

Targeting lipid–protein interaction to treat Syk-mediated acute myeloid leukemia

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SUPPLEMENTARY INFORMATION

Title: Targeting lipid-protein interaction for resistance-defying kinase drug development: A novel Syk inhibitor for acute myeloid leukemia

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Supplementary Notes: Chemical synthesis and characterization of Syk inhibitors

1. Chemical synthesis of Syk inhibitors
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Supplementary Data Files:

1. Source data files for Supplementary Fig. 5a
2. Source data files for Supplementary Fig. 6

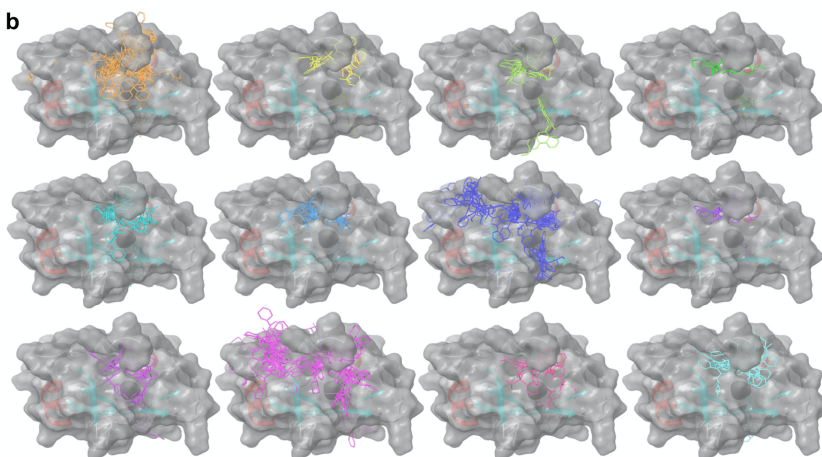
Supplementary Fig. 1. Ensemble docking of various structural scaffolds to Syk-cSH2

a. Structures of 12 scaffolds that bind to the PIP₃-binding sites of SH2 domains are shown in shaded boxes. A representative derivative of each scaffold is shown in the open box below. **b.** Docking of various derivatives of each of 12 scaffolds to Syk-cSH2 in the order shown in **a**. Notice that most of these compounds preferentially bind to the primary PIP₃-binding site (see **Fig. 1b** and **Fig. S1d**) but some bind to other surface grooves preferentially. **c.** Ensemble docking of all 12 scaffolds shown in **Fig. S1b** to Syk-cSH2. **d.** Docking of three best inhibitors, WC35 (red), WC36 (green), and WC38 (blue), to Syk-cSH2. They all preferentially bind to the primary PIP₃-binding site. Syk-cSH2 is shown in the same orientation as in **Fig. 1b** (right panel) in all figures. Top 5 binding poses are shown for each compound.

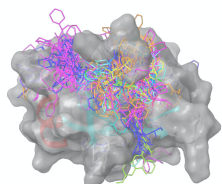
a

Scaffolds						
Examples						
Scaffolds						
Examples						

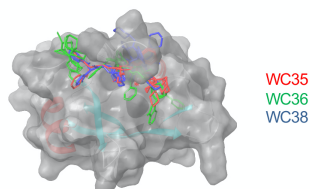
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c



d



Supplementary Fig. 2. Determination of Z' factor for the fluorescence quenching-based assay.

Z'-factor for the high-throughput assay was calculated using 1 % dimethylsulfoxide (DMSO) and 20 μM D-myo-inositol-1,3,4,5-tetraphosphate (IP_4) as negative and positive controls, respectively in the 96-well plates. POPC/POPS/PIP₃/dabsyl-PE (67:20:3:10) vesicles (40 μM) were used for the assay.

Z'-factor was calculated using the equation:

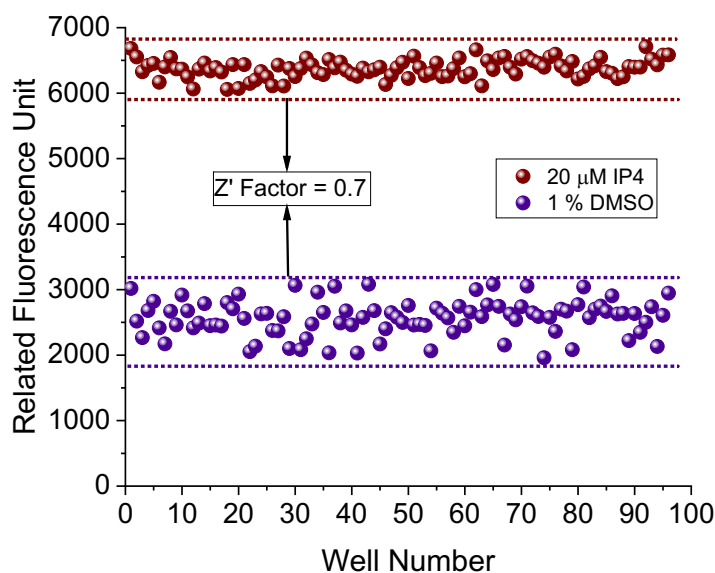
$$Z' = 1 - \frac{3(\sigma_p + \sigma_n)}{\mu_p - \mu_n}$$

where σ_p = standard deviation of the positive control (146)

σ_n = standard deviation of the negative controls (269)

μ_p = mean of the positive control (6370)

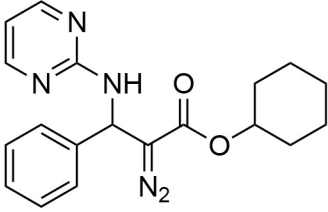
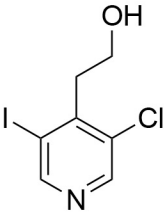
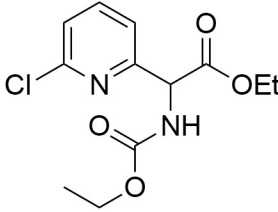
μ_n = mean of the negative control (2566)



Supplementary Fig. 3. Structures and properties of PIP₃-SH2 domain interaction inhibitors.

a. Structures of inhibitors for Syk-cSH2, BLNK-SH2, and PLC γ 2-cSH2. **b.** Efficacy and selectivity of SH2 domain inhibitors determined by the vesicle binding assay using POPC/POPS/PIP₃/dabsyl-PE (67:20:3:10) vesicles (40 μ M). Each data is average \pm SD from 5 independent measurements.

a

			
Name	VG354	VG594	VG370
Target	Syk-cSH2	BLNK-SH2	PLC γ 2-cSH2

b

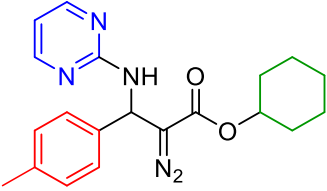
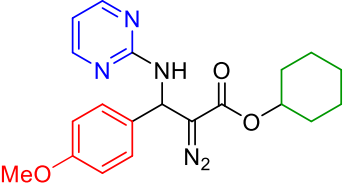
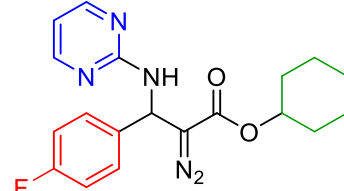
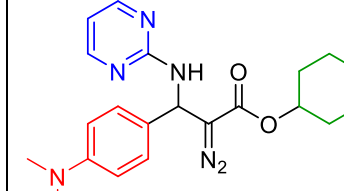
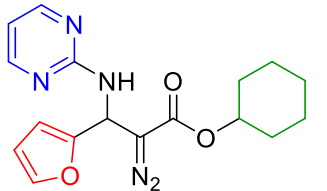
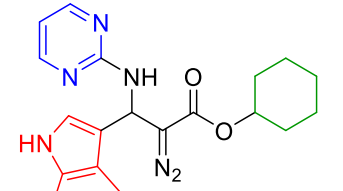
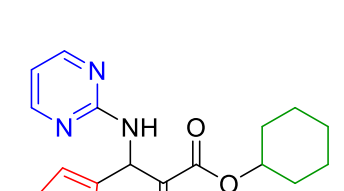
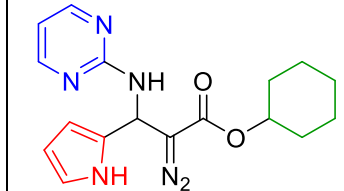
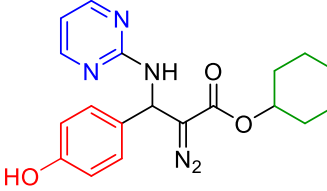
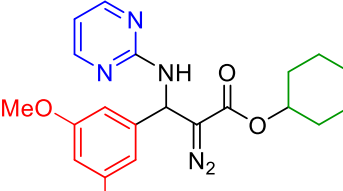
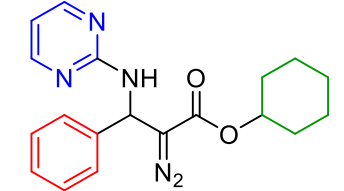
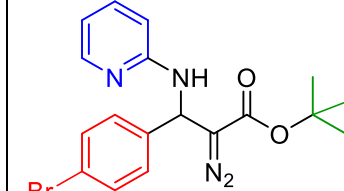
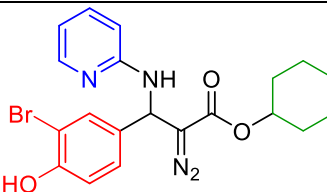
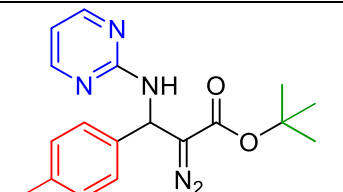
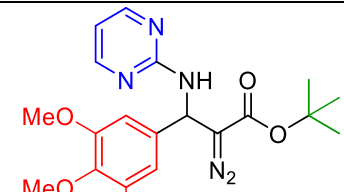
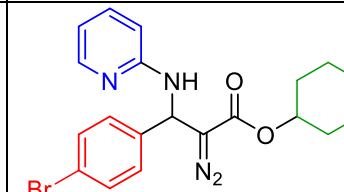
	VG354		VG594		VG370	
	<i>I</i> _{max} (%)	IC ₅₀ (nM)	<i>I</i> _{max} (%)	IC ₅₀ (nM)	<i>I</i> _{max} (%)	IC ₅₀ (nM)
Syk-cSH2	40 \pm 5	350 \pm 20	<10	ND ^a	<10	ND
BLNK-SH2	<10	ND	45 \pm 5	400 \pm 30	<20	ND
PLC γ 2-cSH2	<10	ND	<10	ND	50 \pm 5	680 \pm 50

