Binding affinity calculations using MM-PBSA for acetylated peptide, NAD+ and inhibitors

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Sir2TM/Ac-p53/ NAD+ | | Sir2TM/Ac-p53/NAD+/NAM | | | Sir2TM/ NAD+ (AC) |
| Ac-p53 | NAD+ | Ac-p53 | NAD+ | NAM | NAD+ |
| -22.671 | -23.190 | -23.921 | -16.154 | -5.778 | -42.021 |

G is averaged over the 1000 frames from last 10 ns.

Binding affinity calculations using MM-GBSA for acetylated peptide, NAD+ and inhibitors

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Sir2TM/Ac-p53/ NAD+ | | Sir2TM/Ac-p53/NAD+/NAM | | | Sir2TM/ NAD+ (AC) |
| Ac-p53 | NAD+ | Ac-p53 | NAD+ | NAM | NAD+ |
| -76.783 | -105.477 | -76.540 | -81.780 | -20.738 | -106.391 |

G is averaged over the 1000 frames from last 10 ns.

Binding affinity calculations using MM-PB(GB)SA for acetylated peptide, NAD+ and inhibitors

1) Binding affinity of Acetylated peptide (Ac-p53)\* (MM-PBSA results)

|  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| 2H4F (Sir2TM, Ac-p53, NAD+) | | | | 2H4F (Sir2TM, Ac-p53, NAD+, NAM) | | | | 2H4F (Sir2TM, NAD+) | | | |
|  |  |  | Mean\* |  |  |  | Mean |  |  |  | Mean |
| Ac-p53 Binding: PBSA | | | -22.671 | Ac-p53 Binding: PBSA | | | -23.921 | Ac-p53 Binding: PBSA | | |  |
| ∆G | STD\* | SEM\* |  | ∆G | STD | SEM |  | ∆G | STD | SEM |  |
| -24.205 | 7.587 | 0.535 |  | -10.467 | 6.629 | 0.468 |  |  |  |  |  |
| -18.882 | 4.449 | 0.314 |  | -8.518 | 6.967 | 0.491 |  |  |  |  |  |
| -19.526 | 5.436 | 0.383 |  | -12.776 | 7.244 | 0.511 |  |  |  |  |  |
| -21.340 | 5.778 | 0.408 |  | -8.804 | 5.800 | 0.409 |  |  |  |  |  |
| -15.280 | 5.464 | 0.385 |  | -12.676 | 6.860 | 0.484 |  |  |  |  |  |
| -20.329 | 5.140 | 0.363 |  | -17.714 | 4.447 | 0.314 |  |  |  |  |  |
| -20.550 | 5.506 | 0.388 |  | -18.064 | 4.982 | 0.351 |  |  |  |  |  |
| -22.454 | 5.445 | 0.384 |  | -22.096 | 4.677 | 0.330 |  |  |  |  |  |
| -23.289 | 4.600 | 0.325 |  | -23.524 | 4.923 | 0.347 |  |  |  |  |  |
| -21.266 | 5.437 | 0.384 |  | -27.131 | 6.003 | 0.423 |  |  |  |  |  |

\* ∆G is calculated every 200 frames in 2 ns simulation;

Mean value is averaged over the 1000 frames from last 10 ns;

STD: standard deviation;

SEM: standard error of the mean.

2) Binding affinity of Acetylated peptide (Ac-p53) (MM-GBSA results)

