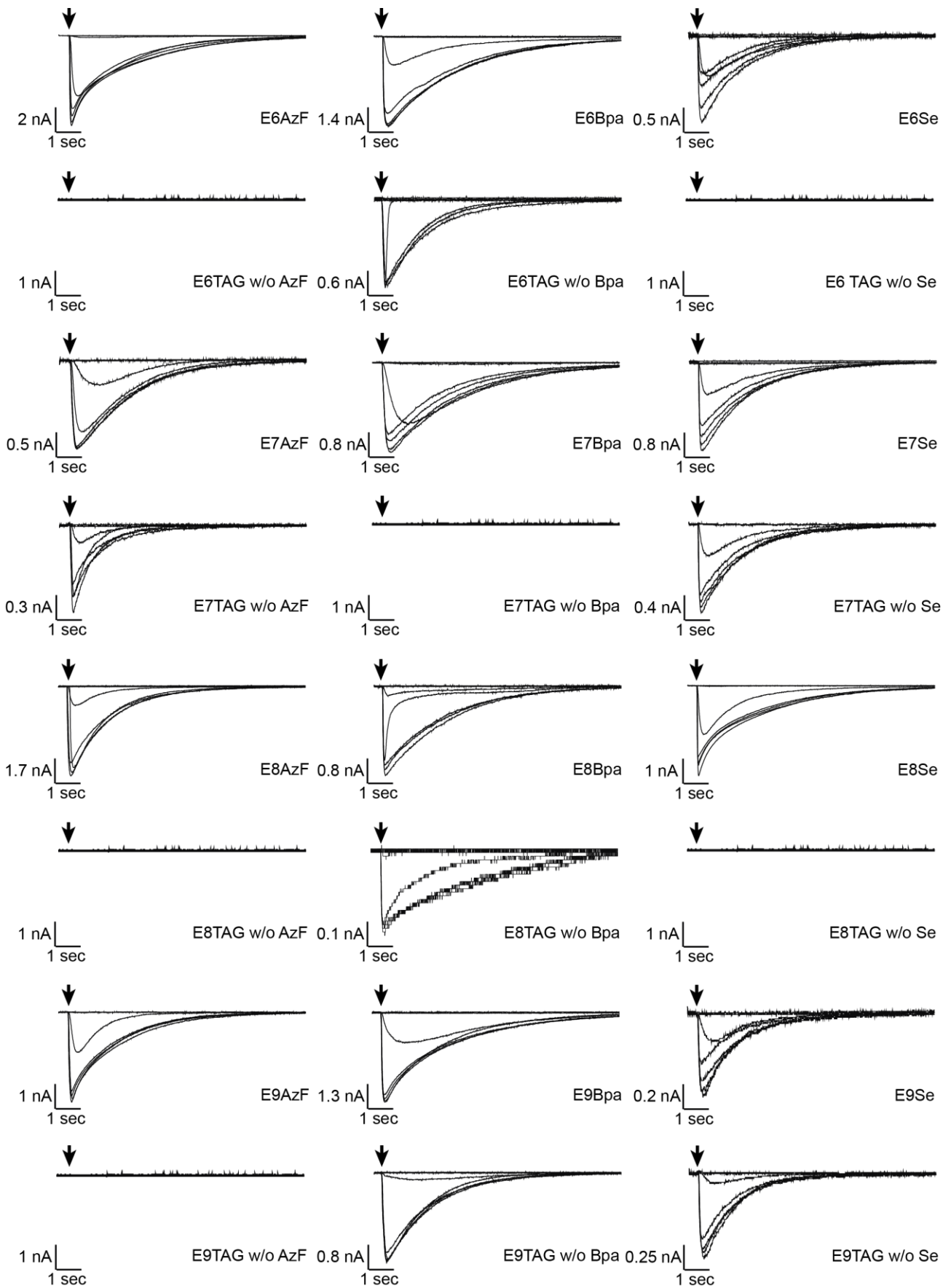


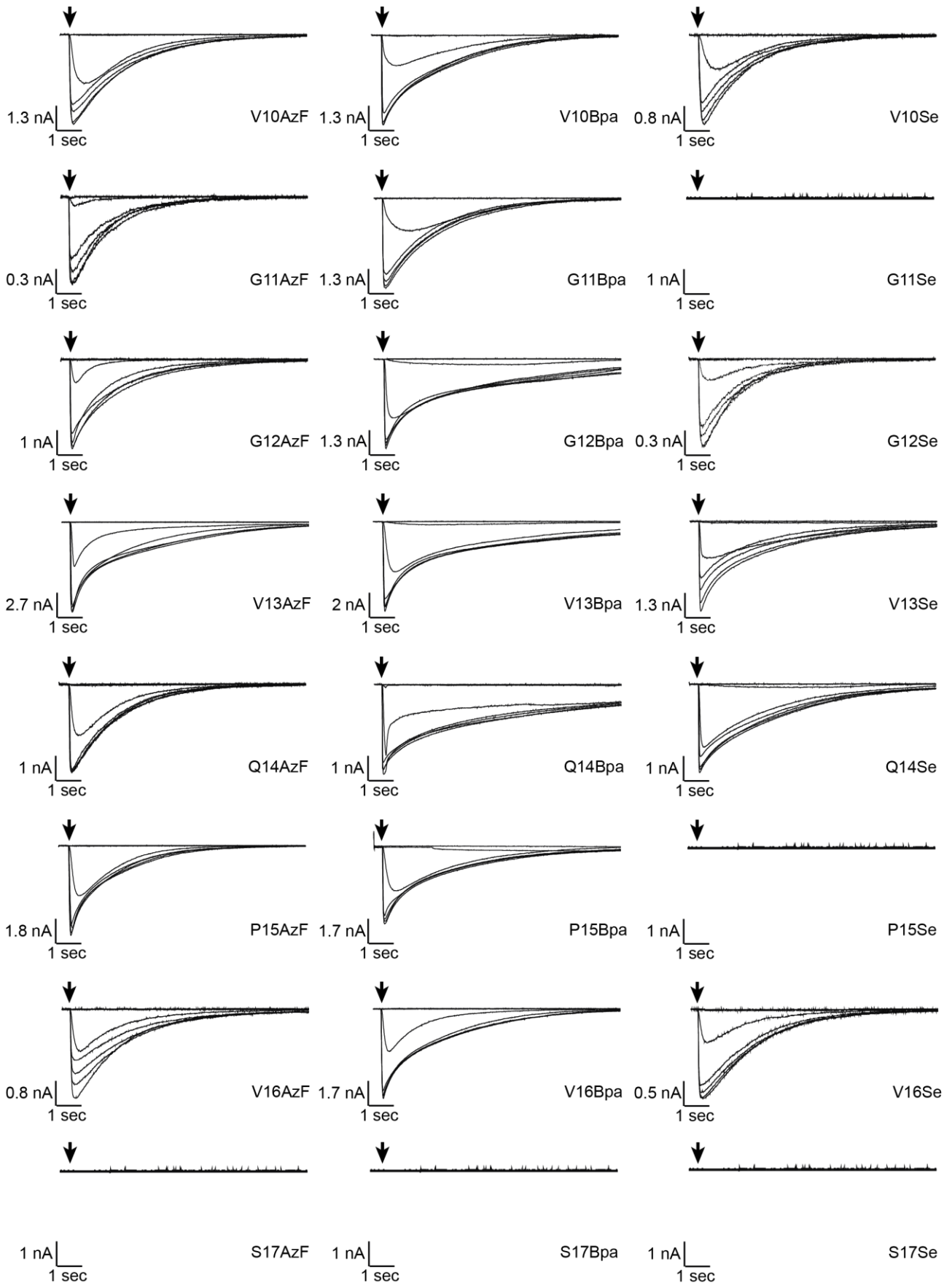
# Collection of example current traces of all 309 ASIC1a TAG variants tested in the APC screen

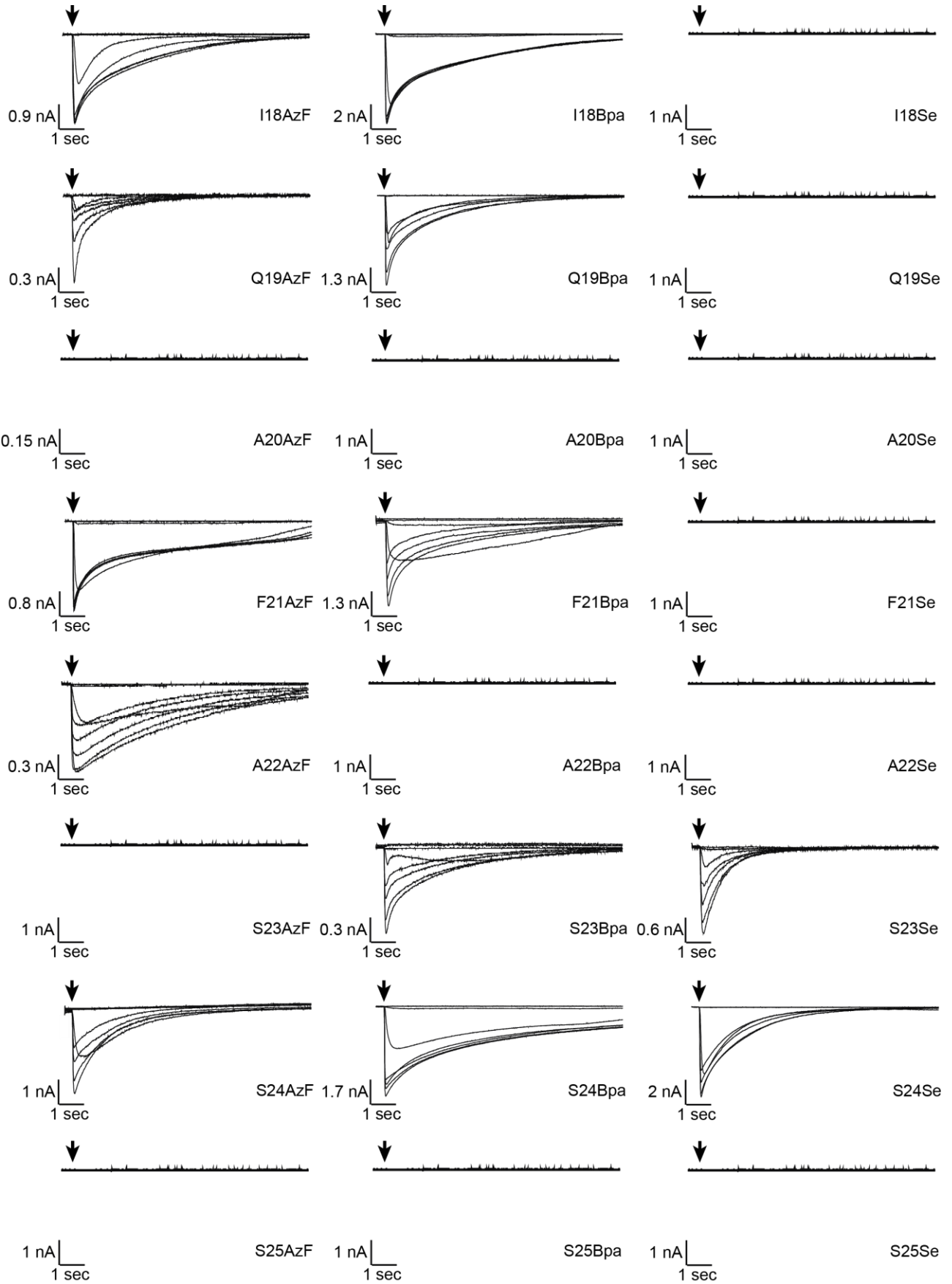
This document contains example current traces for:

