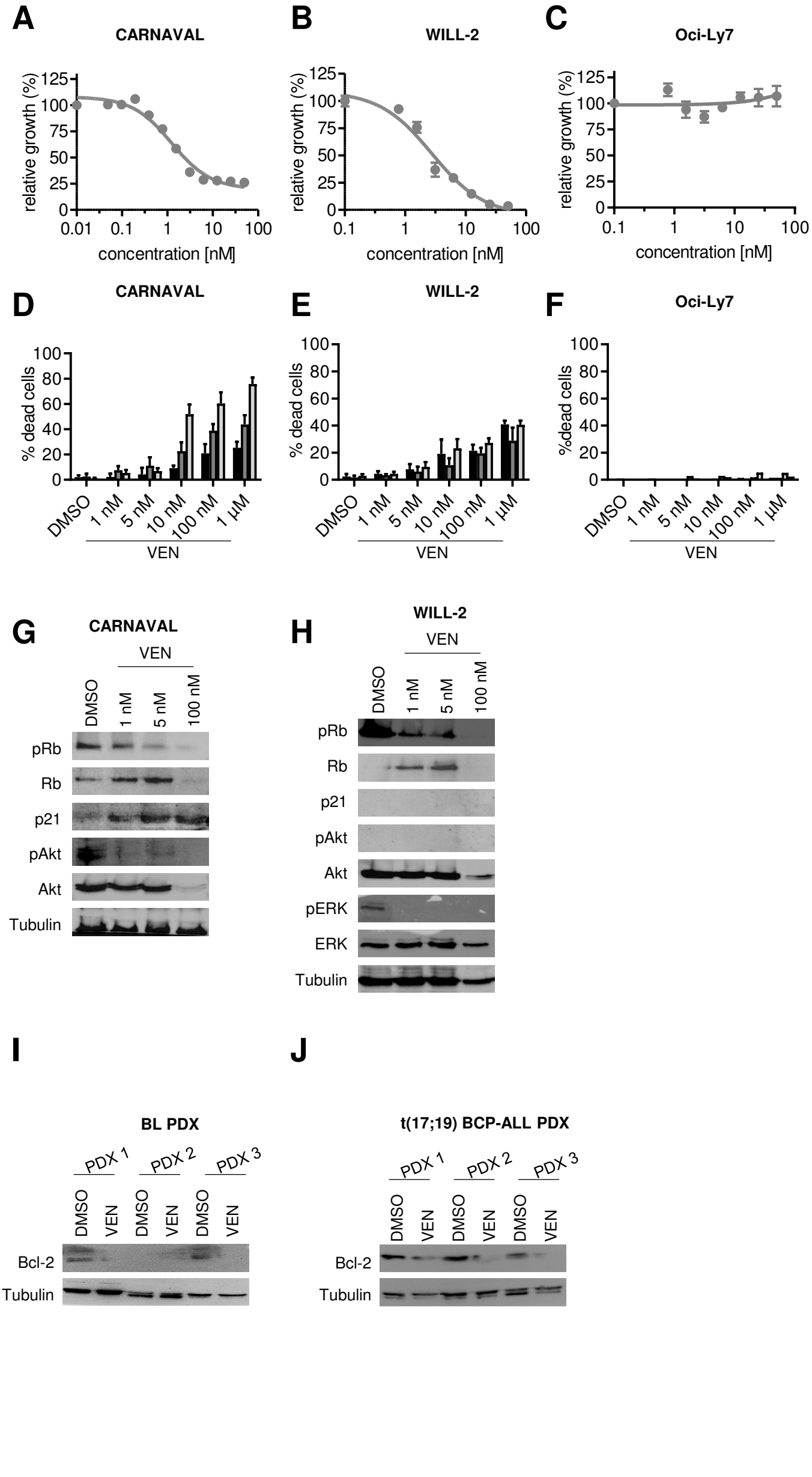
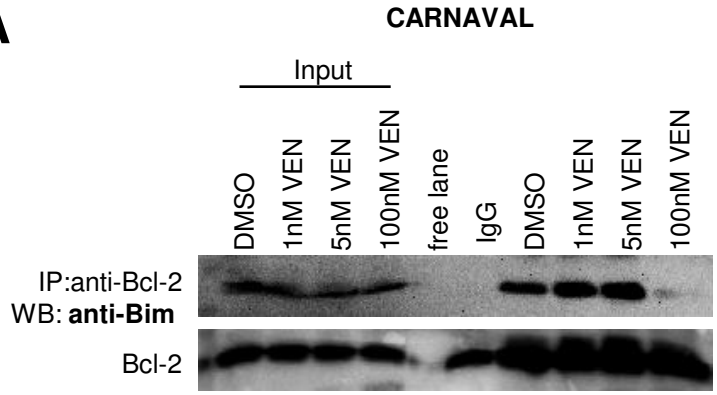
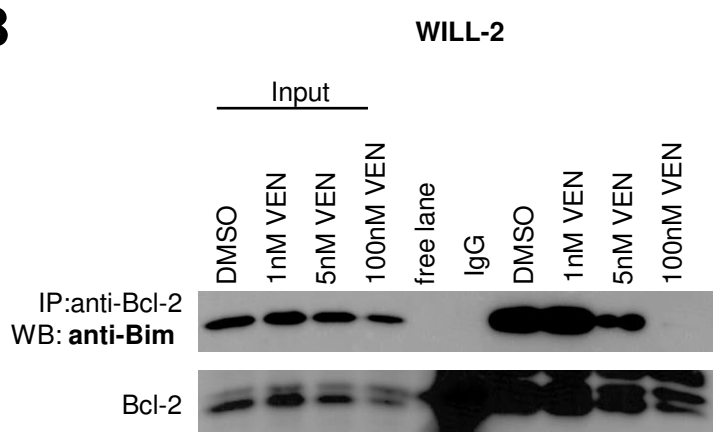

