Supporting Information

Binding motifs in the CBP bromodomain: an analysis of 20 crystal structures of complexes with small molecules

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Crystallization, data collection and refinement of the CBP/ligand complexes.

CBP bromodomain was purified as previously described[1] and was crystallized with ligands by vapor diffusion in hanging drops at 277 K. Co-crystals of CBP bromodomain in complex with compound **1**, **10**, **15**, **16** and **20** were grown under the condition of 0.15 M potassium thiocyanate, 20% PEG3350 and 10% ethylene glycol. CBP bromodomain was co-crystallized with compound **2** using reservoir buffer of 0.1 M MES, pH 6.5, 0.1 M $MgCl_2$, 20% PEG6000 and 10% ethylene glycol. Co-crystals of CBP with compound **3** and **12** were grown from reservoir buffer of 0.1 M HEPES-Na, pH 7.5, 0.2 M potassium thiocyanate, 20% PEG3350 and 10% ethylene glycol. CBP bromodomain was co-crystallized with compound **9** under the condition of 0.1 M sodium cacodylate, pH 6.5, 0.2 M calcium acetate and 18% PEG8000. Co-crystal of CBP and compound **13** was grown under the condition of 0.1 M HEPES-Na, pH 7.5, 0.2 M $MgCl_2$ and 25% P3350. Co-crystal of CBP bromodomain with compound **17** was grown under the condition of 0.1 M HEPES-Na, pH 7.5, 0.2 M $LiSO_4$ and 25% PEG3350. CBP bromodomain with compound **18** was crystallized against the reservoir buffer of 0.1 M Bis-Tris, pH 6.5, 0.2 M $MgCl_2$, 5% ethylene glycol and 23% PEG3350. Co-crystal of CBP bromodomain with compound **19** was obtained using the reservoir buffer of 0.1 M sodium citrate, pH 5.6 and 1.3 M ammonium sulfate.

Diffraction datasets of co-crystals of CBP/ligand complex were collected at beamlines X06DA and X06SA, Swiss Light Source. Data reduction was performed with XDS[2] and scaled with Aimless.[3] Structures were solved by molecular replacement with Molrep[4] or Phaser[5] using the apo CBP structure 3DWY as a start model. Structures were refined with PHENIX[6] and were manually modelled with COOT.[7] Topology files for compounds were generated from the PRODRG server.[8]

BROMOscan assay

The binding constant (Kd) determinations by means of BROMOscan technology were carried out at DiscoverX. An E. coli strain derived from BL21 was used as the host to grow T7 phage strains displaying the bromodomains. *E. coli*, grown to log-phase, were infected with T7 phage (from a frozen stock, being the multiplicity of infection 0.4) and incubated while shaking at 32 °C for 90-150 minutes until lysis. In order to remove cell debris, lysates were centrifuged at 5,000 x g and filtered (0.2 µm). Affinity resins were obtained by treating streptavidin-coated magnetic beads with biotinylated acetylated peptide ligands for 30 minutes at 25°C. Those beads were then blocked with excess of biotin and washed with blocking buffer (SeaBlock (Pierce), 1 % bovine serum albumin (BSA), 0.05% Tween 20, 1 mM dithiothreitol (DTT) to remove the unbound ligand and reduce non-specific phage binding. During the experiment, the bromodomain, ligand-bound affinity beads and test compounds were combined in a buffer composed of 17% SeaBlock, 33% phosphate-buffered solution (PBS), 0.04% Tween 20, 0.02% BSA, 0.004% sodium azide and 7.4 mM DTT. Test compounds were prepared as 50 mM solutions in pure DMSO and diluted to 5 mM with monoethylene gycol, MEG (100× concentrated in respect to the top screening concentration 50 µM). During the assay the DMSO and MEG final concentrations were 0.1% and 0.9%, respectively. The assays were carried out in polystyrene 96-well plates in a final volume of 0.135 mL. The assay plates were incubated at 25 °C with shaking for 1 hour and the affinity beads were washed with a buffer composed of 0.05% Tween 20 in PBS.

The beads were then re-suspended in the elution buffer (1x PBS, 0.05% Tween 20, 2 μM non-biotinylated affinity ligand) and incubated at 25°C with shaking for 30 minutes. The bromodomain concentration in the elutes was measured by qPCR. K_d values were calculated with a standard dose-response curve using the Hill equation and curves were fitted using a non-linear least square fit with the Levenberg-Marquardt algorithm.

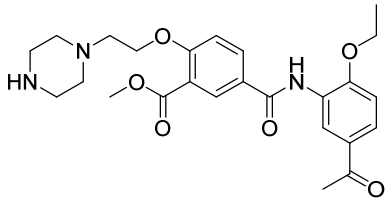
AlphaScreen assay

IC_{50} determinations by means of Amplified Luminescent Proximity Homogeneous Assay (Alpha) Screen technology were carried out at Reaction Biology. Compounds were tested in 10-dose IC_{50} mode with 2 or 3-fold serial dilution starting at varying concentrations. The competitive ligand was H4/4Ac: Histone H4 peptide (1-21) K5/8/12/16Ac-Biotin. Alpha signal (Ex/Em=680/520-620 nm) was detected with an EnSphire plate reader. Data include raw data (signal-background, background was measured without BRD but all other components.), % binding (relative to DMSO controls), and curve fits. An IC_{50} value higher than the starting compound concentration was estimated based on the best curve fitting available. Dose-response curves were fit with GraphPad Prism 6 software.

Synthesized final products characterizations

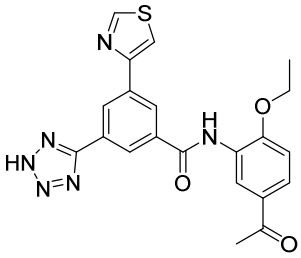
NMR spectra were recorded on AV2 400 or AV2 500 MHz Bruker spectrometers. Chemical shifts are given in ppm. The spectra are calibrated to the residual 1H and 13C signals of the solvents. Multiplicities are abbreviated as follows: singlet (s), doublet (d), triplet (t), quartet (q), doublet-doublet (dd), quintet (quint), multiplet (m), and broad (br). Melting points were determined on a Mettler Toledo MP70 melting point instrument. Infrared spectra were recorded on a JASCO FT/IR-4100 spectrometer. High-resolution electrospray ionization mass spectrometry was performed on a Finnigan MAT 900 (Thermo Finnigan, San Jose, CA, USA) double-focusing magnetic sector mass spectrometer. Ten spectra were acquired. A mass accuracy ≤2 ppm was obtained in the peak matching acquisition mode by using a solution containing 2 μL PEG200, 2 μL PPG450, and 1.5 mg NaOAc (all obtained from Sigma-Aldrich, Buchs, Switzerland) dissolved in 100 mL MeOH (HPLC Supra grade, Scharlau, E-Barcelona) as internal standard.

Methyl 5-((5-acetyl-2-ethoxyphenyl)carbamoyl)-2-(2-(piperazin-1-yl)ethoxy)benzoate (12)



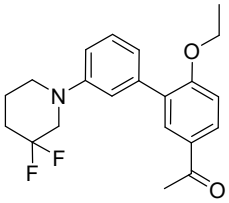
Yellow solid; mp 122-127 °C; ^{1}H NMR (400 MHz, $CDCl_3$): δ = 9.14 (d, J = 2.2 Hz, 1H), 8.56 (s, 1H), 8.35 (d, J = 2.4 Hz, 1H), 8.06 (dd, J = 8.7, 2.5 Hz, 1H), 7.78 (dd, J = 8.6, 2.2 Hz, 1H), 7.09 (d, J = 8.8 Hz, 1H), 6.96 (d, J = 8.7 Hz, 1H), 4.36 – 4.17 (m, 4H), 3.91 (s, 3H), 2.96 – 2.84 (m, 6H), 2.61 – 2.60 (m, 7H), 1.55 (t, J = 7.0 H□, 3H); ^{13}C NMR (101 MHz, $CDCl_3$): δ = 197.2, 165.7, 163.8, 161.1, 151.0, 132.8, 130.6, 130.5, 127.4, 126.6, 124.6, 120.5, 120.3, 113.3, 110.4, 67.8, 64.7, 57.4, 55.1, 52.2, 46.1, 26.6, 14.7; IR (neat): ῡ = 3438, 2927, 2828, 1723, 1677, 1605, 1592, 1540, 1504, 1435, 1258, 1221, 1150, 1116, 1079, 1039, 913, 834, 802, 728, 594, 578, 554 cm^{-1}; HRM□ (E□I), m/□: calcd for $C_{25}H_{32}N_3O_6^+$, 470.2286; found, 470.2288.

***N*-(5-acetyl-2-ethoxyphenyl)-3-(2H-tetrazol-5-yl)-5-(thiazol-4-yl)benzamide (13)**



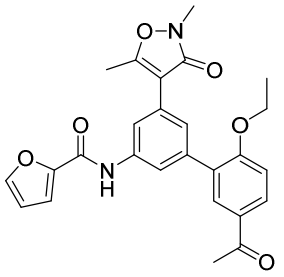
Brown solid; mp 234-235 °C; ^{1}H NMR (400 MHz, DMSO-d_6): δ = 9.97 (s, 1 H), 9.32 (s, 1 H), 8.90 (s, 1 H), 8.75 (s, 1 H), 8.59 (s, 1 H), 8.45 (s, 1 H), 8.32 (s, 1 H), 7.89 (d, *J* = 8.3 Hz, 1 H), 7.24 (d, *J* = 8.3 Hz, 1 H), 4.23 (q, *J* = 7.2 Hz, 2 H), 2.56 (s, 3 H), 1.39 (t, *J* = 7.0 H□, 3 H); ^{13}C NMR (126 MHz, DMSO-d_6): δ = 196.2, 164.5, 155.4, 153.4, 136.3, 135.4, 129.4, 129.2, 127.6, 127.3, 127.2, 127.0, 126.6, 125.7, 125.0, 116.5, 112.0, 64.4, 26.4, 14.4. One carbon is missing due to overlapping; IR (neat): ῡ = 3434, 1671, 1652, 1584, 1538, 1435, 1337, 1267, 1030, 889, 835, 816, 736, 597 cm^{-1}; HRM□(E□I), m/□: calcd for $C_{21}H_{19}N_6O_3S^+$: 435.1239 found: 435.1236.

1-(3'-(3,3-difluoropiperidin-1-yl)-6-ethoxy-[1,1'-biphenyl]-3-yl)ethan-1-one (15)



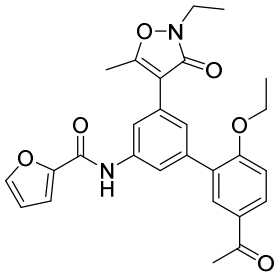
Yellow oil; ^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.97 - 7.93 (m, 2 H), 7.33 (t, *J* = 7.9 Hz, 1 H), 7.15 (t, *J* = 1.9 Hz, 1 H), 7.07 (dt, *J* = 7.5, 1.5 Hz, 1 H), 7.00 (d, *J* = 8.7 Hz, 1 H), 6.94 (dd, *J* = 7.9, 2.3 Hz, 1 H), 4.15 (q, *J* = 7.2 Hz, 2 H), 3.42 (t, *J* = 11.5 Hz, 2 H), 3.25 (t, *J* = 5.3 Hz, 2 H), 2.59 (s, 3 H), 2.11 - 1.99 (m, 2 H), 1.98 - 1.90 (m, 2 H), 1.41 (t, *J* = 7.0 H□, 3 H); ^{13}C NMR (101 MHz, $CDCl_3$): δ = 196.8, 159.8, 150.2, 138.6, 131.4, 130.7, 130.2, 129.6, 128.8, 121.8, 118.4, 120.1 (t, *J* = 243.2 Hz), 116.0, 111.5, 64.2, 55.6 (t, *J* = 29.8 Hz), 49.2, 32.3 (t, *J* = 23.8 Hz), 26.4, 22.2 (t, *J* = 5.4 H□), 14.6; IR (neat): ῡ = 1673, 1597, 1445, 1355, 1264, 1242, 1212, 1100, 1039, 916, 751, 699, 587 cm^{-1}; HRM□(E□I), m/□: calcd for C*21*H*24*F2NO*2*$^{+}$: 360.1775 found: 360.1767.

