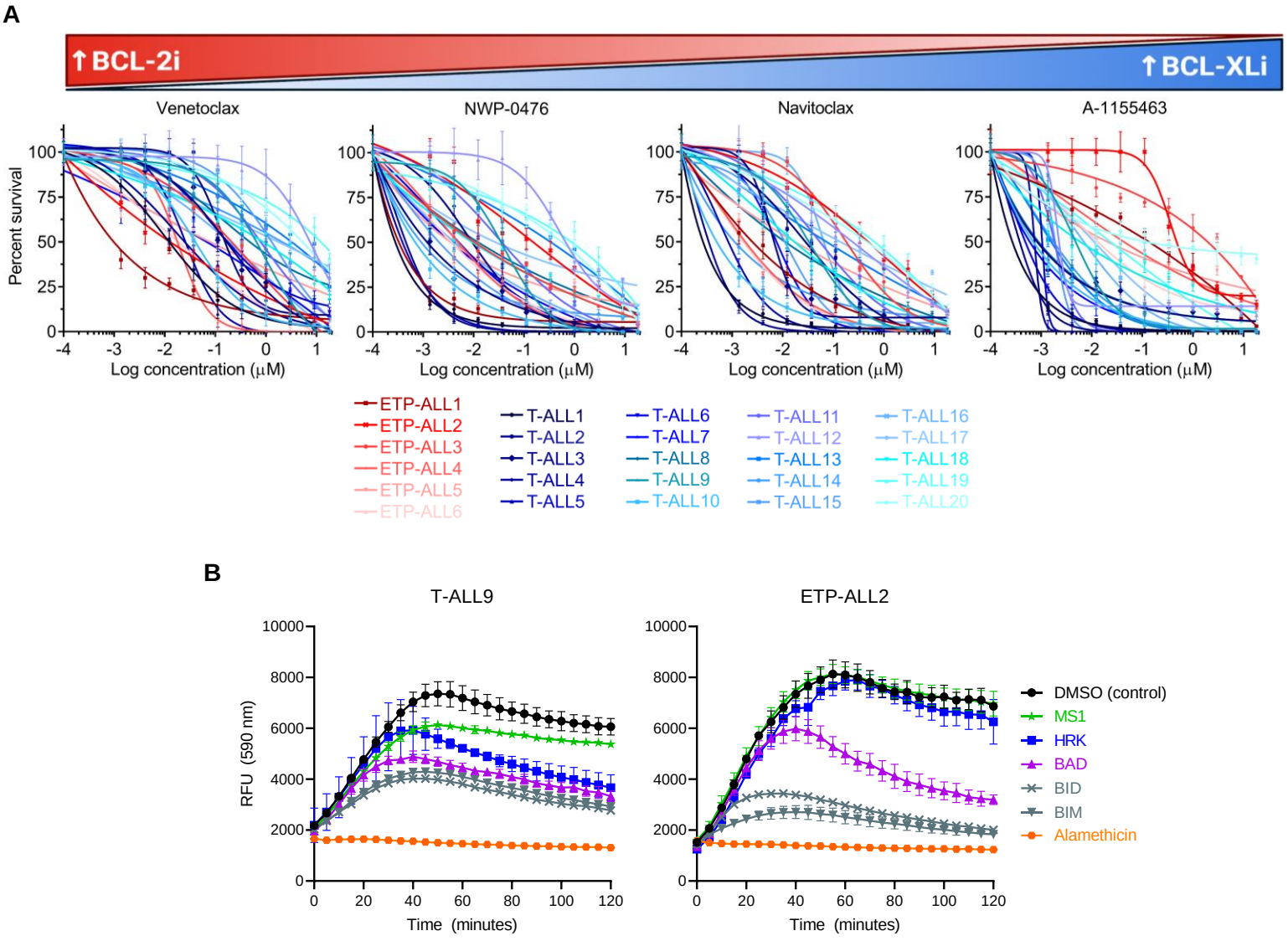
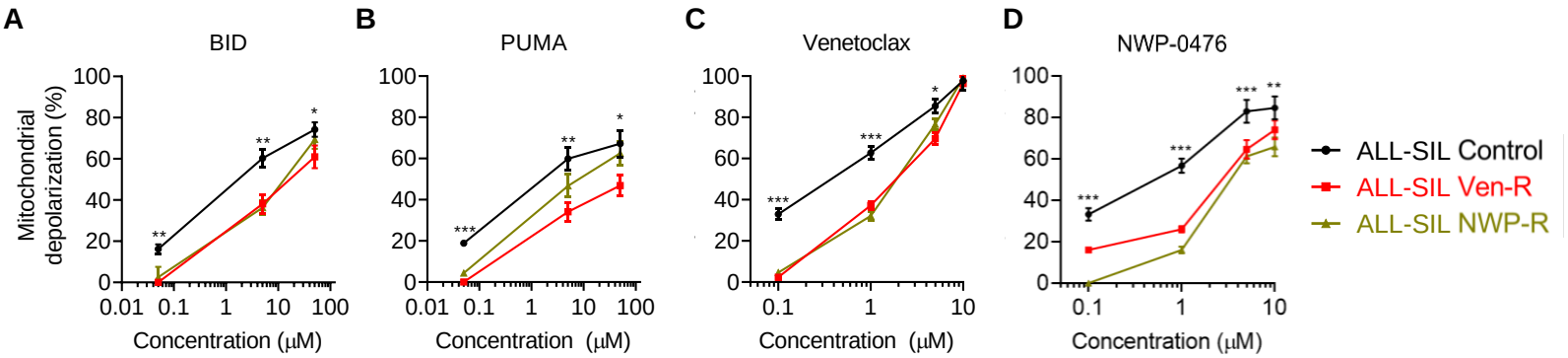
Supplementary Figure 1