A
CARNAVAL
relative growth (%)
125
100
75
50
25
0
0.01
0.1
1
10
100
concentration [nM]
B
WILL-2
relative growth (%)
125
100
75
50
25
0
0.1
1
10
100
concentration [nM]
C
Oci-Ly7
relative growth (%)
125
100
75
50
25
0
0.1
1
10
100
concentration [nM]
D
CARNAVAL
% dead cells
100
80
60
40
20
0
DMSO
1 nM
5 nM
10 nM
100 nM
1 μM
VEN
E
WILL-2
% dead cells
100
80
60
40
20
0
DMSO
1 nM
5 nM
10 nM
100 nM
1 μM
VEN
F
Oci-Ly7
%dead cells
100
80
60
40
20
0
DMSO
1 nM
5 nM
10 nM
100 nM
1 μM
VEN
G
CARNAVAL
VEN
DMSO
1 nM
5 nM
100 nM
pRb
Rb
p21
pAkt
Akt
Tubulin
H
WILL-2
VEN
DMSO
1 nM
5 nM
100 nM
pRb
Rb
p21
pAkt
Akt
pERK
ERK
Tubulin
I
BL PDX
PDX 1
PDX 2
PDX 3
DMSO
VEN
DMSO
VEN
DMSO
VEN
Bcl-2
Tubulin
J
t(17;19) BCP-ALL PDX
PDX 1
PDX 2
PDX 3
DMSO
VEN
DMSO
VEN
DMSO
VEN
Bcl-2
Tubulin

