Electronic Supplementary Information

Heat Shock Protein 90 and Serine/Threonine Kinase B-Raf inhibitors have overlapping chemical space

A. Anighoro,[a] L. Pinzi,[b] G. Marverti,[c] J. Bajorath*[a] and G. Rastelli*[b]

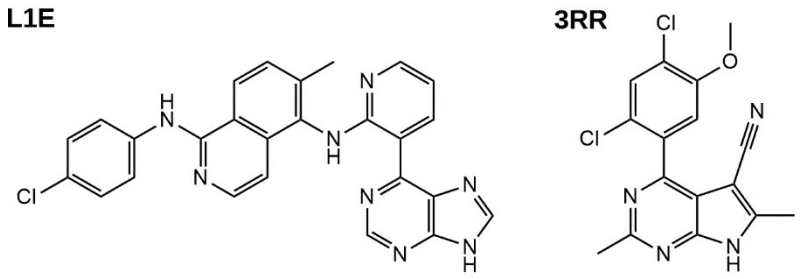
a. Department of Life Science Informatics, B-IT, LIMES Program Unit Chemical Biology and Medicinal Chemistry, Rheinische Friedrich-Wilhelms-Universität, Dahlmannstr. 2, D-53113 Bonn, Germany. E-mail: bajorath@bit.uni-bonn.de

b. Department of Life Sciences, University of Modena and Reggio Emilia, Via Campi 103, 41125, Modena, Italy. E-mail: giulio.rastelli@unimore.it

c. Department of Biomedical, Metabolic and Neurosciences, University of Modena and Reggio Emilia, Via Campi 287, 41125, Modena, Italy.