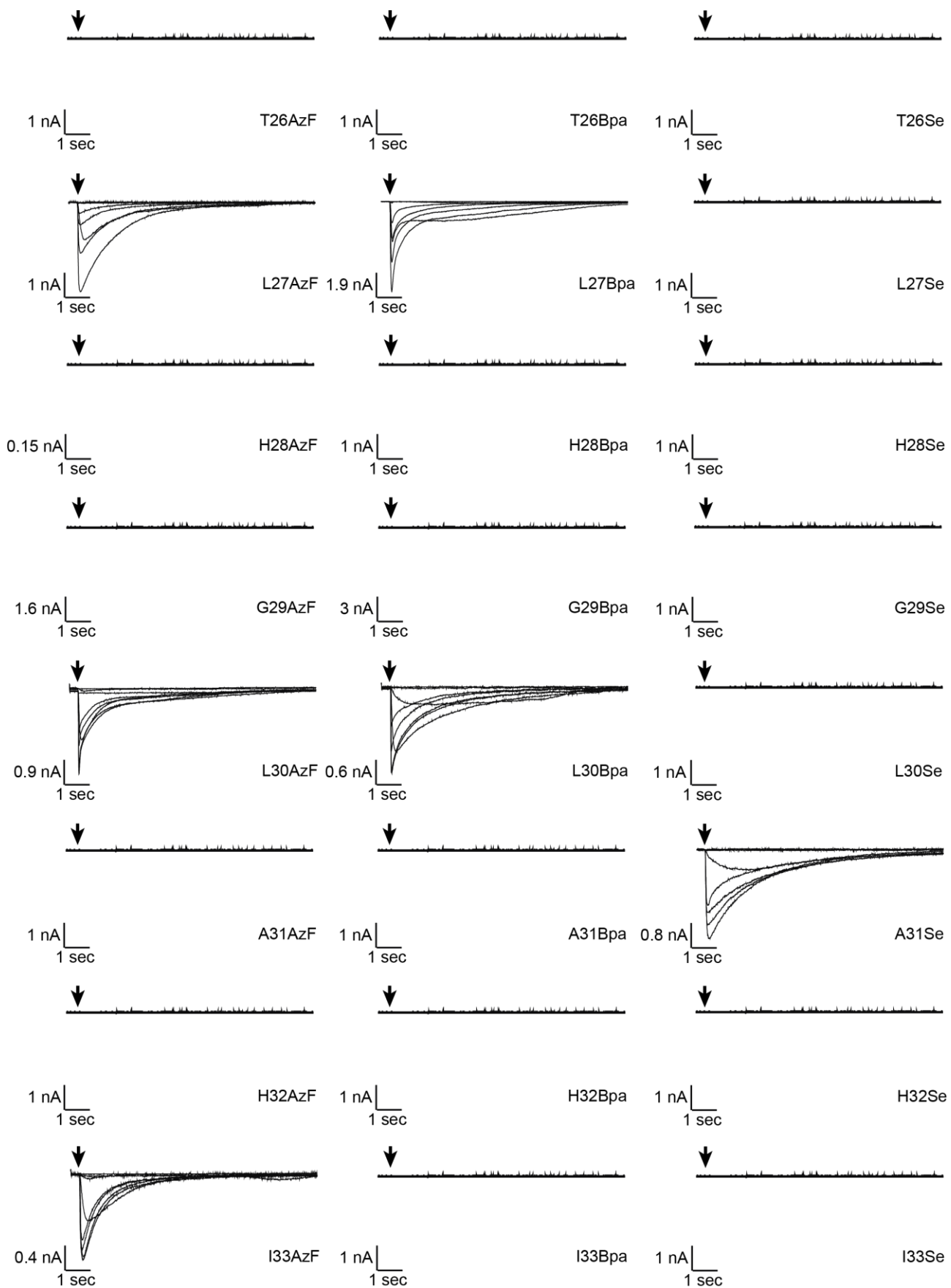
- All 309 ASIC1a TAG variants tested in the APC screen (Figures 2, 3, S2-5, S8)
- Truncated variants shown in Figures S5 and S6
- SSD curve measurements (Figure S10)

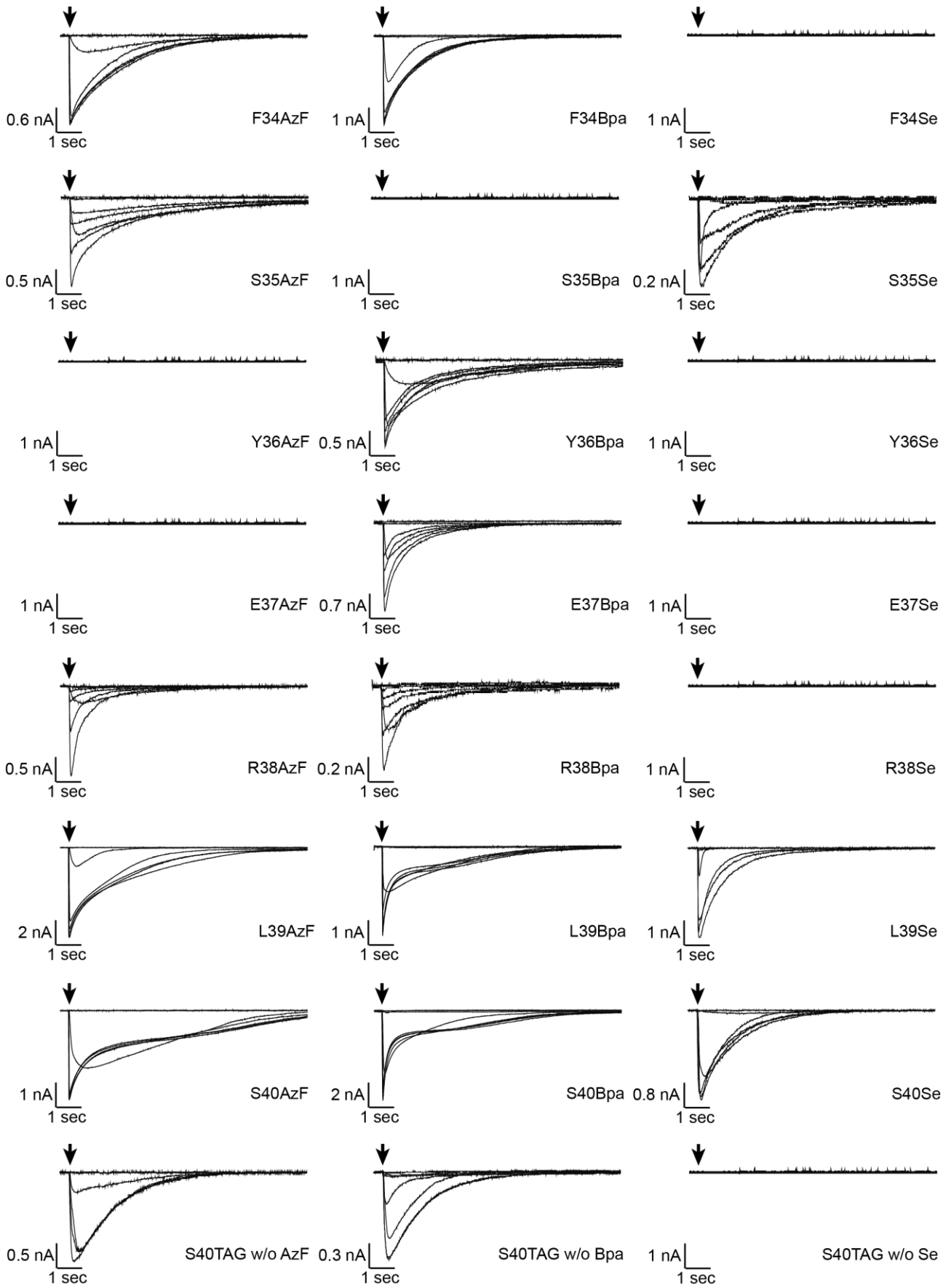
**Figure 1 (next 6 pages):** Representative current traces of all N-terminal ASIC1a variants tested in the APC screen on the SyncroPatch 384PE. All proton-gated responses of the CRC are shown in presence and - in the case of unspecific incorporation - in absence of AzF (left panel), Bpa (middle panel) and Se-AbK (right panel).