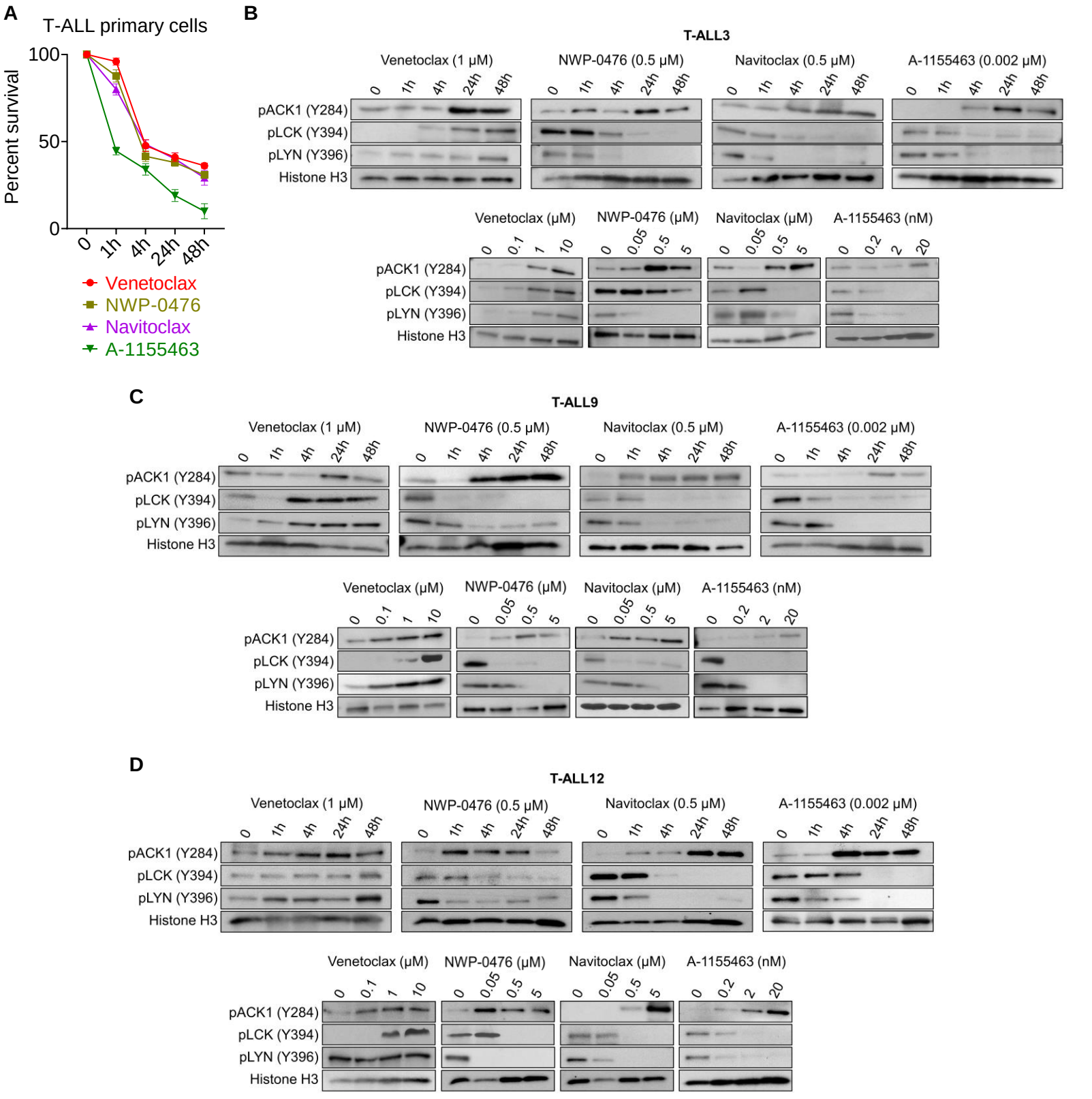
(A) Curves showing the viability of T-ALL primary cells treated with BH3 mimetics. **(B)** Mitochondrial polarization curves were obtained with BH3 profiling of T-lineage ALL primary cells.

