

Supporting Information

Structural Insights of Stereospecific Inhibition of Human

Acetylcholinesterase by VX and Subsequent Reactivation by

HI-6

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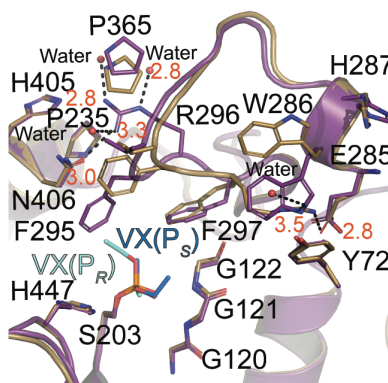
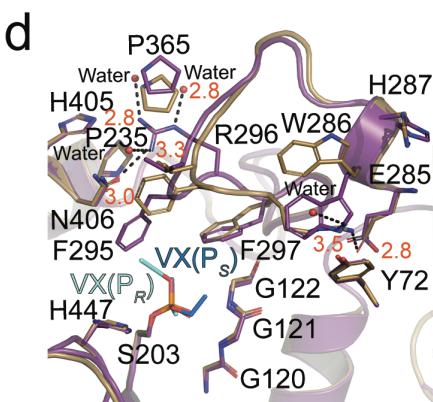
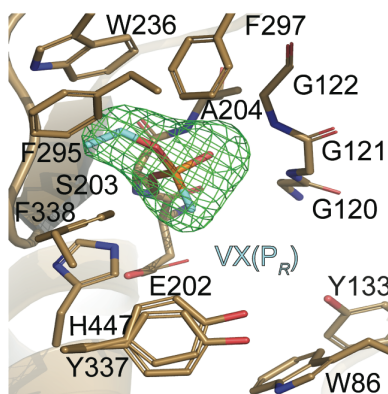
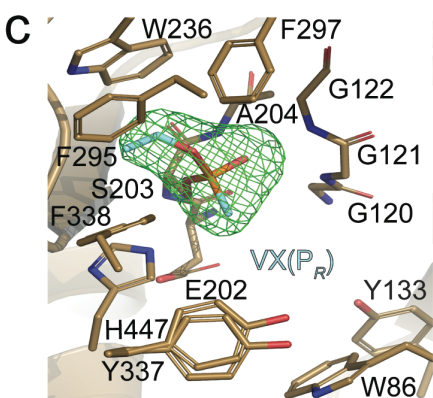
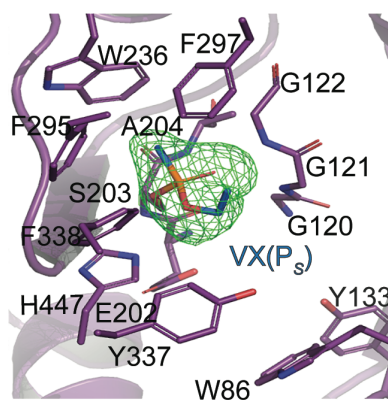
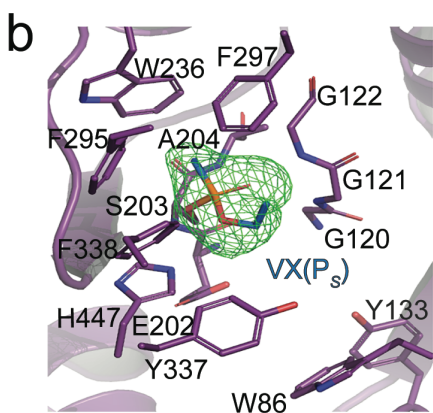
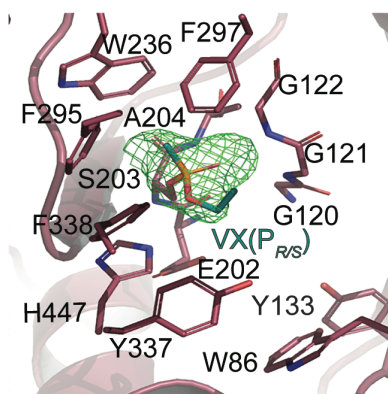
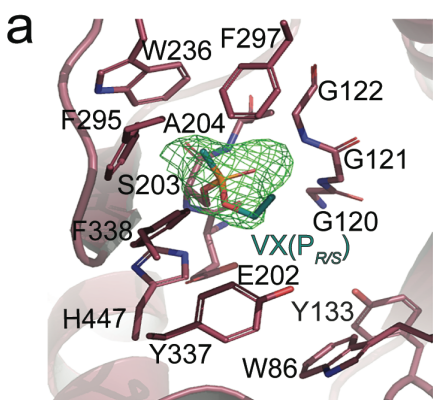
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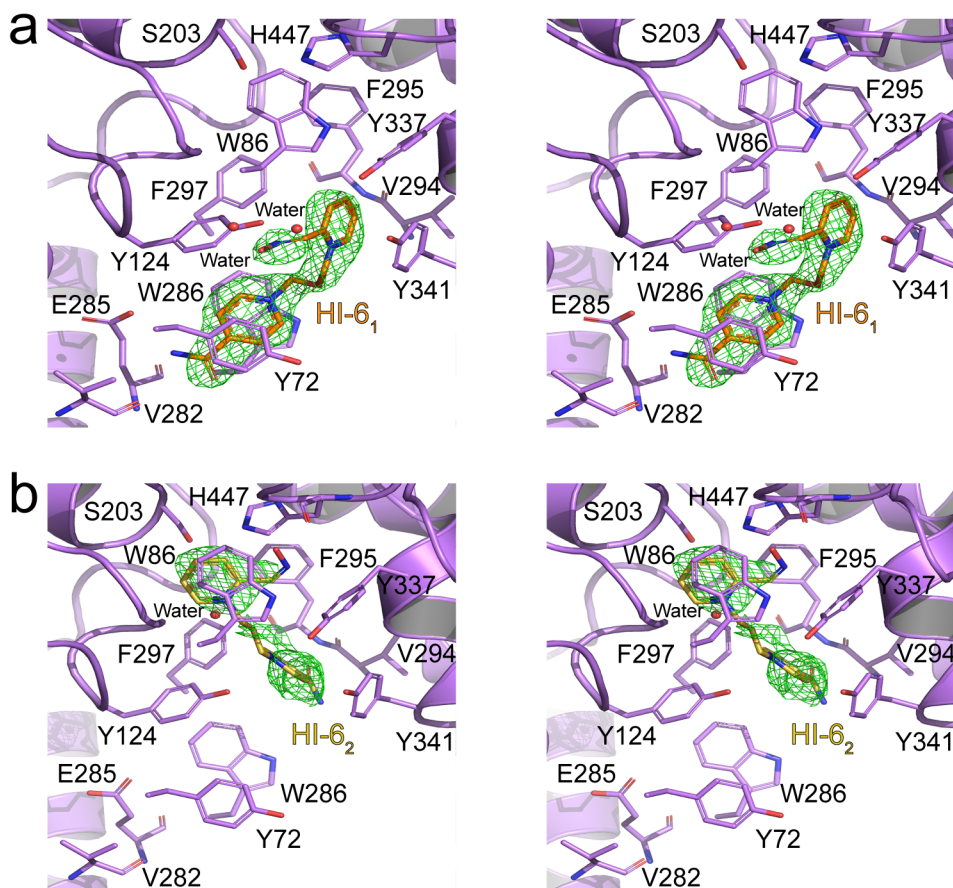