***N*-(5'-acetyl-5-(2,5-dimethyl-3-oxo-2,3-dihydroisoxazol-4-yl)-2'-ethoxy-[1,1'-biphenyl]-3-yl)furan-2-carboxamide (16)**



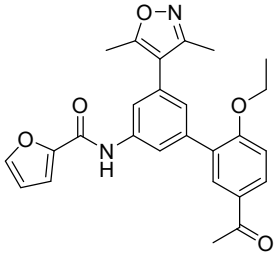
Beige solid; mp 114-115 °C; ^{1}H NMR (400 MHz, $CDCl_3$): δ = 8.22 (br s, 1 H), 8.01 - 7.94 (2 H), 7.88 (d, *J* = 9 4 Hz, 2 H), 7.54 (br s, 2 H), 7.26 (br s, 1 H), 7.01 (d, *J* = 9.0 Hz, 1 H), 6.58 (br s, 1 H), 4.17 (q, *J* = 6.8 Hz, 2 H), 3.58 (br s, 3 H), 2.60 (s, 3 H), 2.49 (s, 3 H), 1.43 (t, *J* = 6.6 H□, 3 H); ^{13}C NMR (101 MHz, $CDCl_3$): δ = 196.8, 166.6, 164.7, 159.8, 156.1, 147.7, 144.3, 139.0, 137.4, 131.7, 130.1, 129.9, 129.5, 125.4, 120.3, 118.4, 115.3, 112.6, 111.6, 109.8, 64.4, 26.4, 14.7, 13.3. Two carbons are missing due to overlapping; IR (neat): ῡ = 1660, 1593, 1435, 1256, 1039, 759, 586 cm^{-1}; HRM□(E□I), m/□: calcd for $C_{26}H_{25}N_2O_6^+$: 461.1713 found: 461.1711.

***N*-(5'-acetyl-2'-ethoxy-5-(2-ethyl-5-methyl-3-oxo-2,3-dihydroisoxazol-4-yl)-[1,1'-biphenyl]-3-yl)furan-2-carboxamide (17)**



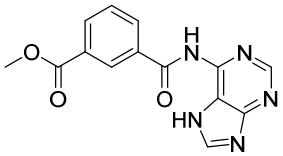
White solid; mp 138-140 °C (decomposition); ^{1}H NMR (400 MHz, $CDCl_3$): δ = 8.26 (br s, 1H), 7.96 - 7.94 (m, 2H), 7.87 - 7.86 (m, 2H), 7.52 - 7.51 (m, 2H), 7.24 (d, *J* = 3.6 Hz, 1H), 6.99 (d, *J* = 9.2 Hz, 1H), 6.56 - 6.55 (m, 1H), 4.15 (q, *J* = 7.0 Hz, 2H), 3.98 (q, *J* = 7.2 Hz, 2H), 2.58 (s, 3H), 2.48 (s, 3H), 1.40 (t, *J* = 7.0 Hz, 3H), 1.35 (t, *J* = 7.2 H□, 3H); ^{13}C NMR (101 MHz, $CDCl_3$): δ = 197.0, 166.4, 165.0, 159.9, 156.3, 147.9, 144.4, 139.1, 137.5, 131.8, 130.2, 130.1, 130.0, 129.8, 125.5, 120.4, 118.6, 115.4, 112.8, 111.7, 110.1, 64.5, 41.4, 26.6, 14.8, 13.4, 12.9; IR (neat): ῡ = 3271, 2984, 2358, 2338, 1672, 1645, 1637, 1608, 1593, 1579, 1551, 1437, 1410, 1355, 1291, 1252, 1226, 1201, 1152, 1039, 1009, 973, 916, 877, 805, 756, 723, 699, 618, 611, 594, 582 cm^{-1}; HRMS (ESI) m/z calcd for $C_{27}H_{27}O_6N_2^+$: 475.1864, found: 475.1866.

_N_-(5'-acetyl-5-(3,5-dimethylisoxazol-4-yl)-2'-ethoxy-[1,1'-biphenyl]-3-yl)furan-2-carboxamide (18)



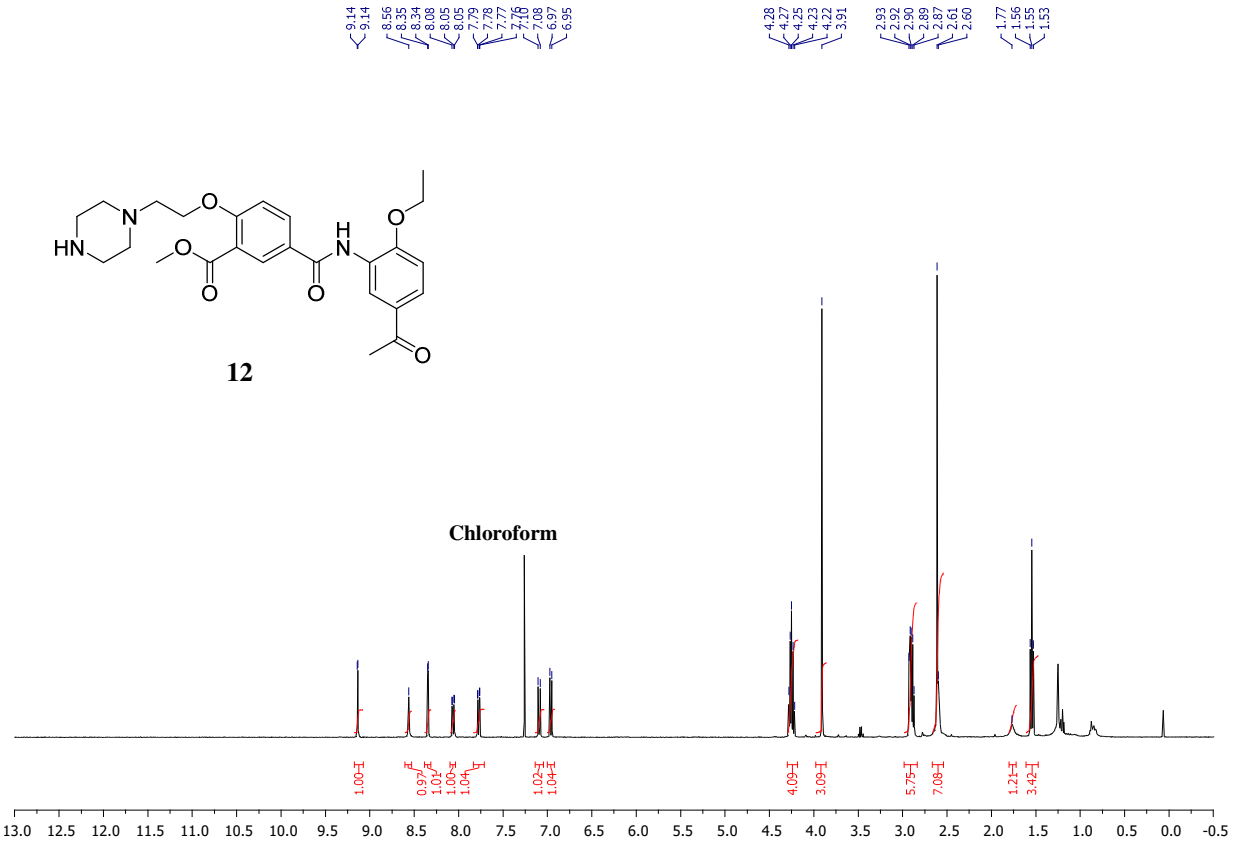
White solid; mp 191-192 °C; ^{1}H NMR (400 MHz, $CDCl_3$): δ = 8.19 (br s, 1H), 7.98 - 7.96 (m, 2H), 7.78 (t, *J* = 1.6 Hz, 1H), 7.62 (t, *J* = 1.6 Hz, 1H), 7.54 - 7.53 (m, 1H), 7.27 - 7.26 (m, 1H), 7.23 (t, *J* = 1.2 Hz, 1H), 7.03 (d, *J* = 9.2 Hz, 1H), 6.58 (m, 1H), 4.17 (q, *J* = 7.0 Hz, 2H), 2.59 (s, 3H), 2.48 (s, 3H), 2.35 (s, 3H), 1.41 (t, *J* = 7.0 H□, 3H); ^{13}C NMR (101 MHz, $CDCl_3$): δ = 196.8, 165.7, 159.9, 158.9, 156.3, 147.8, 144.5, 139.3, 137.9, 131.6, 131.2, 130.4, 130.3, 129.8, 126.6, 120.1, 119.7, 116.5, 115.7, 112.9, 111.7, 64.6, 26.5, 14.8, 11.9, 11.1; IR (neat): ῡ = 3266, 1673, 1644, 1608, 1593, 1580, 1564, 1551, 1438, 1411, 1355, 1318, 1290, 1252, 1226, 1201, 1152, 1039, 1009, 973, 916, 877, 805, 756, 727, 700, 618, 611, 595, 582 cm^{-1}; HRMS (ESI) m/z calcd for [$C_{26}H_{25}N_2O_5^+$]: 445.1758; found: 445.1765.

Methyl 3-((7H-purin-6-yl)carbamoyl)benzoate (20)



Off-white solid; mp 216-221 °C; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 12.39 (s, 1H), 11.79 (s, 1H), 8.75 (s, 1H), 8.69 (s, 1H), 8.53 (s, 1H), 8.38 (d, *J* = 7.4 Hz, 1H), 8.22 (d, *J* = 7.4 Hz, 1H), 7.74 (t, *J* = 7.7 H□, 1H), 3.92 (s, 3H); ^{13}C NMR (126 MHz, DMSO-d_6): δ = 172.0, 166.5, 165.7, 165.6, 162.2, 151.1, 146.3, 144.4, 133.8, 133.5, 133.1, 133.0, 130.0, 129.9, 129.8, 129.5, 129.4, 129.2, 114.6, 52.5; IR (neat): ῡ = 3437, 3362, 2962, 1728, 1695, 1624, 1593, 1531, 1381, 1301, 1247, 1095, 1080, 888, 722, 699 cm^{-1}; HRM□ (E□I): m/□ calcd for $C_{14}H_{10}N_5O_3^-$: 296.0789, found: 296.0789.