Supplementary Figure 2



BH3 profiling of ALL-SIL control, venetoclax-resistant (ven-R), and NWP-0476-resistant (NWP-R) cells, showing mitochondrial depolarization with titration assays for BID **(A)**, PUMA **(B)**, venetoclax **(C)**, and NWP-0476 **(D)**.

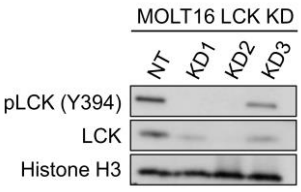
Supplementary Figure 3



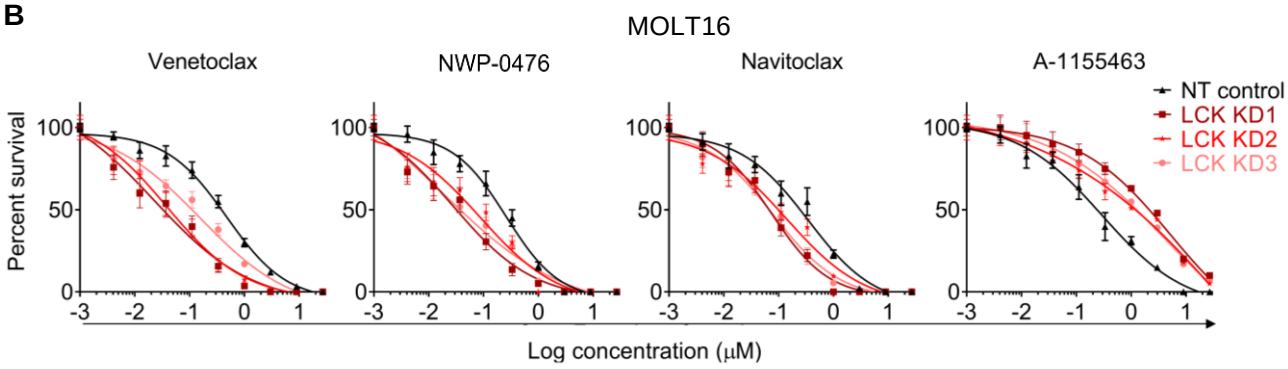
(A) Percent survival curves for the T-ALL primary cells (T-ALL3, T-ALL9 and T-ALL12) treated with venetoclax, NWP-0476, navitoclax and A-1155463 over 48 hours. **(B, C, D)** Immunoblots showing time-dependent and dose-dependent changes in pACK, pLCK, and pLYN levels of T-ALL primary cells treated with BH3 mimetics.

Supplementary Figure 4

A



B



C

MOLT4 LCK KD IC_{50} values (μM)

	NT control	KD1	KD2	KD3
Venetoclax	4.03	0.66	0.92	1.124
NWP-0476	1.6	0.67	0.19	0.09
Navitoclax	0.31	0.06	0.013	0.002
A-1155463	0.012	0.1	0.19	0.14