|  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| 2H4F (Sir2TM, Ac-p53, NAD+) | | | | 2H4F (Sir2TM, Ac-p53, NAD+, NAM) | | | | 2H4F (Sir2TM, NAD+) | | | |
|  |  |  | Mean |  |  |  | Mean |  |  |  | Mean |
| Ac-p53 Binding: GBSA | | | -76.783 | Ac-p53 Binding: GBSA | | | -76.540 | Ac-p53 Binding: GBSA | | |  |
| ∆G | STD | SEM |  | ∆G | STD | SEM |  | ∆G | STD | SEM |  |
| -76.150 | 6.257 | 0.441 |  | -70.978 | 5.536 | 0.391 |  |  |  |  |  |
| -69.241 | 3.858 | 0.272 |  | -69.703 | 4.880 | 0.344 |  |  |  |  |  |
| -72.173 | 4.086 | 0.288 |  | -74.403 | 6.094 | 0.430 |  |  |  |  |  |
| -74.616 | 4.399 | 0.310 |  | -68.840 | 5.566 | 0.393 |  |  |  |  |  |
| -74.633 | 4.838 | 0.341 |  | -71.048 | 4.276 | 0.302 |  |  |  |  |  |
| -76.444 | 4.434 | 0.313 |  | -71.750 | 3.676 | 0.259 |  |  |  |  |  |
| -78.471 | 4.374 | 0.309 |  | -69.857 | 4.483 | 0.316 |  |  |  |  |  |
| -78.188 | 4.081 | 0.288 |  | -76.855 | 3.815 | 0.269 |  |  |  |  |  |
| -76.463 | 4.105 | 0.290 |  | -78.494 | 4.548 | 0.321 |  |  |  |  |  |
| -73.100 | 4.511 | 0.318 |  | -79.385 | 6.126 | 0.432 |  |  |  |  |  |
| -77.692 | 4.189 | 0.296 |  | -78.107 | 4.880 | 0.344 |  |  |  |  |  |

3) Binding affinity of NAD+ (MM-PBSA results)

|  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| 2H4F (Sir2TM, Ac-p53, NAD+) | | | | 2H4F (Sir2TM, Ac-p53, NAD+, NAM) | | | | 2H4F (Sir2TM, NAD+) | | | |
|  |  |  | Mean\* |  |  |  | Mean |  |  |  | Mean |
| NAD+ Binding: PBSA | | | -23.190 | NAD+ Binding: PBSA | | | -16.154 | NAD+ Binding: PBSA | | | -42.021 |
| ∆G | STD\* | SEM\* |  | ∆G | STD | SEM |  | ∆G | STD | SEM |  |
| -26.484 | 6.256 | 0.441 |  | -24.468 | 6.986 | 0.493 |  | -30.654 | 7.525 | 0.531 |  |
| -17.003 | 7.213 | 0.509 |  | -22.570 | 7.284 | 0.514 |  | -31.828 | 6.511 | 0.459 |  |
| -25.781 | 7.557 | 0.533 |  | -12.154 | 7.569 | 0.534 |  | -34.213 | 6.248 | 0.441 |  |
| -26.374 | 7.798 | 0.550 |  | -4.963 | 6.780 | 0.478 |  | -35.933 | 6.430 | 0.454 |  |
| -18.952 | 6.958 | 0.491 |  | -12.833 | 10.658 | 0.752 |  | -38.179 | 7.946 | 0.561 |  |
| -17.746 | 8.059 | 0.568 |  | -19.590 | 7.648 | 0.539 |  | -43.411 | 8.575 | 0.605 |  |
| -20.030 | 6.889 | 0.486 |  | -14.752 | 8.753 | 0.617 |  | -46.652 | 7.494 | 0.529 |  |
| -22.356 | 7.376 | 0.520 |  | -21.081 | 6.820 | 0.481 |  | -47.116 | 7.419 | 0.523 |  |
| -21.974 | 7.350 | 0.518 |  | -17.202 | 7.078 | 0.499 |  | -47.624 | 6.143 | 0.433 |  |
| -25.778 | 6.570 | 0.463 |  | -16.340 | 7.361 | 0.519 |  | -39.625 | 12.219 | 0.862 |  |
| -25.811 | 6.304 | 0.445 |  | -11.397 | 8.451 | 0.596 |  | -29.091 | 8.947 | 0.631 |  |