NMR traces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7.08
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6.95
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4.27
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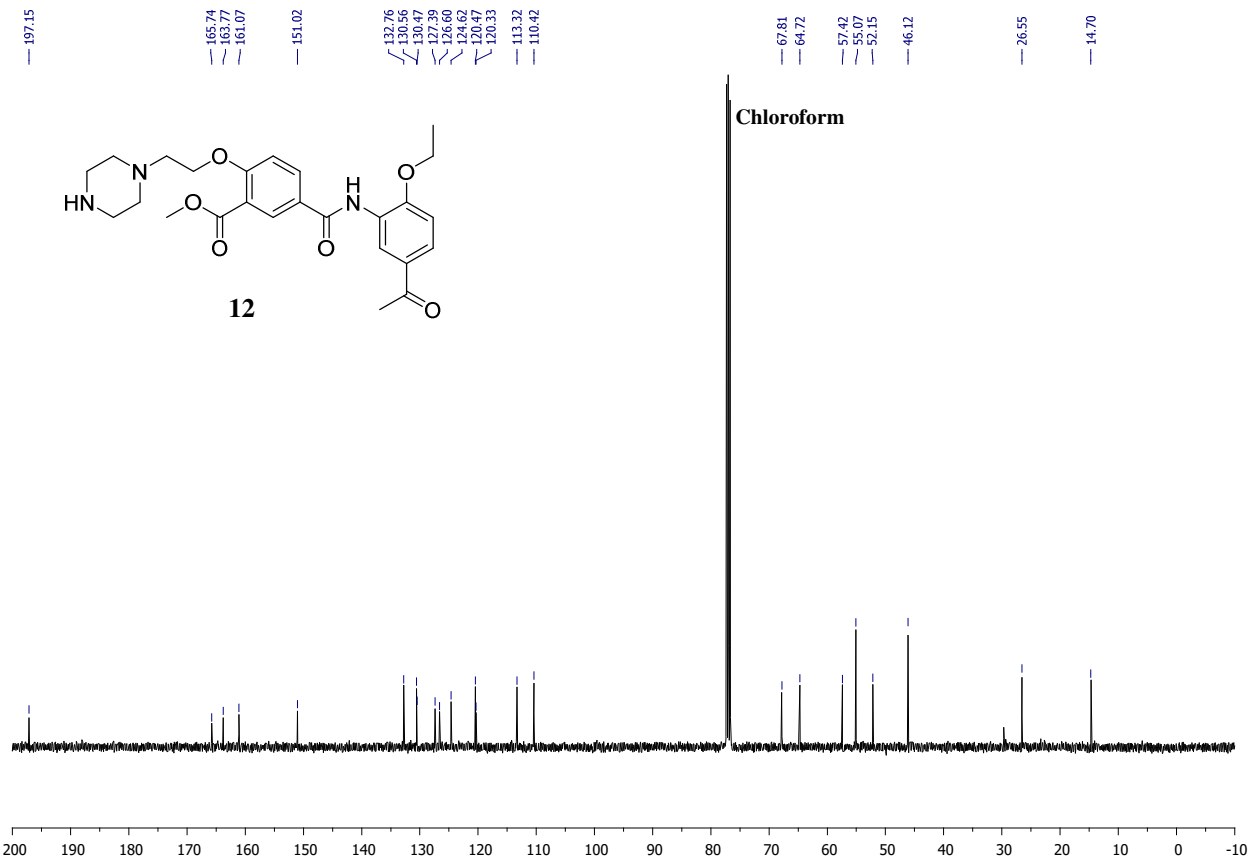
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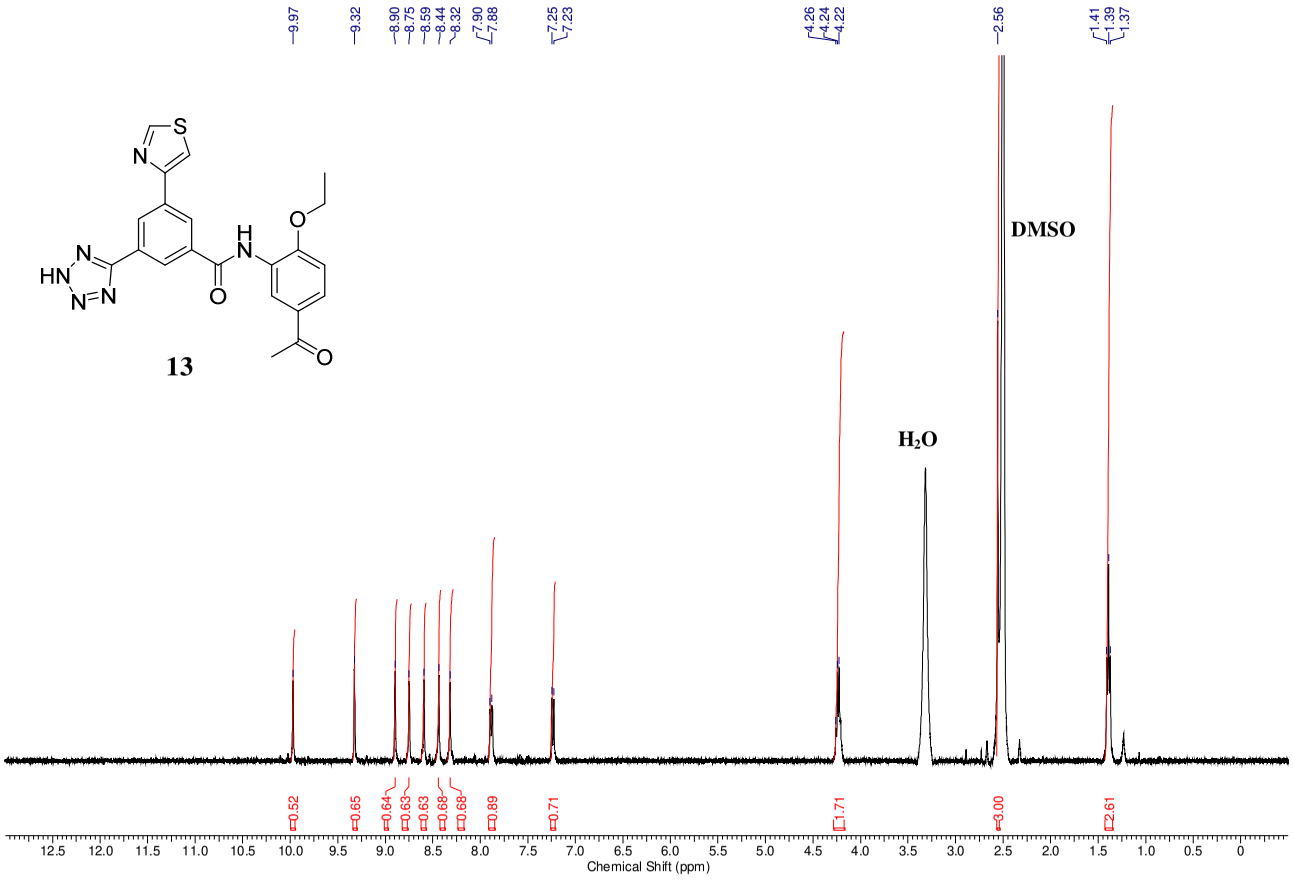
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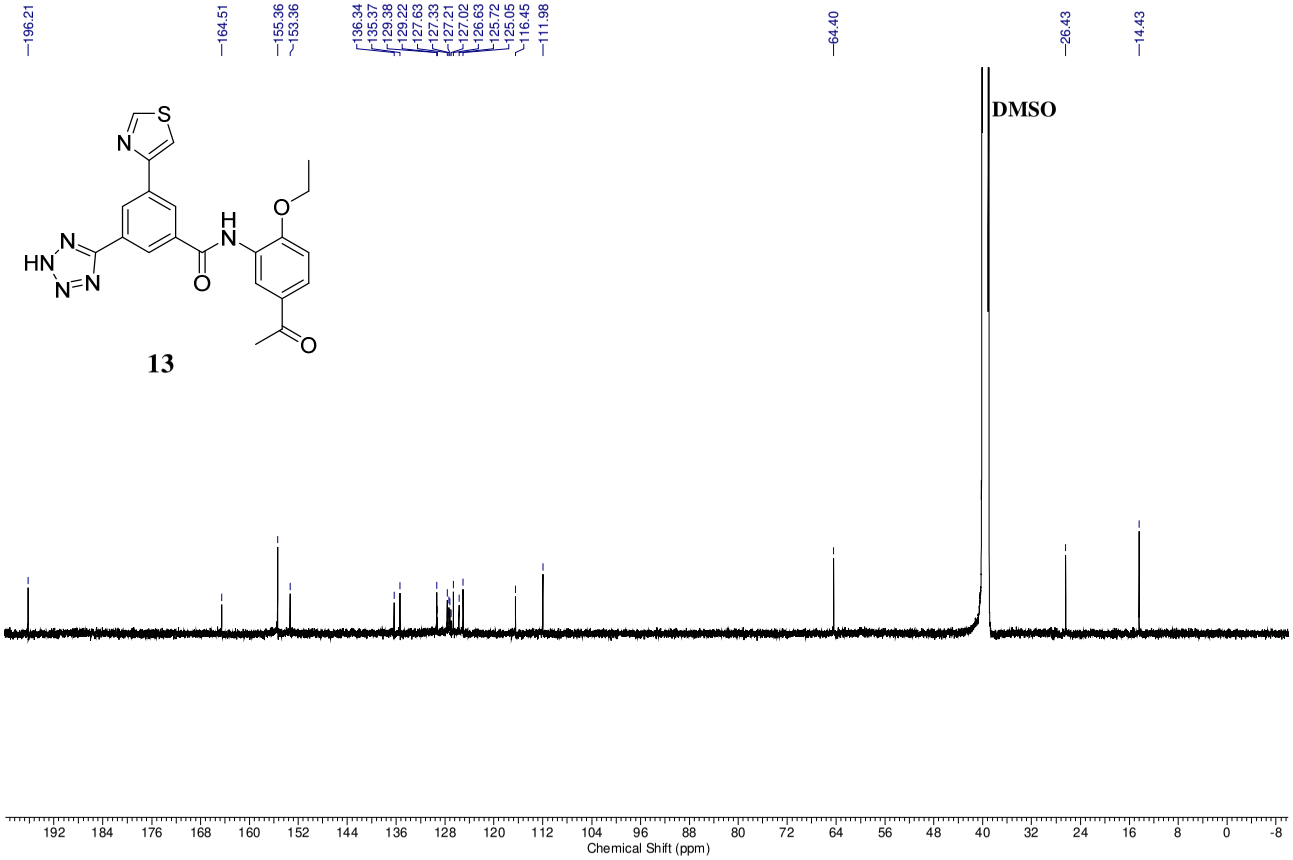
Chemical Shift (ppm)

13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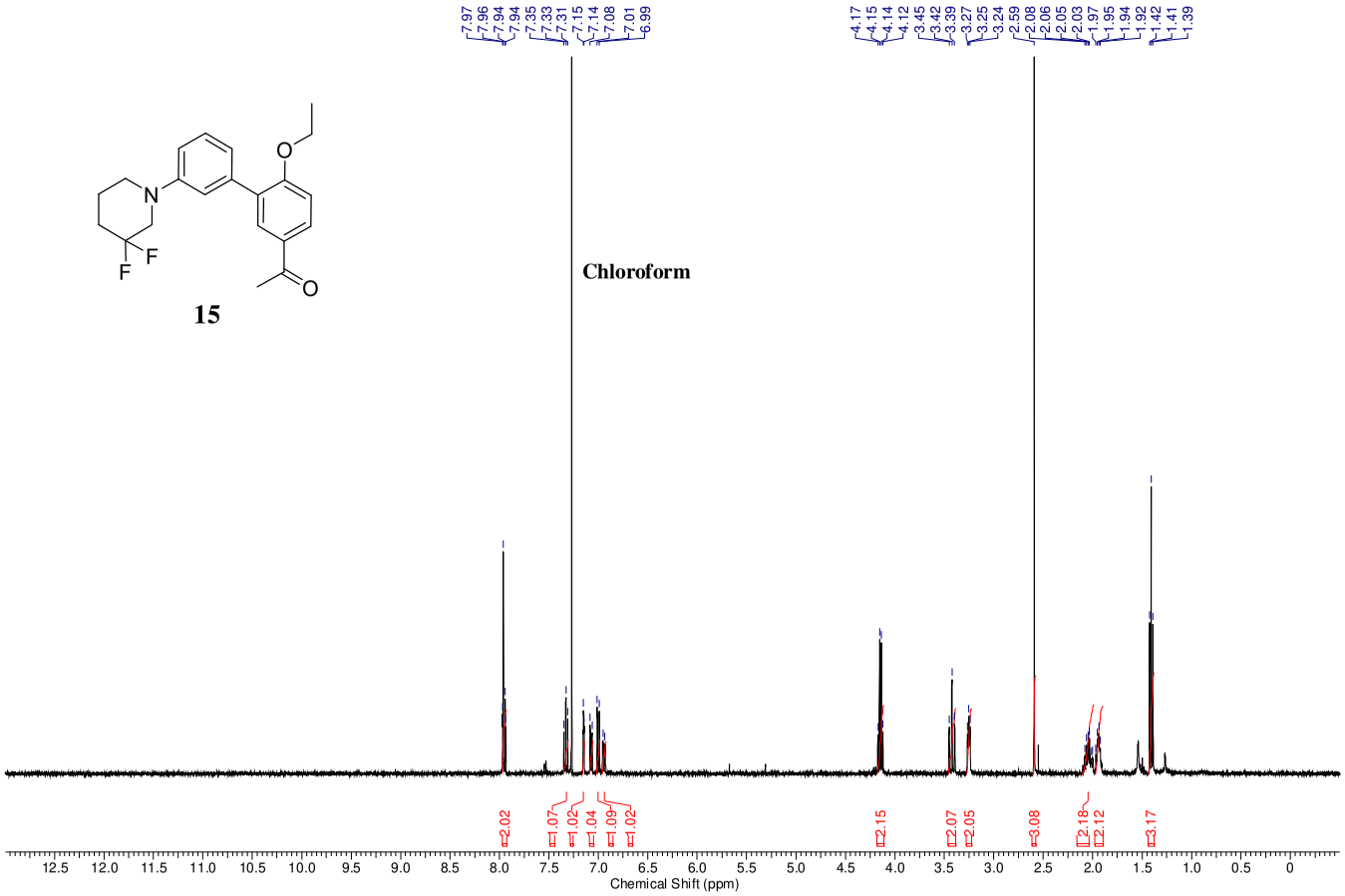
DMSO
Chemical Shift (ppm)