Vogiatzi et al., 2022 Supplementary Figure 2

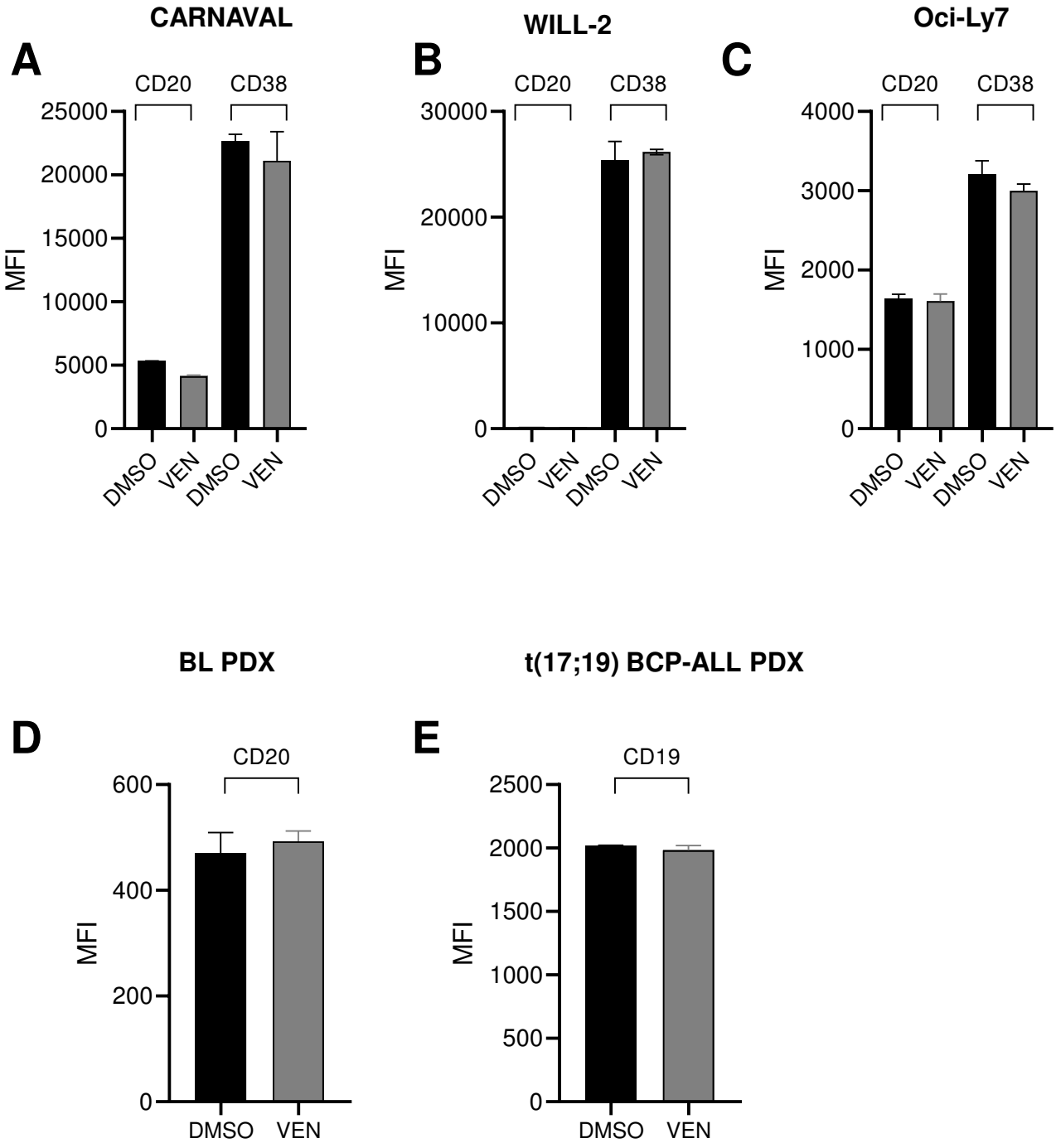
A



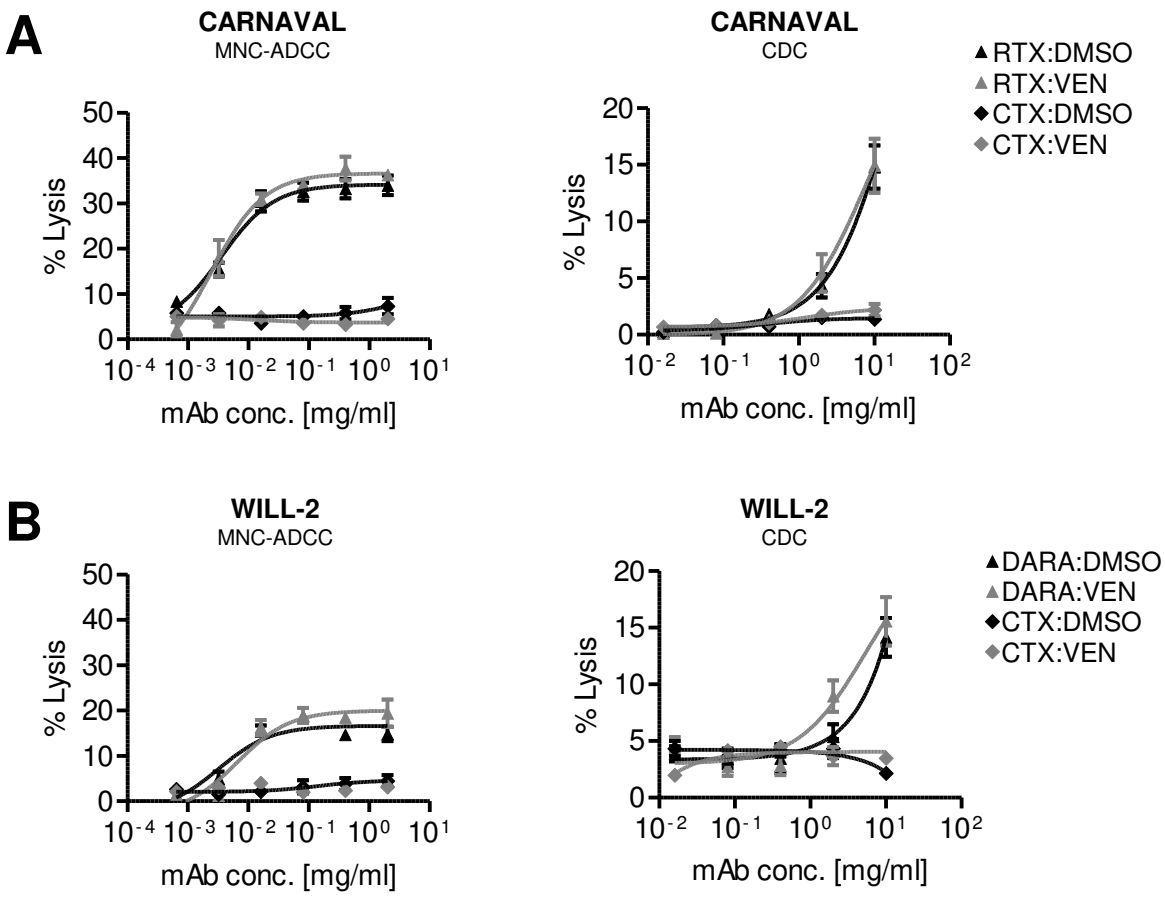