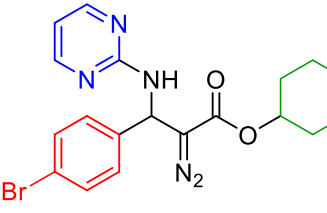
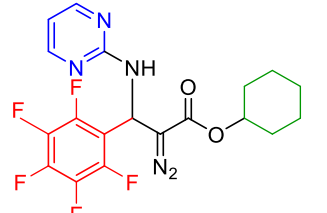
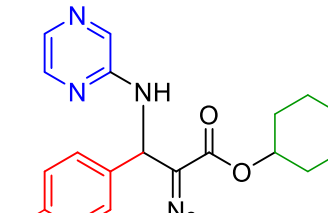
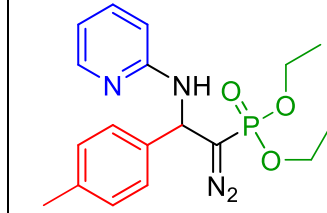
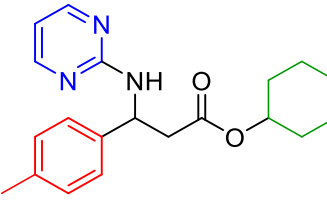
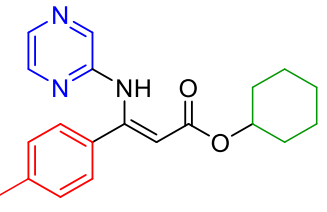
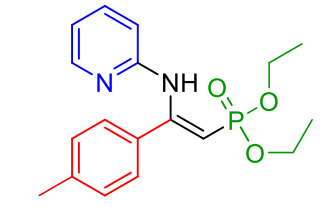
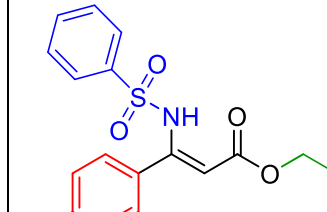
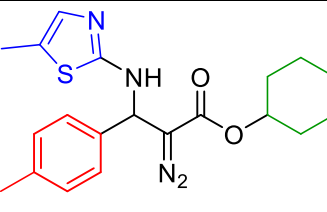
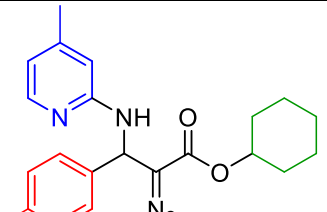
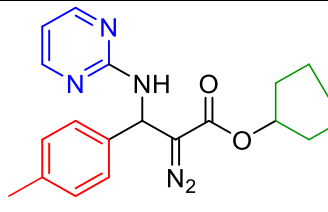
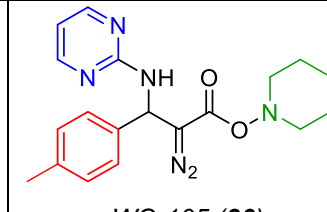
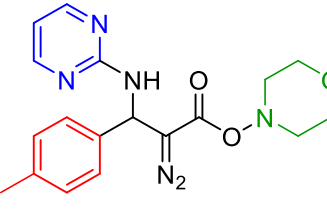
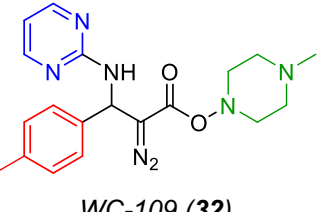
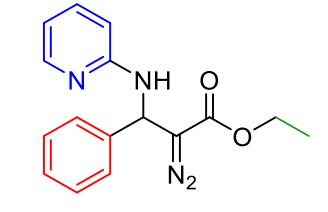
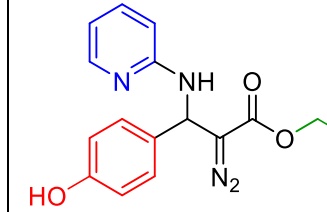
^aNot determined

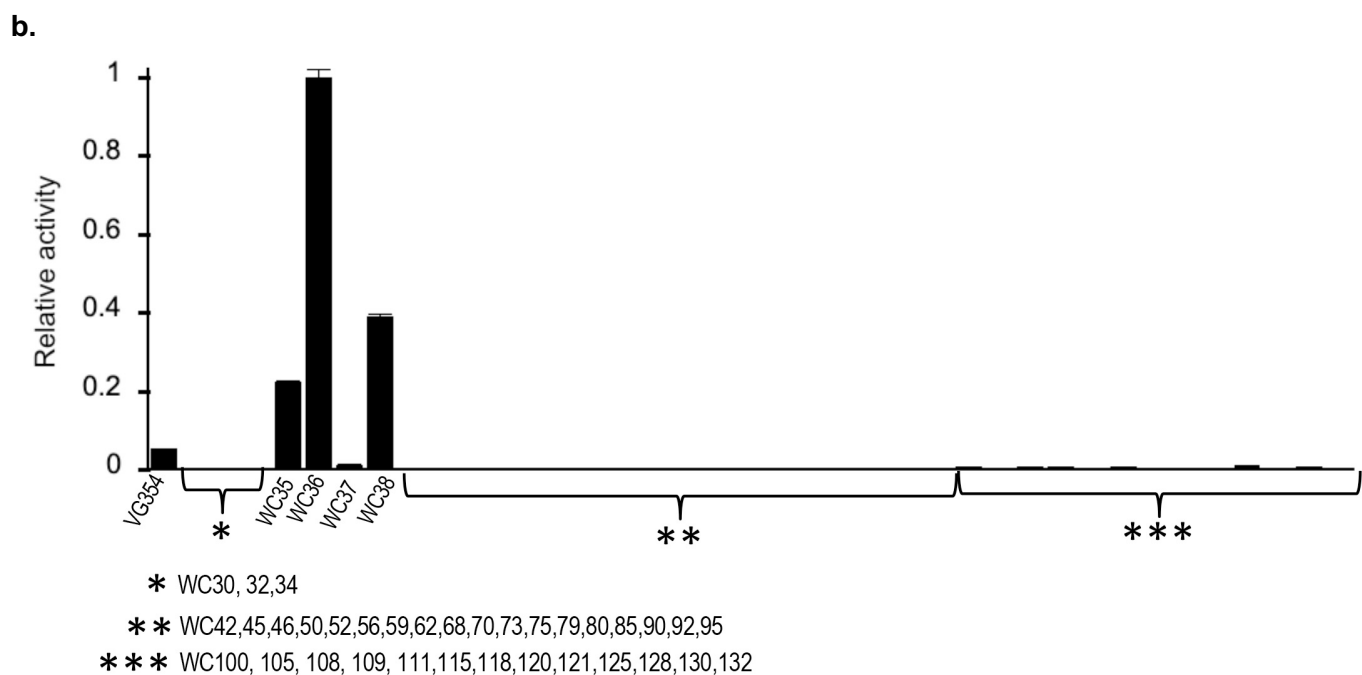
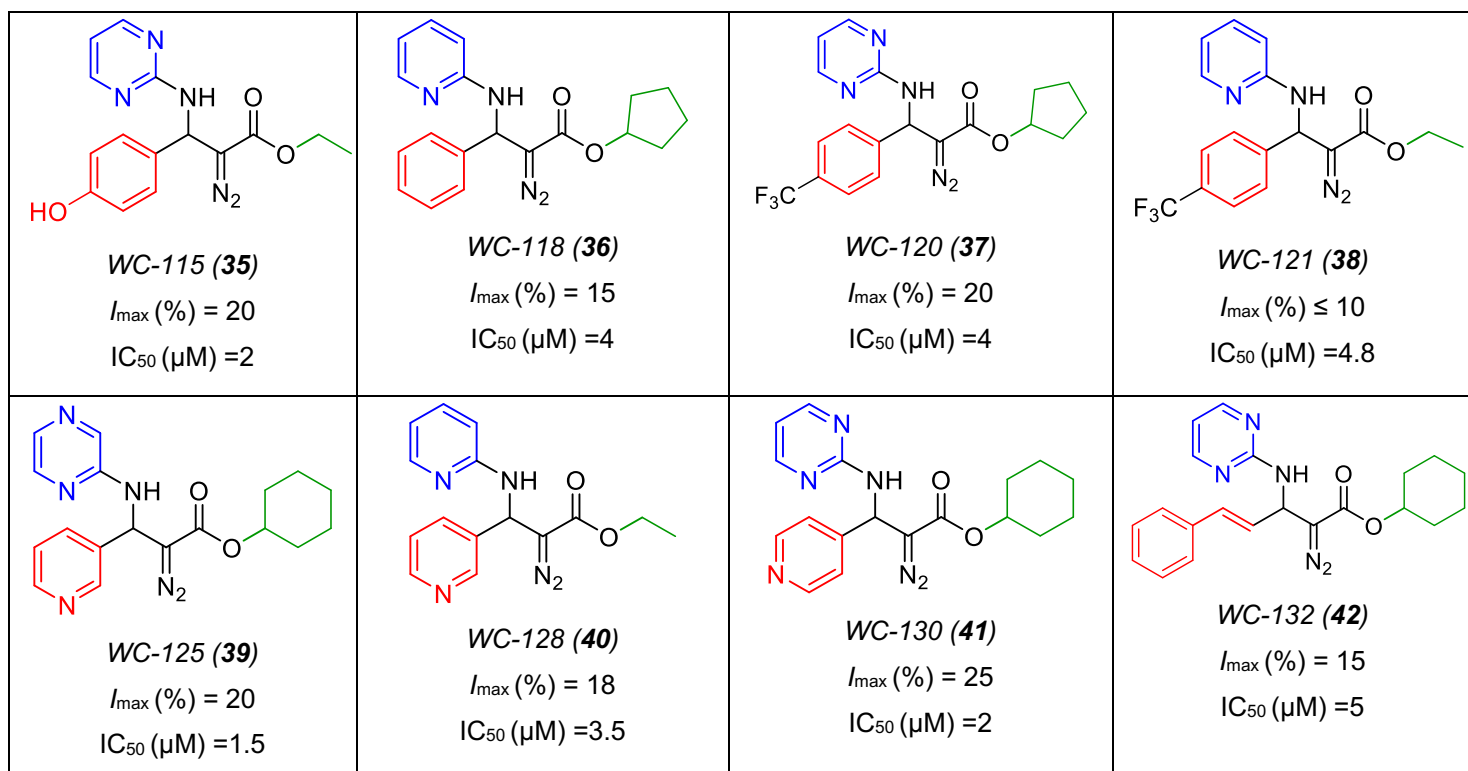
Supplementary Fig. 4. Optimization of Syk-cSH2 inhibitors by the structure-activity relationship analysis.

a. Structures derivatives of VG354 and their efficacy determined by the vesicle binding assay. **b.** Relative activity of VG354 derivatives. I_{max}/IC_{50} values were normalized against that of WC36. Error bars indicate SD's from 3 independent measurements.

a.