13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1.95
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1.92
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1.41
1.39
Chloroform
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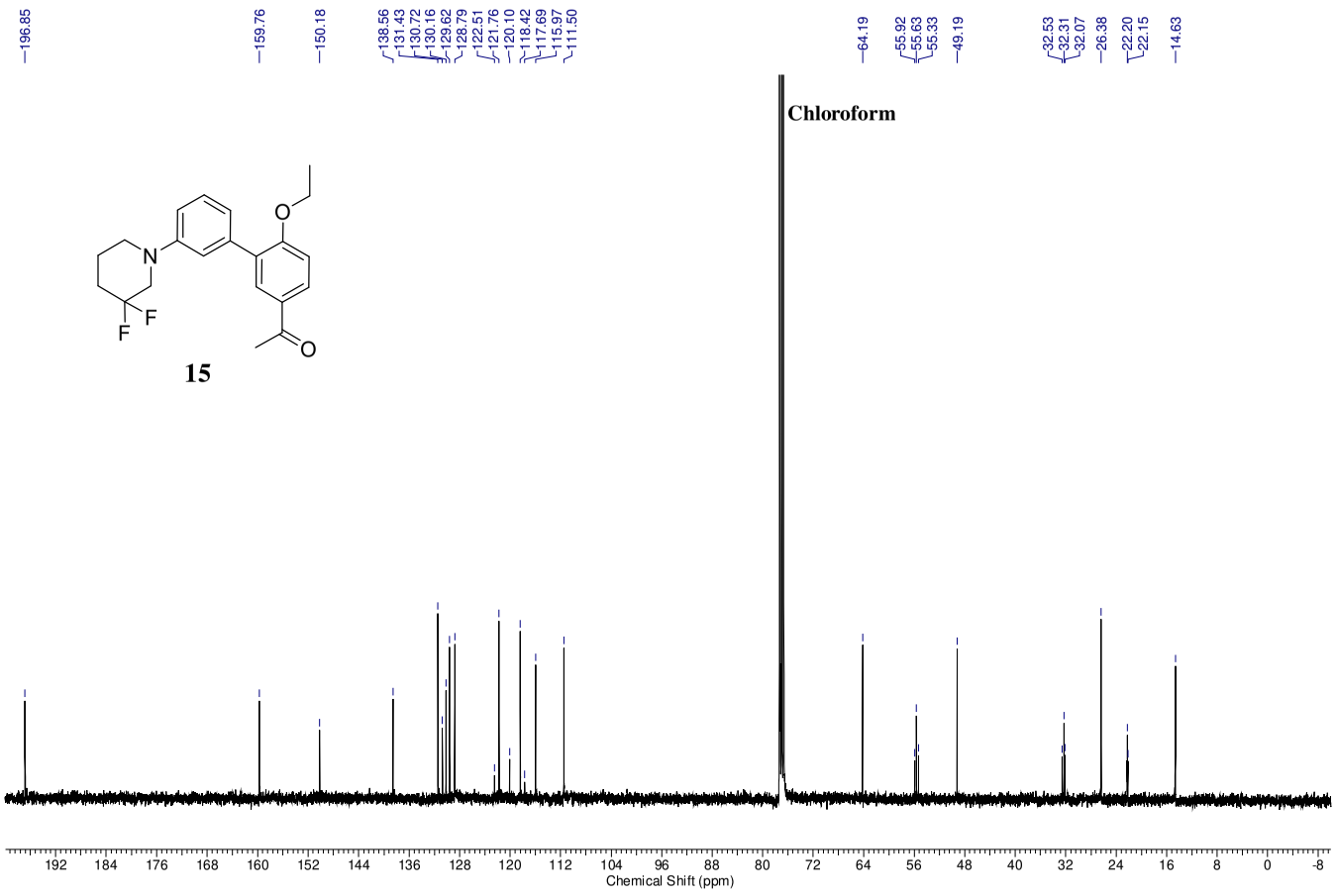
3.17
Chemical Shift (ppm)

15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111.50
64.19
55.92
55.63
55.33
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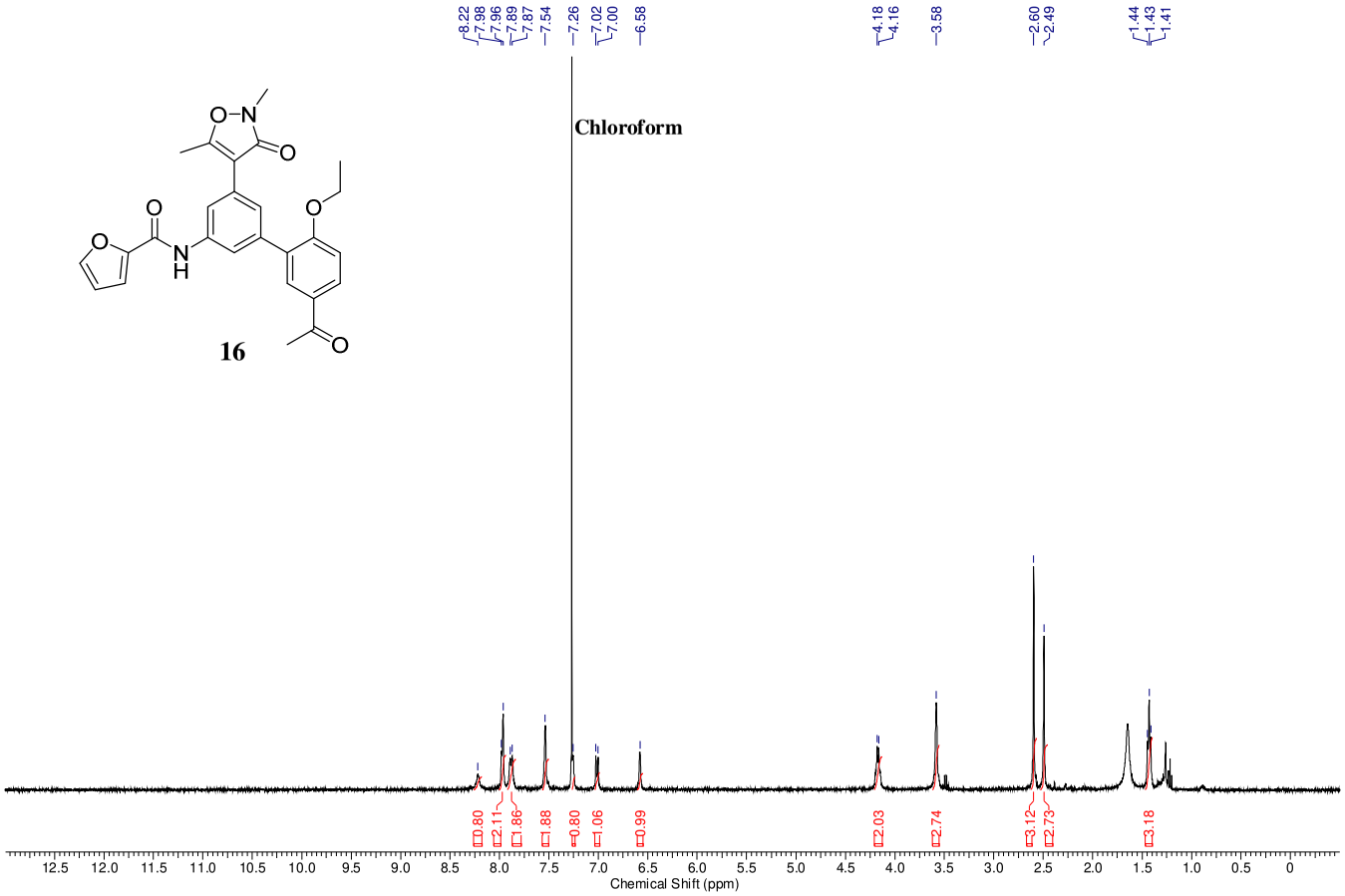
32.31
32.07
26.38
22.20
22.15
14.63
Chloroform
Chemical Shift (ppm)

15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7.54
7.26
7.02
7.00
6.58
4.18
4.16
3.58
2.60
2.49
1.44
1.43
1.41
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O
N
O
O
O
N
H
O
O
16
0.80
2.11
1.86
1.88
0.80
1.06
0.99
2.03
2.74
3.12
2.73
3.18
12.5 12.0 11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0
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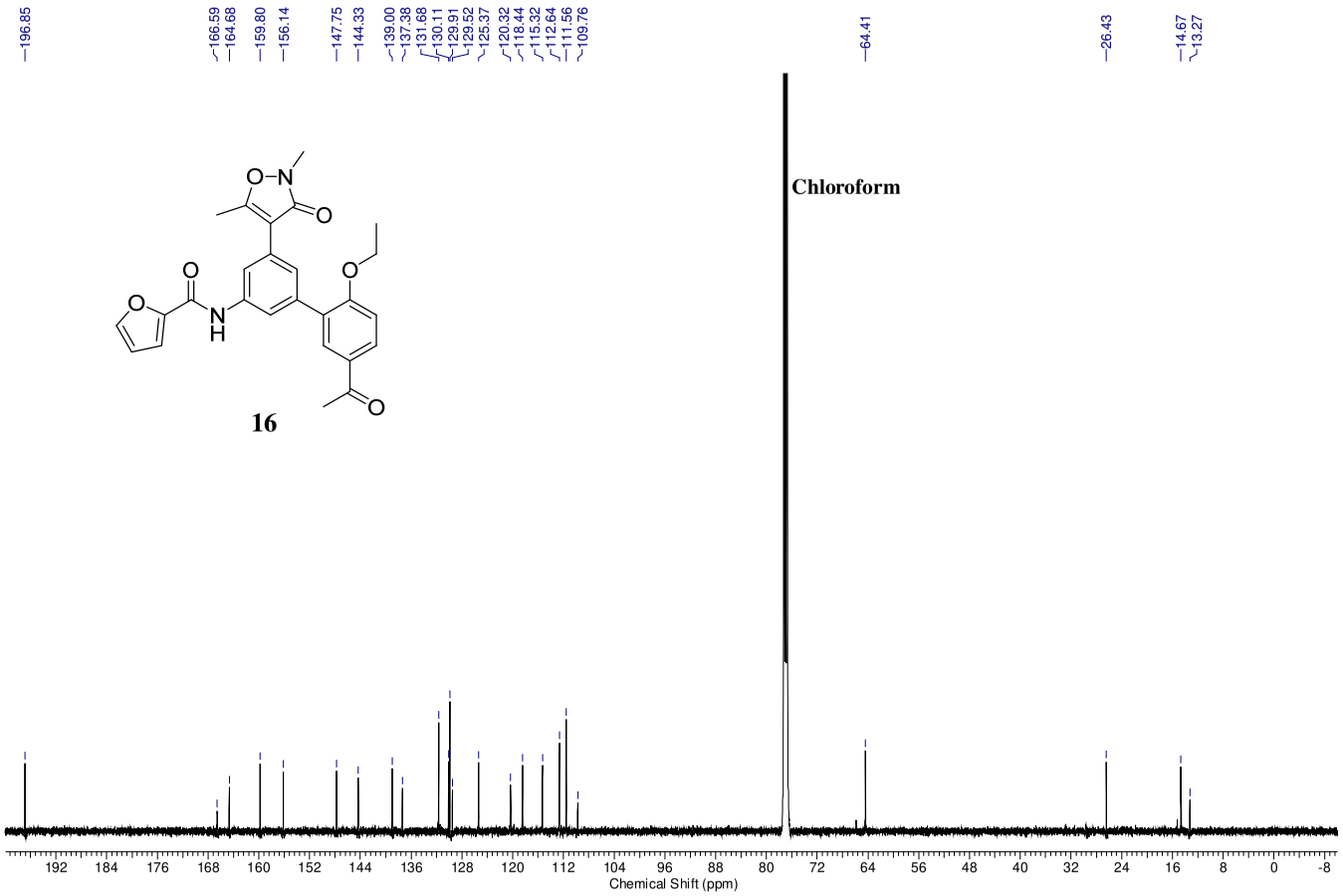
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125.37
120.32
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112.64
111.56
109.76
64.41
26.43
14.67
13.27
Chloroform
O
N
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O
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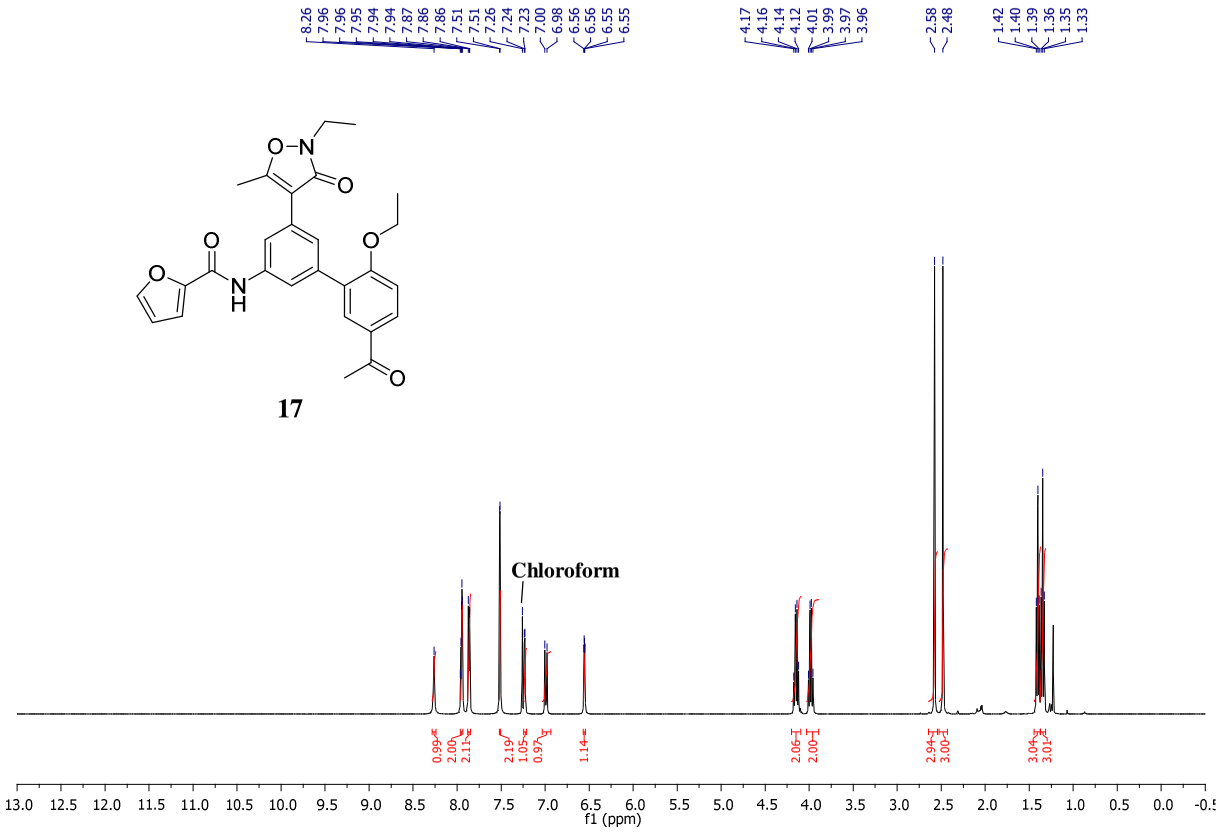
Chemical Shift (ppm)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7.24
7.23
7.00
6.98
6.56
6.56
6.55
6.55
4.17
4.16
4.14
4.12
4.01
3.99
3.97
3.96
2.58
2.48
1.42
1.40
1.39
1.36
1.35
1.33
Chloroform
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2.00
2.11
2.19
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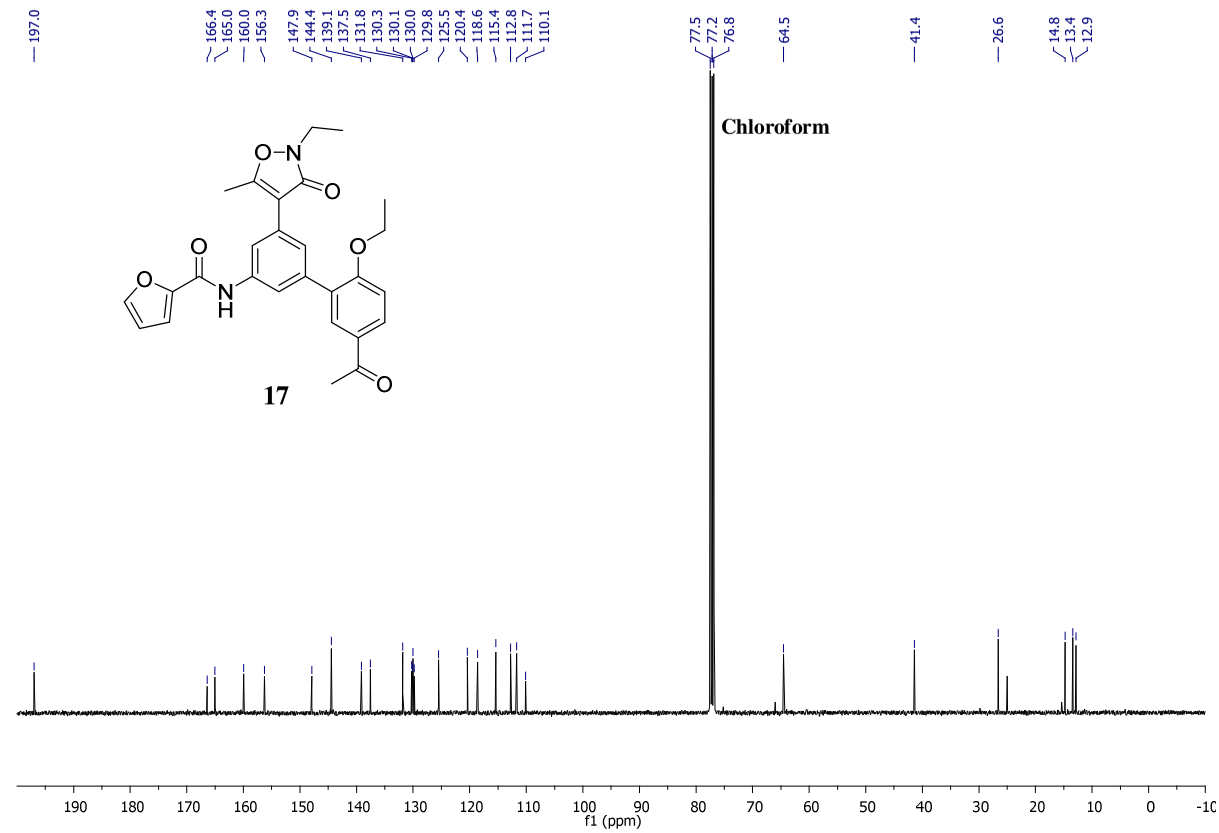
1.14
2.06
2.00
2.94
3.00
3.04
3.01
f1 (ppm)