B

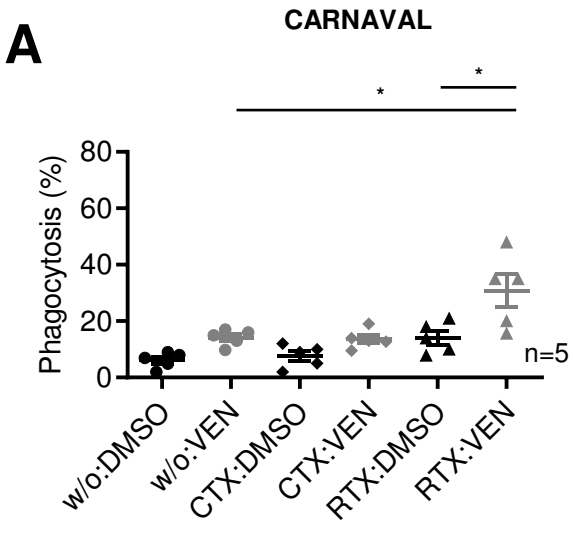


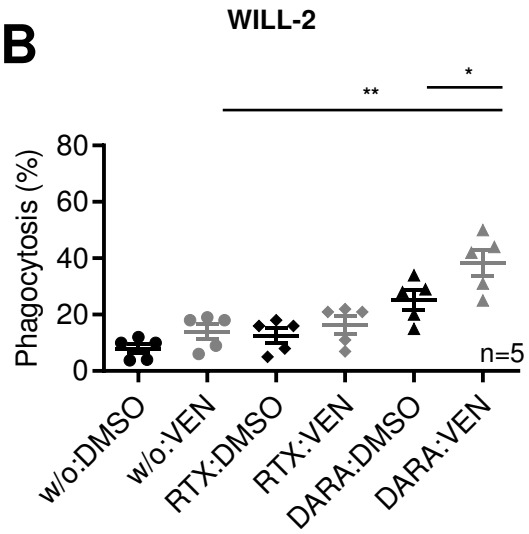
Vogiatzi et al., 2022 Supplementary Figure 3