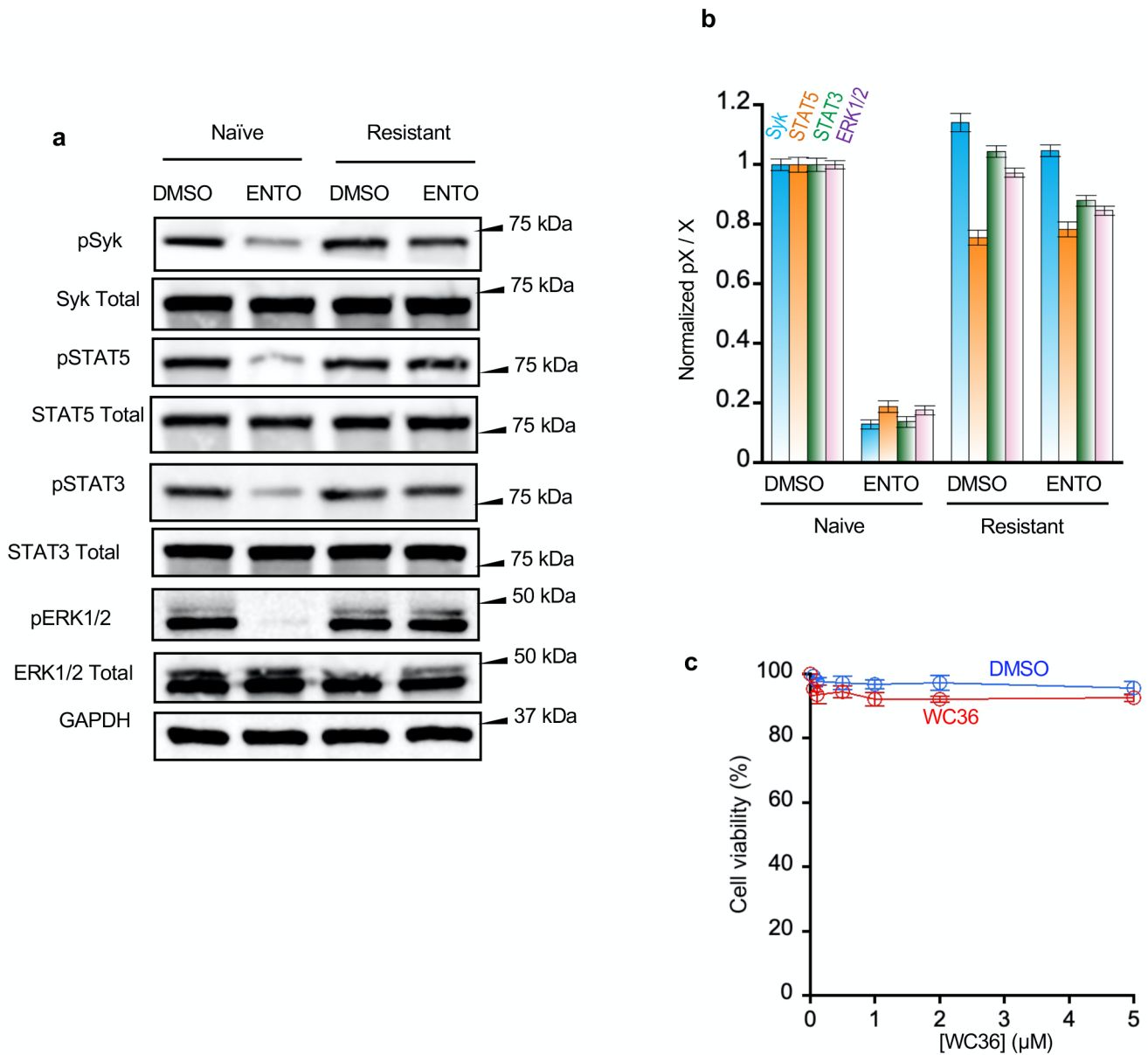
 <p>VG-354 (1) I_{max} (%) = 40 IC_{50} (μM) = 0.5</p>	 <p>WC-30 (7) I_{max} (%) = 10 IC_{50} (μM) = 3.8</p>	 <p>WC-32 (8) I_{max} (%) = 10 IC_{50} (μM) = 5</p>	 <p>WC-34 (9) I_{max} (%) = 10 IC_{50} (μM) = 2.8</p>
 <p>WC-35 (4) I_{max} (%) = 50 IC_{50} (μM) = 0.15</p>	 <p>WC-36 (5) I_{max} (%) = 60 IC_{50} (μM) = 0.04</p>	 <p>WC-37 (10) I_{max} (%) = 15 IC_{50} (μM) = 2.8</p>	 <p>WC-38 (6) I_{max} (%) = 70 IC_{50} (μM) = 0.12</p>
 <p>WC-42 (11) I_{max} (%) = 12 IC_{50} (μM) = 4</p>	 <p>WC-45 (12) I_{max} (%) = 12 IC_{50} (μM) = 3.6</p>	 <p>WC-46 (13) I_{max} (%) = 10 IC_{50} (μM) = 5</p>	 <p>WC-50 (14) I_{max} (%) = 10 IC_{50} (μM) = 4.5</p>
 <p>WC-52 (15) I_{max} (%) = 12 IC_{50} (μM) = 3.5</p>	 <p>WC-56 (16) I_{max} (%) \leq 10 IC_{50} (μM) = 2</p>	 <p>WC-59 (17) I_{max} (%) \leq 10 IC_{50} (μM) = 4</p>	 <p>WC-62 (18) I_{max} (%) \leq 10 IC_{50} (μM) = 5</p>

 <p>WC-68 (19) $I_{max} (\%) \leq 10$ $IC_{50} (\mu M) = 4.5$</p>	 <p>WC-70 (20) $I_{max} (\%) \leq 10$ $IC_{50} (\mu M) = 2.5$</p>	 <p>WC-73 (21) $I_{max} (\%) \leq 10$ $IC_{50} (\mu M) = 8$</p>	 <p>WC-75 (22) $I_{max} (\%) = 12$ $IC_{50} (\mu M) = 7$</p>
 <p>WC-79 (23) $I_{max} (\%) \leq 10$ $IC_{50} (\mu M) = 10$</p>	 <p>WC-80 (24) $I_{max} (\%) = >10$ $IC_{50} (\mu M) = 15$</p>	 <p>WC-85 (25) $I_{max} (\%) \leq 10$ $IC_{50} (\mu M) = 12$</p>	 <p>WC-90 (26) $I_{max} (\%) \leq 10$ $IC_{50} (\mu M) = 20$</p>
 <p>WC-92 (27) $I_{max} (\%) = 15$ $IC_{50} (\mu M) = 10$</p>	 <p>WC-95 (28) $I_{max} (\%) \leq 10$ $IC_{50} (\mu M) = 5$</p>	 <p>WC-100 (29) $I_{max} (\%) = 18$ $IC_{50} (\mu M) = 2$</p>	 <p>WC-105 (30) $I_{max} (\%) = 15$ $IC_{50} (\mu M) = 4$</p>
 <p>WC-108 (31) $I_{max} (\%) = 20$ $IC_{50} (\mu M) = 2$</p>	 <p>WC-109 (32) $I_{max} (\%) = 25$ $IC_{50} (\mu M) = 2$</p>	 <p>WC-110 (33) $I_{max} (\%) \leq 10$ $IC_{50} (\mu M) = 5$</p>	 <p>WC-111 (34) $I_{max} (\%) \leq 10$ $IC_{50} (\mu M) = 7$</p>



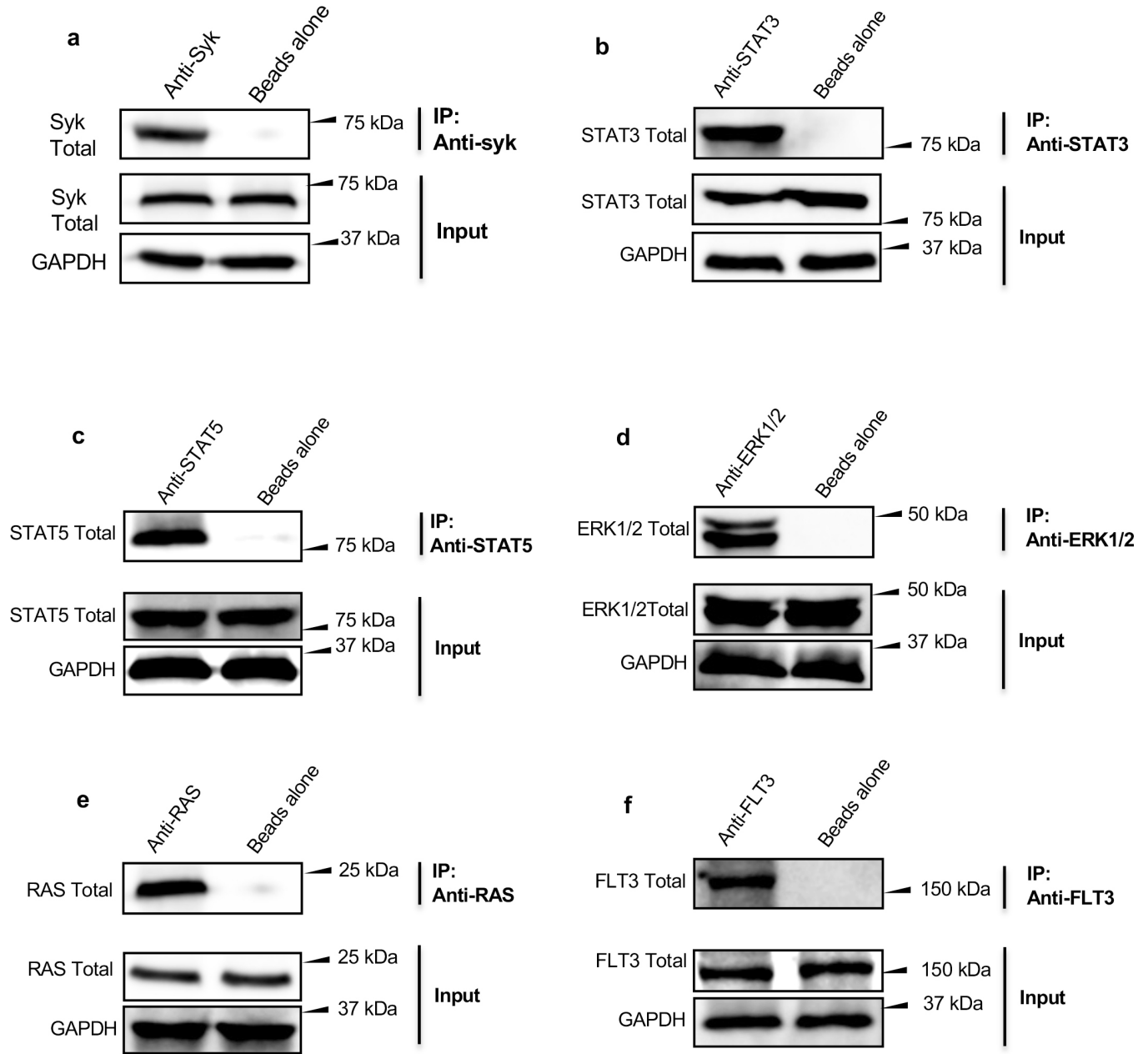
Supplementary Fig. 5. Inhibitory activity of entospletinib in naïve and resistant MV4-11 cells.

a. Inhibition of phosphorylation of Syk, STAT3/5 and ERK1/2 by entospletinib (ENTO: 5 μ M) in naïve and entospletinib-resistant MV4-11 cells. Entospletinib-resistant MV4-11 cells were obtained after chronic exposure of cells to increasing concentrations (0.5 to 5 μ M) of entospletinib over 10 weeks. Cells were stimulated with IgG2 (10 ng/ml for 10 min). DMSO was used as a negative control. The gel image is representative of 3 independent measurements. **b.** The immunoblotting data were quantified as described in **Fig. 3a** and normalized against DMSO-treated naïve cells. (pX/totalX) values were greatly increased for most proteins. Error bars indicate SD. **c.** Effects of WC36 (0.5-5 μ M) on viability of Raji B cells after 72 h incubation measured with the CellTiter-Glo luminescence reagents. Cell viability data was normalized against the cell viability without treatment. DMSO was used as a negative control. Each data is mean \pm SD from 3 independent measurements. For pSyk/Syk, $p = <0.0001$ (naïve), 0.089 (resistant). For pSTAT5/STAT5, $p = 0.00035$ (naïve), 0.25 (resistant). For pSTAT3/STAT3, $p = 0.0032$ (naïve), 0.05 (resistant). For pERK1/2/ERK1/2, $p = 0.00018$ (naïve), 0.65 (resistant).