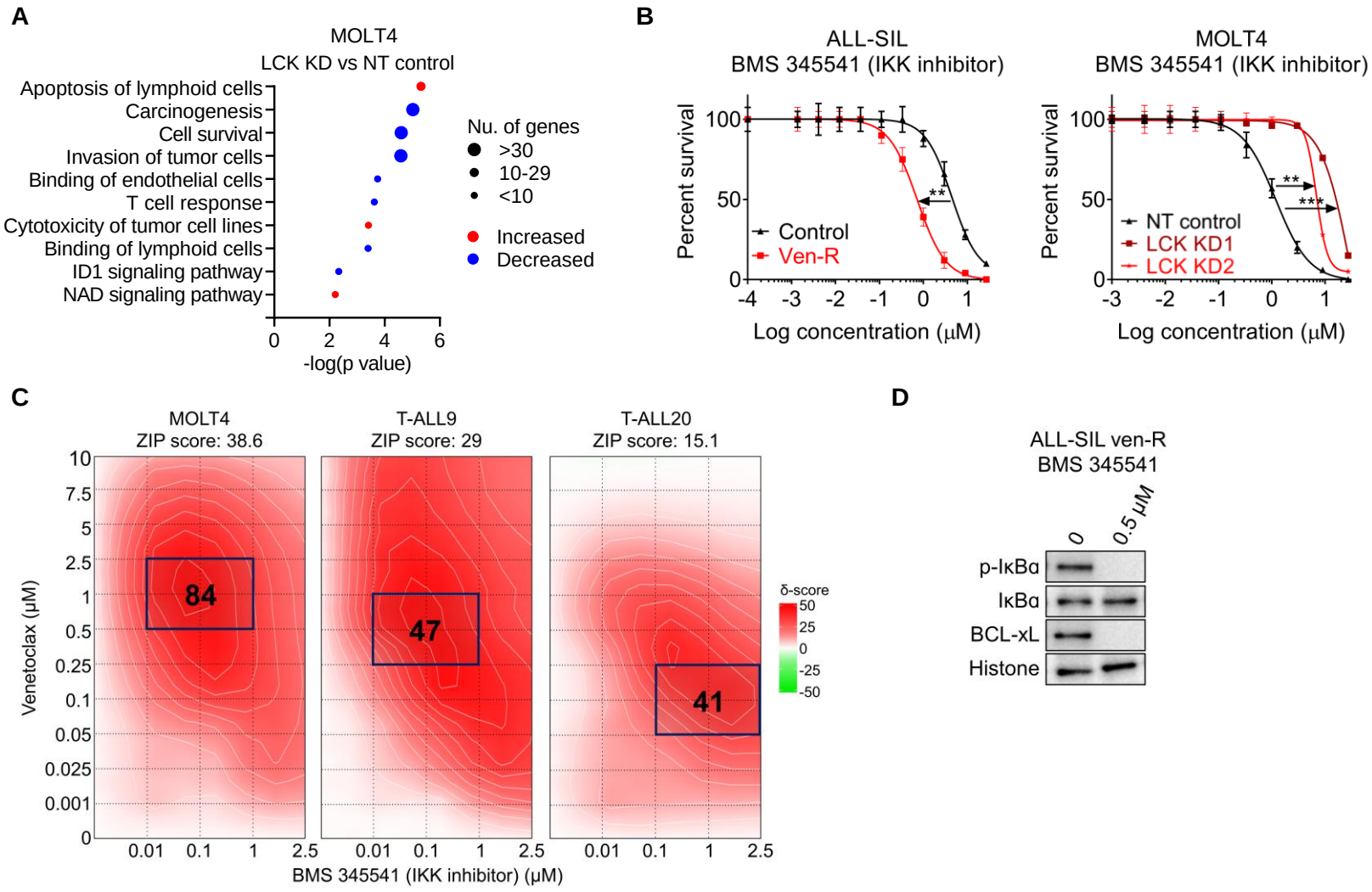
D

MOLT16 LCK KD IC_{50} values (μM)

	NT control	KD1	KD2	KD3
Venetoclax	0.47	0.02	0.03	0.15
NWP-0476	0.26	0.02	0.08	0.01
Navitoclax	0.34	0.07	0.14	0.08
A-1155463	0.29	2.9	1.1	1.9

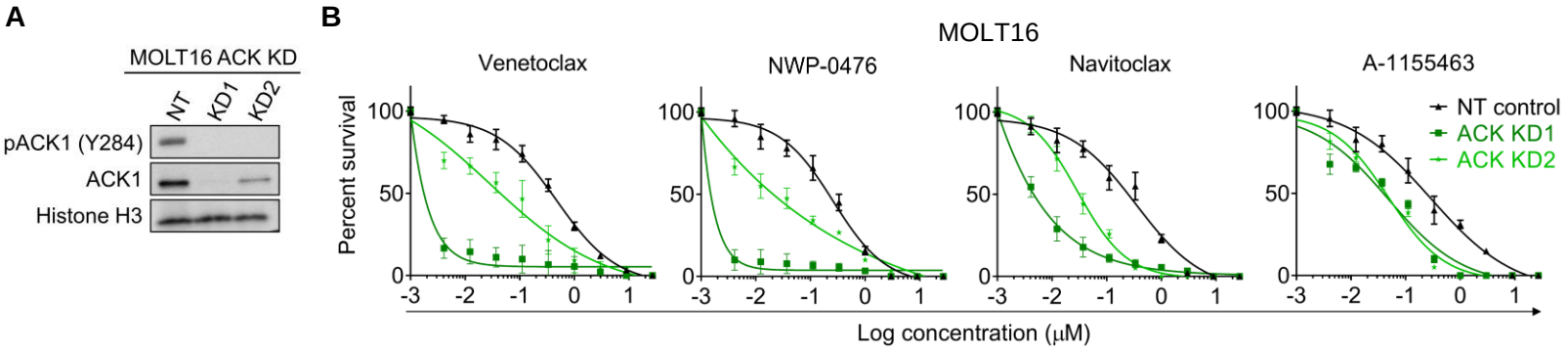
(A) Immunoblot showing LCK knockdown (KD) in MOLT16 T-ALL cell line, using three non-overlapping shRNA constructs. **(B)** Curves showing viability of MOLT16 non-targeting (NT) control and LCK KD cells treated with BH3 mimetics. **(C, D)** Tables showing IC_{50} values for MOLT4 and MOLT16 NT control and LCK KD cells treated with BH3 mimetics.

