SUPPLEMENTARY MATERIAL

Expression, function and trafficking of the human ABCG2 multidrug transporter containing mutations in an unstructured cytoplasmic loop

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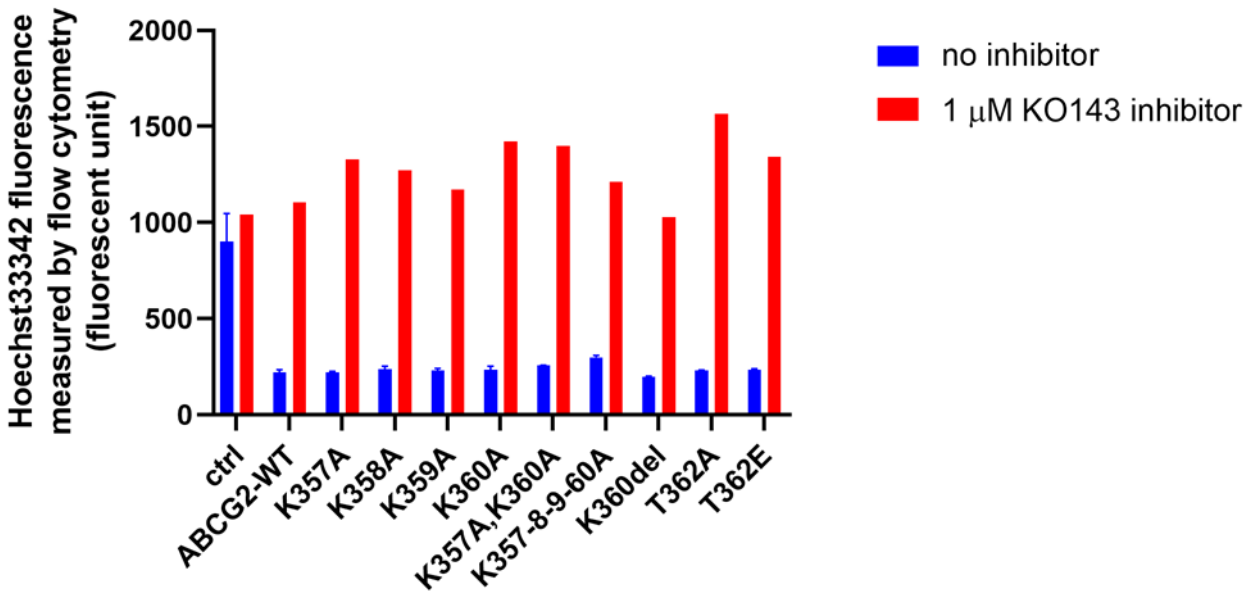
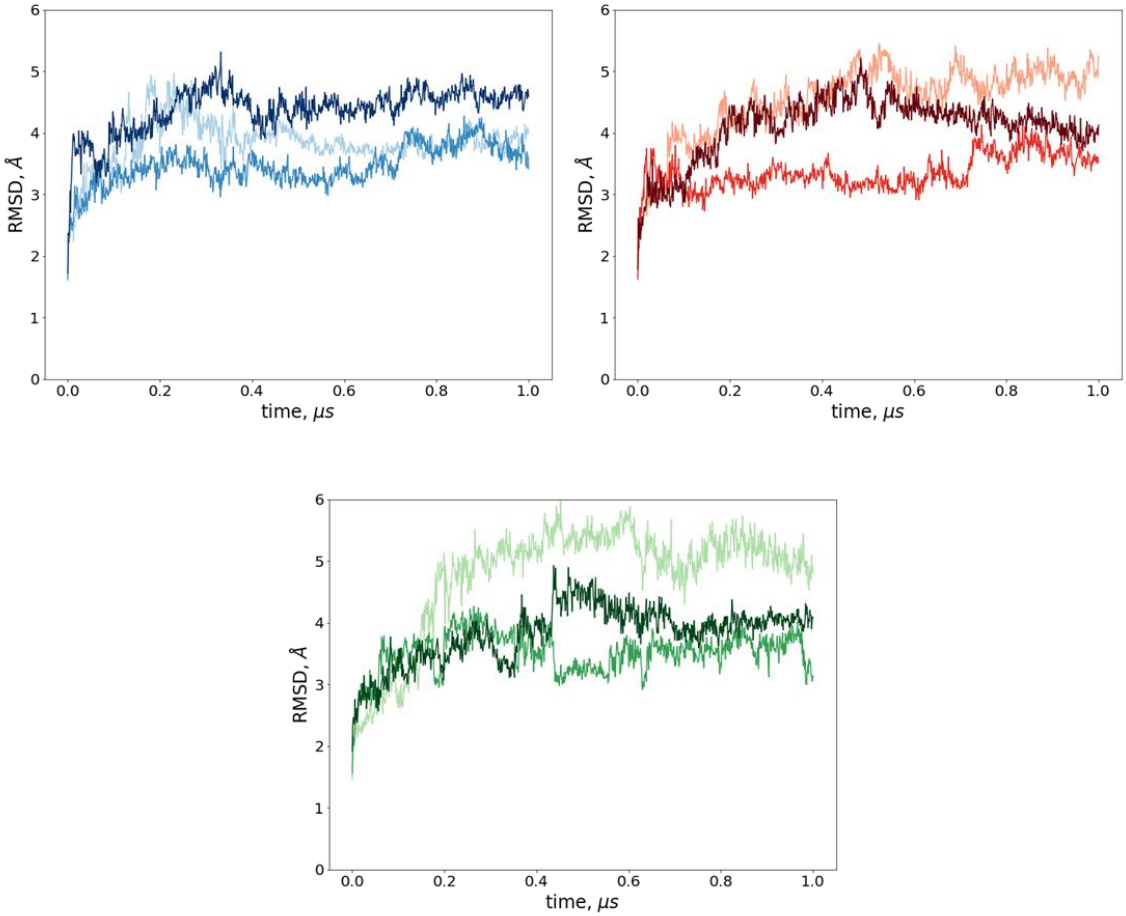
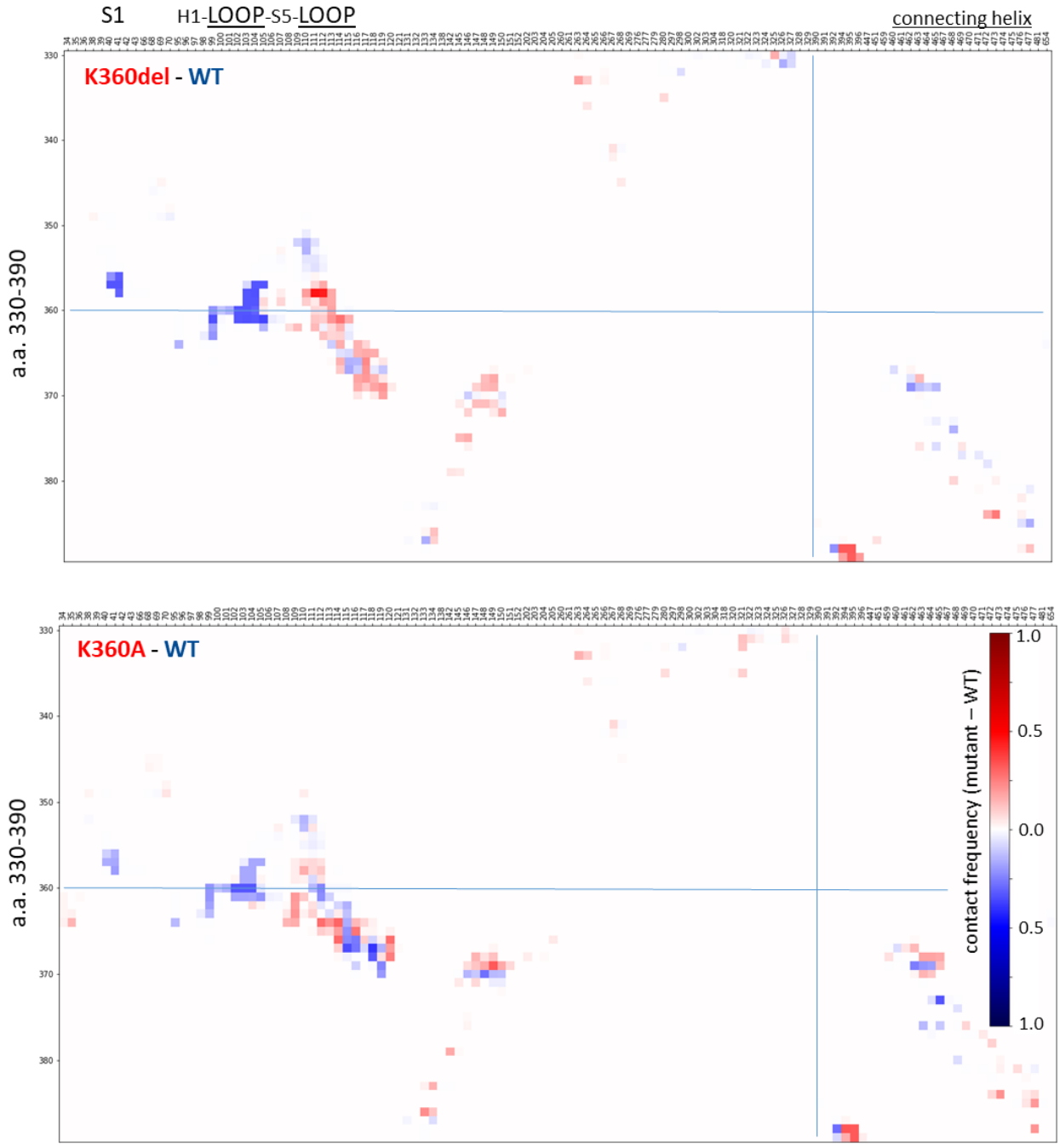