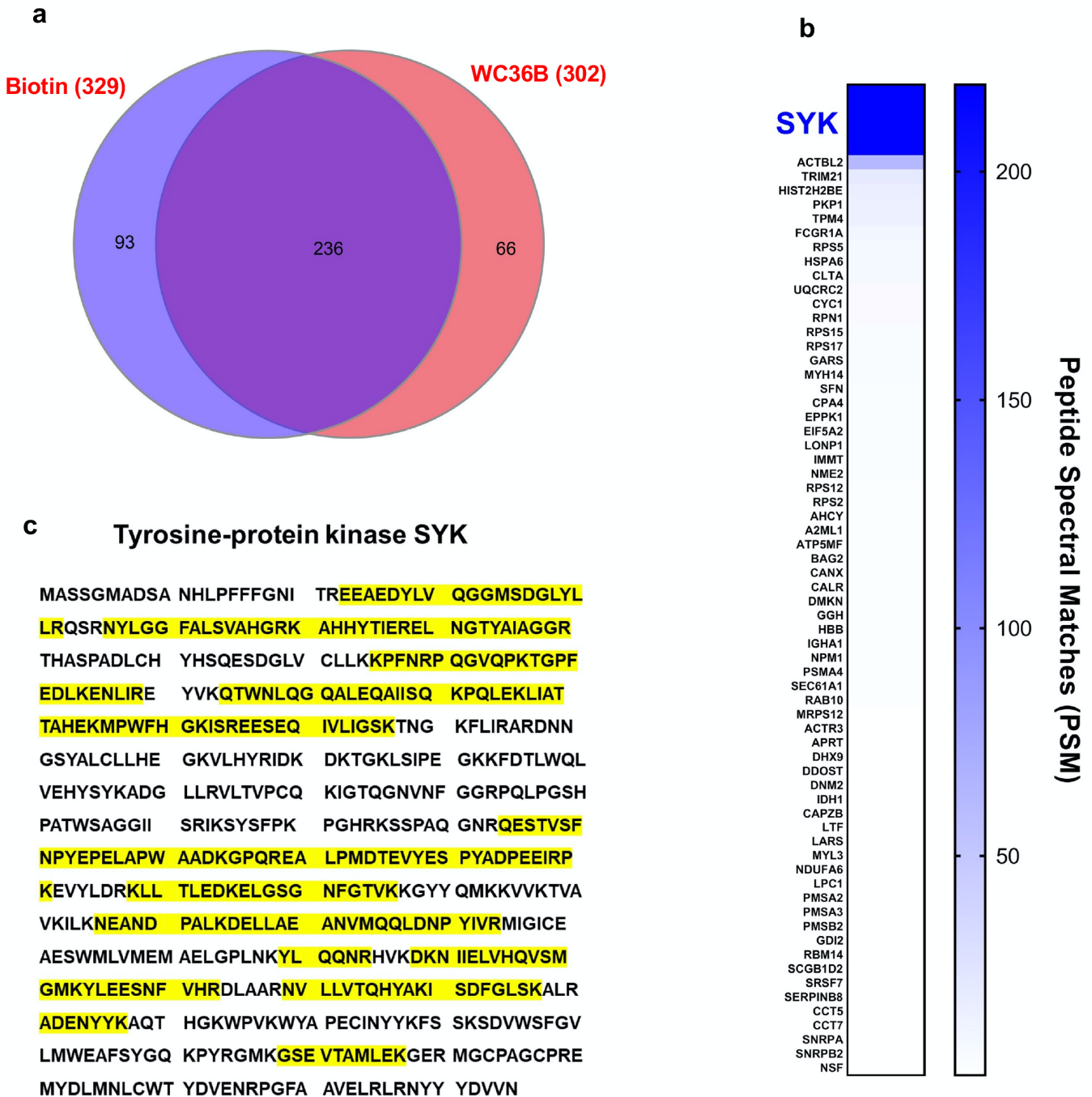
Supplementary Fig. 6. Control immunoprecipitation with beads.

Negative control IP experiments with beads alone show that IP signals for Syk (a), STAT3 (b), STAT5 (c), ERK1/2 (d), RAS (e), and FLT3 (f) in Fig. 5c are not due to non-specific background. All gel images are representative of 3 independent measurements.



Supplementary Fig. 7. Mass spectrometry-based identification of captured proteins.

Proteins captured by WC36B and the biotin control were subject to data-dependent mass spectrometry analysis and identification by searching of the raw data against the Swiss Protein *Homo sapien* database (Table S5). **a.** Mass spectrometry analysis revealed 329 and 302 identified proteins in biotin and WC36B affinity captures, respectively. Venn diagram analysis of these identified proteins show that 66 proteins were unique to the WC36B probe. **b.** Further analysis of the number of mass spectra used to identify these 66 proteins (peptide spectra matches (PSMs)) reveals that the PSMs for Syk is much greater than any of the other 65 proteins, indicating that the Syk protein is much higher in abundance relative to other proteins in the capture. **c.** Sequence coverage map for the 28 unique SYK peptides (highlighted in yellow), which were identified only in the WC36B capture.



Supplementary Table 1. Affinity of SH2 domain wild type and mutants for lipid vesicles and a pY peptide^a

SH2 domains	K_d^b (nM) for POPC/POPS/PIP ₃ vesicles	K_d^c (nM) for the pY peptide
Syk-cSH2 WT	150 ± 30	2.4 ± 0.5
Syk-cSH2 K220A/K220A	1200 ± 400	1.8 ± 0.2
Syk-cSH2 K172A	410 ± 50	ND ^d
Syk-cSH2 R195A	180 ± 23	230 ± 170
BLNK-SH2 WT	280 ± 30	ND
BLNK-SH2 K411A/K412K/K413A	710 ± 250	ND
PLCγ2-cSH2 WT	240 ± 60	ND
PLCγ2-cSH2 K727A/K728A	1250 ± 500	ND

^aDerived from doubly phosphorylated Igα

^bFor SPR data, see **Extended Data Fig. 1h, 3d, and 4d.**

^cFor fluorescence anisotropy data, see **Fig. 1f.**

^dNot determined

Supplementary Table 2. Small molecule screening data

Category	Parameter	Description
Assay	Type of assay	Fluorescence quenching-based high-throughput screening
	Target	Syk-cSH ₂ -PIP ₃ binding
	Primary measurement	Measuring the quenching of EGFP fluorescence of Syk-cSH ₂ protein by dabsyl-phosphatidylethanolamine (PE)
	Key reagents	Syk-cSH ₂ , Lipids (POPC, POPS, PIP ₃ and dabsyl-PE), 20 mM Tris buffer (pH 7.4), library compounds dissolved in DMSO
	Assay protocol	Assay is based on fluorescence quenching of EGFP fused to a SH ₂ domain by a dark quencher containing lipid, dabsyl-PE, incorporated in lipid vesicles. The assay was performed using the Synergy™ Neo spectrofluorometer at 25 °C. Nontreated black polystyrene 96-well plates (Corning) were used. To each row of wells containing 25 nM SH ₂ domain protein, increasing concentrations of lipid vesicles in 20 mM Tris buffer, pH 7.4, containing 0.16 M NaCl was added and the EGFP fluorescence emission at 516 nm measured with excitation set at 485 nm.
	Additional comments	N/A
Library	Library size	~650 Compounds
	Library composition	Planer, Fused/Spiro Bicyclic and Heterocyclic
	Source	Wonhwa Cho's and Vladimir Gevorgyan's Laboratories
	Additional comments	N/A
Screen	Format	96-Well Plate
	Concentration(s) tested	1-10 μM
	Plate controls	<ul style="list-style-type: none"> Fix the concentration producing the midpoint signal over the course of the test The rate of outliers should be less than 2 percent (i.e., on average less than 2 wells on a 96-well plate) Design of chemical screening plate is based on 80 different compounds stored in the middle of 96-well plates and the first and last columns are left empty. Correspondingly, column 1 and column 12 are available for controls and to minimize edge-related bias,
	Reagent/ compound dispensing system	Manual
	Detection instrument and software	Synergy™ Neo spectrofluorometer and Origin
	Assay validation/QC	Z' Factor
	Correction factors	N/A
	Normalization	$X_{\text{normalized}} = (X - X_{\text{min}}) / (X_{\text{max}} - X_{\text{min}})$ X = observed X values. X _{min} = minimum X values X _{max} = maximum X values
	Additional comments	N/A
	Post-HTS analysis	Hit criteria
Hit rate		~5 %
Additional assay(s)		N/A
Confirmation of hit purity and structure		Confirmed by NMR, HRMS and elemental analysis and
Additional comments		N/A

Supplementary Table 3. Inhibitory Activity of Optimized Syk inhibitors in different assays

Assays	Targets	WC35		WC36		WC38	
		I_{max} (%)	IC ₅₀ (nM)	I_{max} (%)	IC ₅₀ (nM)	I_{max} (%)	IC ₅₀ (nM)
Vesicle binding ^a	Syk-cSH2	50 ± 5	150 ± 25	60 ± 5	40 ± 5	70 ± 5	120 ± 10
	BLNK-SH2	<10	ND ^f	<10	ND	<10	ND
	PLCγ2-cSH2	<10	ND	<10	ND	<10	ND
	ZAP70-cSH2	<20	ND	<10	ND	30 ± 5	340 ± 20
Immuno-blotting	pSyk ^b	58 ± 5	180 ± 50	67 ± 5	52 ± 10	65 ± 3	180 ± 30
	pZAP70 ^c	<10	ND	<10	ND	<10	ND
Proliferation	Naïve MV4-11 ^d	85 ± 4	370 ± 84	83 ± 3	240 ± 40	83 ± 3	560 ± 97
	Ento-resistant MV4-11 ^e			71 ± 2	500 ± 10		

^aPOPC/POPS/PIP₃/dabsyl-PE (67:20:3:10) vesicles (40 μM) were used for the assay.