Supplementary Figure 5



(A) Ingenuity pathway analysis results showing pathways that are altered with LCK knockdown (KD) in MOLT4 T-ALL cell line. **(B)** Curves showing viability of ALL-SIL control vs venetoclax-resistant (ven-R), and MOLT4 non-targeting (NT) control vs LCK KD cells treated with the IKK inhibitor, BMS 345541. **(C)** ZIP synergy plots for MOLT4 cell line and T-ALL primary cells treated with venetoclax and BMS 345541. **(D)** Immunoblots showing p-IκBα, IκBα, and BCL-xL levels in BMS 345541-treated ALL-SIL ven-R cells. *p <0.05, **p <0.01, ***p <0.001

Supplementary Figure 6



C

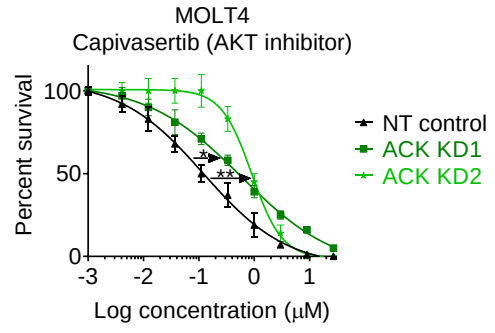
MOLT4 ACK KD IC_{50} values with 95% CI (μM)

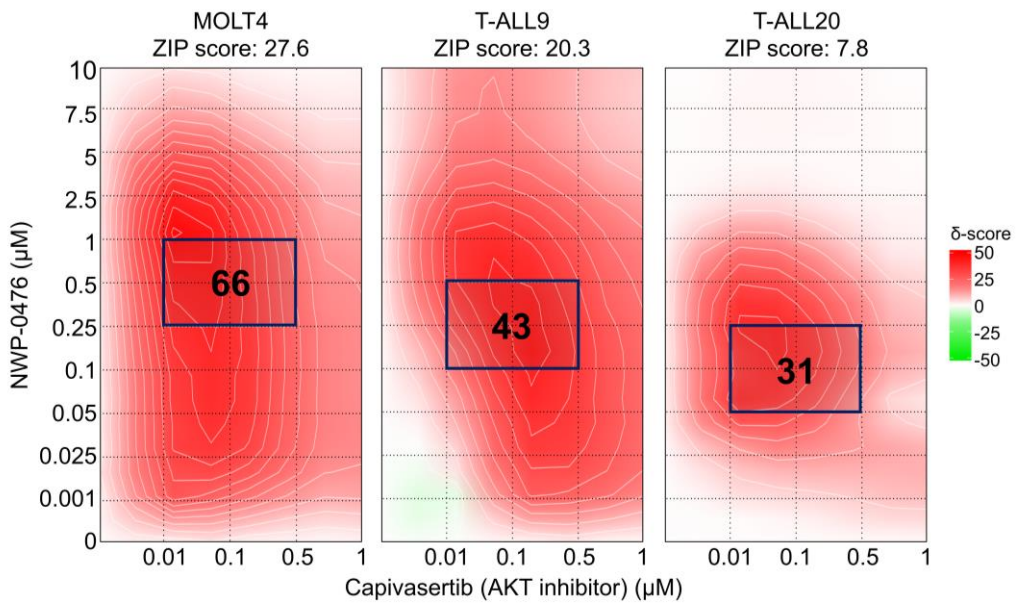
	NT control	KD1	KD2
Venetoclax	4.35 (3.2 – 5.1)	1.1 (0.9 – 1.3)	1.56 (1.4 – 1.7)
NWP-0476	1.01 (0.8 – 1.4)	0.22 (0.1 – 0.3)	0.09 (0.04 – 0.12)
Navitoclax	0.38 (0.2 – 0.5)	0.024 (0.02 – 0.05)	0.021 (0.01 – 0.03)
A-1155463	0.014 (0.01 – 0.02)	0.00017 (0.0001–0.0003)	0.000078 (0.00001-0.0001)

D

MOLT16 ACK KD IC_{50} values (μM)