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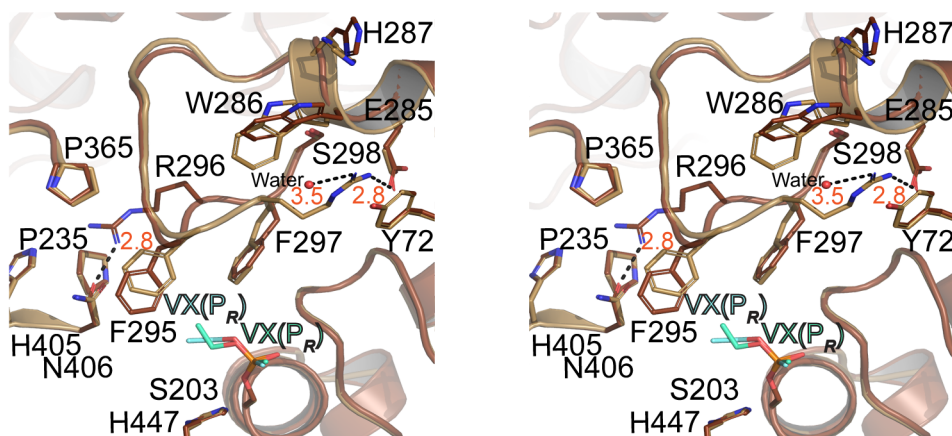
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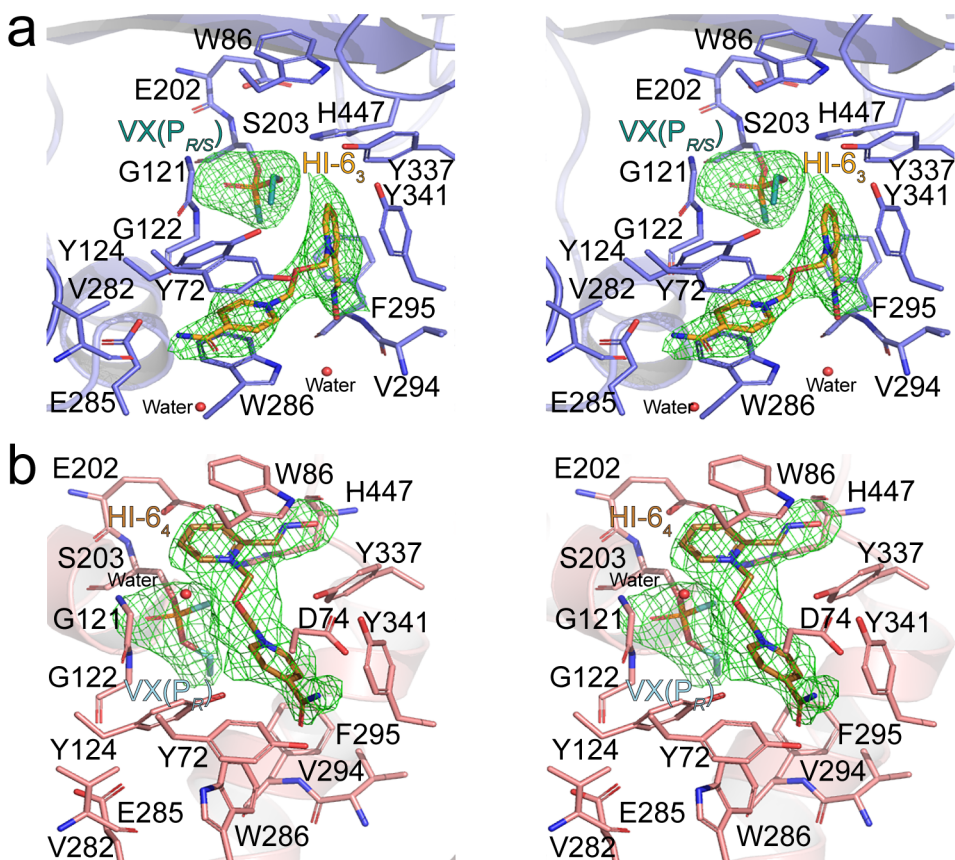
**Figure S1. Crystal Structures of Human Acetylcholinesterase in complex with Stereoisomers of Nerve Agent, VX.** (a) Wall-eyed stereoview of P<sub>R/S</sub>-VX (dark teal) bound to the active site of hAChE (dark pink) with hAChE F<sub>o</sub> – F<sub>c</sub> density scaled to 3 $\sigma$  (green mesh) (PDB 6CQZ). (b) Wall-eyed stereoview of P<sub>S</sub>-VX (marine blue) bound to the active site of hAChE (violet purple) with hAChE F<sub>o</sub> – F<sub>c</sub> density scaled to 3 $\sigma$  (green mesh) (PDB 6CQT). (c) Wall-eyed stereoview of P<sub>R</sub>-VX (aquamarine) bound to the active site of hAChE (sand) with hAChE F<sub>o</sub> – F<sub>c</sub> density scaled to 3 $\sigma$  (green mesh) (PDB 6CQX). (d) Wall-eyed stereo view of an overlay of the acyl loop of hAChE with P<sub>R</sub>-VX (sand brown) and P<sub>S</sub>-VX (violet purple). The black dashed lines represent hydrogen bonds. The AChE residues are represented by black letters, while the red letters are the distances.