^bDetermined from the Western blot assay in Raji B cells. See **Extended Data Fig. 7b**

^cDetermined from the Western blot assay in Jurkat T cells. See **Extended Data Fig. 7c**

^dDetermined from the XTT assay in naïve MV4-11 cells. See **Fig 3b**

^eDetermined from the XTT assay in entospletinib-resistant MV4-11 cells. See **Fig 3d**

^fNot determined.

Supplementary Table. 4. The genetic background and clinical history of four AML patients

	Patient1	Patient2	Patient3	Patient4
Mutations	CEBPA, KIT, STAT5B, WT1	NPM1, TET2, DNMT3A, KRAS	FLT3 ITD, CEBPA A170Pfs, DNMT3A R320, NPM1 W88Cfs, TET2 Q962, H1881R	FLT3 ITD, NPM1 W88Cfs, IDH2 R140Q
Therapy history	Remission for over 1 year	Relapsed after first line of therapy	Died prior to initiating therapy	Achieved complete remission after intensive chemotherapy

Supplementary Table 5. Mass spectrometry-based proteomics analysis.

Proteins captured by WC36B and biotin were subject to identification by data-dependent mass spectrometry analysis. Raw data was searched against the Swiss Protein *Homo sapien* database using the Proteome Discoverer software employing the Sequest HT algorithm. Shown herein are proteins identified in the capture by WC36B and biotin. In this analysis, Syk as only detected in the WC36B fluorophore capture. Peptide sequences for the 28 unique peptides (47% sequence coverage) for Syk are also included. In our approach, the more intense a peptide, the more it is selected for fragmentation (peptide spectral matches: PSM). Although peptides may have different ionization efficiencies, PSM values can be in general related to the abundance of proteins in the capture. In the WC36B-captured proteins, Syk has 219 spectra whereas the next highest one is 79, 71, 61, 15, 11 and many at only 2 PSMs. Many of these proteins were also identified in the biotin capture whereas not a single peptide was identified for Syk in the biotin capture. Collectively, these results show Syk is the major protein species captured by WC36B (see Supplementary Table 5 MS data.xlsx).

Supplementary Notes: Chemical synthesis and characterization of Syk inhibitors

1. Chemical Synthesis of Syk inhibitors

An oven dried flask containing a magnetic stirrer bar was charged with 2-aminopyrimidine (104.5 mg, 1.1 mmol), an aldehyde (1 mmol), 4Å activated molecular sieves (125 mg), and yttrium(III) trifluoromethanesulfonate (56.8 mg, 0.1 mmol) under N₂ atmosphere. Dry dichloromethane (2.5 mL) was added, and the mixture was stirred 10 min at room temperature and then cooled to 4°C. *c*-Hex-diazoacetate⁴⁷ (201.6 mg, 1.2 mmol) in dichloromethane (1 mL) was added, and the reaction mixture was stirred at this temperature and monitored by the thin layer chromatography until complete conversion. The resulting mixture was filtered on Celite and concentrated under a reduced pressure. The crude was purified by column chromatography on a silica gel (hexane/ethylacetate) to afford the corresponding diazo-compounds, WC35, WC36, and WC38. These compounds were tested for their biological activities as racemic mixtures.

Cyclohexyl 2-diazo-3-(furan-2-yl)-3-(pyrimidin-2-ylamino)propanoate (WC35): 57% yield; ¹H NMR (500 MHz, CDCl₃): δ 8.29 (d, *J* = 4.6 Hz, 2H), 7.37 (s, 1H), 6.60-6.63 (m, 1H), 6.33-6.38 (m, 3H), 6.22-6.25 (m, 1H), 4.83-4.88 (m, 1H), 1.79-1.81 (m, 2H), 1.64-1.66 (m, 2H), 1.24-1.47 (m, 4H); ¹³C NMR (125 MHz, CDCl₃): δ 161.1, 158.1, 151.7, 142.4, 111.9, 110.5, 1-7.0, 46.6, 31.7, 31.6, 25.3, 23.5; HRMS (ESI): calculated for C₁₇H₂₀N₅O₃ [M+H]⁺ 342.1566, found 342.1568.

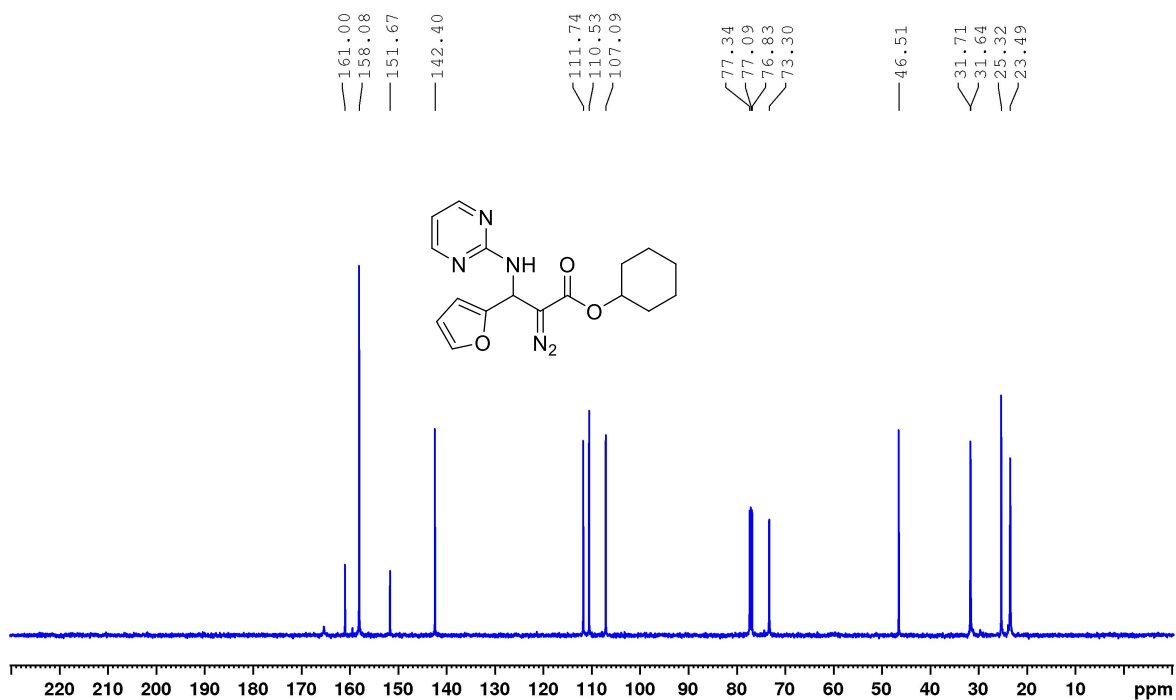
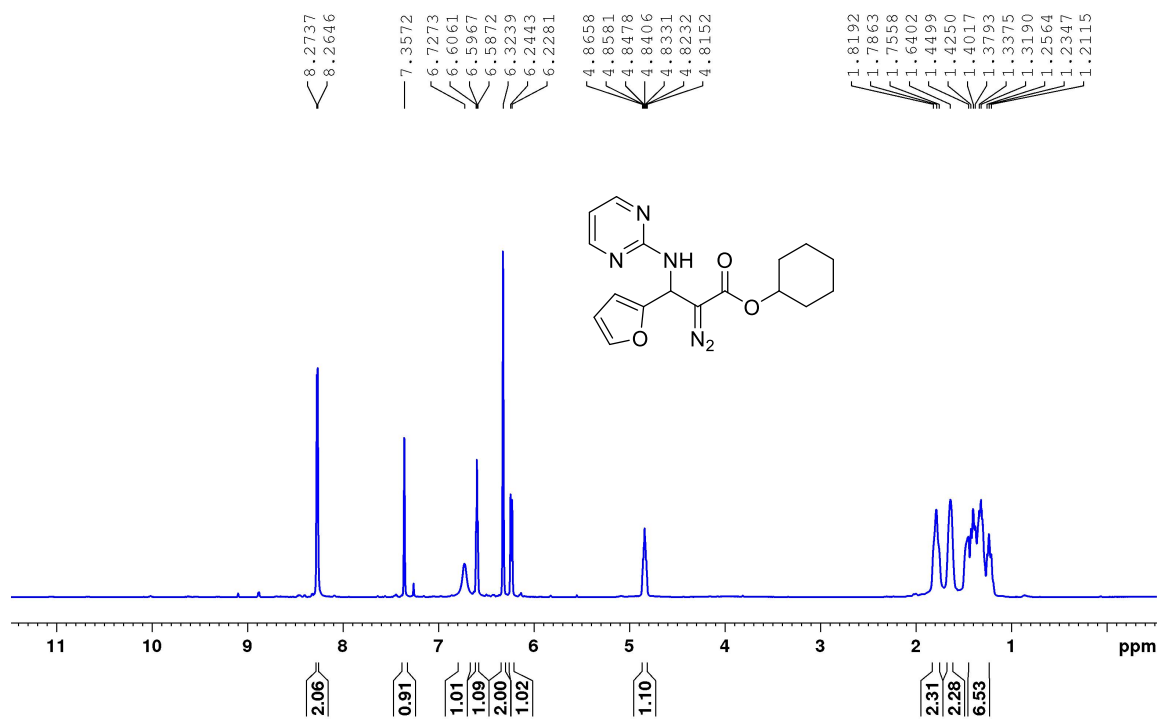
Cyclohexyl 2-diazo-3-(1H-indol-3-yl)-3-(pyrimidin-2-ylamino)propanoate (WC36): 55% yield; ¹H NMR (500 MHz, CDCl₃) 10.67 (d, *J* = 12 Hz, 1H), 8.43 (d, *J* = 4.7 Hz, 2H), 8.24-8.30 (m, 2H), 7.70 (d, *J* = 7.85 Hz, 1H), 7.34 (d, *J* = 8 Hz, 1H), 7.12-7.22 (m, 3H), 6.79 (t, *J* = 4.75 Hz, 1H), 4.97-5.02 (m, 1H), 1.90-1.93 (m, 2H), 1.64-1.68 (m, 2H), 1.33-1.53 (m, 6H); ¹³C NMR (125 MHz, CDCl₃) δ 168.6, 158.3, 138.9, 135.9, 127.3, 127.3, 121.8, 120.2, 119.5, 114.4, 112.9, 111.2, 100.5, 72.8, 31.7, 25.4, 23.8; HRMS (ESI) calculated for C₂₁H₂₃N₆O₂ [M+H]⁺ 391.1882, found 391.1886.