17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129.8
125.5
120.4
118.6
115.4
112.8
111.7
110.1
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64.5
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13.4
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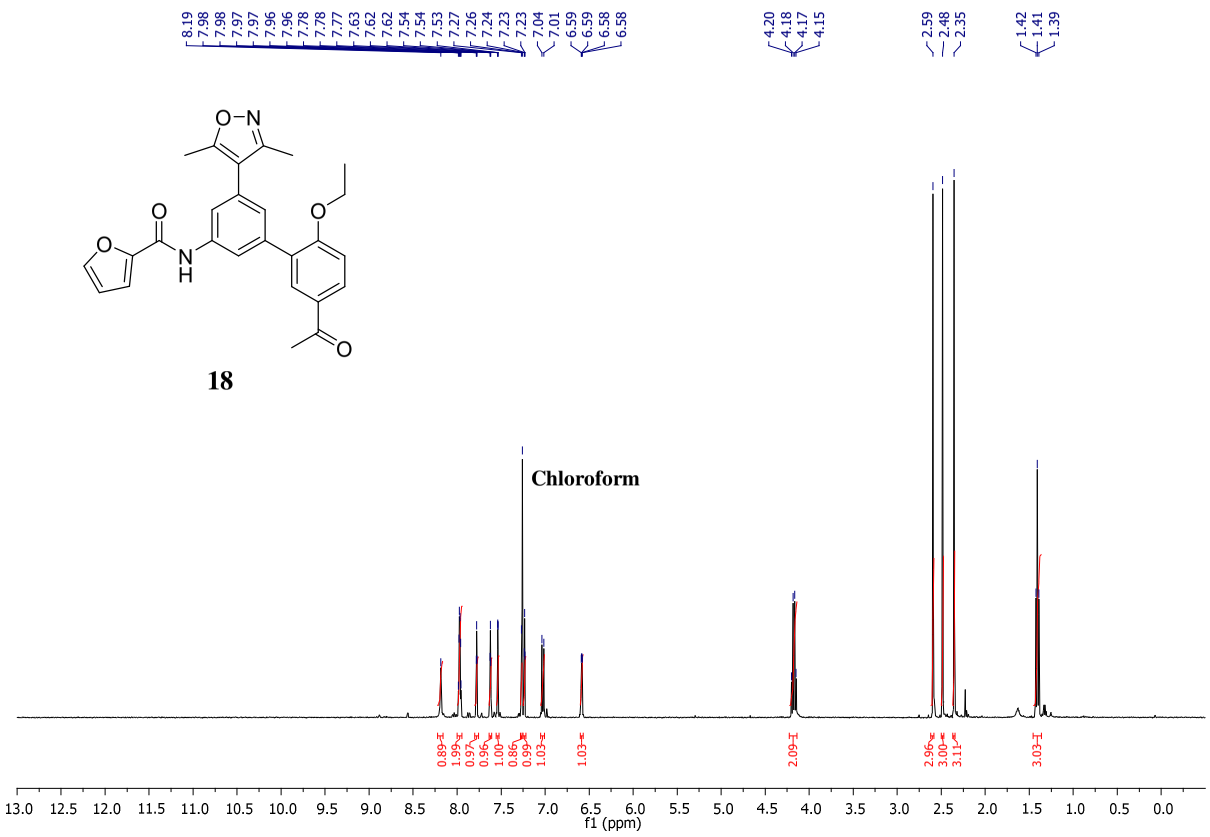
Chloroform
f1 (ppm)