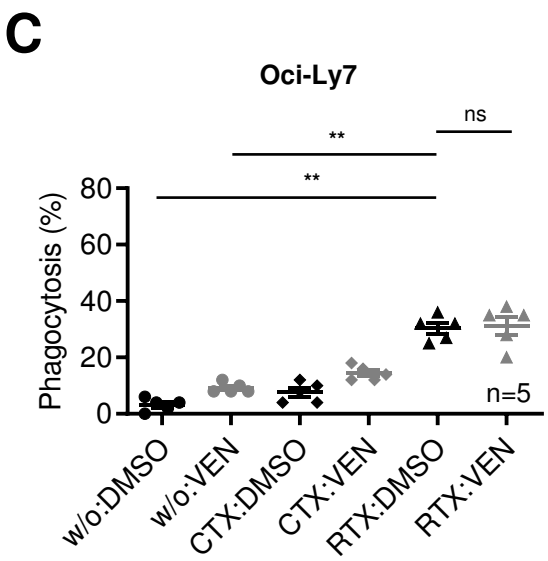


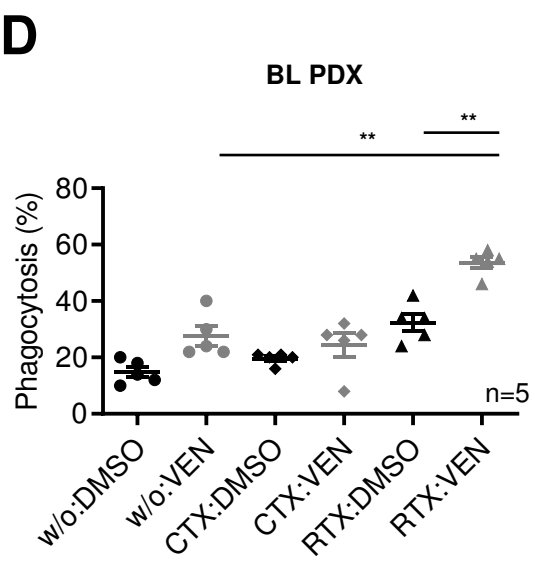
Vogiatzi et al., 2022 Supplementary Figure 4

