## SUPPLEMENTARY MATERIAL

**A Boron Delivery Antibody (BDA) with boronated specific residues: new perspectives in Boron Neutron Capture Therapy.**

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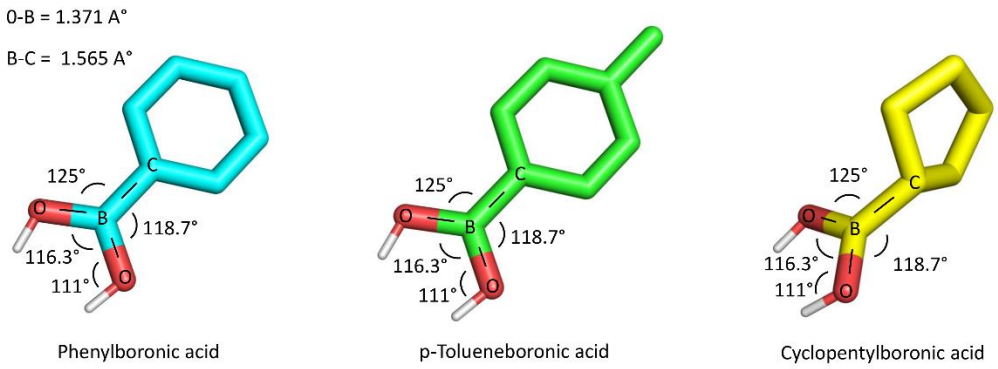
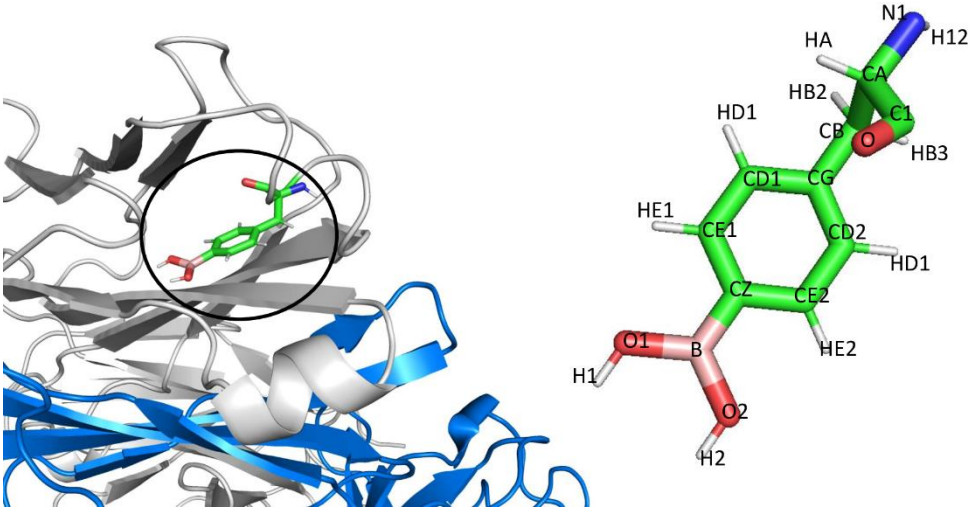



**Supplementary Figure S1.** Fragment probes parametrization. Carbon-boron-oxygen and boron-oxygen angle degrees and carbon -boron and boron-oxygen bond lengths are reported for each fragment probe.



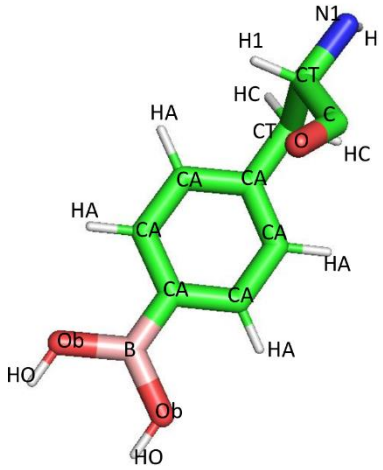

**Supplementary Figure S2.** Chain A BPA186. Graphical representation of BPA inserted within Cetuximab (left). Atom names of the new modified residue (right).

**Supplementary Table S1.** Bond length: atom name referred to Supplementary Figure 2.

| Bond | Å |
|---|---|
| O1-B | 1.371 |
| O2-B | 1.371 |
| B-CZ | 1.565 |

**Supplementary Table S2.** Angles: atom names referred to Supplementary Figure 2.

| Angle | (◦) |
|---|---|
| O1-B-CZ | 125 |
| CZ-B-O2 | 118.7 |
| O1-B-O2 | 116.3 |
| B-O1-H1 | 111 |
| B-O2-H2 | 111 |




**Supplementary Figure S3.** BPA atom names used for parameterization in Amber18 Molecular Dynamics package.

**Supplementary Table S3.** Bond parameters (*This study, ** Ref [18]).

| Bond | Kr (kcal (mol·Å$^2$ ) $^{-1}$) | req (Å) | Source |
|---|---|---|---|
| H -N | 434.00 | 1.010 | * |
| CT-N | 337.00 | 1.449 | * |
| CT-CT | 310.00 | 1.526 | * |
| CT-H1 | 340.00 | 1.090 | * |
| C -CT | 317.00 | 1.522 | * |
| CA-CT | 317.00 | 1.510 | * |
| CT-HC | 340.00 | 1.090 | * |
| CA-CA | 469.00 | 1.400 | * |
| CA-HA | 367.00 | 1.080 | * |
| C -O | 570.00 | 1.229 | * |
| B -CA | 218.951 | 1.559 | ** |
| HO-Ob | 546.139 | 0.970 | ** |
| Ob-B | 350.221 | 1.372 | ** |

**Supplementary Table S4.** Angle parameters (*This study, ** Ref [18]).

| Angle | Kθ (kcal/(mol·radian$^2$ )) | θeq (◦) | Source |
|---|---|---|---|
| CT-CT-N | 80.000 | 109.700 | * |
| H1-CT-N | 50.000 | 109.500 | * |
| C -CT-N | 63.000 | 110.100 | * |
| CT-N -H | 50.000 | 118.040 | * |
| C -CT-H1 | 50.000 | 109.500 | * |
| CA-CT-CT | 63.000 | 114.000 | * |
| CT-CT-HC | 50.000 | 109.500 | * |
| CT-C -O | 80.000 | 120.400 | * |
| CT-CT-H1 | 50.000 | 109.500 | * |
| C -CT-CT | 63.000 | 111.100 | * |
| HC-CT-HC | 35.000 | 109.500 | * |
| CA-CA-CT | 70.000 | 120.000 | * |
| CA-CT-HC | 50.000 | 109.500 | * |
| CA-CA-CA | 63.000 | 120.000 | * |
| CA-CA-HA | 50.000 | 120.000 | * |
| CA-CA-B | 127.380 | 120.970 | ** |
| CA-B -Ob | 89.005 | 118.100 | ** |
| HO-Ob-B | 50.184 | 110.550 | ** |
| Ob-B -Ob | 99.046 | 122.400 | ** |