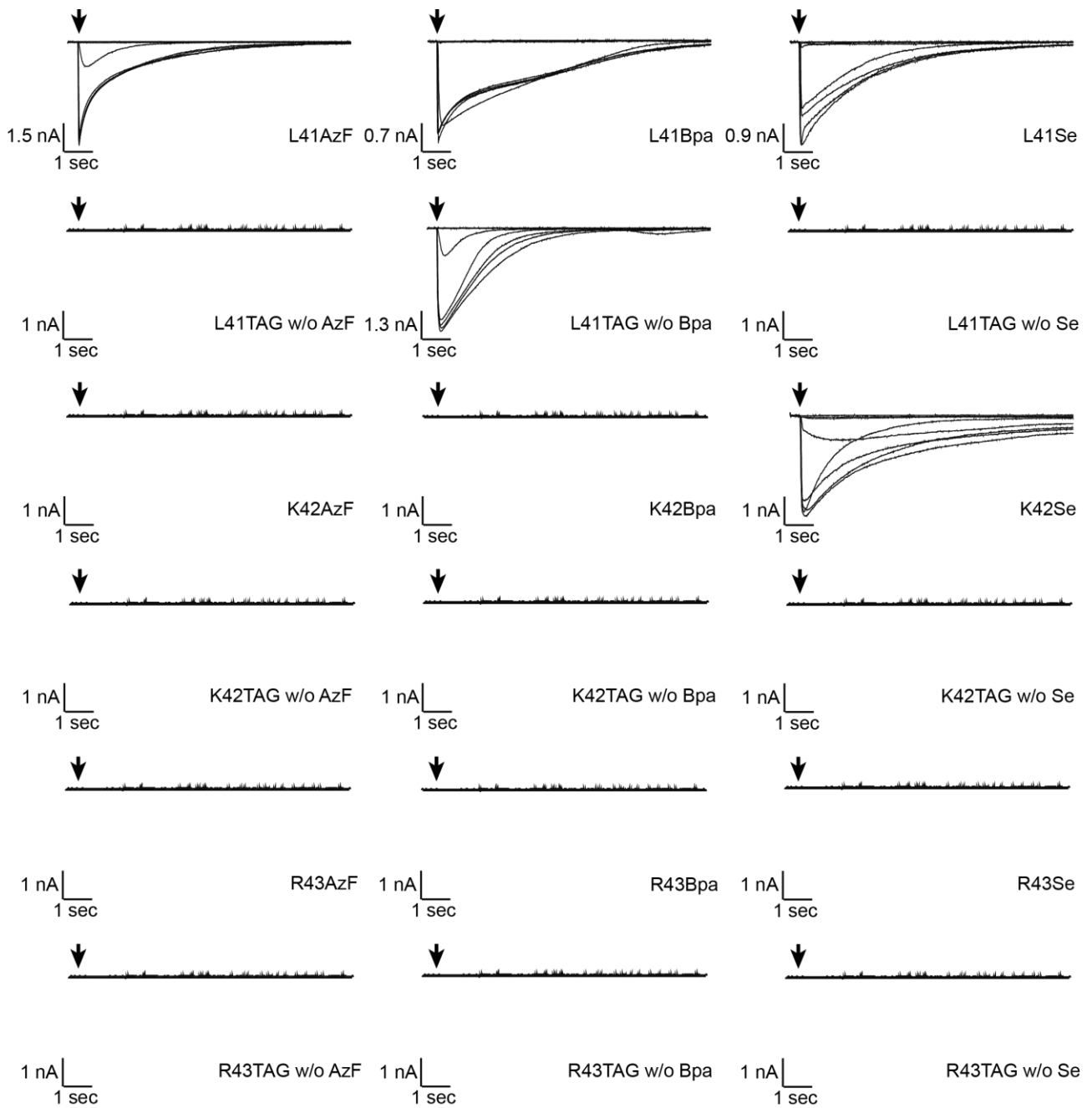




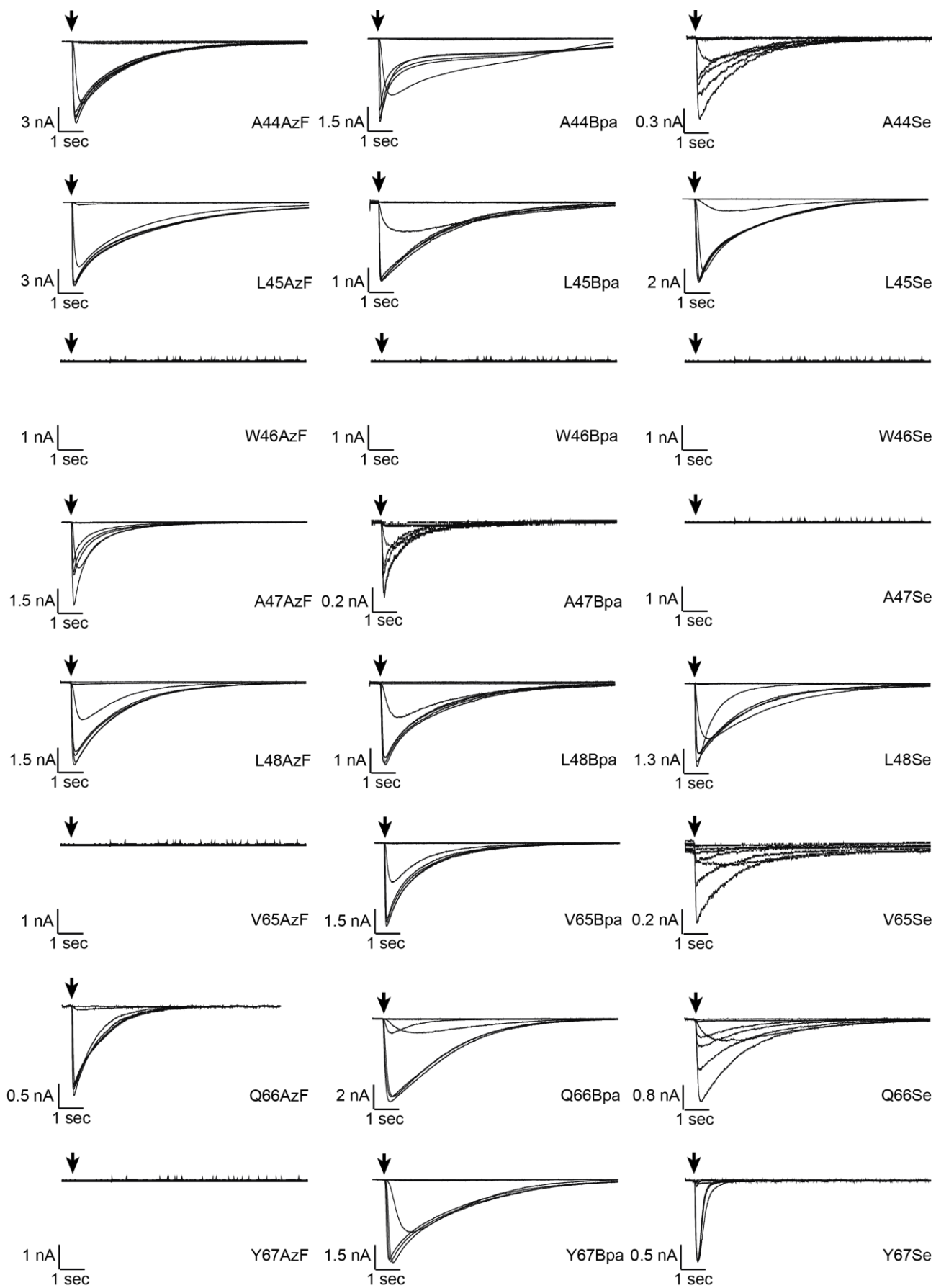




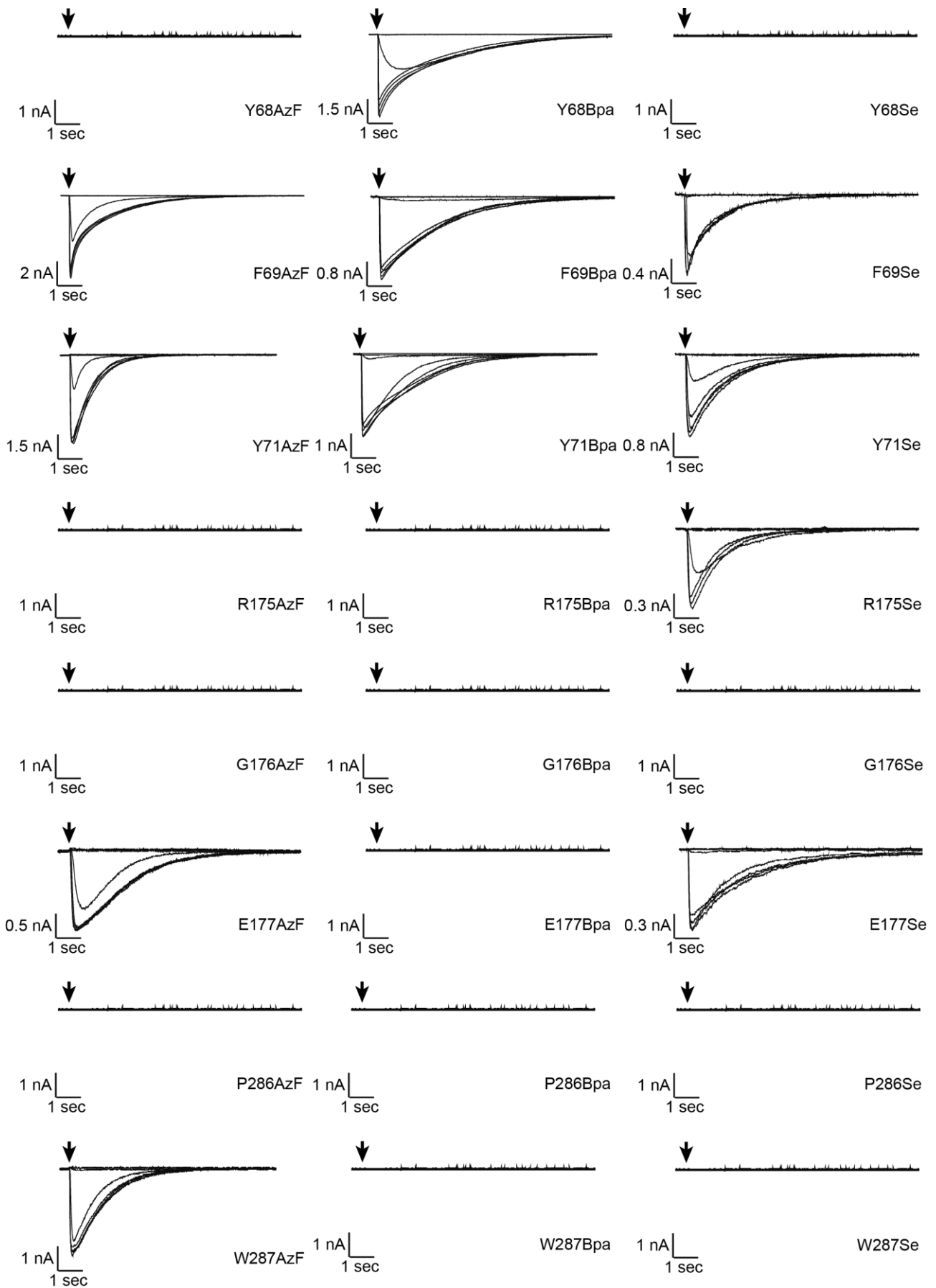


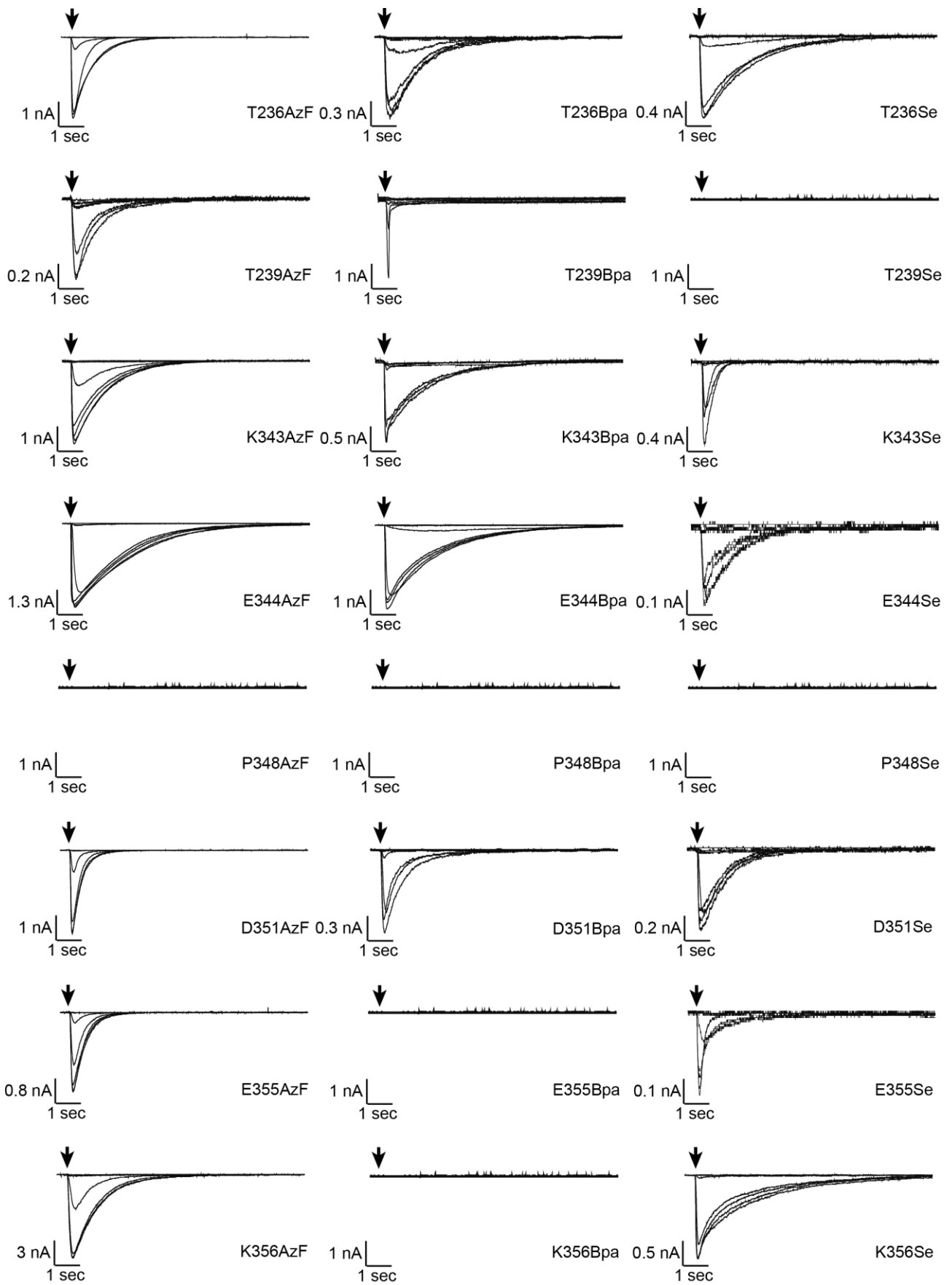


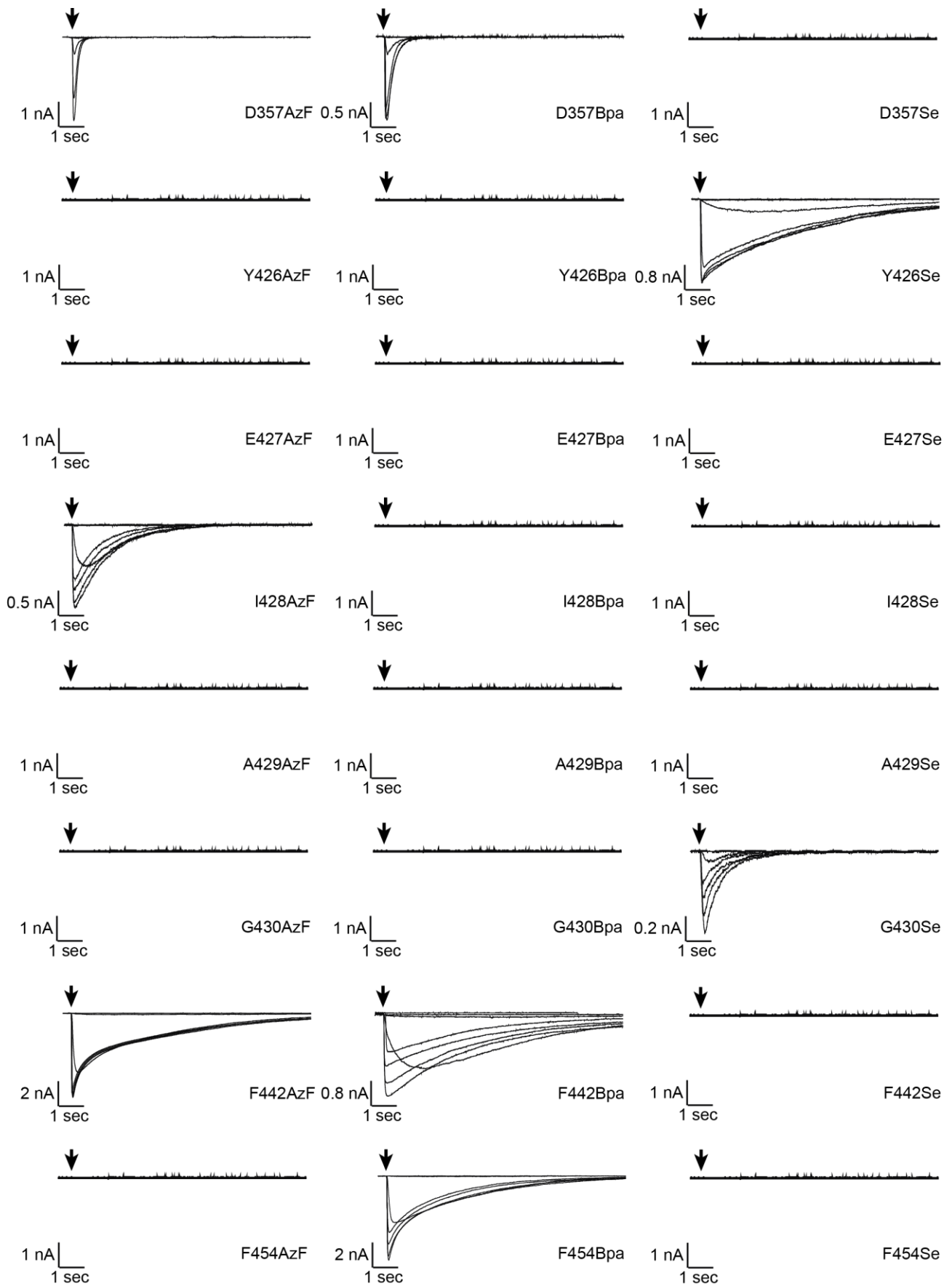
**Figure 2 (next 5 pages):** Representative current traces of all TMD and ECD ASIC1a variants tested in the APC screen on the SyncroPatch 384PE. All proton-gated responses of the CRC are shown in presence of AzF (left panel), Bpa (middle panel) and Se-AbK (right panel).