**Figure S2. Crystal structures of HI-6 bound in the active site of hAChE.** (A/B) Wall-eyed stereoview of the active sites of hAChE (bright purple) on chains A & B bound to HI-6 (orange and dark yellow respectively).  $F_o - F_c$  density scaled to  $3\sigma$  (green mesh) (PDB 6CQU). Waters are shown as red spheres and black dashed lines indicate hydrogen bond interactions.

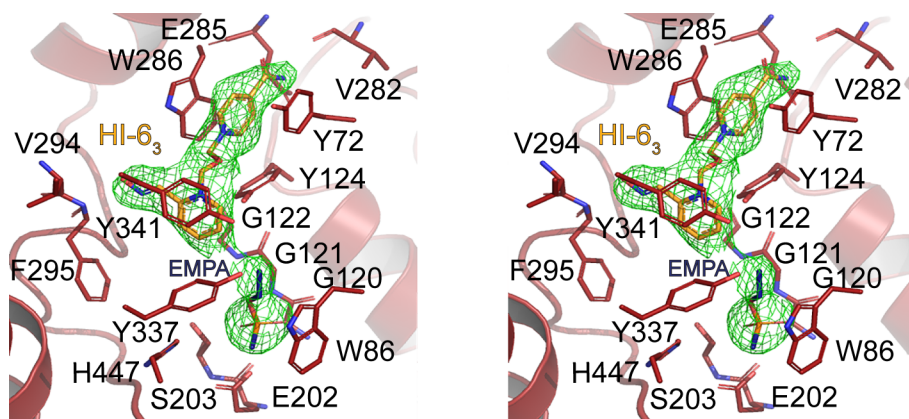


**Figure S3. Overlay of the Acyl Loop Region of Human Acetylcholinesterase Inhibited by VX- $P_R$ .** Wall-eyed stereo view of an overlay of the acyl loop of hAChE chain B (sand brown) and Chain A (chocolate) inhibited by  $P_R$ -VX (aquamarine; sea foam green). The black dashed lines represent hydrogen bonds. The AChE residues are represented by black letters, while the red letters are the distances.



**Figure S4. The Active Site of human Acetylcholinesterase with Stereoisomers of VX and the Reactivator, HI-6.** (a) Wall-eyed stereoview of P<sub>R/S</sub>-VX (dark teal) with reactivator HI-6 (bright orange) bound to the active site of hAChE (slate) with hAChE F<sub>o</sub> – F<sub>c</sub> density scaled to 3σ (green mesh) (PDB 6CQW). (b) Wall-eyed stereoview of P<sub>R</sub>-VX (aquamarine) with the reactivator HI-6 (copper-colored) bound to the active site of hAChE (salmon pink) with hAChE F<sub>o</sub> – F<sub>c</sub> density scaled to 3σ (green mesh) (PDB 6CQV).





**Figure S5. The Active Site of human Acetylcholinesterase with Post-Catalysis HI-6 and a by-product, EMPA.** Wall-eyed stereoview of hAChE active site with reactivator, HI-6 (bright orange), and unbound VX (indigo), with hAChE  $F_o - F_c$  density scaled to  $3\sigma$  (green mesh) (PDB 6CQY).