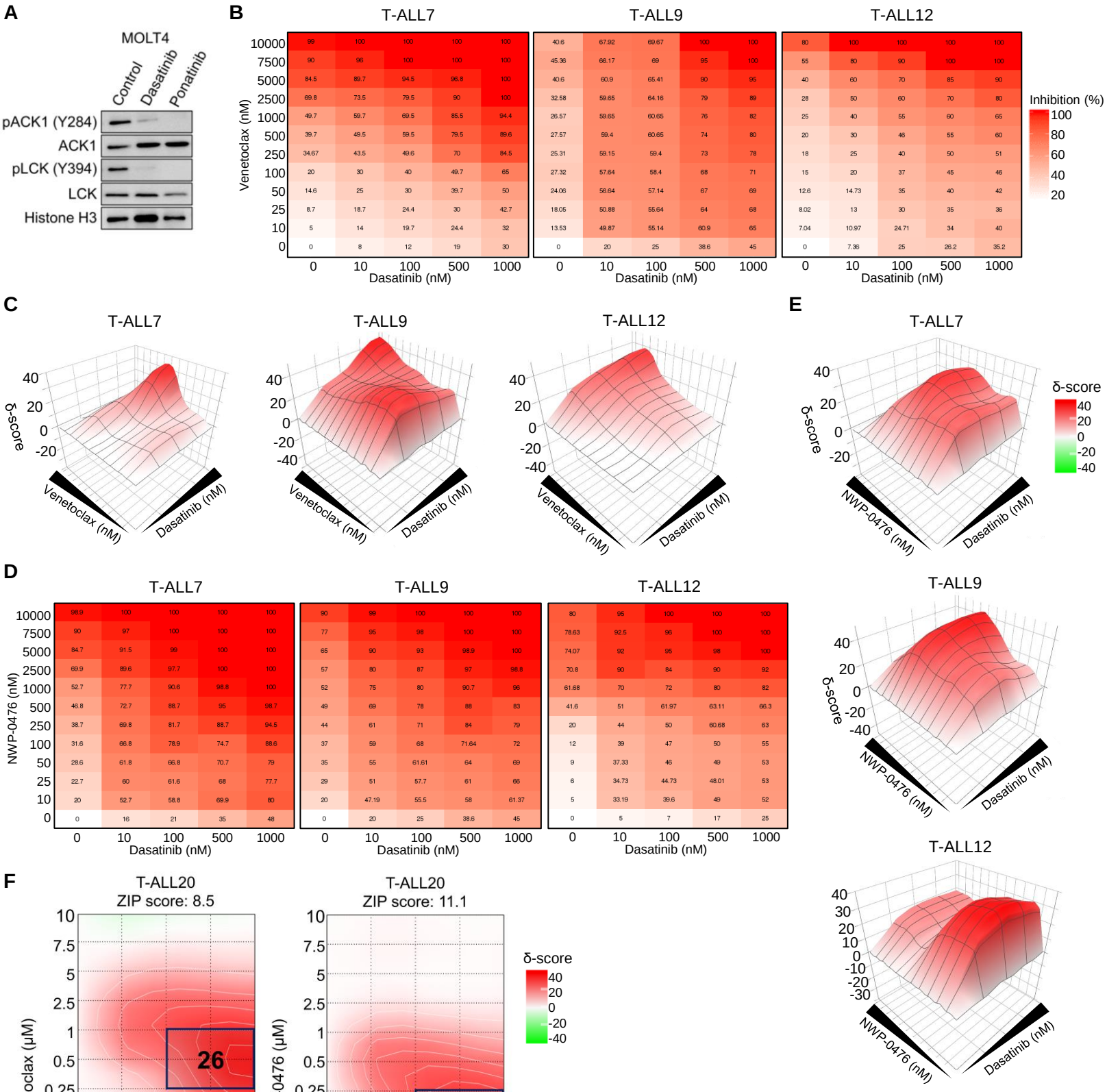
	NT control	KD1	KD2
Venetoclax	0.47 (0.3 – 0.62)	0.003 (0.001– 0.004)	0.02 (0.01 – 0.03)
NWP-0476	0.26 (0.1 – 0.4)	0.002 (0.001 – 0.005)	0.03 (0.01 – 0.05)
Navitoclax	0.34 (0.28 – 0.51)	0.005 (0.001 – 0.01)	0.02 (0.01 – 0.04)
A-1155463	0.29 (0.14 – 0.42)	0.04 (0.01 – 0.9)	0.04 (0.02 – 0.07)