Cyclohexyl 2-diazo-3-(pyrimidin-2-ylamino)-3-(1H-pyrrol-2-yl)propanoate (WC38): 50% yield; ¹H NMR (500 MHz, CDCl₃): δ 8.89 (d, *J* = 2.35 Hz, 2H), 8.16 (s, 1H), 7.71-7.73 (m, 1H), 7.37-7.40 (m, 1H), 6.97-6.98 (m, 1H), 6.69-6.71 (m, 1H), 5.00-5.07 (m, 1H), 1.95-1.99 (m, 2H), 1.76-1.79 (m, 2H), 1.32-1.60 (m, 6H); ¹³C NMR (125 MHz, CDCl₃): δ 163.3, 159.0, 158.0, 145.1, 130.5, 127.0, 120.2, 116.6, 115.7, 108.3, 106.5, 73.8, 31.7, 25.4, 23.8; HRMS (ESI): calculated for C₁₇H₂₁N₆O₂ [M+H]⁺ 341.1726, found 341.1729.

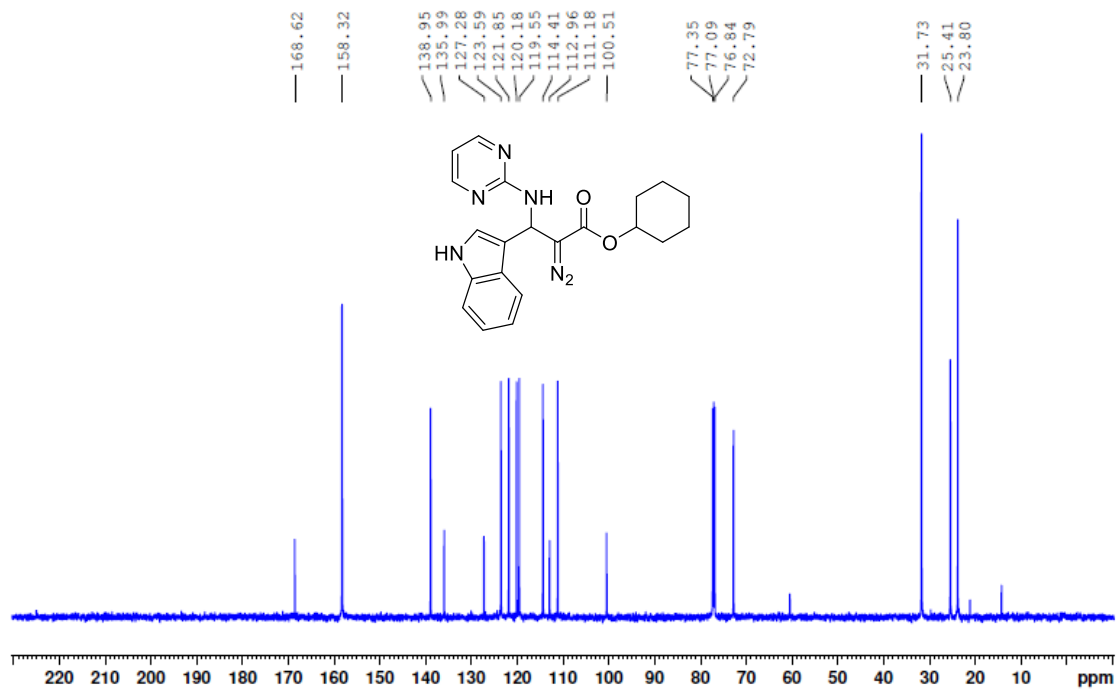
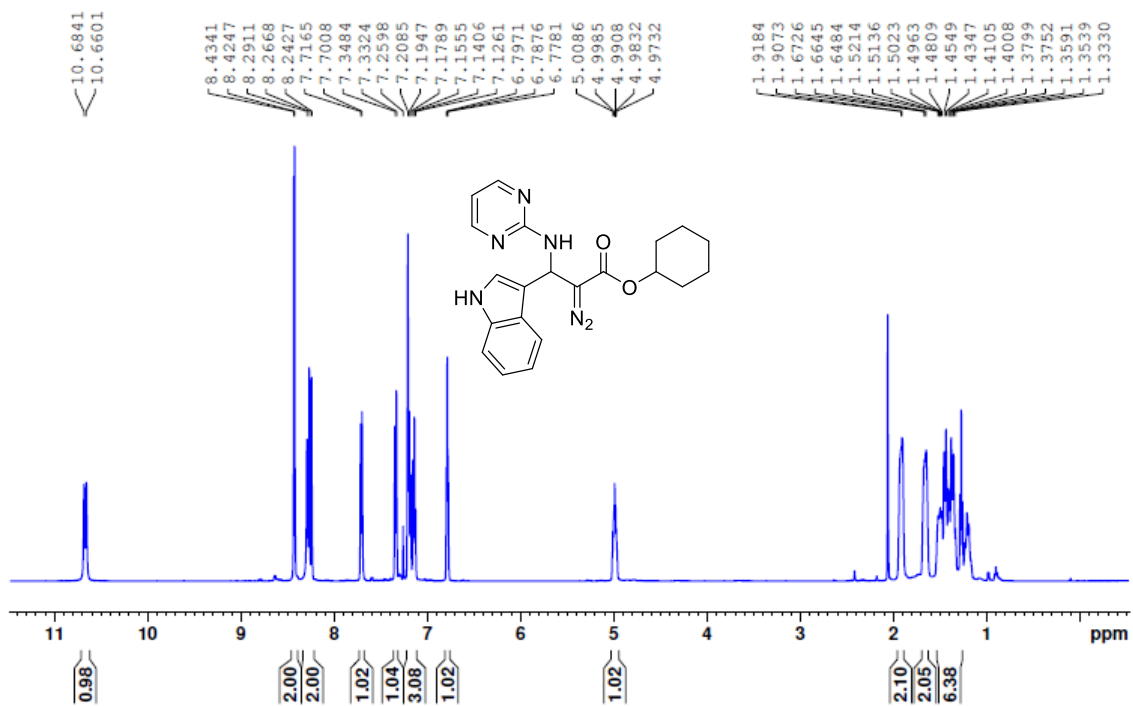
Synthesis of biotinylated WC36 (WC36B; N-(2-(2-diazo-3-(1H-indol-3-yl)-3-(pyrimidin-2-ylamino)propanamido)ethyl)-5-((3aR,4R,6aS)-2-oxohexahydro-1H-thieno[3,4d]imidazol-4-yl)pentanamide): Flask containing a magnetic stirrer bar was charged with WC-36 (195 mg, 0.5 mmol), and biotinamide (121.5 mg, 0.5 mmol) under N₂ atmosphere. Methanol (5 mL) and triethylamine (100 ml) was added, and the mixture was stirred overnight at the room temperature. The resulting mixture was concentrated under a reduced pressure. The crude was purified by column chromatography on a silica gel (hexane/ethylacetate) to afford the corresponding WC36B. 42% yield; ¹H NMR (500 MHz, CDCl₃): δ 9.71 (brs, 1H), 8.20-8.23 (m, 2H), 7.47-7.50 (m, 1H), 7.26-7.33 (m, 2H), 6.98-7.10 (m, 3H), 6.78-6.83 (m, 1H), 6.60-6.67 (m, 1H), 6.54 (t, 1H, *J* = 4.70 Hz), 6.03 (brs, 1H), 5.50-5.52 (m, 2H), 4.76-4.98 (m, 2H), 4.02-4.21 (m, 2H), 2.89-3.22 (m, 4H), 2.50-2.67 (m, 2H), 1.27-2.01 (m, 12H). ¹³C NMR (125 MHz, CDCl₃): δ 174.1, 169.8, 168.8, 164.5, 163.1, 158.2, 153.0, 148.6, 136.3, 136.0, 127.6, 126.8, 125.0, 122.8, 121.5, 121.3, 120.2, 119.1, 118.8, 111.4, 111.3, 108.4, 93.2, 90.5, 71.6, 61.8, 60.2, 55.6, 47.5, 40.3, 35.8, 35.6, 32.1, 31.9, 28.1, 27.9, 25.5, 23.9; HRMS (ESI): calculated for C₂₇H₃₃N₁₀O₃S [M]⁺ 577.2630, found 577.2645.