**Supplementary Table S5.** Dihedral parameters (*This study, ** Ref [18]).

| Dihedral | Divider | Vn (kcal/ mol) | γ | n | Source |
|---|---|---|---|---|---|
| CA-CT-CT-N | 9 | 1.400 | 0.000 | 3.000 | * |
| HC-CT-CT-N | 9 | 1.400 | 0.000 | 3.000 | * |
| O -C -CT-N | 6 | 0.000 | 0.000 | 2.000 | * |
| CT-CT-N -H | 6 | 0.000 | 0.000 | 2.000 | * |
| H1-CT-N -H | 6 | 0.000 | 0.000 | 2.000 | * |
| C -CT-N -H | 6 | 0.000 | 0.000 | 2.000 | * |
| CA-CA-CT-CT | 6 | 0.000 | 0.000 | 2.000 | * |
| O -C -CT-CT | 6 | 0.000 | 0.000 | 2.000 | * |
| CA-CA-CA-CT | 4 | 4.500 | 180.000 | 2.000 | * |
| CT-CA-CA-HA | 4 | 4.500 | 180.000 | 2.000 | * |
| CA-CT-CT-H1 | 9 | 1.400 | 0.000 | 3.000 | * |
| C -CT-CT-CA | 9 | 1.400 | 0.000 | 3.000 | * |
| CA-CA-CA-CA | 4 | 4.500 | 180.000 | 2.000 | * |
| CA-CA-CA-HA | 4 | 4.500 | 180.000 | 2.000 | * |
| CA-CA-CT-HC | 6 | 0.000 | 0.000 | 2.000 | * |
| HA-CA-CA-HA | 4 | 4.500 | 180.000 | 2.000 | * |
| H1-CT-CT-HC | 9 | 1.400 | 0.000 | 3.000 | * |
| C -CT-CT-HC | 9 | 1.400 | 0.000 | 3.000 | * |
| O -C -CT-H1 | 1 | 0.800 | 0.000 | -1.000 | * |
| O -C -CT-H1 | 1 | 0.000 | 0.000 | -2.000 | * |
| O -C -CT-H1 | 1 | 0.080 | 180.000 | 3.000 | * |
| CA-CA-B -Ob | 1 | 0.500 | 180.000 | 2.000 | ** |

| | | | | | |
|---|---|---|---|---|---|
| HO-Ob-B -CA | 1 | 2.700 | 180.000 | 2.000 | ** |
| HO-Ob-B -Ob | 1 | 2.700 | 180.000 | 2.000 | ** |
| Ob-B -Ob-HO | 1 | 2.700 | 180.000 | 2.000 | ** |

**Supplementary Table S6.** Blind docking results for Phe mutated to Gly, fragment probe *p*-toluene boronic acid.

| pose number | affinity (kcal/mol) | nearest Phe (in WT protein) | Distance (Å) | Angle (°) |
|---|---|---|---|---|
| 1 | -5.60 | Chain B Phe128 | 4.19 | 135.49 |
| 2 | -5.20 | Chain B Phe128 | 2.26 | 45.78 |
| 3 | -5.10 | Chain A Phe209 | 1.06 | 112.90 |
| 4 | -5.00 | Chain B Phe106 | 13.90 | 111.47 |
| 5 | -5.00 | Chain B Phe106 | 13.90 | 111.09 |
| 6 | -4.90 | Chain B Phe128 | 10.15 | 99.44 |
| 7 | -4.80 | Chain A Phe98 | 12.61 | 87.51 |
| 8 | -4.80 | Chain B Phe172 | 18.47 | 79.41 |
| 9 | -4.60 | Chain B Phe106 | 14.78 | 104.37 |
| 10 | -4.60 | Chain B Phe128 | 9.19 | 78.61 |
| 11 | -4.60 | Chain A Phe21 | 13.47 | 133.27 |
| 12 | -4.50 | Chain B Phe172 | 13.57 | 133.33 |
| 13 | -4.50 | Chain B Phe152 | 10.93 | 104.66 |
| 14 | -4.50 | Chain B Phe152 | 4.57 | 127.16 |
| 15 | -4.50 | Chain A Phe98 | 13.36 | 119.93 |
| 16 | -4.40 | Chain B Phe80 | 14.54 | 133.36 |
| 17 | -4.40 | Chain A Phe139 | 14.94 | 67.23 |
| 18 | -4.30 | Chain B Phe63 | 12.98 | 111.00 |
| 19 | -4.30 | Chain B Phe152 | 13.91 | 112.21 |
| 20 | -4.20 | Chain B Phe172 | 10.11 | 99.86 |

**Supplementary Table S7.** Blind docking results for Phe mutated to Gly, fragment probe cyclopentilboronic acid.

| pose number | affinity (kcal/mol) | nearest Phe (in WT protein) | Distance (Å) | Angle (°) |
|---|---|---|---|---|
| 1 | -5.40 | Chain A Phe98 | 3.95 | 135.65 |
| 2 | -4.90 | Chain B Phe106 | 2.79 | 107.63 |
| 3 | -4.50 | Chain B Phe106 | 2.19 | 63.48 |
| 4 | -4.40 | Chain A Phe98 | 12.29 | 32.07 |
| 5 | -4.10 | Chain B Phe172 | 17.74 | 69.00 |
| 6 | -4.10 | Chain B Phe152 | 2.93 | 123.62 |
| 7 | -4.00 | Chain B Phe106 | 12.06 | 92.30 |
| 8 | -3.90 | Chain B Phe152 | 10.79 | 94.58 |
| 9 | -3.90 | Chain B Phe172 | 13.98 | 126.45 |
| 10 | -3.80 | Chain B Phe152 | 4.58 | 154.87 |
| 11 | -3.80 | Chain B Phe172 | 14.48 | 98.93 |
| 12 | -3.80 | Chain B Phe106 | 13.58 | 51.76 |
| 13 | -3.80 | Chain B Phe152 | 14.39 | 31.79 |

| | | | | |
|---|---|---|---|---|
| 14 | -3.80 | Chain A Phe21 | 12.04 | 101.71 |
| 15 | -3.70 | Chain B Phe172 | 14.04 | 80.85 |
| 16 | -3.60 | Chain B Phe172 | 10.07 | 74.32 |
| 17 | -3.60 | Chain B Phe79 | 3.50 | 144.92 |
| 18 | -3.60 | Chain A Phe116 | 14.10 | 93.56 |
| 19 | -3.50 | Chain B Phe152 | 14.46 | 95.41 |
| 20 | -3.40 | Chain B Phe172 | 10.08 | 97.49 |

**Supplementary Table S8.** Blind docking results for Phe mutated to Ala, fragment probe phenylboronic acid.

| pose number | affinity (kcal/mol) | nearest Phe (in WT protein) | Distance (Å) | Angle (°) |
|---|---|---|---|---|
| 1 | -4.80 | Chain B Phe128 | 3.03 | 65.95 |
| 2 | -4.80 | Chain A Phe98 | 12.32 | 38.52 |
| 3 | -4.70 | Chain B Phe128 | 10.13 | 99.39 |
| 4 | -4.60 | Chain B Phe106 | 13.94 | 112.47 |
| 5 | -4.50 | Chain B Phe172 | 18.52 | 78.22 |
| 6 | -4.40 | Chain B Phe152 | 14.42 | 69.73 |
| 7 | -4.40 | Chain B Phe106 | 13.66 | 25.79 |
| 8 | -4.40 | Chain A Phe209 | 7.36 | 86.94 |
| 9 | -4.40 | Chain A Phe21 | 13.75 | 133.24 |
| 10 | -4.30 | Chain A Phe98 | 12.11 | 72.53 |
| 11 | -4.30 | Chain A Phe209 | 5.30 | 26.12 |
| 12 | -4.20 | Chain B Phe152 | 13.53 | 80.00 |
| 13 | -4.20 | Chain A Phe98 | 8.80 | 30.37 |
| 14 | -4.20 | Chain B Phe27 | 14.28 | 65.03 |
| 15 | -4.00 | Chain B Phe172 | 10.15 | 128.92 |
| 16 | -4.00 | Chain B Phe106 | 10.09 | 144.34 |
| 17 | -3.80 | Chain B Phe172 | 7.83 | 139.84 |
| 18 | -3.80 | Chain B Phe128 | 17.20 | 118.44 |
| 19 | -3.80 | Chain B Phe106 | 12.65 | 55.98 |
| 20 | -3.80 | Chain B Phe152 | 13.91 | 92.01 |