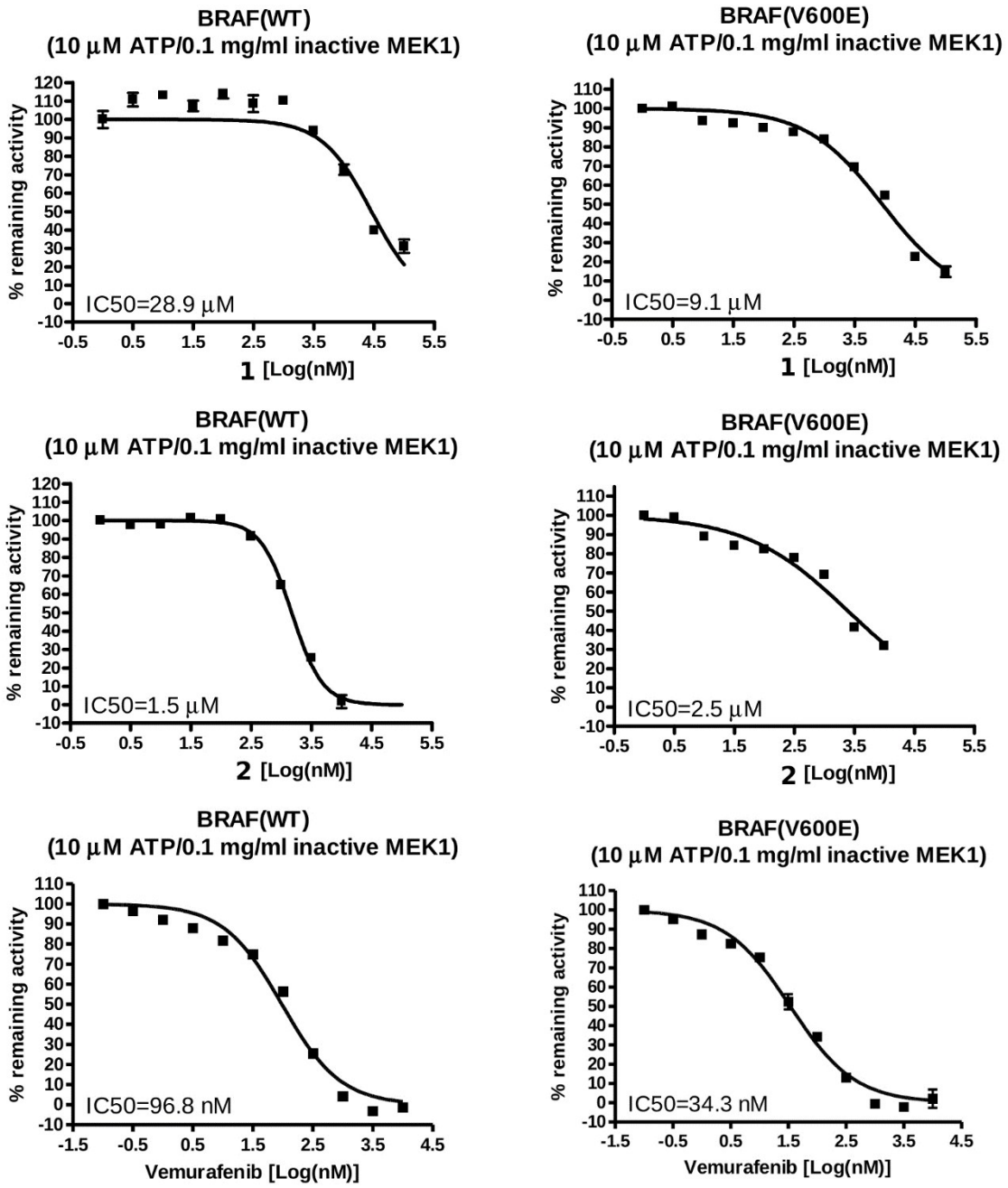
Contents

Figure S1. Structure of compounds L1E and 3RR.



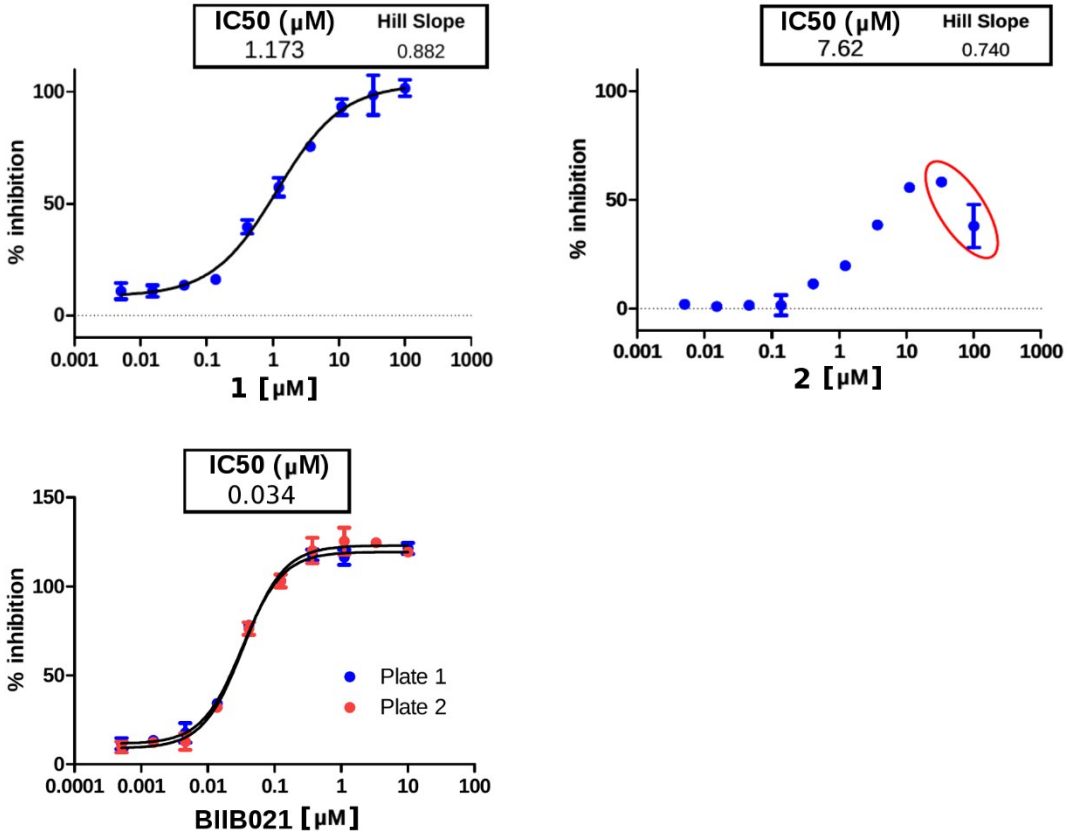
Chemical structures of the two cocrystallized inhibitors L1E (B-Raf) and 3RR (Hsp90) that were identified as a potential starting point for the computational design of dual inhibitors.