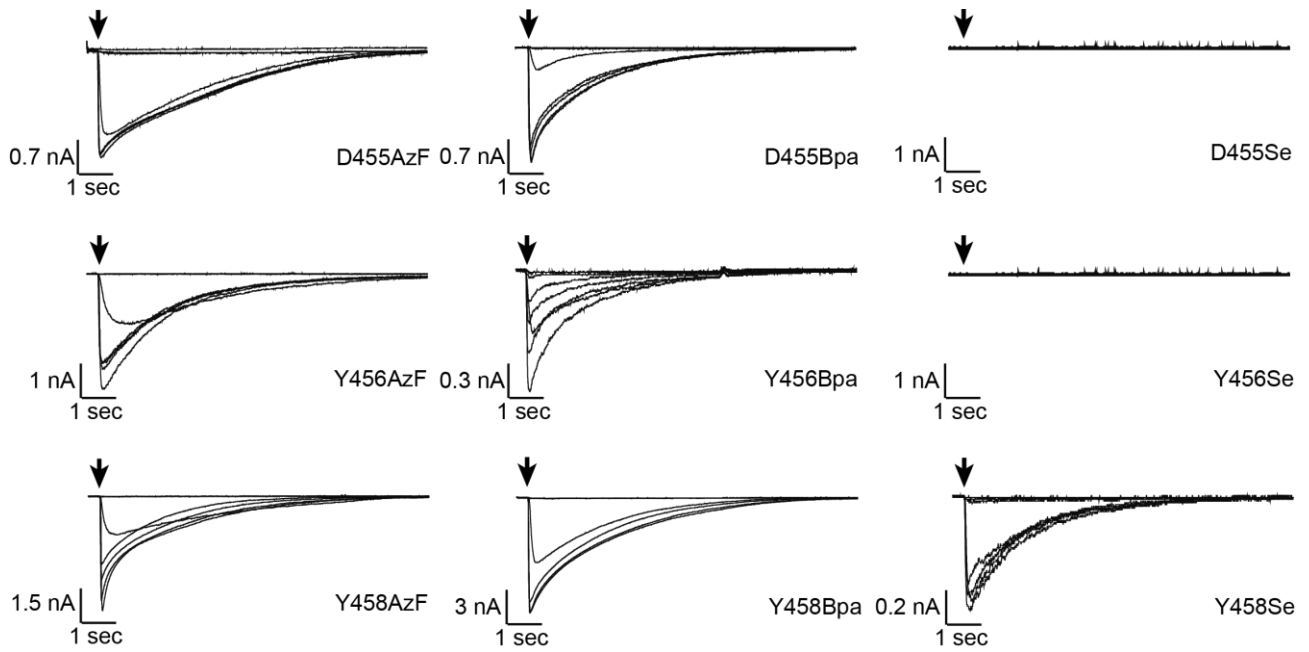




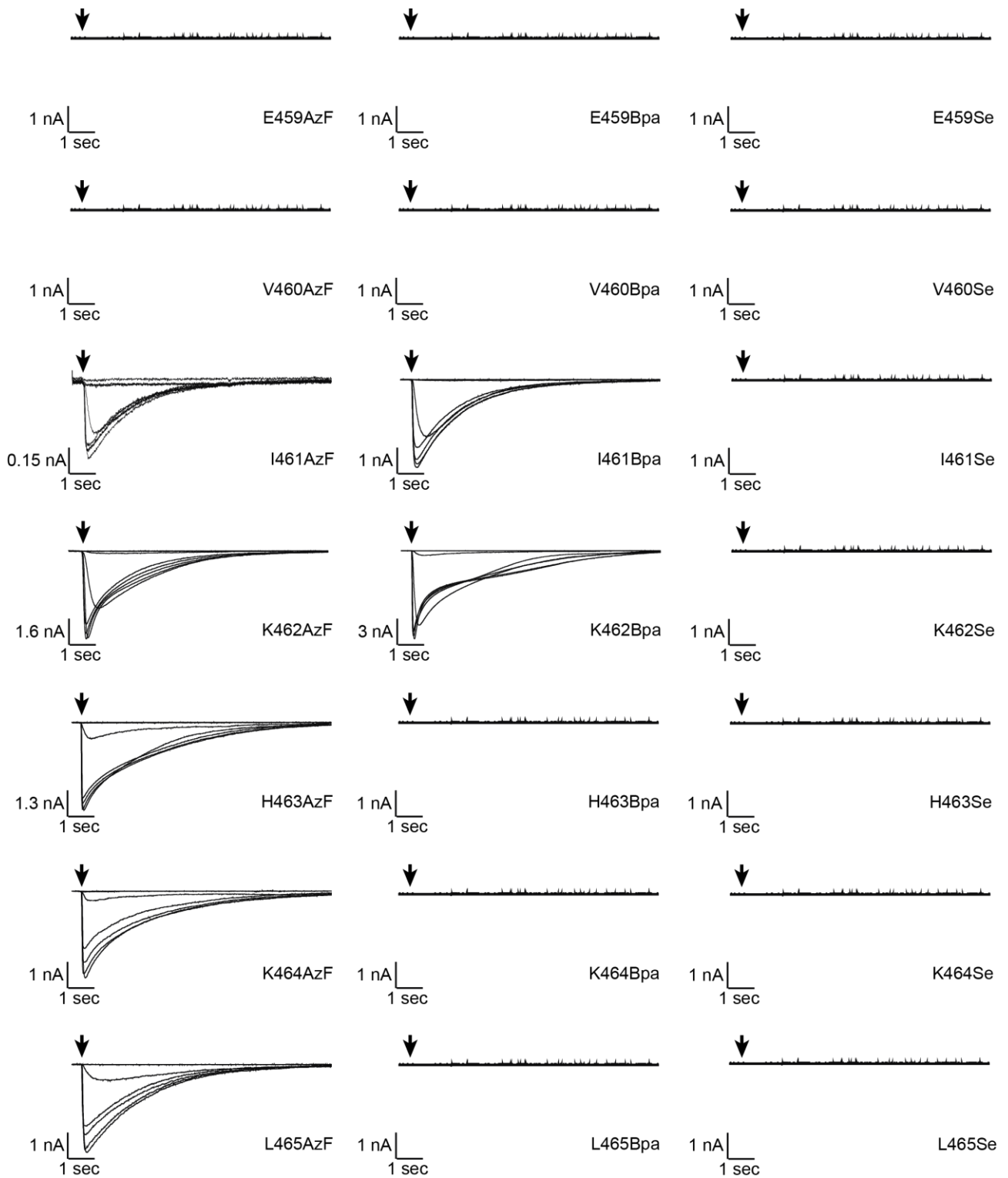


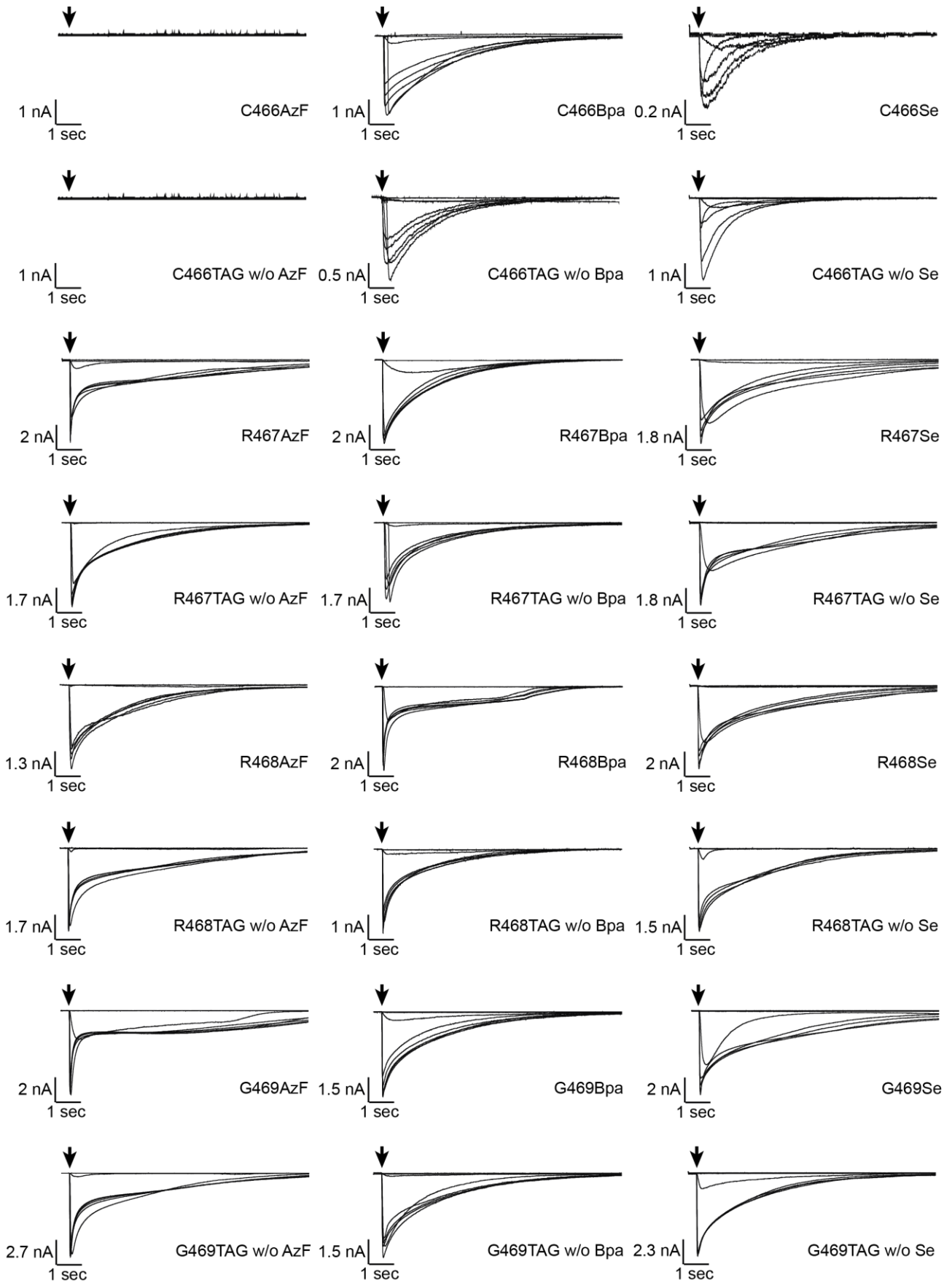


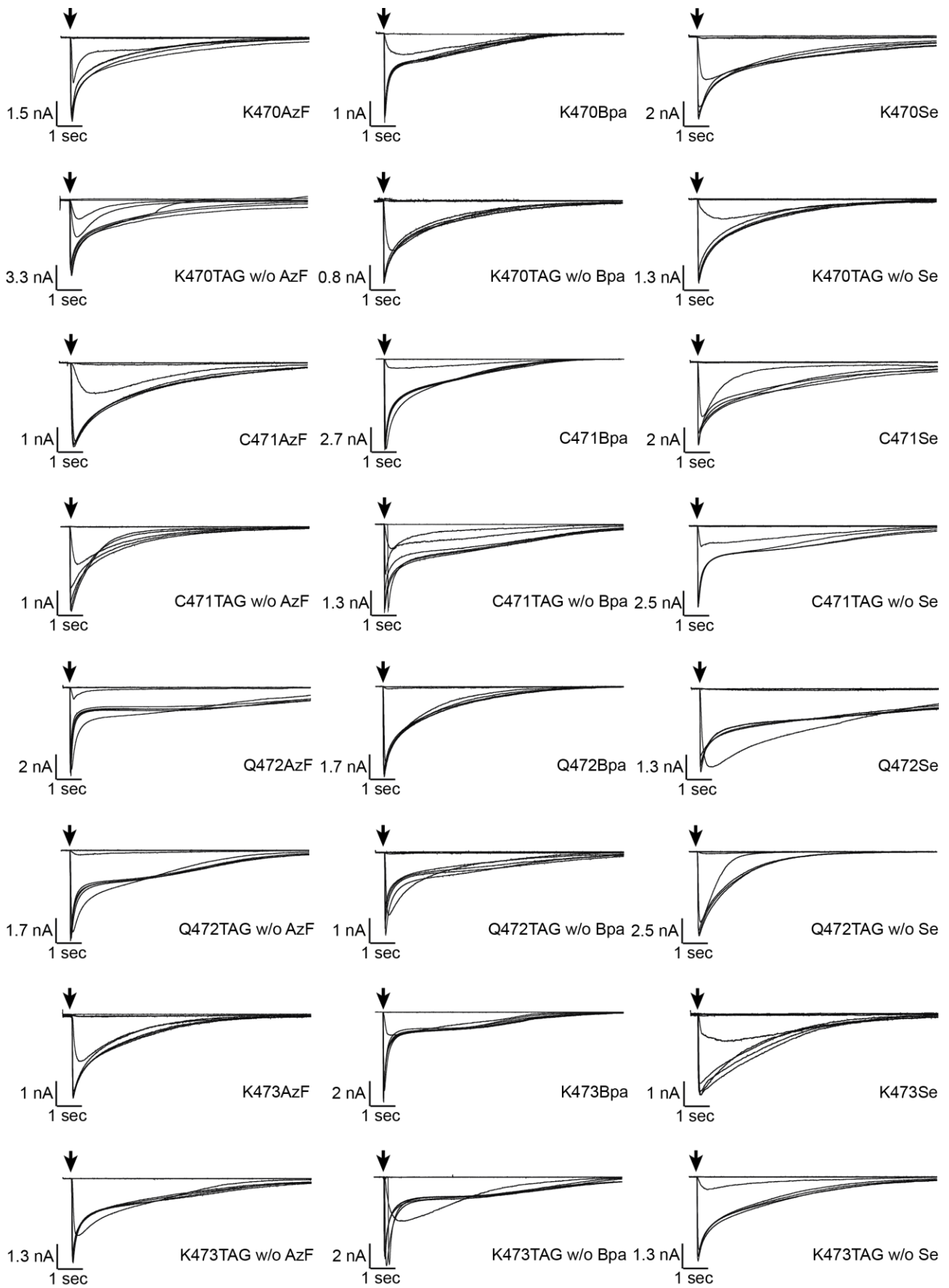


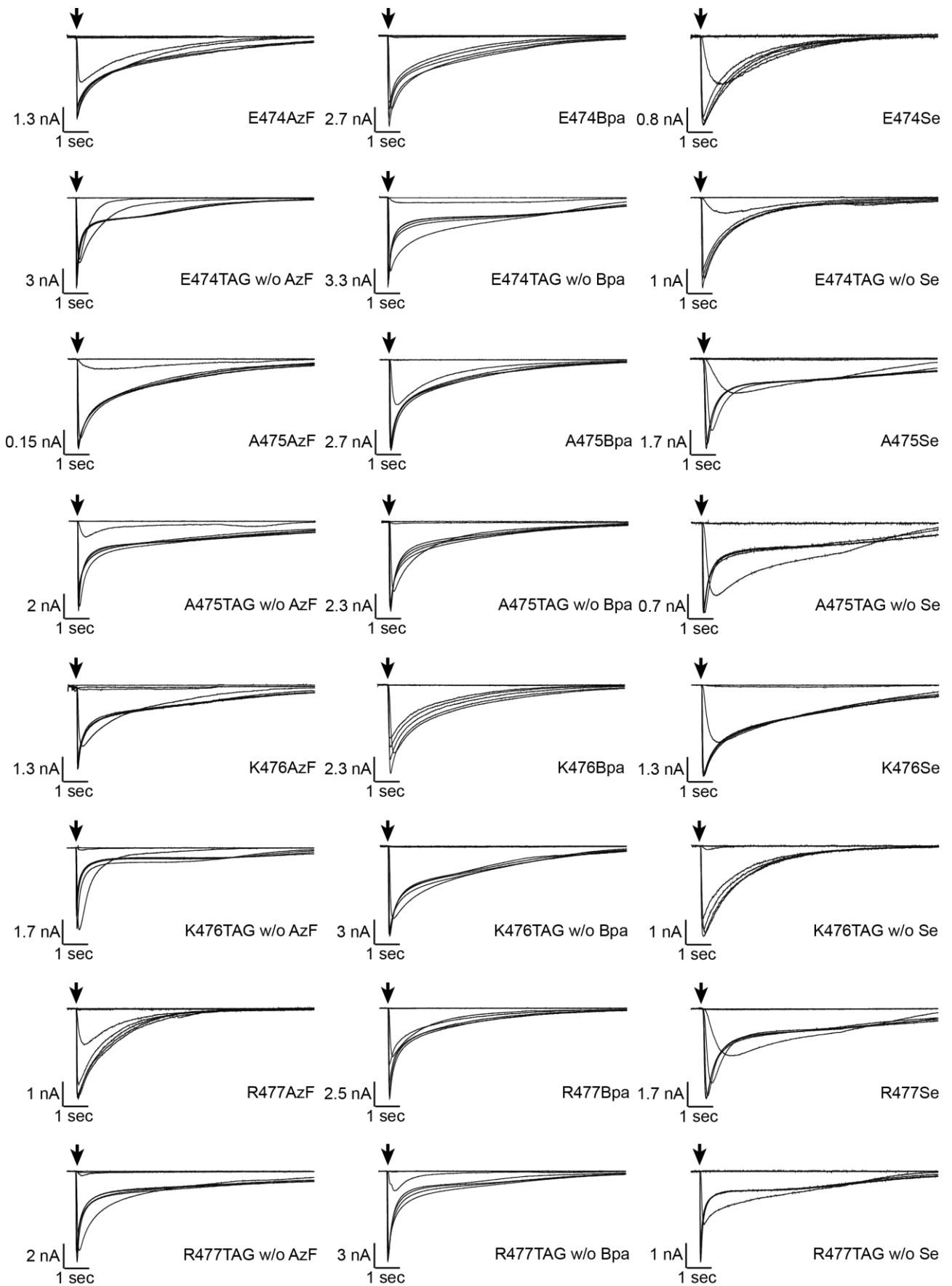


**Figure 3 (next 7 pages):** Representative current traces of all C-terminal ASIC1a variants tested in the APC screen on the SyncroPatch 384PE. All proton-gated responses of the CRC are shown in presence and - in the case of unspecific incorporation - in absence of AzF (left panel), Bpa (middle panel) and Se-AbK (right panel).