**Supplementary Table S9.** Blind docking results for Phe mutated to Ala, fragment probe cyclopentilboronic acid.

| pose number | affinity (kcal/mol) | nearest Phe (in WT protein) | Distance (Å) | Angle (°) |
|---|---|---|---|---|
| 1 | -4.30 | Chain B Phe106 | 14.67 | 118.69 |
| 2 | -4.20 | Chain B Phe128 | 9.64 | 88.90 |
| 3 | -4.20 | Chain A Phe139 | 9.71 | 124.89 |
| 4 | -4.10 | Chain A Phe98 | 12.57 | 26.08 |
| 5 | -4.10 | Chain A Phe98 | 14.55 | 43.43 |
| 6 | -4.10 | Chain A Phe21 | 13.43 | 119.53 |
| 7 | -4.00 | Chain B Phe106 | 12.01 | 92.78 |
| 8 | -4.00 | Chain B Phe172 | 17.54 | 84.77 |
| 9 | -4.00 | Chain B Phe172 | 14.99 | 85.75 |
| 10 | -4.00 | Chain A Phe98 | 7.97 | 30.86 |
| 11 | -3.90 | Chain A Phe98 | 13.45 | 117.13 |

| | | | | |
|---|---|---|---|---|
| 12 | -3.90 | Chain A Phe98 | 12.47 | 57.51 |
| 13 | -3.90 | Chain B Phe152 | 10.71 | 94.16 |
| 14 | -3.80 | Chain B Phe128 | 9.70 | 86.54 |
| 15 | -3.80 | Chain B Phe80 | 10.36 | 108.60 |
| 16 | -3.80 | Chain A Phe98 | 12.73 | 110.16 |
| 17 | -3.80 | Chain A Phe139 | 16.92 | 100.41 |
| 18 | -3.80 | Chain A Phe98 | 14.01 | 130.68 |
| 19 | -3.80 | Chain A Phe139 | 18.49 | 102.19 |
| 20 | -3.80 | Chain B Phe152 | 13.41 | 91.58 |

**Supplementary Table S10.** Blind docking results for Tyr mutated to Gly, fragment probe *p*-toluene boronic acid.

| pose number | affinity (kcal/mol) | nearest Phe (in WT protein) | Distance (Å) | Angle (°) |
|---|---|---|---|---|
| 1 | -5.80 | Chain A Tyr173 | 3.87 | 129.31 |
| 2 | -5.40 | Chain B Tyr94 | 1.68 | 38.31 |
| 3 | -5.40 | Chain A Tyr173 | 2.23 | 61.76 |
| 4 | -5.30 | Chain A Tyr173 | 1.80 | 55.81 |
| 5 | -5.30 | Chain A Tyr140 | 5.14 | 154.86 |
| 6 | -5.30 | Chain A Tyr173 | 3.50 | 83.84 |
| 7 | -5.20 | Chain A Tyr173 | 3.62 | 14.74 |
| 8 | -5.00 | Chain A Tyr186 | 13.85 | 109.12 |
| 9 | -5.00 | Chain B Tyr59 | 15.18 | 147.16 |
| 10 | -4.90 | Chain B Tyr104 | 4.91 | 159.48 |
| 11 | -4.80 | Chain B Tyr200 | 5.52 | 118.14 |
| 12 | -4.80 | Chain B Tyr151 | 14.24 | 130.90 |
| 13 | -4.70 | Chain B Tyr182 | 3.96 | 106.03 |
| 14 | -4.50 | Chain B Tyr200 | 5.61 | 141.93 |
| 15 | -4.40 | Chain A Tyr87 | 11.39 | 155.55 |
| 16 | -4.40 | Chain B Tyr200 | 12.45 | 105.15 |
| 17 | -4.40 | Chain B Tyr182 | 1.65 | 65.12 |
| 18 | -4.40 | Chain B Tyr182 | 10.59 | 74.00 |
| 19 | -4.40 | Chain B Tyr182 | 4.21 | 109.78 |
| 20 | -4.30 | Chain B Tyr182 | 10.34 | 86.85 |

**Supplementary Table S11.** Blind docking results for Tyr mutated to Gly, fragment probe cyclopentilboronic acid.

| pose number | affinity (kcal/mol) | nearest Phe (in WT protein) | Distance (Å) | Angle (°) |
|---|---|---|---|---|
| 1 | -5.10 | Chain B Tyr94 | 1.67 | 50.75 |
| 2 | -4.80 | Chain B Tyr94 | 3.24 | 75.19 |
| 3 | -4.60 | Chain A Tyr186 | 2.24 | 80.13 |
| 4 | -4.60 | Chain A Tyr173 | 5.43 | 135.78 |
| 5 | -4.60 | Chain A Tyr192 | 2.26 | 86.59 |
| 6 | -4.50 | Chain B Tyr94 | 2.24 | 31.05 |
| 7 | -4.50 | Chain A Tyr140 | 4.79 | 157.82 |
| 8 | -4.40 | Chain A Tyr173 | 1.88 | 11.24 |
| 9 | -4.30 | Chain A Tyr186 | 2.70 | 114.18 |

| | | | | |
|---|---|---|---|---|
| 10 | -4.30 | Chain A Tyr192 | 2.37 | 67.09 |
| 11 | -4.20 | Chain A Tyr140 | 3.44 | 98.78 |
| 12 | -4.20 | Chain B Tyr151 | 13.80 | 117.54 |
| 13 | -4.20 | Chain A Tyr140 | 2.91 | 121.08 |
| 14 | -4.20 | Chain A Tyr140 | 5.07 | 99.50 |
| 15 | -4.10 | Chain A Tyr140 | 5.44 | 135.87 |
| 16 | -4.00 | Chain A Tyr87 | 9.68 | 120.89 |
| 17 | -3.90 | Chain B Tyr93 | 14.32 | 46.04 |
| 18 | -3.90 | Chain B Tyr182 | 1.57 | 49.30 |
| 19 | -3.80 | Chain A Tyr140 | 1.50 | 65.13 |
| 20 | -3.70 | Chain B Tyr94 | 10.28 | 152.16 |

**Supplementary Table S12.** Blind docking results for Tyr mutated to Ala, fragment probe phenylboronic acid.

| pose number | affinity (kcal/mol) | nearest Phe (in WT protein) | Distance (Å) | Angle (°) |
|---|---|---|---|---|
| 1 | -5.30 | Chain A Tyr192 | 2.22 | 81.05 |
| 2 | -5.10 | Chain A Tyr140 | 7.62 | 136.46 |
| 3 | -5.10 | Chain A Tyr192 | 2.14 | 81.08 |
| 4 | -4.80 | Chain A Tyr173 | 2.48 | 66.41 |
| 5 | -4.80 | Chain A Tyr173 | 3.47 | 70.56 |
| 6 | -4.70 | Chain A Tyr186 | 13.86 | 109.09 |
| 7 | -4.70 | Chain A Tyr173 | 2.49 | 10.07 |
| 8 | -4.70 | Chain B Tyr93 | 15.02 | 55.02 |
| 9 | -4.60 | Chain A Tyr140 | 3.06 | 40.90 |
| 10 | -4.60 | Chain A Tyr186 | 1.91 | 18.49 |
| 11 | -4.60 | Chain B Tyr200 | 5.50 | 116.97 |
| 12 | -4.60 | Chain A Tyr140 | 4.29 | 141.66 |
| 13 | -4.50 | Chain A Tyr87 | 12.67 | 128.60 |
| 14 | -4.30 | Chain A Tyr87 | 13.61 | 117.97 |
| 15 | -4.30 | Chain A Tyr87 | 11.49 | 166.07 |
| 16 | -4.20 | Chain B Tyr200 | 6.90 | 150.40 |
| 17 | -4.20 | Chain A Tyr86 | 12.46 | 136.99 |
| 18 | -4.20 | Chain A Tyr87 | 10.06 | 83.51 |
| 19 | -4.20 | Chain B Tyr182 | 10.60 | 73.29 |
| 20 | -4.10 | Chain A Tyr140 | 3.11 | 67.16 |