4) Binding affinity of NAD+ (MM-GBSA results)

|  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| 2H4F (Sir2TM, Ac-p53, NAD+) | | | | 2H4F (Sir2TM, Ac-p53, NAD+, NAM) | | | | 2H4F (Sir2TM, NAD+) | | | |
|  |  |  | Mean\* |  |  |  | Mean |  |  |  | Mean |
| NAD+ Binding: GBSA | | | -105.477 | NAD+ Binding: GBSA | | | -81.780 | NAD+ Binding: GBSA | | | -106.391 |
| ∆G | STD\* | SEM\* |  | ∆G | STD | SEM |  | ∆G | STD | SEM |  |
| -104.799 | 5.639 | 0.398 |  | -97.840 | 5.816 | 0.410 |  | -106.608 | 7.050 | 0.497 |  |
| -102.452 | 5.643 | 0.398 |  | -97.792 | 5.887 | 0.415 |  | -104.284 | 5.781 | 0.408 |  |
| -101.931 | 6.486 | 0.458 |  | -91.252 | 5.198 | 0.367 |  | -105.740 | 5.745 | 0.405 |  |
| -101.113 | 6.385 | 0.450 |  | -88.668 | 5.284 | 0.373 |  | -107.797 | 5.325 | 0.376 |  |
| -96.338 | 5.904 | 0.417 |  | -88.822 | 6.507 | 0.459 |  | -104.650 | 7.400 | 0.522 |  |
| -96.267 | 6.056 | 0.427 |  | -87.823 | 8.668 | 0.611 |  | -110.253 | 7.035 | 0.496 |  |
| -101.986 | 6.024 | 0.425 |  | -84.056 | 7.869 | 0.555 |  | -110.292 | 6.966 | 0.491 |  |
| -104.826 | 5.645 | 0.398 |  | -86.284 | 6.930 | 0.489 |  | -110.303 | 7.129 | 0.503 |  |
| -104.244 | 5.642 | 0.398 |  | -78.257 | 6.036 | 0.426 |  | -109.455 | 6.034 | 0.426 |  |
| -105.359 | 5.939 | 0.419 |  | -81.595 | 7.871 | 0.555 |  | -107.327 | 8.401 | 0.593 |  |
| -110.972 | 5.514 | 0.389 |  | -78.708 | 8.082 | 0.570 |  | -94.577 | 8.976 | 0.633 |  |

5) Binding affinity of Inhibitor (NAM) (MM-PBSA results)

|  |  |  |  |
| --- | --- | --- | --- |
| 2H4F (Sir2TM, Ac-p53, NAD+, NAM) | | | |
|  |  |  | Mean |
| NAM Binding: PBSA | | | -5.778 |
| ∆G | STD | SEM |  |
| -1.086 | 2.785 | 0.196 |  |
| -0.928 | 3.362 | 0.237 |  |
| -0.033 | 3.161 | 0.223 |  |
| -1.023 | 3.371 | 0.238 |  |
| -2.241 | 3.565 | 0.251 |  |
| -6.352 | 2.659 | 0.188 |  |
| -6.088 | 2.964 | 0.209 |  |
| -6.035 | 3.203 | 0.226 |  |
| -6.560 | 3.133 | 0.221 |  |
| -4.163 | 4.373 | 0.308 |  |
| -6.046 | 3.178 | 0.224 |  |

6) Binding affinity of Inhibitor (NAM) (MM-GBSA results)

|  |  |  |  |
| --- | --- | --- | --- |
| 2H4F (Sir2TM, Ac-p53, NAD+, NAM) | | | |
|  |  |  | Mean |
| NAM Binding: GBSA | | | -20.738 |
| ∆G | STD | SEM |  |
| -21.198 | 2.245 | 0.158 |  |
| -21.562 | 1.939 | 0.137 |  |
| -20.148 | 1.563 | 0.110 |  |
| -19.518 | 1.839 | 0.130 |  |
| -20.402 | 1.939 | 0.137 |  |
| -20.805 | 1.988 | 0.140 |  |
| -20.692 | 1.961 | 0.138 |  |
| -20.671 | 2.077 | 0.147 |  |
| -20.786 | 1.992 | 0.141 |  |
| -20.745 | 2.187 | 0.154 |  |
| -20.795 | 2.013 | 0.142 |  |