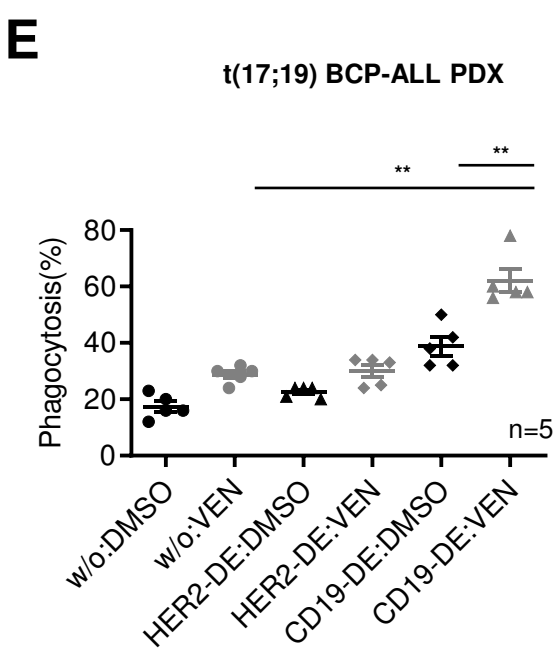


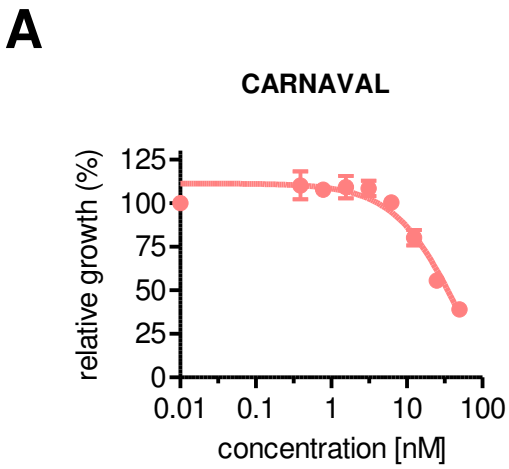

A
CARNAVAL
Phagocytosis (%)
n=5
w/o:DMSO
w/o:VEN
CTX:DMSO
CTX:VEN
RTX:DMSO
RTX:VEN

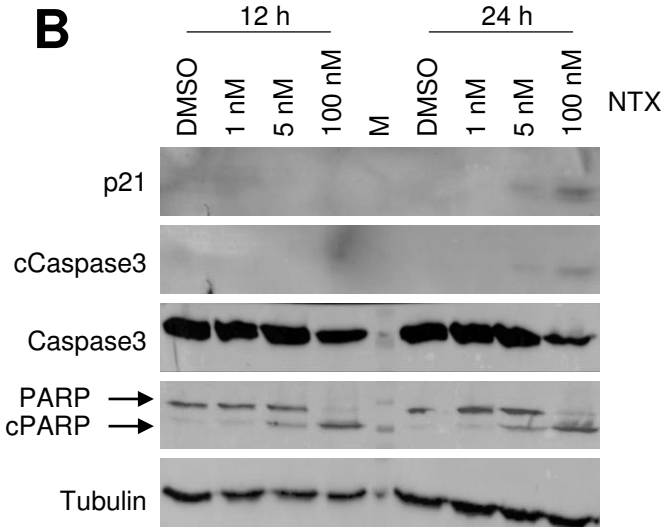

B
WILL-2
Phagocytosis (%)
n=5
w/o:DMSO
w/o:VEN
RTX:DMSO
RTX:VEN
DARA:DMSO
DARA:VEN