**Supplementary Table S13.** Blind docking results for Tyr mutated to Ala, fragment probe cyclopentilboronic acid.

| pose number | affinity (kcal/mol) | nearest Phe (in WT protein) | Distance (Å) | Angle (°) |
|---|---|---|---|---|
| 1 | -5.00 | Chain A Tyr186 | 3.01 | 120.10 |
| 2 | -4.80 | Chain A Tyr173 | 4.03 | 76.30 |
| 3 | -4.70 | Chain A Tyr192 | 2.44 | 97.85 |
| 4 | -4.40 | Chain A Tyr173 | 2.32 | 11.72 |
| 5 | -4.30 | Chain A Tyr140 | 3.66 | 117.56 |
| 6 | -4.30 | Chain A Tyr140 | 4.84 | 123.18 |
| 7 | -4.20 | Chain B Tyr151 | 13.84 | 117.52 |

| | | | | |
|---|---|---|---|---|
| 8 | -4.10 | Chain A Tyr140 | 5.98 | 132.64 |
| 9 | -4.10 | Chain A Tyr87 | 13.50 | 113.99 |
| 10 | -4.10 | Chain A Tyr173 | 4.00 | 25.64 |
| 11 | -3.90 | Chain B Tyr94 | 9.13 | 58.56 |
| 12 | -3.90 | Chain A Tyr186 | 2.26 | 51.01 |
| 13 | -3.90 | Chain A Tyr87 | 9.78 | 130.57 |
| 14 | -3.90 | Chain A Tyr87 | 10.85 | 166.51 |
| 15 | -3.90 | Chain B Tyr93 | 9.35 | 148.60 |
| 16 | -3.80 | Chain A Tyr192 | 15.92 | 83.81 |
| 17 | -3.80 | Chain A Tyr87 | 10.78 | 120.11 |
| 18 | -3.80 | Chain A Tyr 87 | 8.72 | 118.57 |
| 19 | -3.80 | Chain B Tyr200 | 1.93 | 68.88 |
| 20 | -3.70 | Chain B Tyr94 | 10.25 | 152.44 |

**Supplementary Table S14.** Blind docking results for Trp mutated to Gly, fragment probe *p*-toluene boronic acid.

| pose number | affinity (kcal/mol) | nearest Phe (in WT protein) | Distance (Å) | Angle (°) |
|---|---|---|---|---|
| 1 | -6.50 | Chain B Trp109 | 4.87 | 122.28 |
| 2 | -5.60 | Chain B Trp36 | 3.32 | 89.74 |
| 3 | -5.50 | Chain A Trp94 | 3.97 | 64.97 |
| 4 | -5.20 | Chain A Trp148 | 3.47 | 112.14 |
| 5 | -5.20 | Chain B Trp47 | 2.23 | 76.88 |
| 6 | -5.00 | Chain B Trp160 | 17.64 | 61.51 |
| 7 | -4.90 | Chain B Trp36 | 12.71 | 147.37 |
| 8 | -4.80 | Chain B Trp52 | 1.50 | 73.32 |
| 9 | -4.80 | Chain A Trp148 | 16.46 | 118.88 |
| 10 | -4.80 | Chain B Trp160 | 11.61 | 78.24 |
| 11 | -4.60 | Chain B Trp109 | 14.68 | 62.13 |
| 12 | -4.60 | Chain A Trp35 | 18.02 | 75.78 |
| 13 | -4.60 | Chain B Trp109 | 17.78 | 131.32 |
| 14 | -4.60 | Chain B Trp109 | 12.49 | 31.46 |
| 15 | -4.50 | Chain B Trp109 | 14.97 | 87.63 |
| 16 | -4.50 | Chain A Trp35 | 16.29 | 16.42 |
| 17 | -4.40 | Chain A Trp35 | 16.18 | 116.23 |
| 18 | -4.40 | Chain A Trp94 | 8.42 | 130.99 |
| 19 | -4.40 | Chain B Trp47 | 8.43 | 115.86 |
| 20 | -4.30 | Chain B Trp109 | 11.92 | 29.55 |

**Supplementary Table S15.** Blind docking results for Trp mutated to Gly, fragment probe cyclopentilboronic acid.

| pose number | affinity (kcal/mol) | nearest Phe (in WT protein) | Distance (Å) | Angle (°) |
|---|---|---|---|---|
| 1 | -5.10 | Chain B Trp47 | 4.60 | 101.61 |
| 2 | -4.20 | Chain A Trp148 | 15.26 | 97.15 |
| 3 | -4.20 | Chain B Trp52 | 4.09 | 142.24 |
| 4 | -4.20 | Chain B Trp47 | 2.99 | 82.92 |
| 5 | -4.20 | Chain B Trp52 | 1.58 | 61.79 |

| | | | | |
|---|---|---|---|---|
| 6 | -4.10 | Chain B Trp160 | 10.77 | 89.48 |
| 7 | -4.10 | Chain B Trp109 | 13.52 | 110.31 |
| 8 | -4.10 | Chain B Trp109 | 12.40 | 106.13 |
| 9 | -4.00 | Chain B Trp109 | 17.91 | 119.05 |
| 10 | -4.00 | Chain B Trp47 | 7.34 | 127.11 |
| 11 | -3.80 | Chain B Trp36 | 14.51 | 96.09 |
| 12 | -3.80 | Chain B Trp109 | 15.14 | 22.60 |
| 13 | -3.80 | Chain B Trp109 | 15.33 | 46.91 |
| 14 | -3.80 | Chain B Trp109 | 14.55 | 109.40 |
| 15 | -3.80 | Chain B Trp47 | 9.13 | 23.32 |
| 16 | -3.70 | Chain B Trp36 | 13.63 | 58.01 |
| 17 | -3.70 | Chain B Trp160 | 18.17 | 82.94 |
| 18 | -3.70 | Chain B Trp109 | 13.32 | 48.23 |
| 19 | -3.60 | Chain B Trp160 | 12.26 | 66.57 |
| 20 | -3.60 | Chain B Trp109 | 14.06 | 73.74 |

**Supplementary Table S16.** Blind docking results for Trp mutated to Ala, fragment probe phenylboronic acid.

| pose number | affinity (kcal/mol) | nearest Phe (in WT protein) | Distance (Å) | Angle (°) |
|---|---|---|---|---|
| 1 | -6.70 | Chain B Trp109 | 5.21 | 103.46 |
| 2 | -5.70 | Chain A Trp94 | 4.90 | 75.20 |
| 3 | -5.20 | Chain B Trp109 | 5.31 | 151.94 |
| 4 | -5.00 | Chain A Trp94 | 3.03 | 45.12 |
| 5 | -4.90 | Chain A Trp148 | 15.69 | 89.71 |
| 6 | -4.70 | Chain B Trp160 | 17.60 | 60.90 |
| 7 | -4.70 | Chain B Trp36 | 12.73 | 146.91 |
| 8 | -4.70 | Chain B Trp109 | 13.10 | 102.78 |
| 9 | -4.50 | Chain B Trp160 | 11.62 | 77.81 |
| 10 | -4.40 | Chain B Trp109 | 13.16 | 53.43 |
| 11 | -4.40 | Chain A Trp35 | 18.25 | 66.91 |
| 12 | -4.30 | Chain B Trp109 | 14.08 | 111.01 |
| 13 | -4.30 | Chain B Trp160 | 12.30 | 94.20 |
| 14 | -4.20 | Chain A Trp35 | 16.36 | 17.75 |
| 15 | -4.20 | Chain A Trp35 | 15.59 | 129.87 |
| 16 | -4.10 | Chain B Trp52 | 4.31 | 172.29 |
| 17 | -4.10 | Chain B Trp36 | 14.10 | 89.20 |
| 18 | -4.10 | Chain B Trp36 | 14.21 | 61.44 |
| 19 | -4.00 | Chain B Trp160 | 15.79 | 73.41 |
| 20 | -4.00 | Chain B Trp160 | 17.38 | 171.00 |