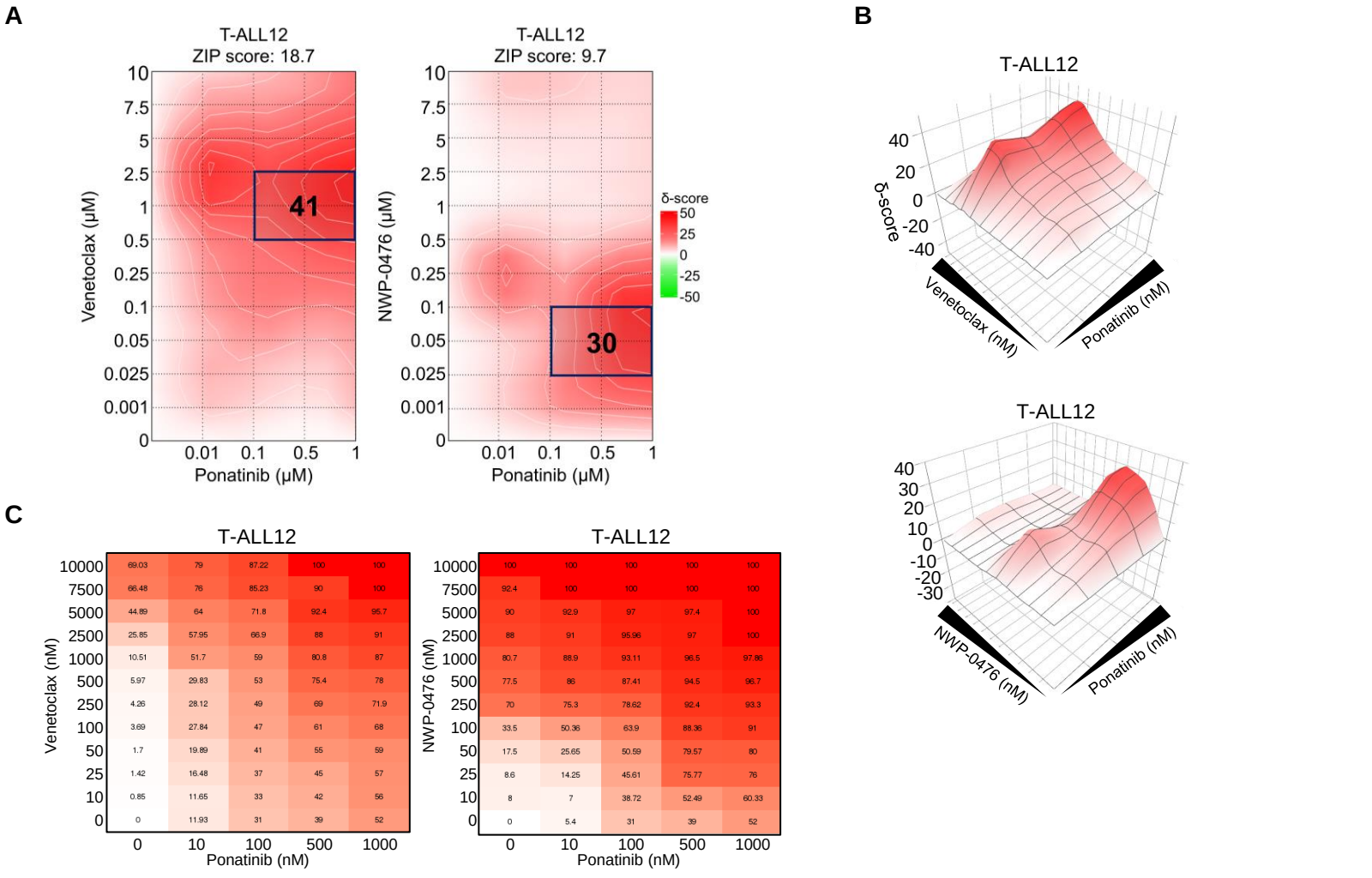
(A) Immunoblot showing ACK1 knockdown (KD) in MOLT16 T-ALL cell line, using two non-overlapping shRNA constructs. **(B)** Curves showing viability of MOLT16 non-targeting (NT) control and ACK1 KD cells treated with BH3 mimetics. **(C, D)** Tables showing IC_{50} values and their 95% confidence intervals (CI) for MOLT4 and MOLT16 NT control and ACK1 KD cells treated with BH3 mimetics. Experiments were done in triplicates. **(E)** Curves showing viability of MOLT4 NT control and ACK1 KD cells treated with capivasertib. **(F)** ZIP synergy plots for MOLT4 cell line and T-ALL primary cells treated with NWP-0476 and capivasertib. . *p <0.05, **p <0.01, ***p <0.001

Supplementary Figure 7



(A) Immunoblot showing changes in pACK1 and pLCK levels in MOLT4 T-ALL cells after 24-hours of treatment with dasatinib or ponatinib at 0.5 μM dose. **(B, D)** Tables showing percent death of T-ALL primary cells treated with combinations of BH3 mimetics and dasatinib. **(C, E, F)** ZIP synergy plots of T-ALL primary cells treated with BH3 mimetics and dasatinib.