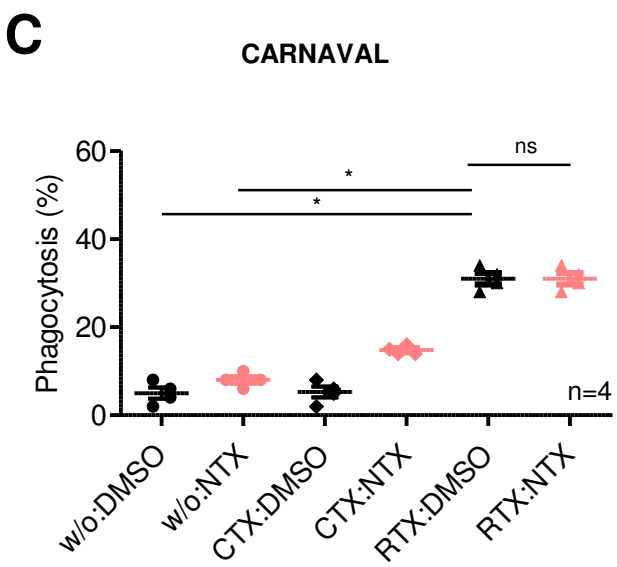

C
Oci-Ly7
Phagocytosis (%)
n=5
w/o:DMSO
w/o:VEN
CTX:DMSO
CTX:VEN
RTX:DMSO
RTX:VEN