**Su pplementary Table S17.** Blind docking results for Trp mutated to Ala, fragment probe cyclopentilboronic acid.

| pose number | affinity (kcal/mol) | nearest Phe (in WT protein) | Distance (Å) | Angle (°) |
|---|---|---|---|---|
| 1 | -4.40 | Chain B Trp52 | 2.51 | 75.69 |
| 2 | -4.30 | Chain B Trp160 | 17.61 | 57.22 |
| 3 | -4.20 | Chain B Trp109 | 12.64 | 110.66 |

| | | | | |
|---|---|---|---|---|
| 4 | -4.20 | Chain B Trp160 | 10.80 | 87.76 |
| 5 | -4.10 | Chain B Trp109 | 13.30 | 107.29 |
| 6 | -4.00 | Chain B Trp109 | 17.93 | 118.77 |
| 7 | -3.90 | Chain A Trp35 | 15.68 | 62.59 |
| 8 | -3.90 | Chain B Trp47 | 9.24 | 35.32 |
| 9 | -3.90 | Chain A Trp94 | 10.08 | 114.68 |
| 10 | -3.80 | Chain B Trp160 | 13.04 | 10.23 |
| 11 | -3.80 | Chain B Trp109 | 14.50 | 110.20 |
| 12 | -3.80 | Chain B Trp47 | 10.59 | 89.18 |
| 13 | -3.70 | Chain B Trp109 | 15.12 | 25.80 |
| 14 | -3.70 | Chain B Trp109 | 14.72 | 91.03 |
| 15 | -3.70 | Chain B Trp109 | 12.21 | 9.59 |
| 16 | -3.60 | Chain B Trp160 | 15.68 | 59.08 |
| 17 | -3.60 | Chain B Trp160 | 13.68 | 49.29 |
| 18 | -3.60 | Chain B Trp36 | 12.50 | 150.50 |
| 19 | -3.50 | Chain A Trp148 | 8.07 | 95.21 |
| 20 | -3.40 | Chain A Trp148 | 11.28 | 63.95 |

**Supplementary Table S18.** Blind docking results for His mutated to Gly, fragment probe *p*-toluene boronic acid.

| pose number | affinity (kcal/mol) | nearest Phe (in WT protein) | Distance (Å) | Angle (°) |
|---|---|---|---|---|
| 1 | -5.00 | Chain A His189 | 20.86 | 56.59 |
| 2 | -5.00 | Chain B His35 | 18.42 | 66.14 |
| 3 | -5.00 | Chain B His35 | 9.97 | 87.65 |
| 4 | -4.80 | Chain B His206 | 18.28 | 136.59 |
| 5 | -4.80 | Chain B His170 | 10.95 | 110.06 |
| 6 | -4.70 | Chain A His189 | 5.26 | 62.99 |
| 7 | -4.60 | Chain B His206 | 14.88 | 91.62 |
| 8 | -4.60 | Chain B His206 | 19.04 | 81.73 |
| 9 | -4.60 | Chain B His206 | 13.28 | 75.18 |
| 10 | -4.50 | Chain A His189 | 11.03 | 72.77 |
| 11 | -4.50 | Chain A His189 | 14.31 | 89.69 |
| 12 | -4.50 | Chain A His189 | 15.84 | 111.04 |
| 13 | -4.50 | Chain B His206 | 18.97 | 53.65 |
| 14 | -4.40 | Chain B His35 | 12.69 | 98.15 |
| 15 | -4.40 | Chain A His189 | 15.62 | 112.59 |
| 16 | -4.40 | Chain B His35 | 20.36 | 127.20 |
| 17 | -4.40 | Chain B His170 | 5.69 | 83.96 |
| 18 | -4.30 | Chain B His170 | 16.60 | 76.95 |
| 19 | -4.00 | Chain B His170 | 13.49 | 60.40 |
| 20 | -4.00 | Chain A His34 | 15.04 | 141.80 |

**Supplementary Table S19.** Blind docking results for His mutated to Gly, fragment probe cyclopentilboronic acid.

| pose number | affinity (kcal/mol) | nearest Phe (in WT protein) | Distance (Å) | Angle (°) |
|---|---|---|---|---|
| 1 | -4.20 | Chain B His206 | 17.99 | 161.73 |

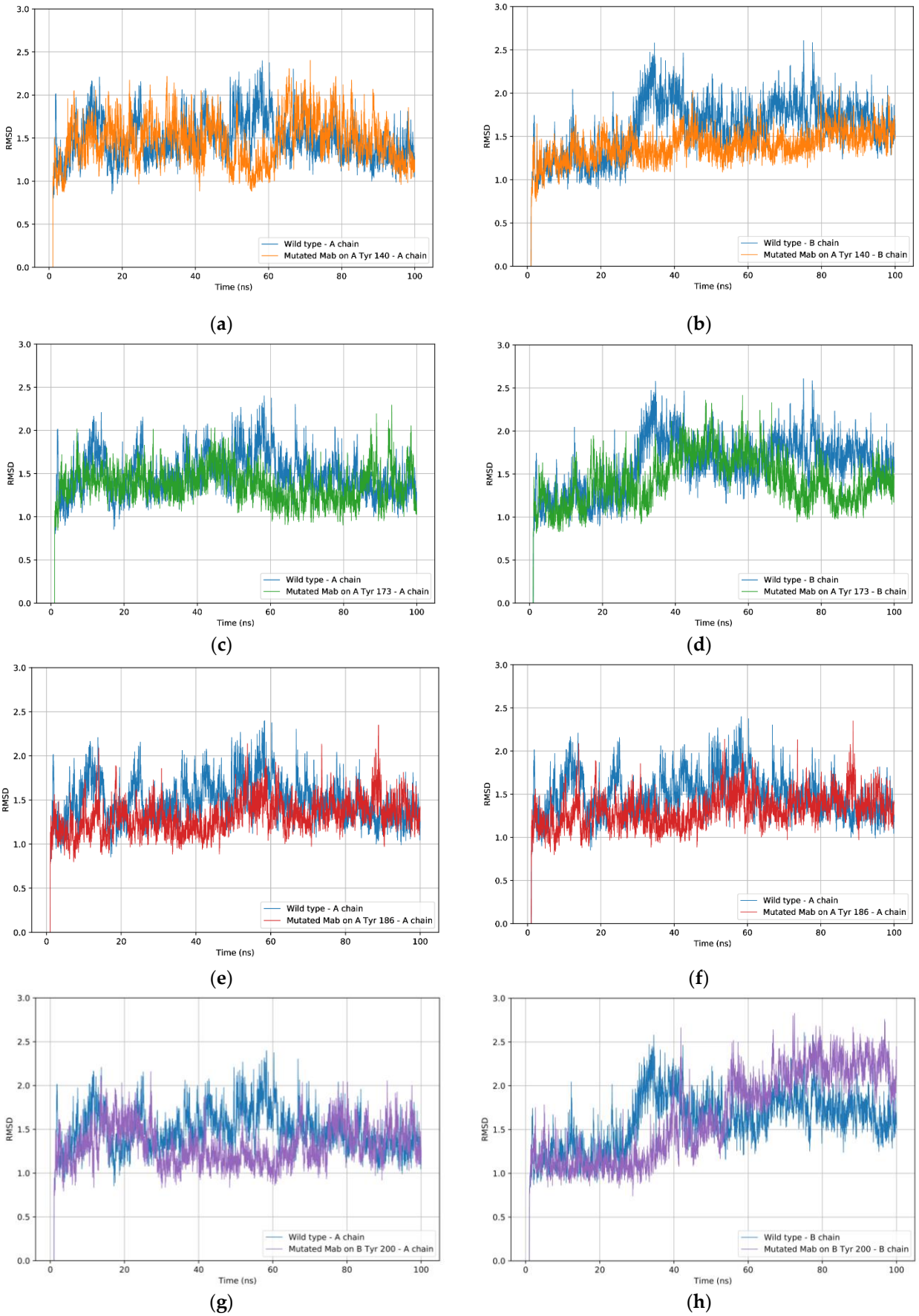
| | | | | |
|---|---|---|---|---|
| 2 | -4.20 | Chain B His35 | 10.68 | 121.61 |
| 3 | -4.20 | Chain B His206 | 10.82 | 123.70 |
| 4 | -4.20 | Chain A His34 | 14.49 | 50.00 |
| 5 | -4.10 | Chain B His170 | 3.96 | 72.23 |
| 6 | -4.10 | Chain B His206 | 18.00 | 166.24 |
| 7 | -4.10 | Chain B His206 | 17.67 | 20.97 |
| 8 | -4.00 | Chain B His170 | 18.89 | 58.10 |
| 9 | -4.00 | Chain B His35 | 18.31 | 59.82 |
| 10 | -3.90 | Chain B His206 | 11.24 | 88.04 |
| 11 | -3.80 | Chain A His34 | 18.90 | 44.99 |
| 12 | -3.80 | Chain B His206 | 15.65 | 77.65 |
| 13 | -3.80 | Chain B His35 | 9.90 | 116.10 |
| 14 | -3.80 | Chain B His35 | 10.61 | 117.27 |
| 15 | -3.70 | Chain A His189 | 13.05 | 59.97 |
| 16 | -3.70 | Chain B His170 | 11.90 | 146.89 |
| 17 | -3.60 | Chain B His170 | 15.34 | 85.93 |
| 18 | -3.60 | Chain A His189 | 12.95 | 149.20 |
| 19 | -3.60 | Chain B His170 | 15.94 | 77.64 |
| 20 | -3.50 | Chain A His189 | 19.68 | 116.09 |