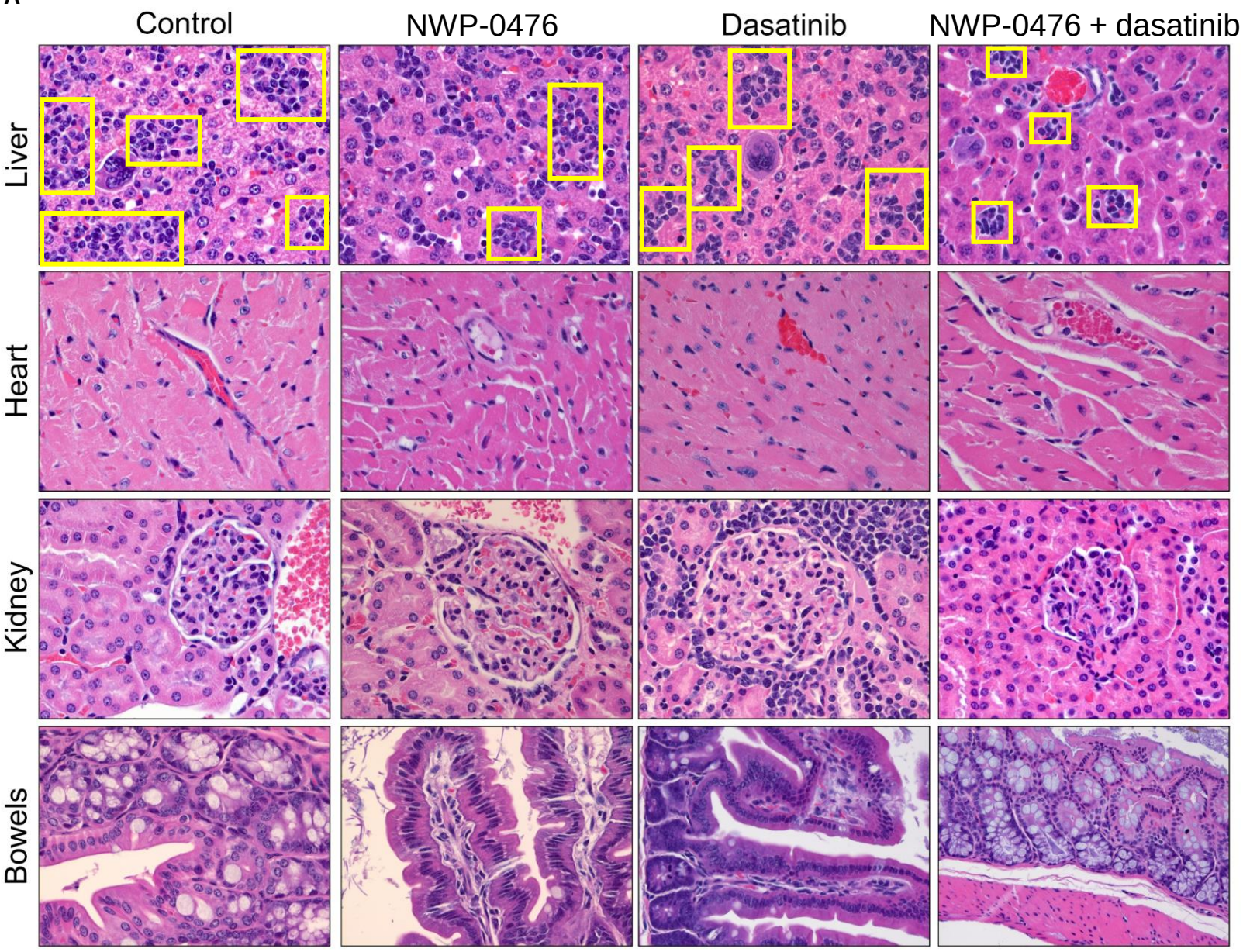
Supplementary Figure 8



(A, B) ZIP synergy plots of T-ALL primary cells treated with BH3 mimetics and ponatinib. **(C)** Tables showing percent death of T-ALL primary cells treated with combinations of BH3 mimetics and ponatinib.

Supplementary Figure 9

A



(A) Histopathologic examination of organs (liver, heart, kidney and bowels) from T-ALL PDX-engrafted NSG mice treated with vehicle control, NWP-0476, dasatinib, and NWP-0476 + dasatinib. Squares in liver images highlight the leukemic cell infiltrates.