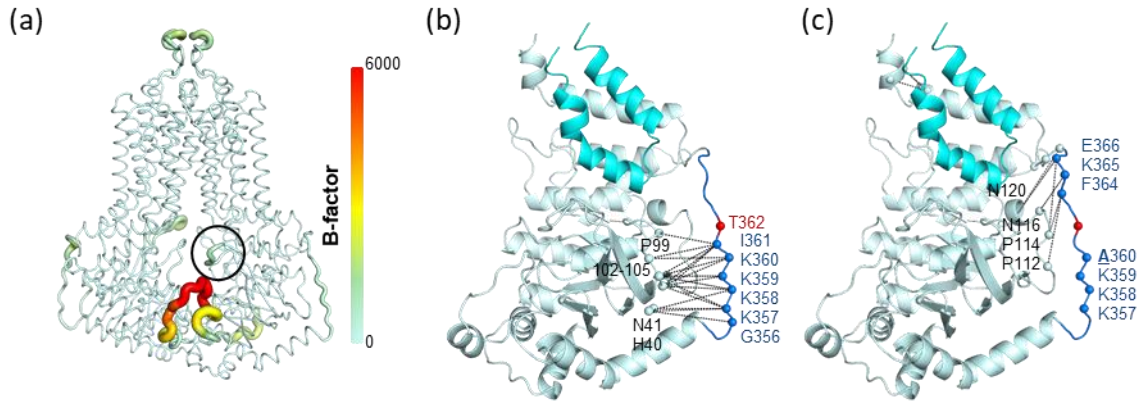
Supplementary Figure S1. Hoechst33344 dye accumulation in HEK293 cells expressing the ABCG2 variants.



Supplementary Figure S2. Root mean square deviation (RMSD) of conformations from the initial structure. The values indicate stable systems of the size of ABCG2, with a mobile loop between NBD and TMD, which contribute to the majority of differences from the starting conformation (see Figure 6a and Figure S4a).



Supplementary Figure S3. Characteristic pairwise residue contacts between the a.a. 330-390 and the rest of the protein. Contact maps were calculated (between Cα with 7.5 Å cutoff) for each construct. To display the most characteristic interactions, the WT map was subtracted from the mutant and only those residues are shown which contact the loop. The contacts present with higher frequencies in the WT are blue and the contacts present more in the K360del and K360A are red. The horizontal and blue lines labels indicate the position 360 (or 361 in K360del) and the location of a.a. 330-390, respectively. This latter region is not shown in the X axis to hide self-interactions for clarity. Both mutations decrease the interactions of the disordered loop (a.a. 354-370) with the first β-strand of NBD1 (S1) and with the loop between the H1 helix containing Walker A and β-strand S5. The interactions of this 354-370 region with the loop next to S5 are characteristic more for K360del than for the K360A or WT.



Supplementary Figure S4. (**a**) B-factors calculated from simulations are displayed in the context of the K360A structure. Although the flexibility of the linker region did not increase in K360A ABCG2, the RI exhibited higher dynamics (black circle) than in the K360del mutant (Figure 6). (**b, c**) The most characteristic contacts of WT (b) and K360A (c) when compared to each other (|difference|>0.25) are shown as black lines between the Cα (spheres) of residue pairs. Contacts were averaged along simulations in protomers, thus indicated only on a single NBD-linker part of the structure. Pale cyan: NBD; cyan: coupling helix region (a.a. 441-483).