**Supplementary Table S20.** Blind docking results for His mutated to Ala, fragment probe phenylboronic acid.

| **pose number** | **affinity (kcal/mol)** | **nearest Phe (in WT protein)** | **Distance (Å)** | **Angle (°)** |
|---|---|---|---|---|
| 1 | -4.70 | Chain A His189 | 20.87 | 56.46 |
| 2 | -4.70 | Chain B His35 | 19.33 | 77.66 |
| 3 | -4.70 | Chain B His35 | 18.47 | 65.48 |
| 4 | -4.60 | Chain B His206 | 18.83 | 155.78 |
| 5 | -4.60 | Chain B His170 | 5.28 | 58.52 |
| 6 | -4.60 | Chain A His189 | 16.36 | 103.62 |
| 7 | -4.50 | Chain B His206 | 10.99 | 109.43 |
| 8 | -4.40 | Chain B His 206 | 11.93 | 100.20 |
| 9 | -4.30 | Chain B His 206 | 14.86 | 93.30 |
| 10 | -4.30 | Chain A His189 | 12.90 | 157.11 |
| 11 | -4.10 | Chain A His189 | 18.22 | 39.57 |
| 12 | -4.10 | Chain A His189 | 17.29 | 104.73 |
| 13 | -4.00 | Chain B His206 | 14.65 | 160.47 |
| 14 | -4.00 | Chain B His35 | 16.02 | 55.41 |
| 15 | -4.00 | Chain A His189 | 2.21 | 88.68 |
| 16 | -4.00 | Chain B His35 | 18.36 | 128.47 |
| 17 | -4.00 | Chain B His170 | 9.36 | 53.77 |
| 18 | -3.90 | Chain B His170 | 6.41 | 49.46 |
| 19 | -3.90 | Chain B His170 | 22.32 | 43.84 |
| 20 | -3.80 | Chain A His34 | 12.85 | 101.20 |

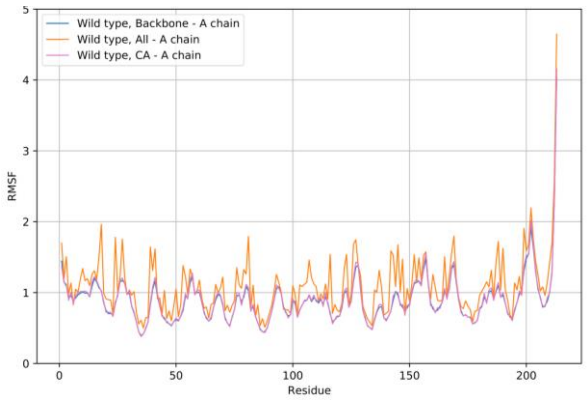
**Supplementary Table S21.** Blind docking results for His mutated to Ala, fragment probe cyclopentilboronic acid.

| pose number | affinity (kcal/mol) | nearest Phe (in WT protein) | Distance (Å) | Angle (°) |
|---|---|---|---|---|
| 1 | -4.70 | Chain B His170 | 4.22 | 47.45 |
| 2 | -4.20 | Chain B His206 | 17.93 | 159.58 |
| 3 | -4.20 | Chain B His35 | 10.64 | 121.49 |
| 4 | -4.10 | Chain B His206 | 18.25 | 129.73 |
| 5 | -4.00 | Chain A His34 | 13.66 | 25.62 |
| 6 | -4.00 | Chain B His35 | 18.03 | 64.52 |
| 7 | -3.90 | Chain B His206 | 13.37 | 45.79 |
| 8 | -3.80 | Chain B His170 | 5.29 | 41.99 |
| 9 | -3.80 | Chain B His 206 | 9.44 | 61.78 |
| 10 | -3.80 | Chain B His170 | 11.70 | 91.89 |
| 11 | -3.80 | Chain B His206 | 11.42 | 99.27 |
| 12 | -3.70 | Chain B His170 | 17.26 | 129.17 |
| 13 | -3.70 | Chain B His206 | 21.24 | 112.58 |
| 14 | -3.50 | Chain B His35 | 10.08 | 77.16 |
| 15 | -3.50 | Chain B His170 | 12.08 | 76.51 |
| 16 | -3.40 | Chain A His198 | 14.23 | 97.00 |
| 17 | -3.40 | Chain A His34 | 15.49 | 38.77 |
| 18 | -3.40 | Chain A His189 | 19.17 | 135.25 |
| 19 | -3.40 | Chain A His189 | 3.53 | 103.52 |
| 20 | -3.40 | Chain B His170 | 10.02 | 152.83 |

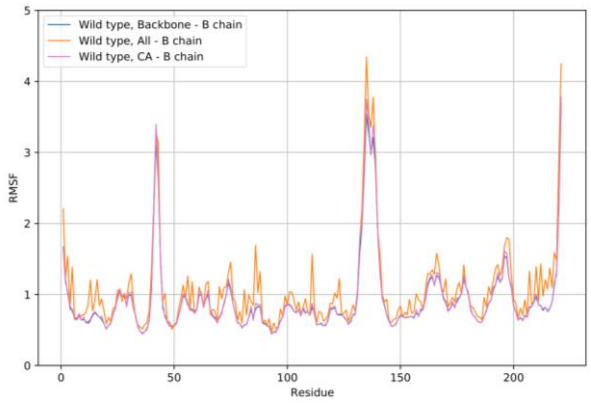



**Supplementary Figure S4.** The root means square deviation (RMSD) analysis. RMSD comparison between wild type and boronated residues on protein chain A and B. On the

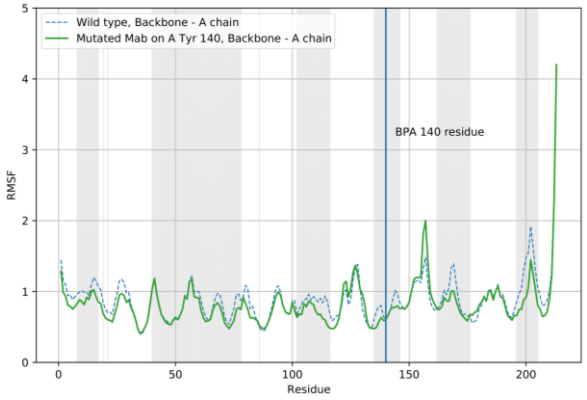
left, RMSD values wild type chain A versus mutated chain A; on the right RMSD values wild type chain B and chain B after chain A (or chain B) mutation. From the top wild-type chain A (blue) and boronated Tyr140 (orange), wild type chain A (blue) and boronated Tyr173 (green), wild type chain A (blue) and boronated Tyr186 (red), wild type chain B (blue) and boronated Tyr200 (purple).