17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7.53
7.27
7.26
7.24
7.23
7.23
7.04
7.01
6.59
6.59
6.58
6.58
4.20
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4.17
4.15
2.59
2.48
2.35
1.42
1.41
1.39
O–N
O
O
N
H
O
O
18
Chloroform
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0.97
0.96
1.00
0.86
0.99
1.03
1.03
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2.96
3.00
3.11
3.03
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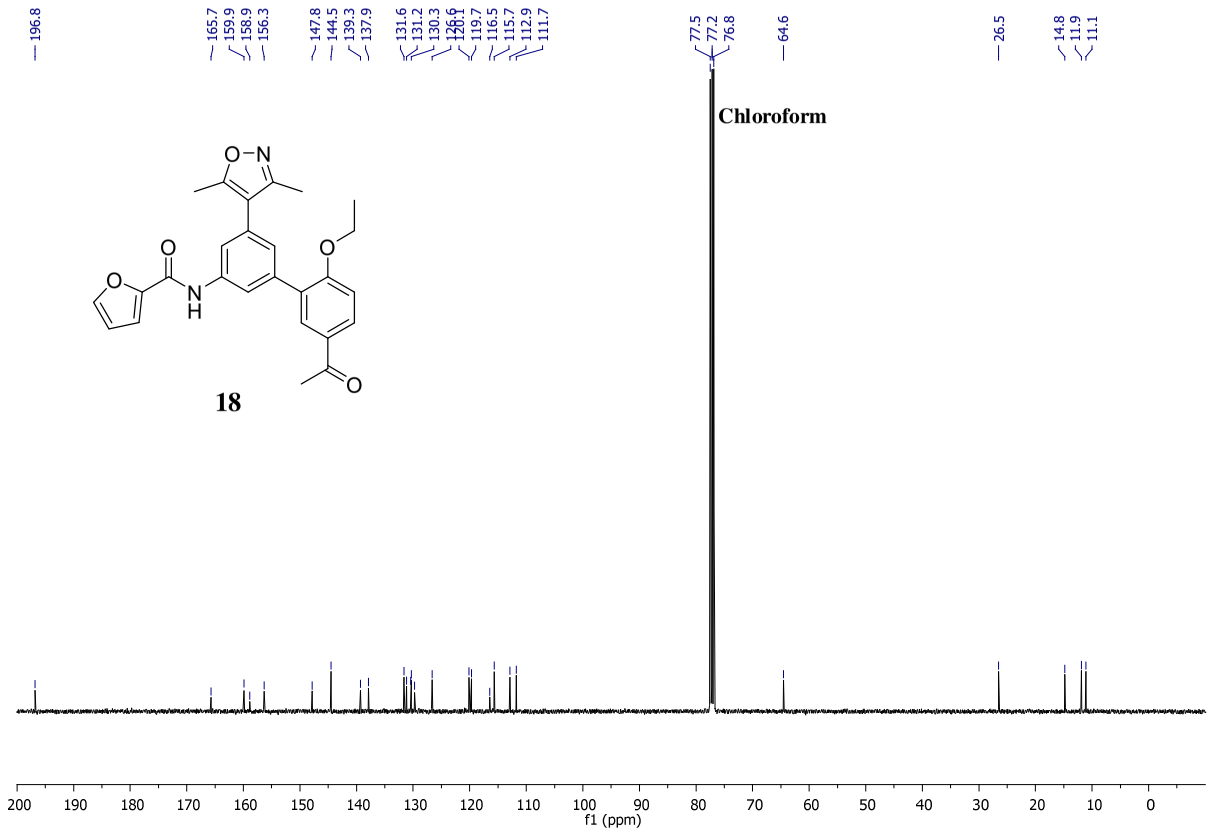
f1 (ppm)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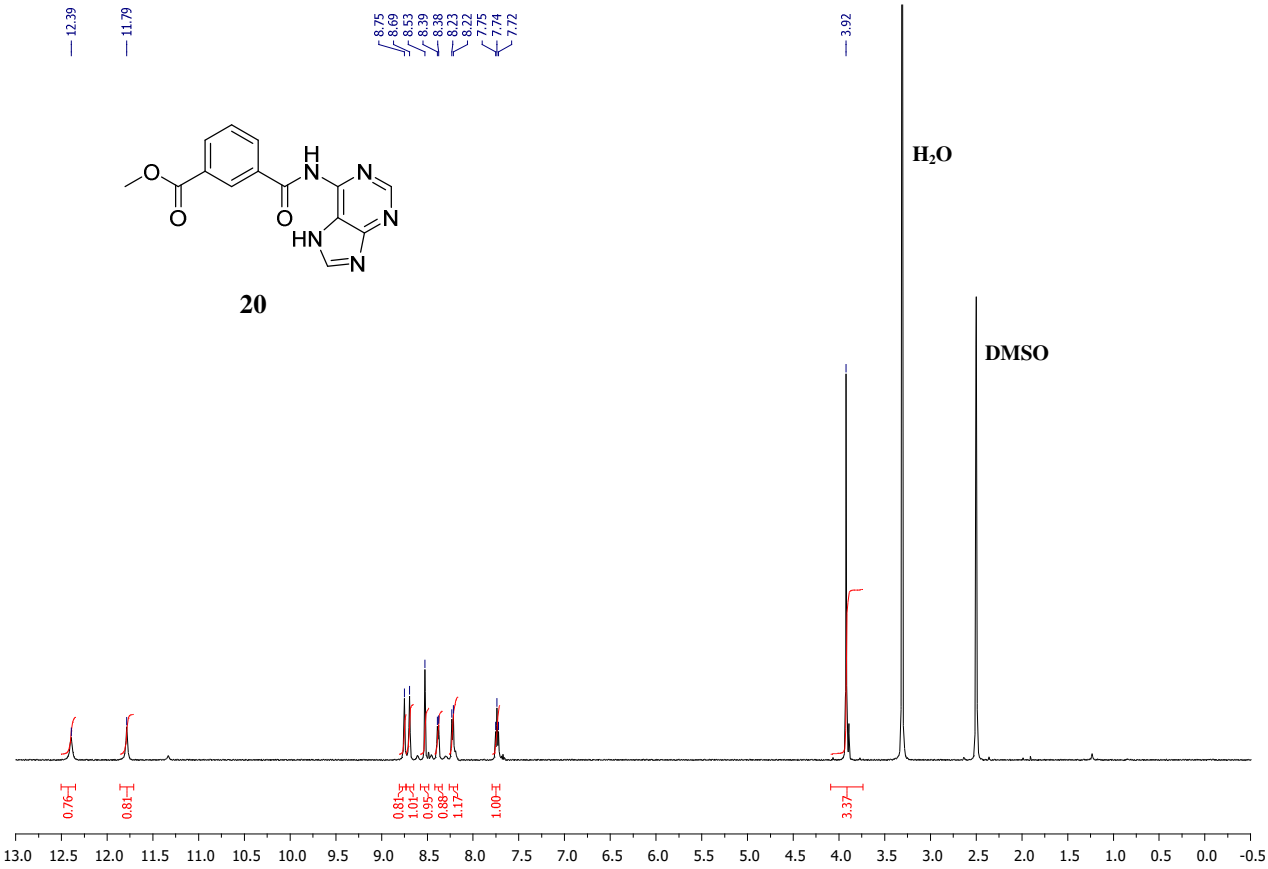
115.7
112.9
111.7
77.5
77.2
76.8
64.6
26.5
14.8
11.9
11.1
Chloroform
O–N
O
O
N
H
O
O
18
200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0
f1 (ppm)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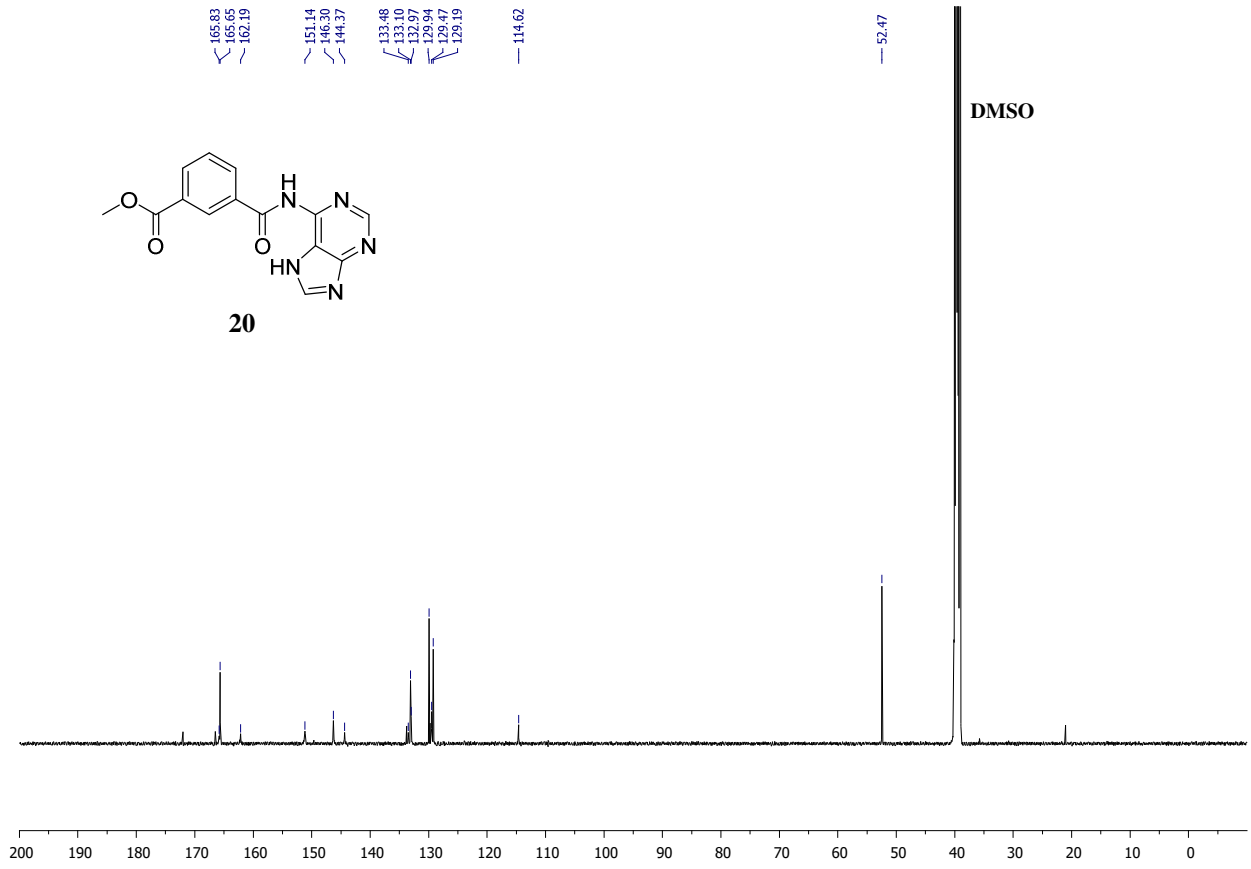
3.92
H2O
DMSO
0.76
0.81
0.81
1.01
0.95
0.89
1.17
1.00
3.37
13.0 12.5 12.0 11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0 -0.5

20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151.14
146.30
144.37
133.48
133.10
132.97
129.94
129.47
129.19
114.62
52.47
DMSO
200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0

20

Table S1. X-ray data collection and refinement statistics for complex structures of the CBP bromodomain and compounds.

PDB ID	**5EIC**	**5OWK**	**5EP7**	**5MME**
Compound	**1**	**2**	**3**	**9**
Data Collection				
Space group	H3	$P2_12_12_1$	$P2_12_12_1$	$P2_12_12_1$
Cell dimensions a, b, c (Å)	121.26, 121.26, 40.78	44.34, 45.07, 61.15	24.35, 24.35, 104.23	53.37, 54.26, 81.05
Cell dimensions α, β, γ (°)	90.00, 90.00, 120.00	90.00, 90.00, 90.00	90.00, 90.00, 90.00	90.00, 90.00, 90.00
Resolution (Å)	38.01 - 1.50	45.07 - 1.25	45.36 - 1.20	45.09 - 1.35
Unique observations*	35709 (5211)	33757 (1613)	37308 (5255)	51995 (7336)
Completeness*	99.8 (99.7)	97.9 (96.0)	99.7 (98.1)	99.2 (97.2)
Redundancy*	5.1 (5.0)	6.0 (5.8)	11.6 (10.6)	6.4 (5.6)
Rmerge*	0.055 (0.310)	0.041 (0.089)	0.036 (0.154)	0.048 (0.609)
CC (1/2)	0.998 (0.937)	0.997 (0.951)	1.000 (0.991)	0.999 (0.786)
I/σI*	15.6 (5.5)	28.8 (14.2)	37.4 (13.3)	17.2 (2.8)
Refinement				
R_{work}/R_{free}*	0.173/0.209 (0.201/0.244)	0.162/0.176 (0.162/0.179)	0.166/0.176 (0.170/0.185)	0.164/0.176 (0.246/0.266)
R.m.s deviations bond (Å)	0.006	0.005	0.011	0.005
R.m.s deviations angles (°)	0.892	0.955	1.158	0.884
B-factors(P/L/O) ($Å^2$) **	12.0/32.0/24.4	11.4/15.7/26.4	12.0/12.2/24.4	18.3/15.5/30.8
Ramanchandran Favored	100	100	100	99.11
Ramanchandran Allowed	0	0	0	0.89
Ramanchandran Disallowed	0	0	0	0

PDB ID	**5MMG**	**5ENG**	**5MPK**	**6FQU**
Compound	**10**	**12**	**13**	**15**
Data Collection				
Space group	$P2_12_12_1$	$P2_12_12_1$	$P2_12_12_1$	$P2_12_12_1$
Cell dimensions a, b, c (Å)	34.18, 49.30, 80.50	34.24, 49.71, 80.35	45.08, 60.20, 87.64	35.26, 49.76, 80.58
Cell dimensions α, β, γ (°)	90.00, 90.00, 90.00	90.00, 90.00, 90.00	90.00, 90.00, 90.00	90.00, 90.00, 90.00
Resolution (Å)	42.04 - 1.23	42.27 - 1.30	45.08 - 1.90	42.34 - 1.43
Unique observations*	39623 (5607)	34079 (4672)	19464 (2783)	26946 (1291)
Completeness*	98.4 (97.0)	98.7 (94.4)	99.9 (99.9)	99.9 (99.4)
Redundancy*	6.2 (5.6)	6.8 (5.5)	8.8 (9.1)	12.5 (8.0)
Rmerge*	0.032 (0.163)	0.031 (0.162)	0.091 (0.967)	0.060 (0.469)
CC (1/2)	0.999 (0.980)	1.000 (0.983)	0.999 (0.811)	0.999 (0.864)
I/σI*	28.2 (9.9)	33.2 (9.5)	12.0 (2.4)	24.3 (4.2)
Refinement				
R_{work}/R_{free}*	0.157/0.163 (0.170/0.167)	0.165/0.173 (0.167/0.176)	0.183/0.239 (0.312/0.343)	0.170/0.194 (0.313/0.346)
R.m.s deviations bond (Å)	0.005	0.008	0.011	0.007
R.m.s deviations angles (°)	0.872	1.113	0.955	1.126
B-factors(P/L/O) ($Å^2$) **	13.4/13.6/29.9	12.1/27.4/25.1	41.7/44.3/41.3	14.6/19.0/30.3
Ramanchandran Favored	98.9	99.00	99.09	99.02
Ramanchandran Allowed	1.10	1.00	0.91	0.98
Ramanchandran Disallowed	0	0	0	0

PDB ID	**6FQO**	**6FR0**	**6FRF**	**5MPN**
Ligand	**16**	**17**	**18**	**19**
Data Collection				
Space group	$P2_1$	$P2_1$	$P2_1$	$P2_12_12_1$
Cell dimensions a, b, c (Å)	50.74, 43.66, 51.30	51.31, 43.88, 51.42	54.64, 93.88, 62.53	44.58, 44.72, 60.76
Cell dimensions α, β, γ (°)	90.00, 99.42, 90.00	90.00, 99.21, 90.00	90.00, 94.22, 90.00	90.00, 90.00, 90.00
Resolution (Å)	43.66 - 1.35	43.88 - 1.50	47.13 - 2.10	44.72 -1.23
Unique observations*	48359 (2388)	35942 (1776)	36396 (2948)	34316 (4552)
Completeness*	99.0 (96.1)	98.8 (98.9)	99.1 (98.6)	95.6 (88.4)
Redundancy*	6.6 (5.8)	6.6 (6.6)	4.7 (4.2)	6.9 (6.8)
Rmerge*	0.033 (0.452)	0.062 (0.417)	0.058 (0.602)	0.058 (0.364)
CC (1/2)	1.000 (0.898)	0.998 (0.920)	0.999 (0.769)	0.999 (0.924)
I/σI*	26.5 (3.4)	14.5 (3.9)	15.1 (2.2)	16.5 (5.1)
Refinement				
R_{work}/R_{free}*	0.161/0.199 (0.171/0.219)	0.178/0.202 (0.202/0.223)	0.175/0.228 (0.241/0.302)	0.158/0.167 (0.192/0.219)
R.m.s deviations bond (Å)	0.006	0.007	0.008	0.014
R.m.s deviations angles (°)	0.832	1.062	1.051	1.000
B-factors(P/L/O) ($Å^2$) **	18.5/12.2/29.0	26.6/21.7/33.8	39.9/33.0/40.9	15.1/14.3/28.2
Ramanchandran Favored	100	100	99.78	100
Ramanchandran Allowed	0	0	0.22	0
Ramanchandran Disallowed	0	0	0	0

* Statistics for the highest resolution shell is shown in parentheses.
* * P/L/O indicate protein, ligand in the active site and solvent molecules, respectively.