Figure S2. Dose-response curves of compounds 1 and 2 on B-Raf and B-Raf^{V600E}



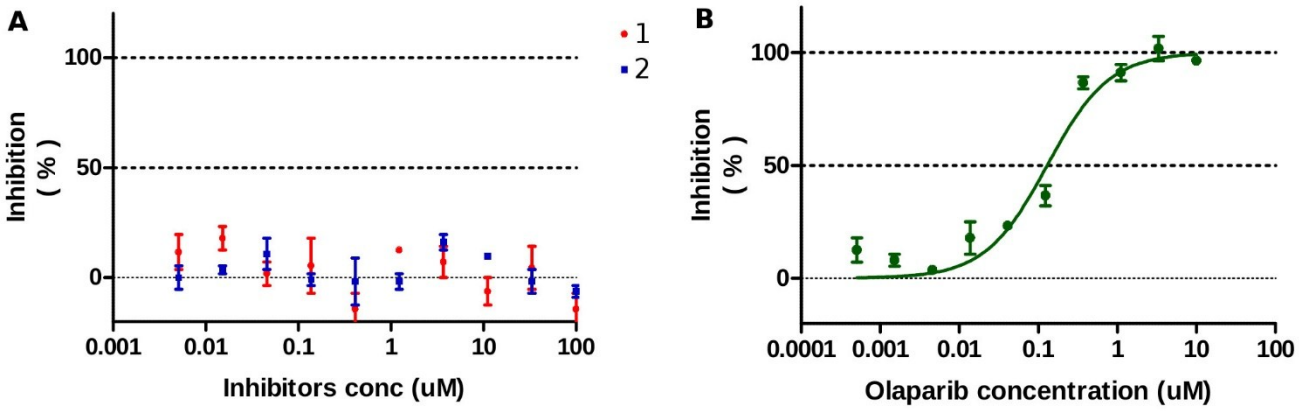
Dose-response curves obtained from the enzymatic assays performed to determine the IC_{50} of compounds **1** (top panels) and **2** (middle panels) on wild type (WT) and mutant (V600E) B-Raf. Dose-response curves of the known inhibitor Vemurafenib against wt B-Raf and V600E B-Raf are shown in the lower panels.

Figure S3. Dose-response curves of compounds 1 and 2 on Hsp90



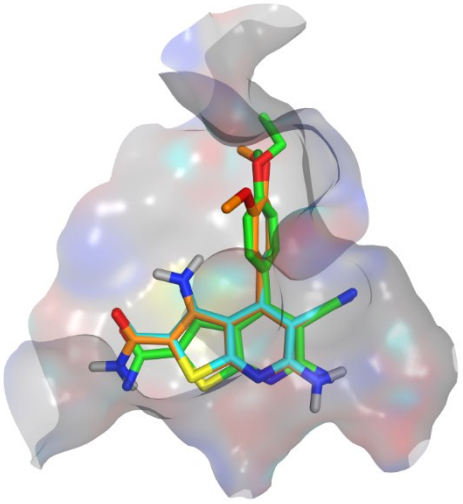
Dose-response curves obtained from the enzymatic assays performed to determine the IC_{50} of compounds **1** (top-left panel) and **2** (top-right panel) on Hsp90. The two highest concentrations (33 and 100 µM, marked in red) of compound **2** were excluded in the calculation of the IC_{50}. This behavior can be due to lower solubility at high concentrations. The dose-response curve (two plates) obtained for the reference compound BIIB021 is shown in the lower panel.

Figure S4. Dose-response curves on PARP-1



Panel A shows the displacement data obtained from the enzymatic assays performed to determine the activity of compounds **1** (red circles, panel A) and **2** (blue squares, panel A) on PARP-1. Panel B shows the dose-response curve obtained for the reference PARP-1 inhibitor Olaparib (IC_{50} of 0.126 µM)

Figure S5. Superposition of compounds 1 and 2 docked in Hsp90 with 2WI5



An overlay of compounds **1** (cyan) and **2** (orange) docked in Hsp90 with an experimental co-crystallized inhibitor of Hsp90 (green) (PDB code: 2WI5) is shown.

Table S1. Protein structures used for pharmacophore analysis

Protein	Ligand (ID)	PDB code	Resolution
B-Raf	215	2FB8	2.90
B-Raf	SM5	3D4Q	2.80
B-Raf	**L1E**	**3IDP**	**2.70**
B-Raf	831	3II5	2.79
B-Raf	032	3OG7	2.45
B-Raf	FNI	3PPK	3.00
B-Raf	FP3	3PRF	2.90
B-Raf	734	4E26	2.55
Hsp90	A56	2QF6	3.10
Hsp90	A94	2QG0	1.85
Hsp90	A91	2QG2	1.80
Hsp90	2GJ	2VCI	2.00
Hsp90	WOE	2XDX	2.42
Hsp90	PYU	3EKO	1.55
Hsp90	PY9	3EKR	2.00
Hsp90	BD0	3HEK	1.95
Hsp90	4CD	3K97	1.95
Hsp90	1RC	3K98	2.40
Hsp90	PFT	3K99	2.10
Hsp90	94M	3QDD	1.79
Hsp90	WOE	3R4M	1.70
Hsp90	FU5	3R4N	2.00
Hsp90	FU3	3R4O	2.65
Hsp90	FU7	3R4P	1.70
Hsp90	3RQ	3RLQ	1.90
Hsp90	**3RR**	**3RLR**	**1.70**
Hsp90	2N6	4FCQ	2.15

List of X-ray crystal structures used in the analysis. The structures selected for docking calculations are highlighted in bold.