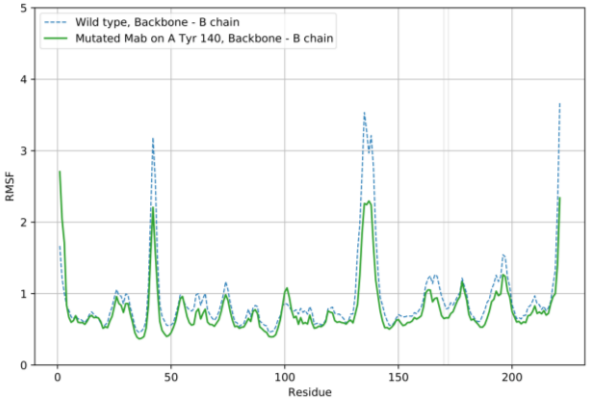

(**a**)



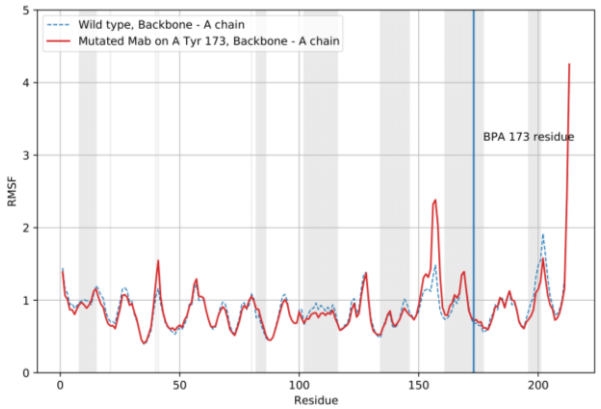

(**b**)



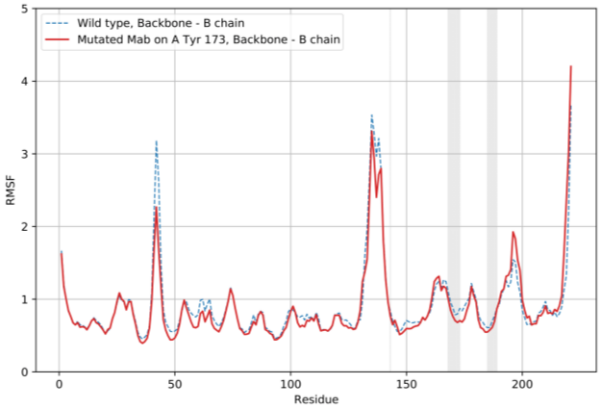

(**c**)



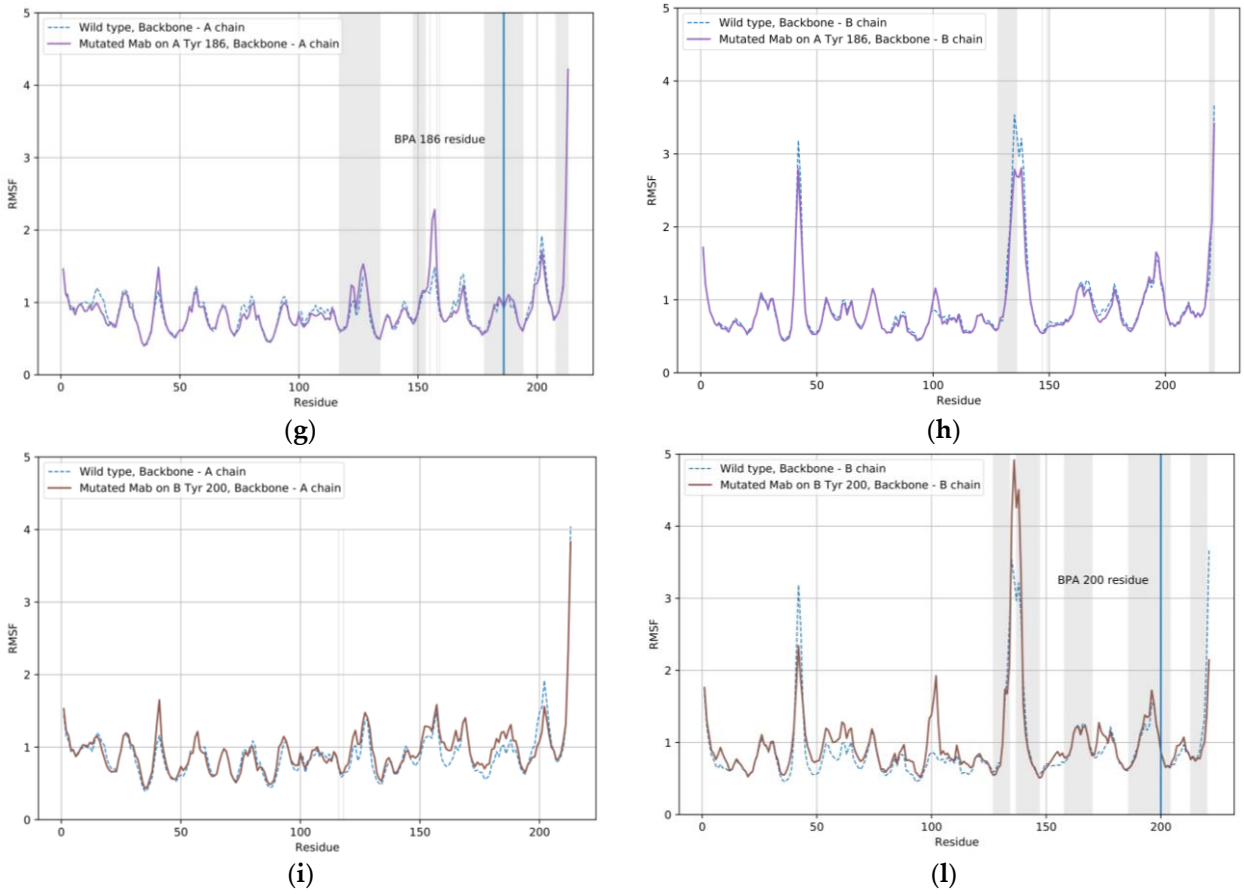

(**d**)



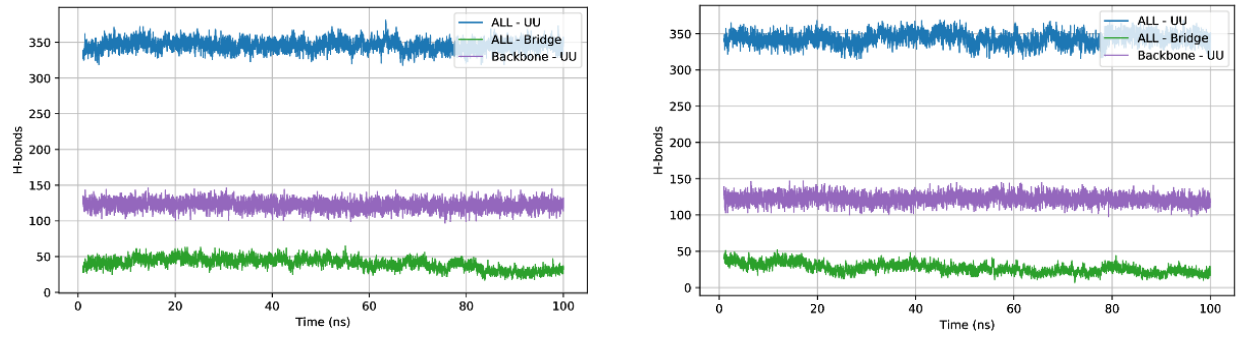

(**e**)