PDB ID	**5H85**			
Compound	**20**			
Data Collection				
Space group	$P2_12_12_1$			
Cell dimensions a, b, c (Å)	24.41, 45.31, 104.66			
Cell dimensions α, β, γ (°)	90.00, 90.00, 90.00			
Resolution (Å)	45.31-1.70			
Unique observations*	13297 (1783)			
Completeness*	98.8 (93.9)			
Redundancy*	11.4 (9.1)			
Rmerge*	0.074 (0.400)			
CC (1/2)	0.999 (0.945)			
I/σI*	24.3 (5.7)			
Refinement				
R_{work}/R_{free}*	0.161/0.199 (0.171/0.219)			
R.m.s deviations bond (Å)	0.006			
R.m.s deviations angles (°)	0.832			
B-factors(P/L/O) ($Å^2$) **	14.4/17.4/26.0			
Ramanchandran Favored	100			
Ramanchandran Allowed	0			
Ramanchandran Disallowed	0			

* Statistics for the highest resolution shell is shown in parentheses.
* * P/L/O indicate protein, ligand in the active site and solvent molecules, respectively.

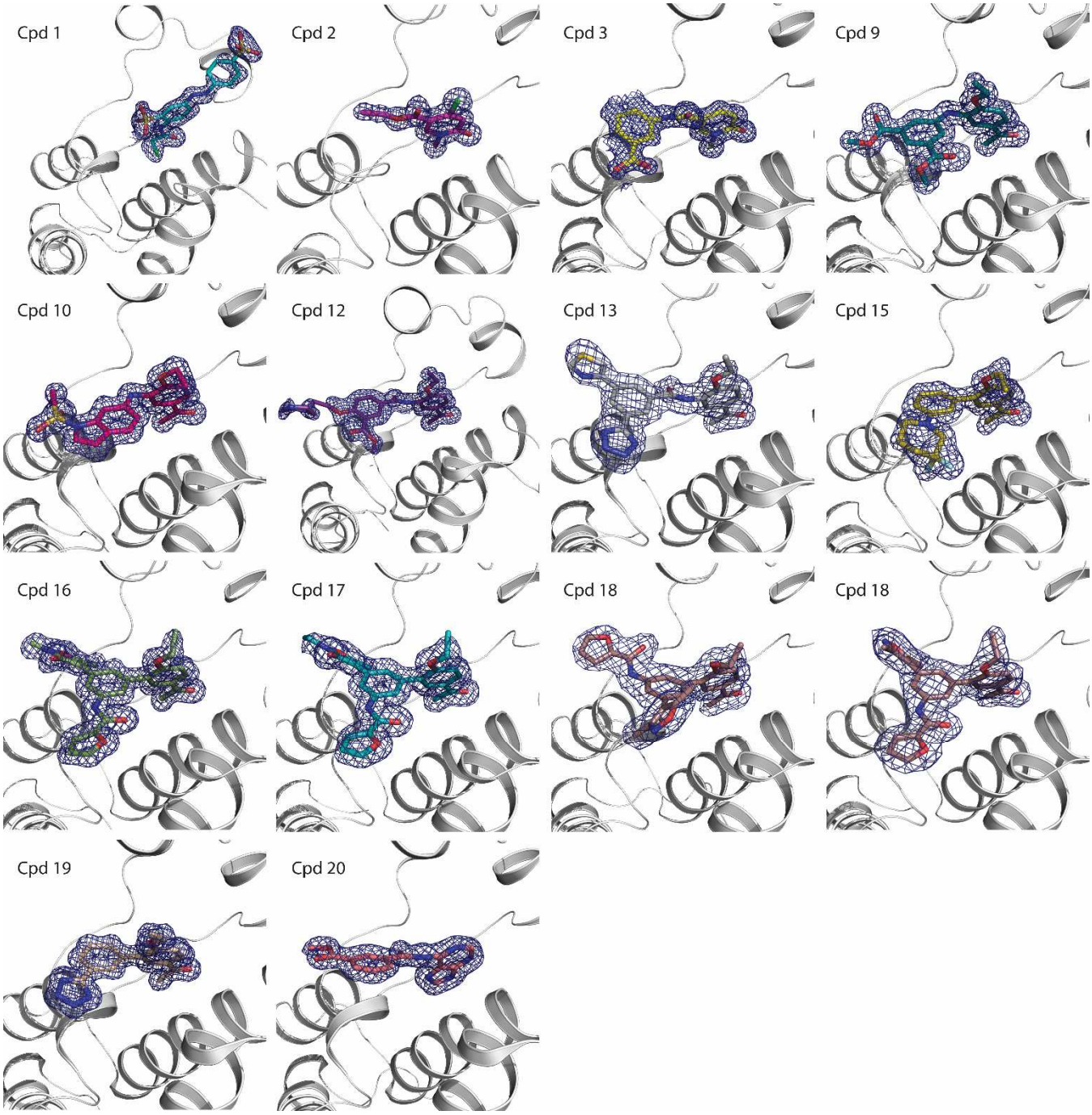

Figure S1. 2Fo-Fc electron density maps of compounds **1**, **2**, **3**, **9**, **10**, **12**, **13**, **15**, **16**, **17**, **18**, **19** and **20** bound to CBP bromodomain at a contour level of 1.0 sigma.

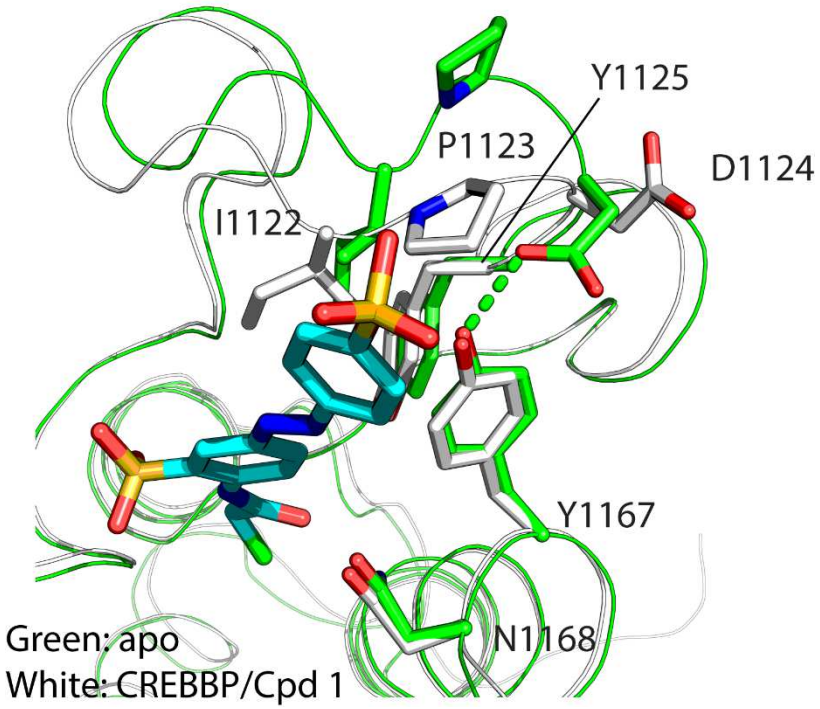

Figure S2. Alignment of complex structure of CBP/**1** with apo CBP bromodomain structure 3DWY. Compound **1** is shown in cyan stick. Hydrogen bond interaction between Asp1124 and Tyr1167 in the apo crystal structure are shown in green dash.

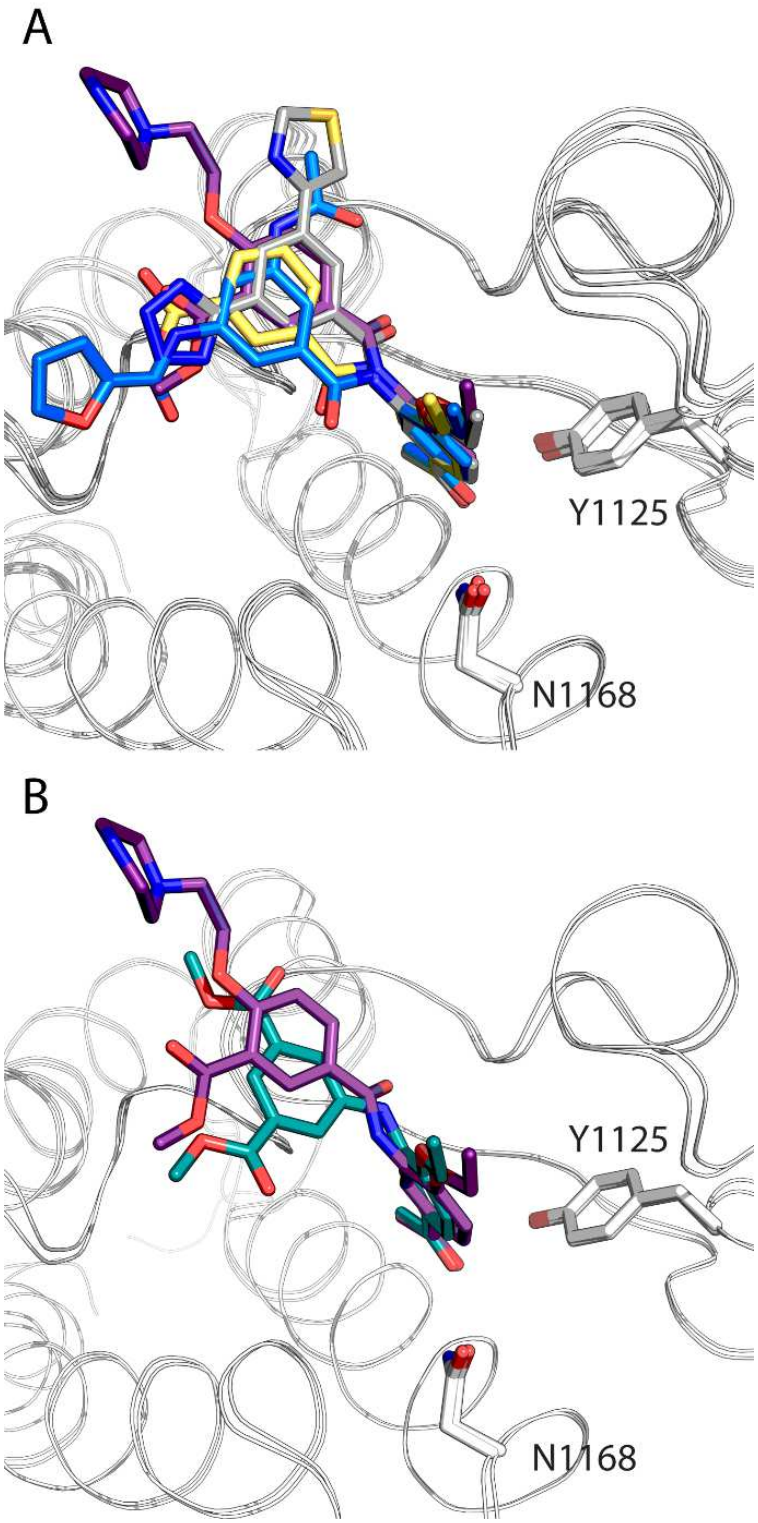

Figure S3. (A) Alignment of complex structures of CBP bromodomain with compounds **11** (yelloworange), **12** (vilotpurple), **13** (grey) and **14** (marine). (B) Structural comparison of the binding mode of compounds **9** (deepteal) and **12** (vilotpurple).