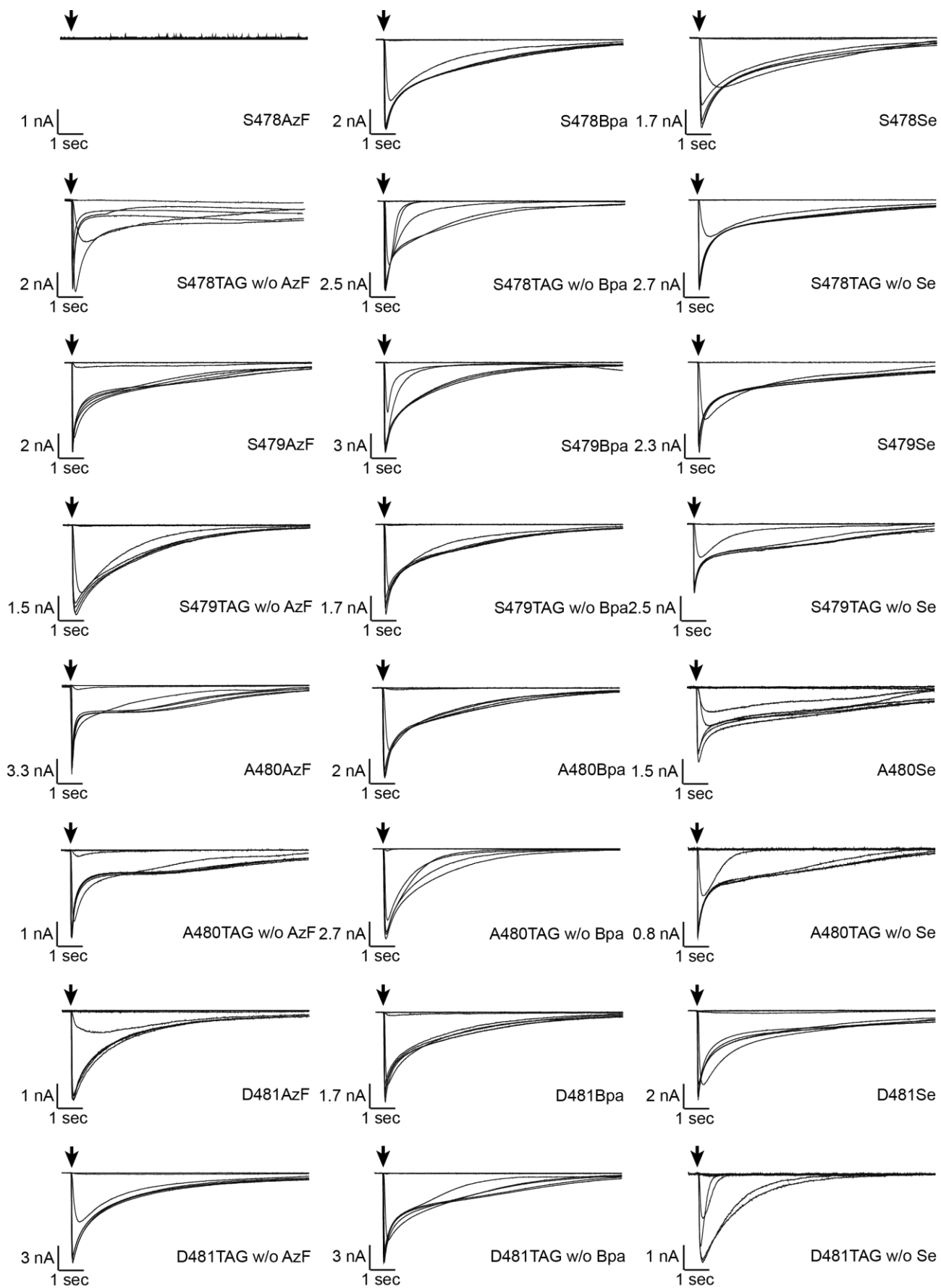


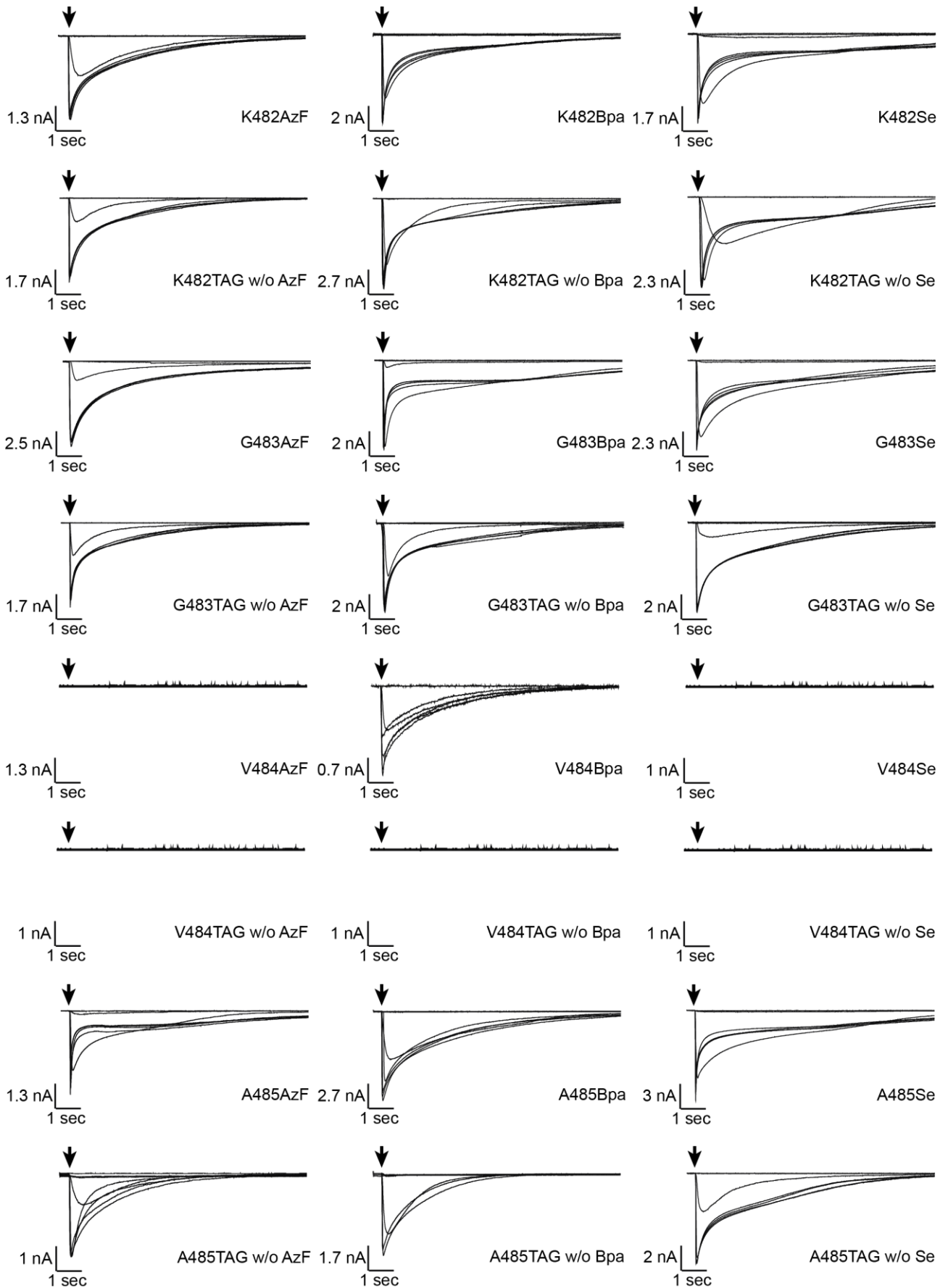


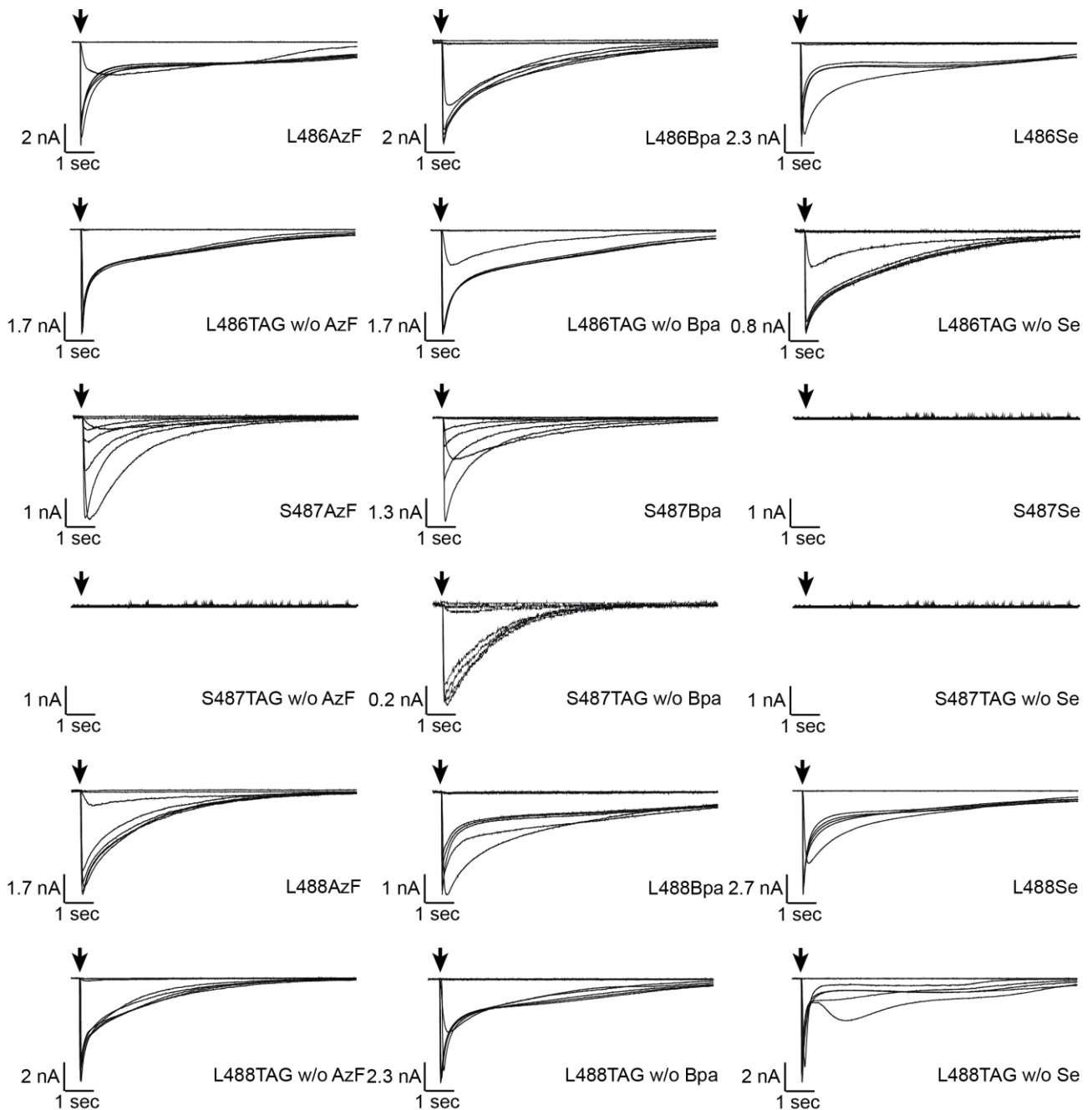




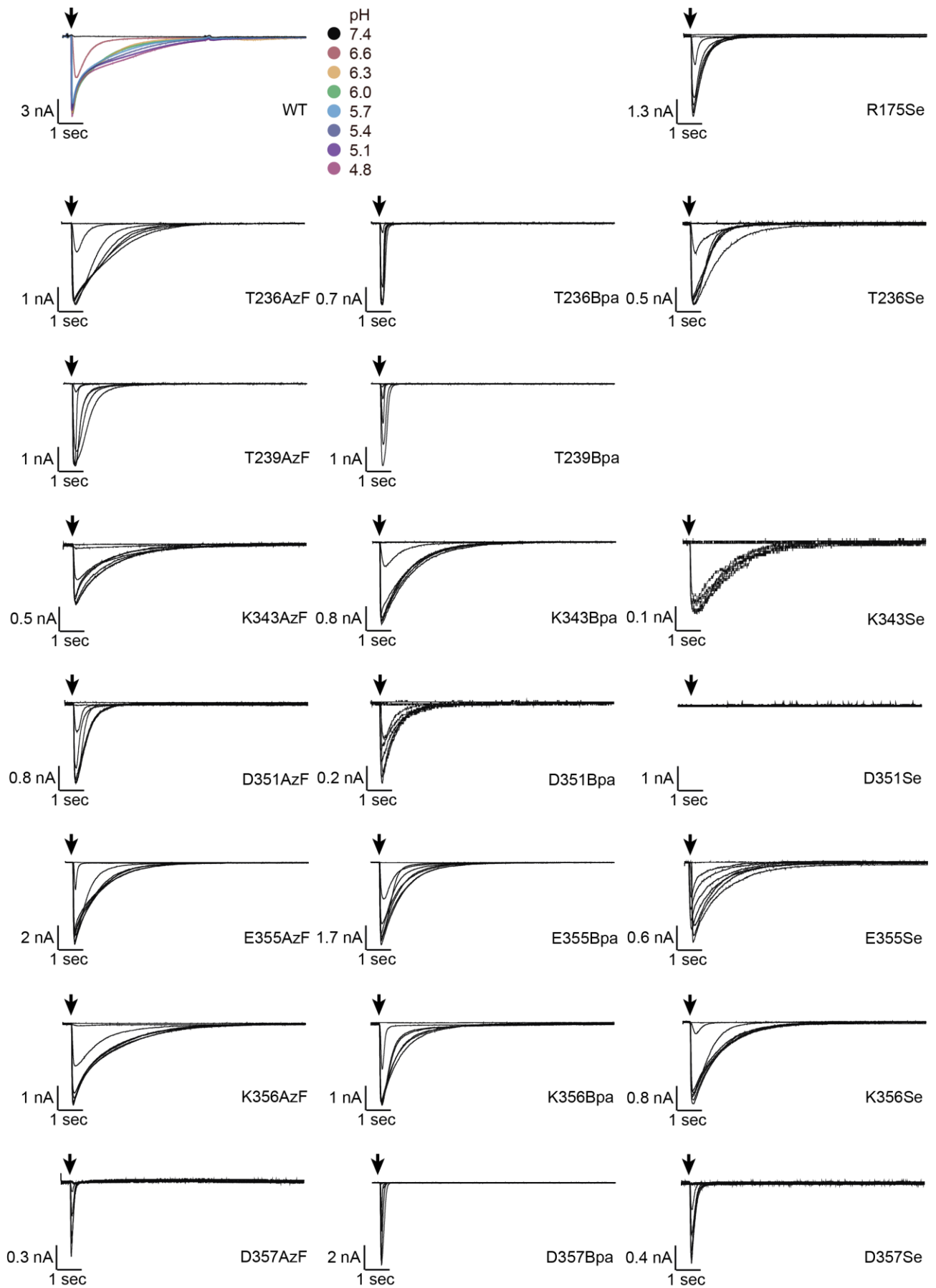


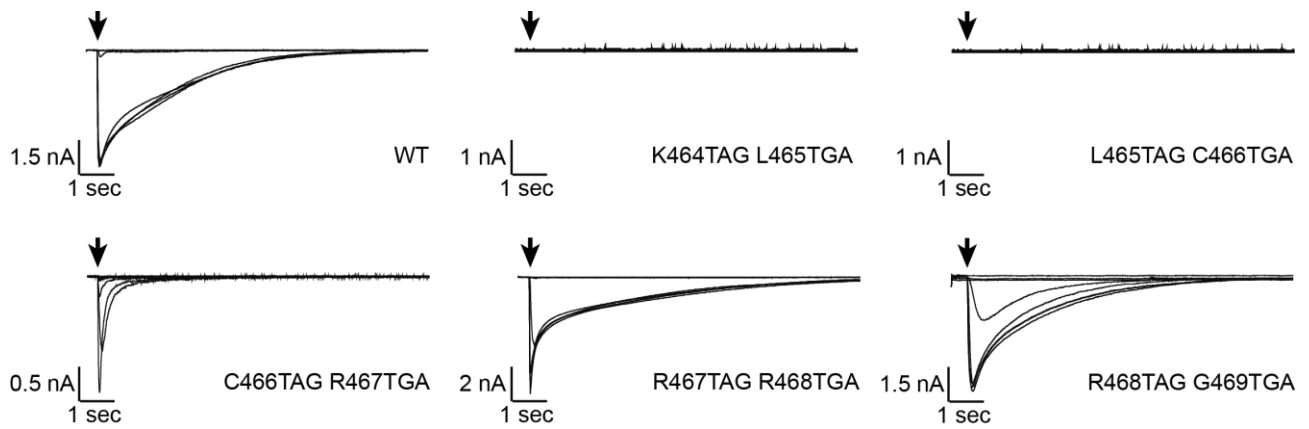




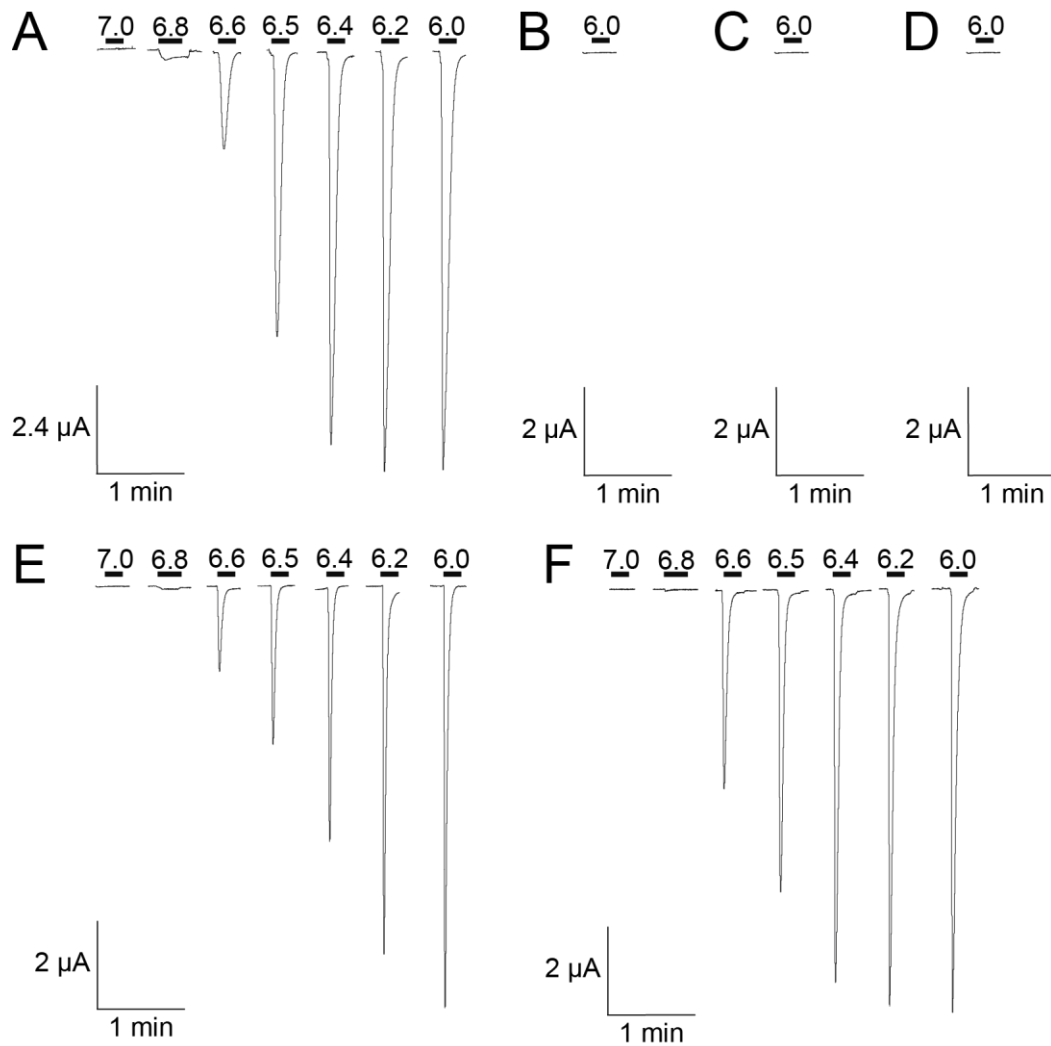


**Figure 4 (next page):** Representative current traces of ASIC1a WT and selected ECD variants that showed shifted  $pH_{50}$  values in the APC screen on the SyncroPatch 384PE and were re-evaluated at lower pH (see upper left panel). All proton-gated responses of the CRC are shown in presence of AzF (left panel), Bpa (middle panel) and Se-AbK (right panel).





**Figure 5:** Representative current traces for the pH-dependent activation of ASIC1a WT and different C-terminally truncated constructs in HEK 293 cells. Traces show all responses of the CRC, cells were kept at pH 7.4 between applications (whole-cell patch-clamp, SyncroPatch 384PE).



**Figure 6:** Representative current traces for the pH-dependent activation of ASIC1a WT and different C-terminally truncated constructs in *X. laevis* oocytes.  $\text{Ca}^{2+}$ -free ND96 at the indicated pH-values was applied for 15 seconds. The basal pH was kept at 7.4 for one minute between applications. For each receptor variant, experiments were performed on at least seven individual oocytes from a minimum of two batches. **A:** WT, **B:** K464TAG L465TGA, **C:** L465TAG C466TGA, **D:** C466TAG R467TGA, **E:** R467TAG R468TGA, **F:** R468TAG G469TGA.

**Figure 7 (next page):** Representative current traces of all ASIC1a variants tested for desensitization on the SyncroPatch 384PE. Shown are all proton-gated responses of the SSD curve with the final control sweep after conditioning with pH 7.6 coloured in red.