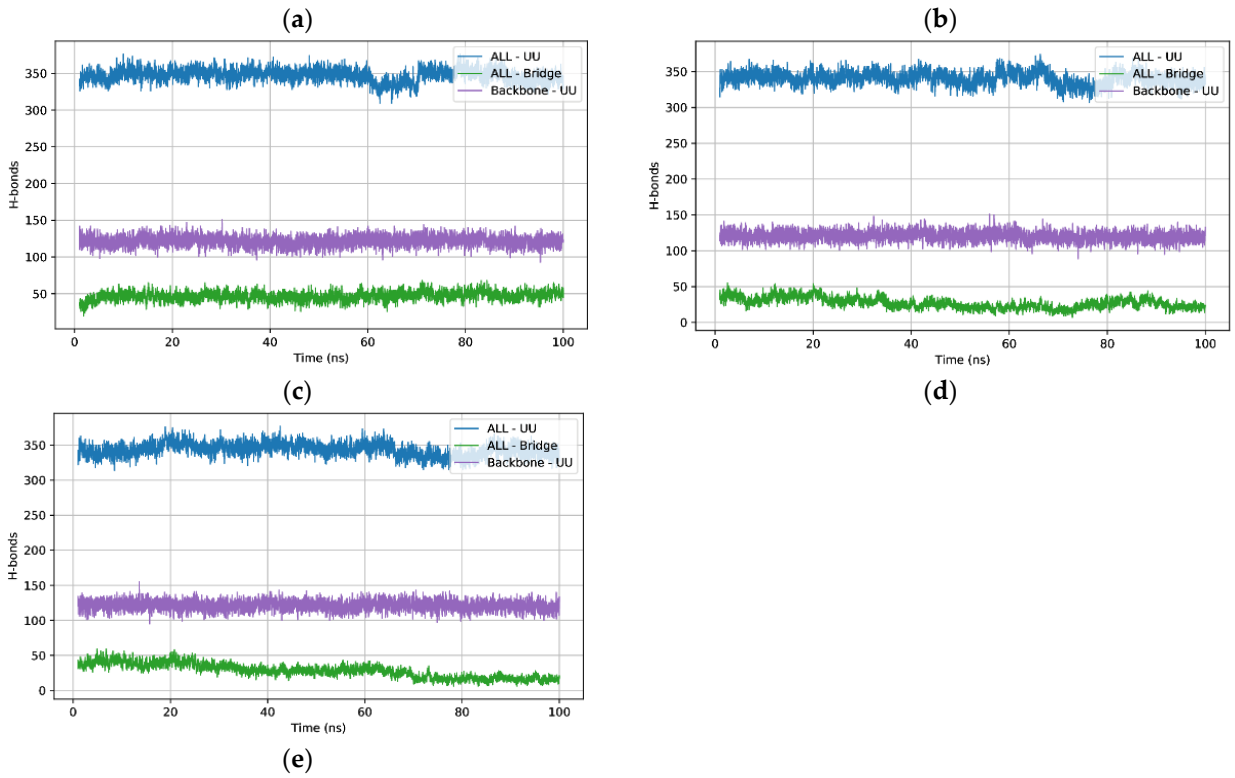



(**f**)




**Supplementary Figure S5.** Mean square fluctuations (RMSF) analysis, the greyed areas represent all residues within 12 Å of the mutation, while the blue vertical lines address the new mutated residue (BPA) : (**a**) RMSF calculation (total, backbone and alpha carbons) of the wild type protein (A chain); (**b**) RMSF calculation (total, backbone and alpha carbons) of the wild type protein (B chain); (**c**) Comparison of backbone fluctuations between wild type and mutated protein on residue A Tyr140 (A chain); (**d**) Comparison of backbone fluctuations between wild type and mutated protein on residue A Tyr140 (B chain); (**e**) Comparison of backbone fluctuations between wild type and mutated protein on residue A Tyr173 (A chain); (**f**) Comparison of backbone fluctuations between wild type and mutated protein on residue A Tyr173 (B chain); (**g)** Comparison of backbone fluctuations between wild type and mutated protein on residue A Tyr186 (A chain);(**h**) Comparison of backbone fluctuations between wild type and mutated protein on residue A Tyr186 (B chain); (**i**) Comparison of backbone fluctuations between wild type and mutated protein on residue B Tyr200 (A chain); (**l**) Comparison of backbone fluctuations between wild type and mutated protein on residue B Tyr200 (B chain).






**Supplementary Figure S6.** H-bond analysis, comparison between the total number of hydrogen bonds in wild type Mab and mutations. the graphs show the total number of solute-solute interactions (ALL - UU), the number of Backbone UU interactions (Backbone - UU) and finally the number of hydrogen bridges (ALL - Bridge) established over the MDs simulations: (**a**) Hydrogen bonds in Wild type; (**b**) Hydrogen bonds in mutated protein (mutation on chain A Tyr140); (**c**) Hydrogen bonds in mutated protein (mutation on chain A Tyr173); (**d**) Hydrogen bonds in mutated protein (mutation on chain A Tyr186); (**e**) Hydrogen bonds in mutated protein (mutation on chain B Tyr200).

**Supplementary Table S22.** H-bonds analysis: comparison between the mutated residue hydrogen bonds and the counterpart residue in the wild type, lasting for more than 70% of MDs. The table below shows the donor and acceptor atom involved in the H-bond, together with information about the time fraction in which the bond lasts, the angle and the average distance of the latter.

| Sample | Acceptor | Donor H | Donor | Frac | Avg Dist (Å) | Avg Ang (°) |
|---|---|---|---|---|---|---|
| **Wild Type** | Tyr 140/O | Ala 111/H | Ala 111/N | 99% | 2.96 | 164.27 |
| | Ala 111/O | Tyr 140/H | Tyr 140/N | 99% | 2.99 | 161.46 |
| **Mutated Mab on chain A Tyr140** | BPA 140/O | Ala 111/H | Ala 111/N | 97% | 3.00 | 162.67 |
| | Ala 111/O | BPA 140/H12 | BPA 140/N1 | 97% | 3.02 | 165.15 |

**Supplementary Table S23.** H-bonds analysis: comparison between the mutated residue hydrogen bonds and the counterpart residue in the wild type, lasting for more than 70% of MDs. The table below shows the donor and acceptor atom involved in the H-bond, together with information about the time fraction in which the bond lasts, the angle and the average distance of the latter.

| Sample | Acceptor | Donor H | Donor | Frac | Avg Dist (Å) | Avg Ang (°) |
|---|---|---|---|---|---|---|
| **Wild Type** | Tyr 173/O | Phe 139/H | Phe 139/N | 85% | 3.07 | 159.12 |
| | Phe 139/O | Tyr 173/H | Tyr 173/N | 95% | 2.86 | 160.94 |
| **Mutated Mab on chain A Tyr173** | BPA 173/O | Phe 139/H | Phe 139/N | 86% | 3.08 | 159.59 |
| | Ala 111/O | BPA 140/H12 | BPA 140/N1 | 97% | 3.02 | 165.15 |

**Supplementary Table S24.** H-bonds analysis: comparison between the mutated residue hydrogen bonds and the counterpart residue in the wild type, lasting for more than 70% of MDs. The table below shows the donor and acceptor atom involved in the H-bond, together with information about the time fraction in which the bond lasts, the angle and the average distance of the latter.

| Sample | Acceptor | Donor H | Donor | Frac | Avg Dist (Å) | Avg Ang (°) |
|---|---|---|---|---|---|---|
| **Wild Type** | Ser 182/O | Tyr 186/H | Tyr 186/N | 95% | 2.99 | 158.77 |
| **Mutated Mab on chain A Tyr186** | Ser 182/O | BPA 186/H12 | BPA 186/N1 | 97% | 3.00 | 163.39 |

**Supplementary Table S25.** H-bonds analysis: comparison between the mutated residue hydrogen bonds and the counterpart residue in the wild type, lasting for more than 70% of MDs. The table below shows the donor and acceptor atom involved in the H-bond, together with information about the time fraction in which the bond lasts, the angle and the average distance of the latter.

| Sample | Acceptor | Donor H | Donor | Frac | Avg Dist (Å) | Avg Ang (°) |
|---|---|---|---|---|---|---|
| **Wild Type** | Tyr 413/O | Val 430/H | Val 430/N | 98% | 2.94 | 161.96 |
| | Ser 407/OG | Tyr 413/HH | Tyr 413/OH | 89% | 2.86 | 158.48 |
| **Mutated Mab on chain B Tyr200** | BPA 413/O | Val 430/H | Val 430/N | 95% | 2.93 | 160.17 |
| | Ser 407/OG | BPA 413/H1 | BPA 413/O1 | 76% | 2.71 | 162.04 |