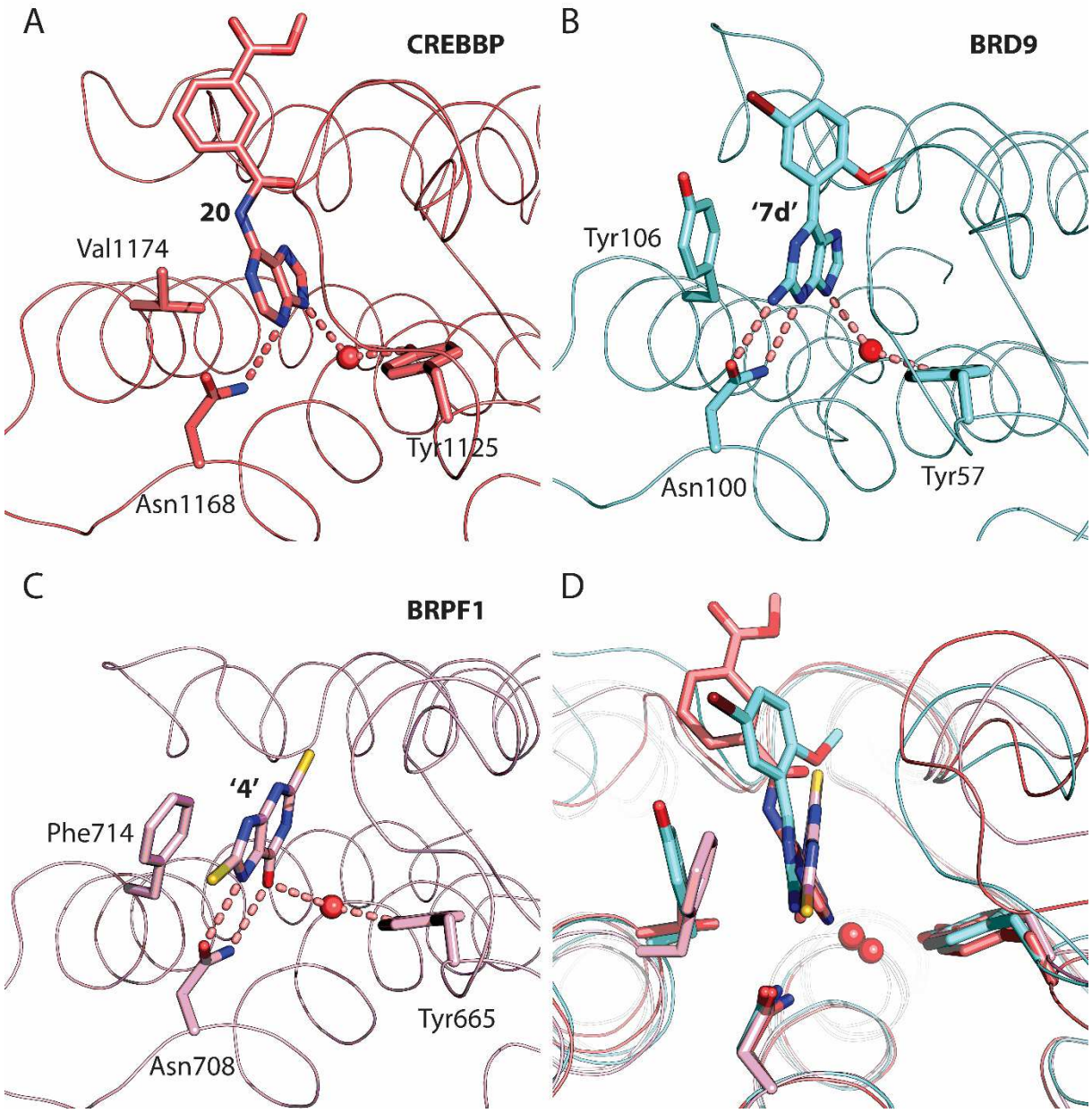

Figure S4. Binding mode comparison of purine-containing compounds of (A) **20** in CBP, (B) '**7d**' in BRD9[9] and (C) '**4**' in BRPF1[10] bromodomains. (D) Alignment of three complex structures. Ligands are shown in stick and CBP, BRD9 and BRPF1 bromodomain are shown in deepsalmon, cyan and pink, respectively.

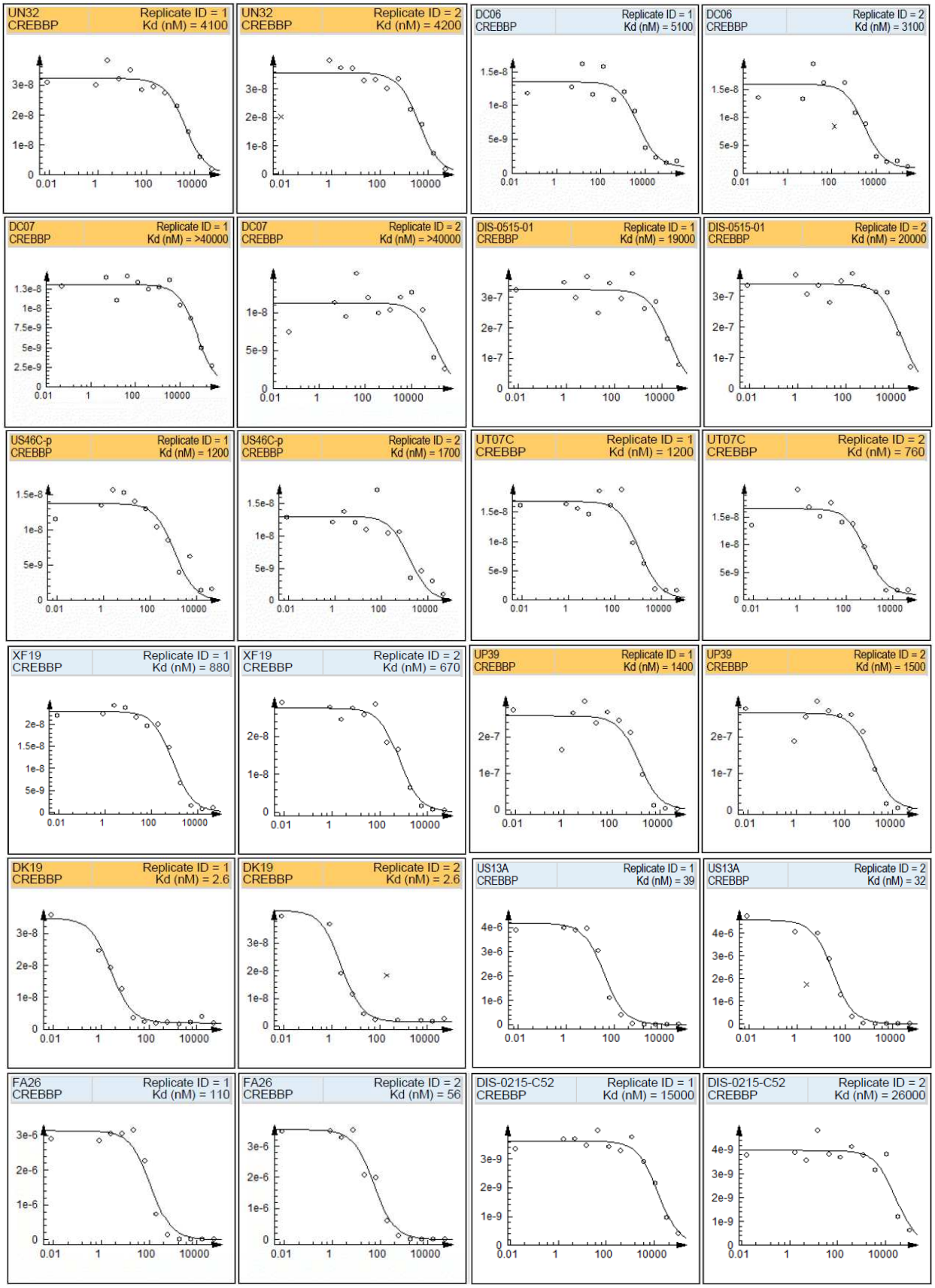

Figure S5. Dose-response curves in duplicates for compounds **3** (UN32), **4** (DC06), **5** (DC07), **7** (DIS-0515-1), **9** (US46C-p), **10** (UT07C), **11** (XF19), **12** (UP39), **13** (DK19), **14** (US13A), **19** (FA26) and **20** (DIS-0215-C52) in BROMOscan assays.

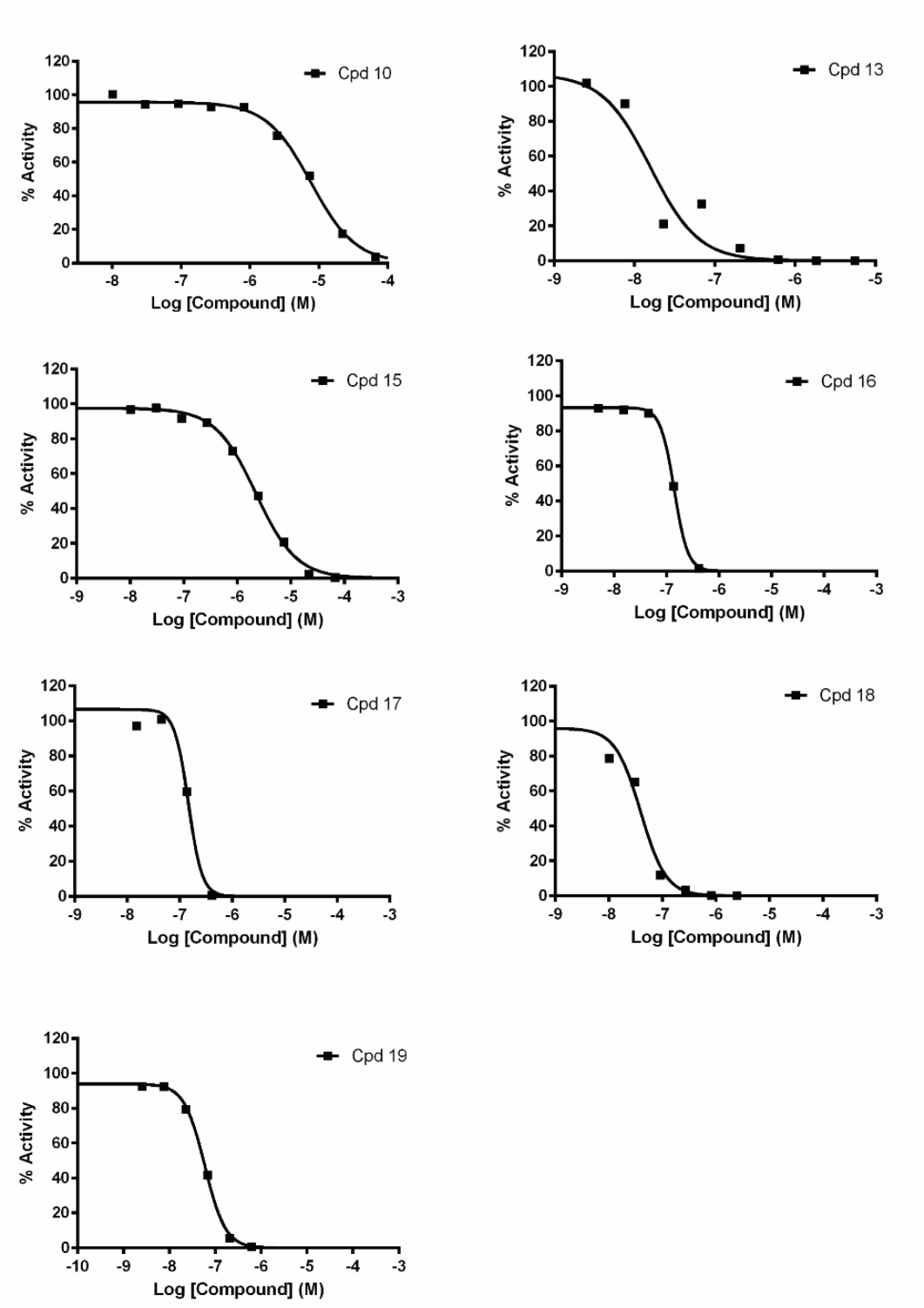

Figure S6. Dose-response curves of compounds **10**, **13**, **15**, **16**, **17**, **18** and **19** in AlphaScreen binding assays.

References:

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