2. NMR spectra of Syk inhibitors

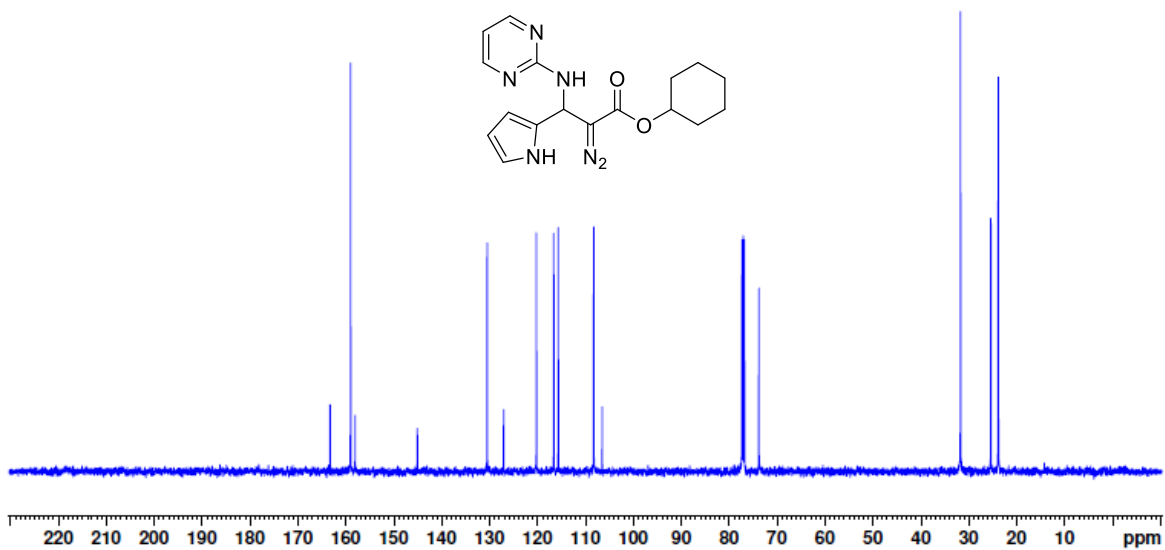
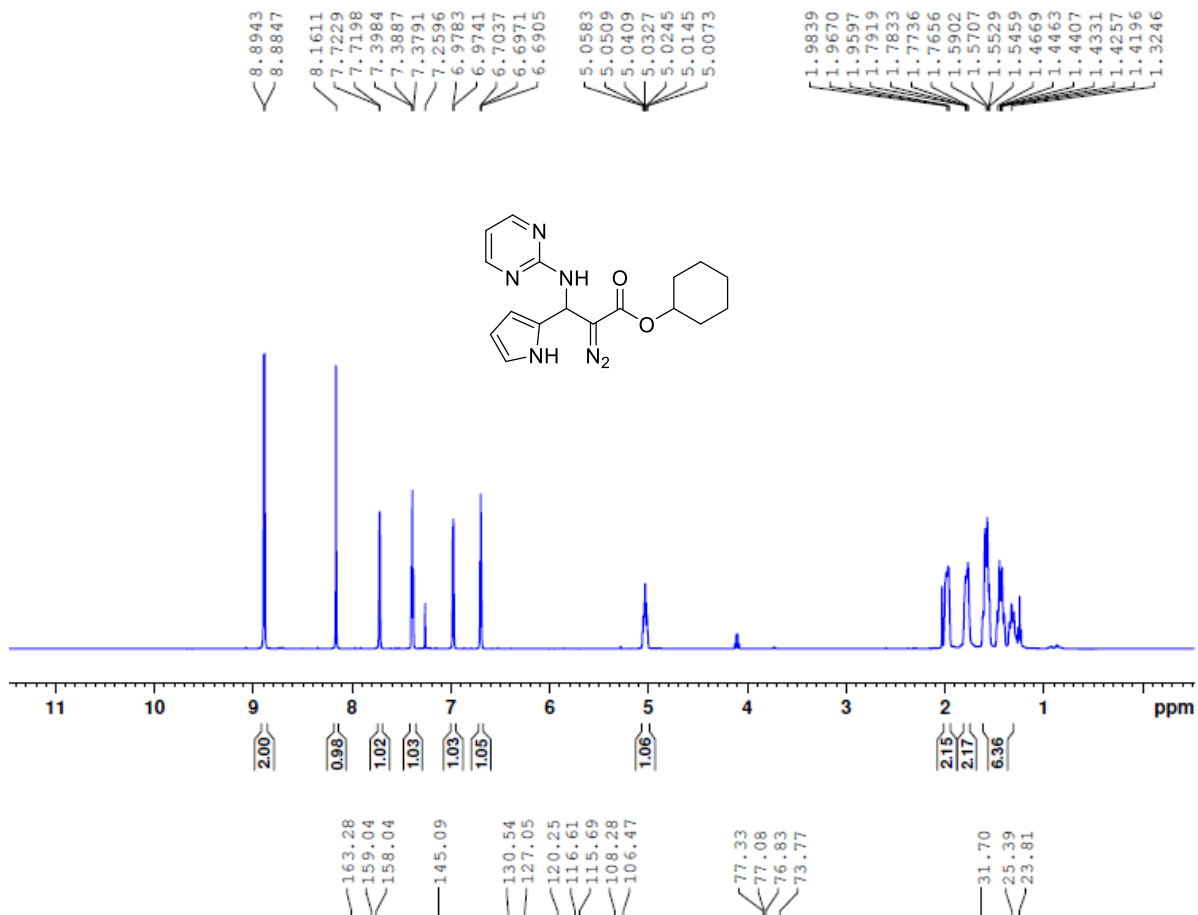
^1H and ^{13}C NMR Spectra of **WC35** (CDCl_3 ; 500 MHz)



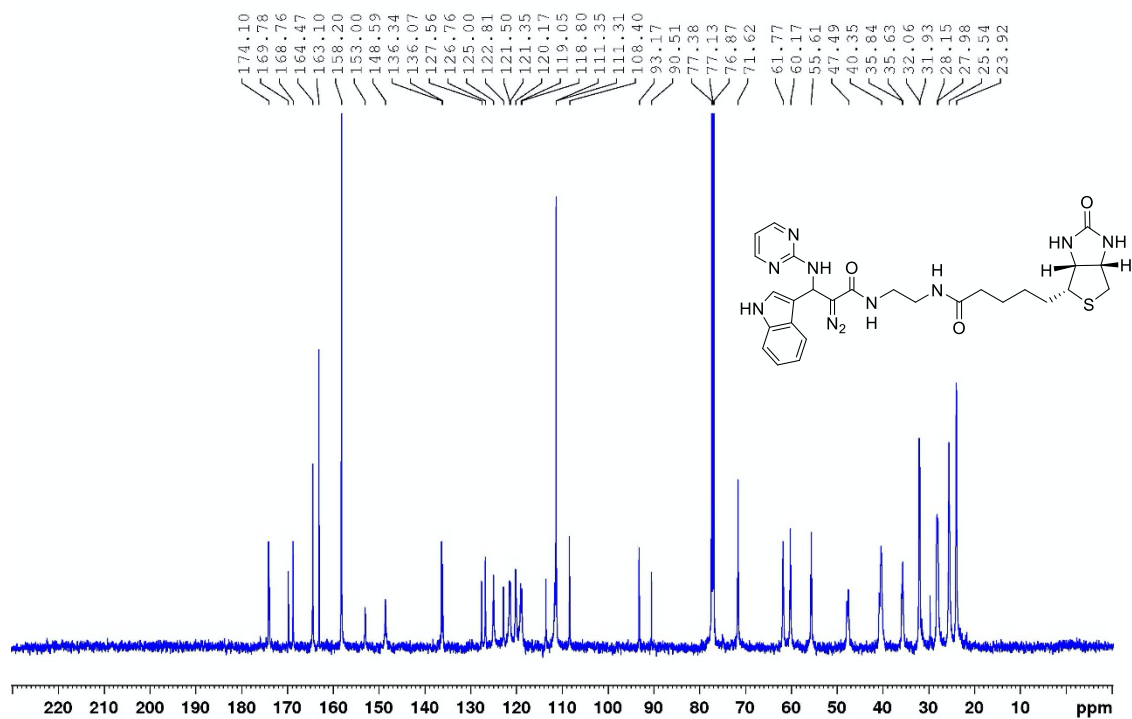
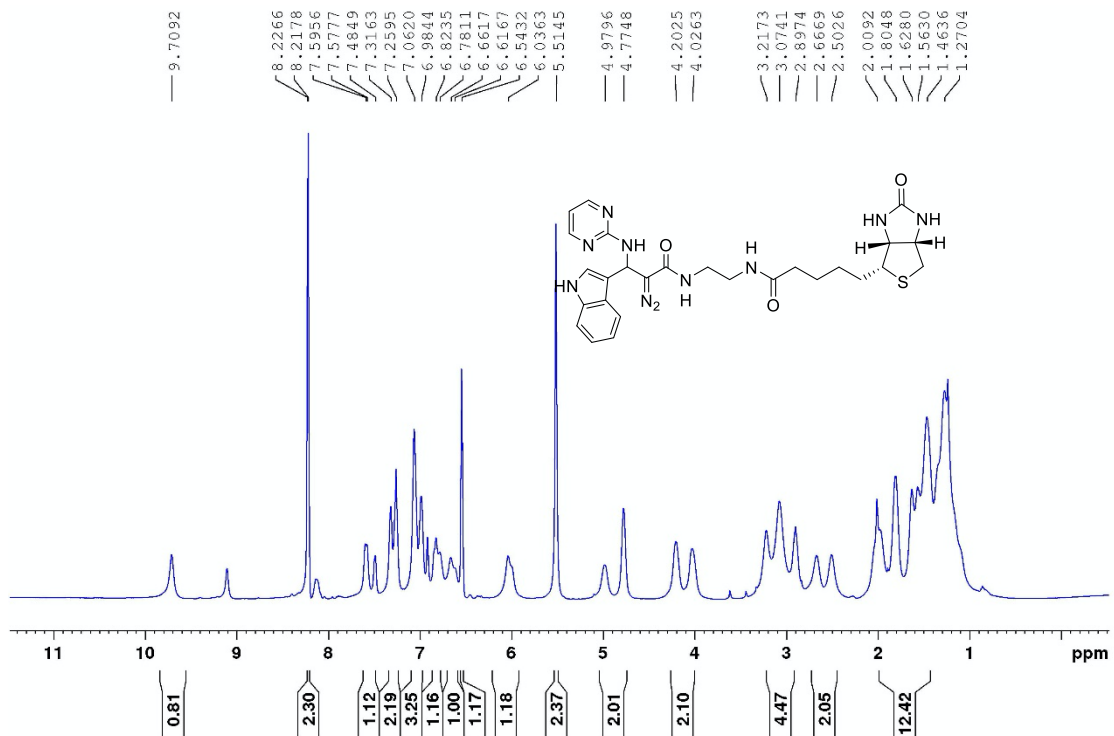
^1H and ^{13}C NMR Spectra of **WC36** (CDCl_3 ; 500 MHz)



^1H and ^{13}C NMR Spectra of **WC38** (CDCl_3 ; 500 MHz)



¹H and ¹³C NMR Spectra of **WC36B** (CDCl₃; 500 MHz)



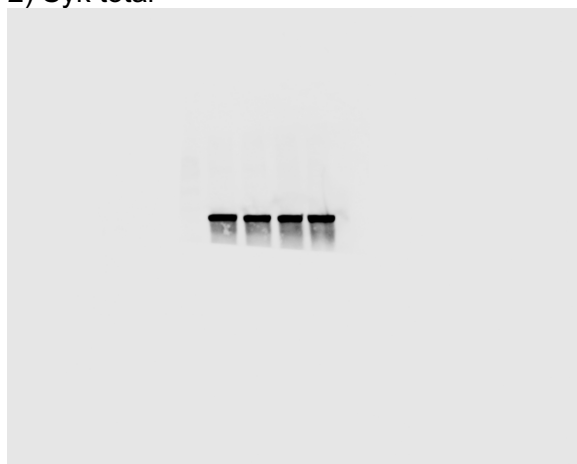
Supplementary Data Files:

1. Source data files for Supplementary Fig. 5a

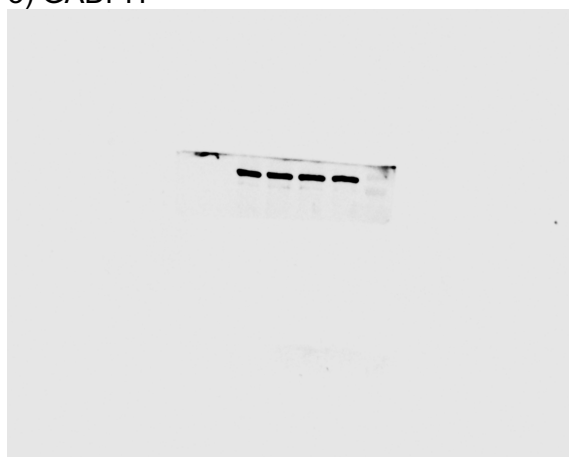
1) pSyk



2) Syk total



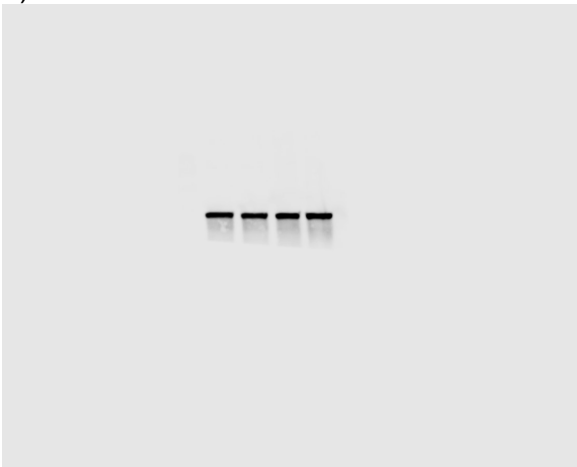
3) GADPH



4) pSTAT5



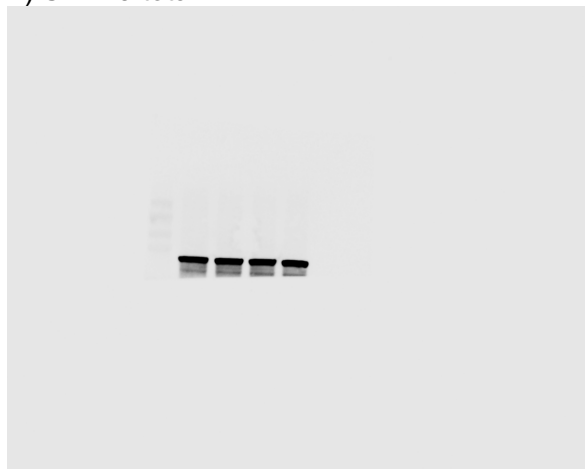
5) STAT5 total



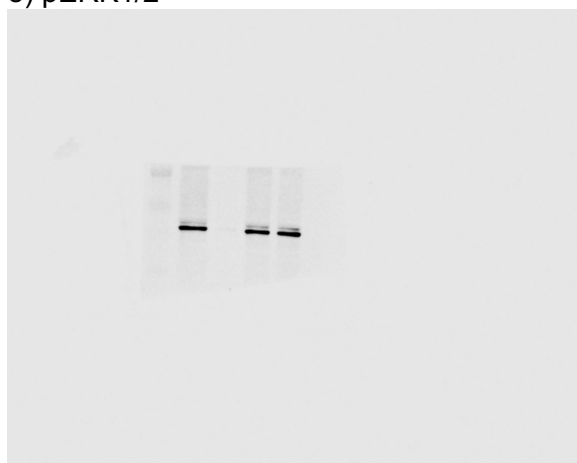
6) pSTAT3



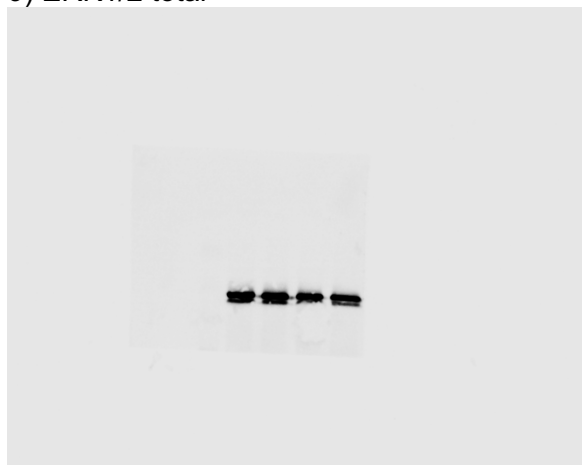
7) STAT3 total



8) pERK1/2



9) ERK1/2 total



2. Source data files for Supplementary Fig. 6
Supplementary Fig 6a

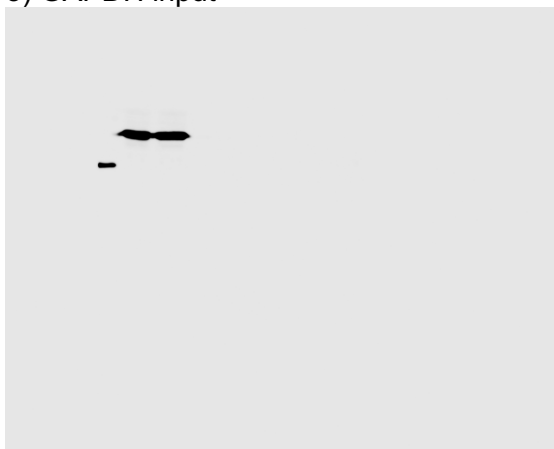
1) Syk IP



2) Syk input



3) GAPDH input



Supplementary Fig 6b

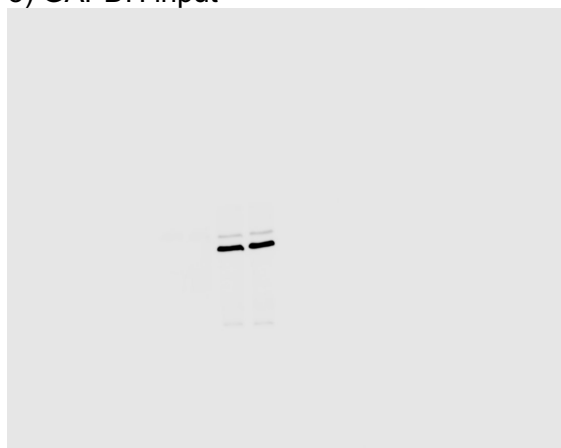
1) STAT3 IP



2) STAT3 input



3) GAPDH input

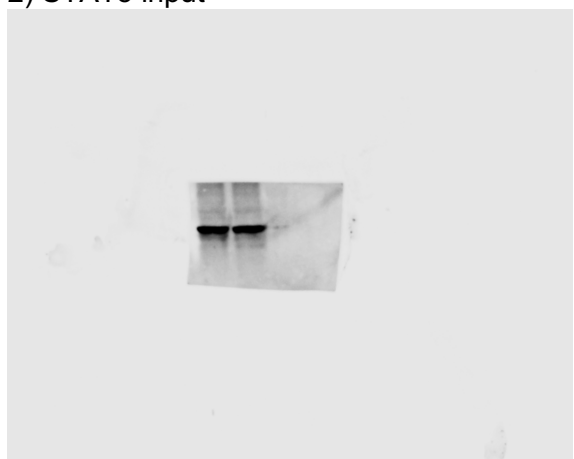


Supplementary Fig 6c

1) STAT5 IP



2) STAT5 input



3) GAPDH input



Supplementary Fig 6d

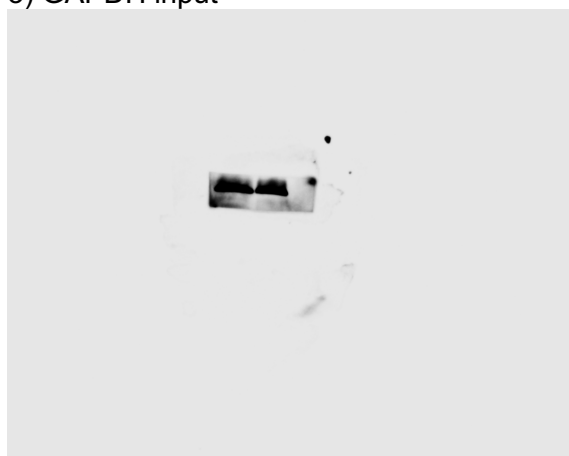
1) ERK1/2 IP



2) ERK1/2 input

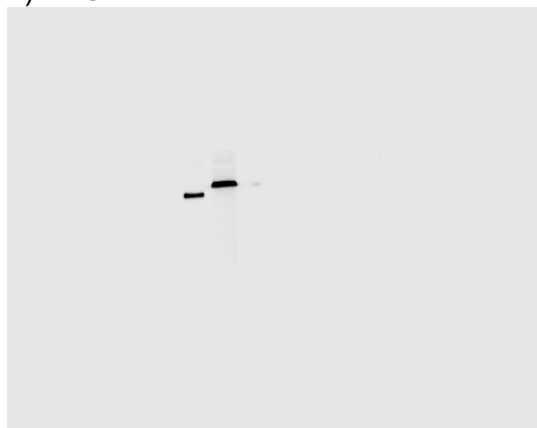


3) GAPDH input

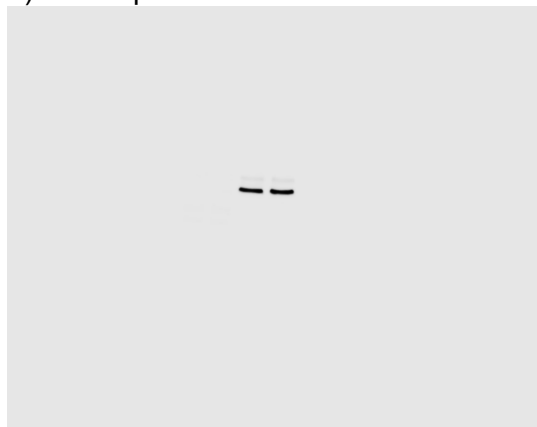


Supplementary Fig 6e

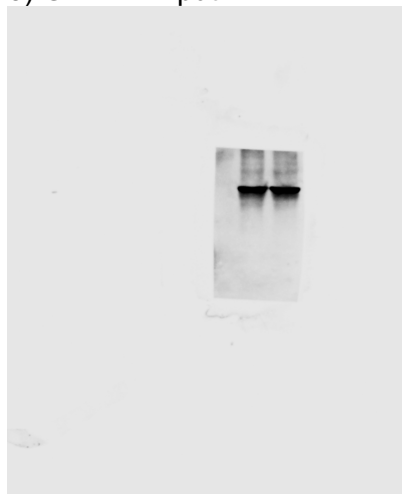
1) RAS IP



2) RAS input

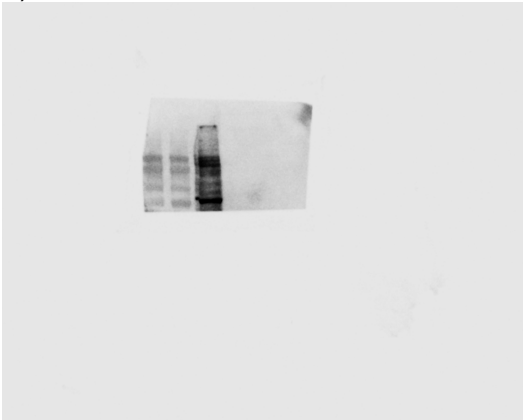


3) GAPDH input

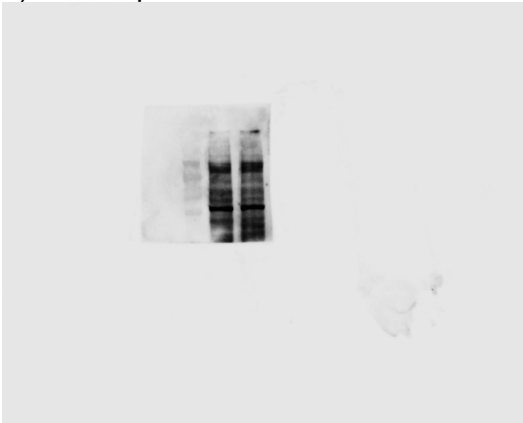


Supplementary Fig 6f

1) FLT3 IP



2) FLT3 input



3) GAPDH input