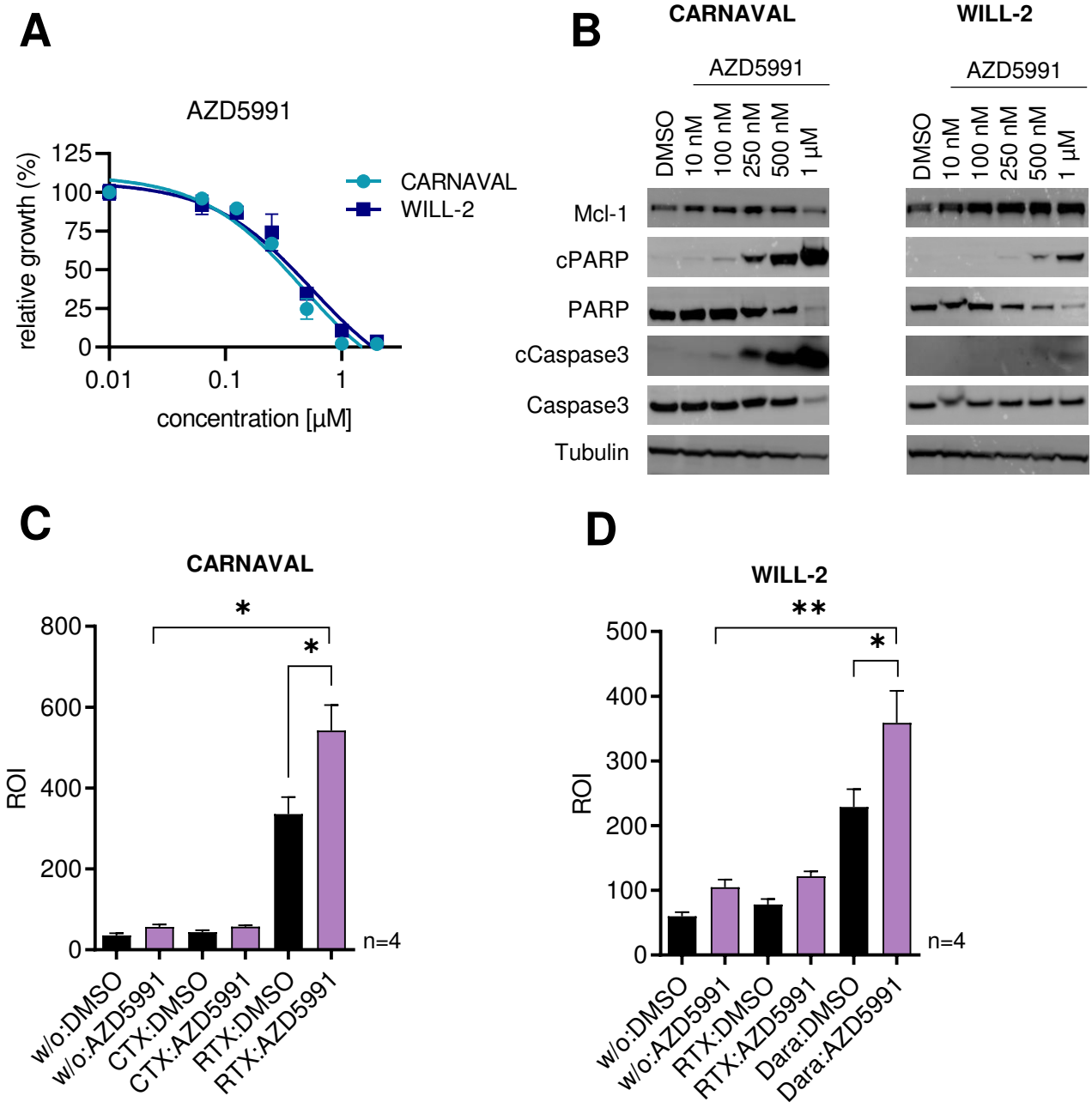

D
BL PDX
Phagocytosis (%)
n=5
w/o:DMSO
w/o:VEN
CTX:DMSO
CTX:VEN
RTX:DMSO
RTX:VEN


E
t(17;19) BCP-ALL PDX
Phagocytosis(%)
n=5
w/o:DMSO
w/o:VEN
HER2-DE:DMSO
HER2-DE:VEN
CD19-DE:DMSO
CD19-DE:VEN

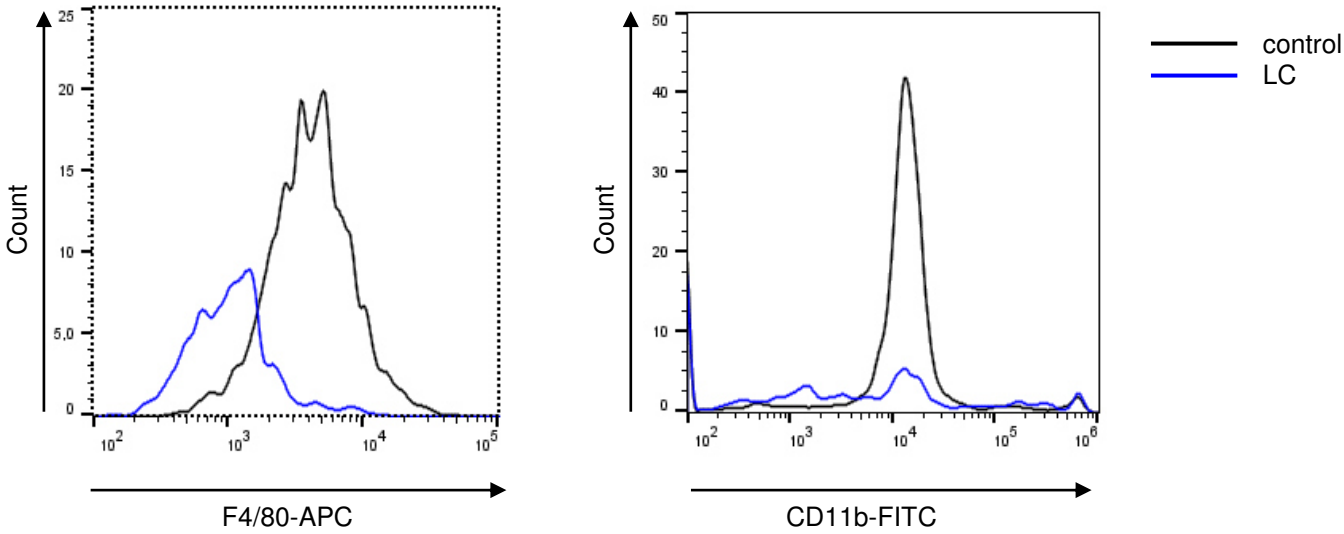

A
CARNAVAL
relative growth (%)
125
100
75
50
25
0
0.01
0.1
1
10
100
concentration [nM]

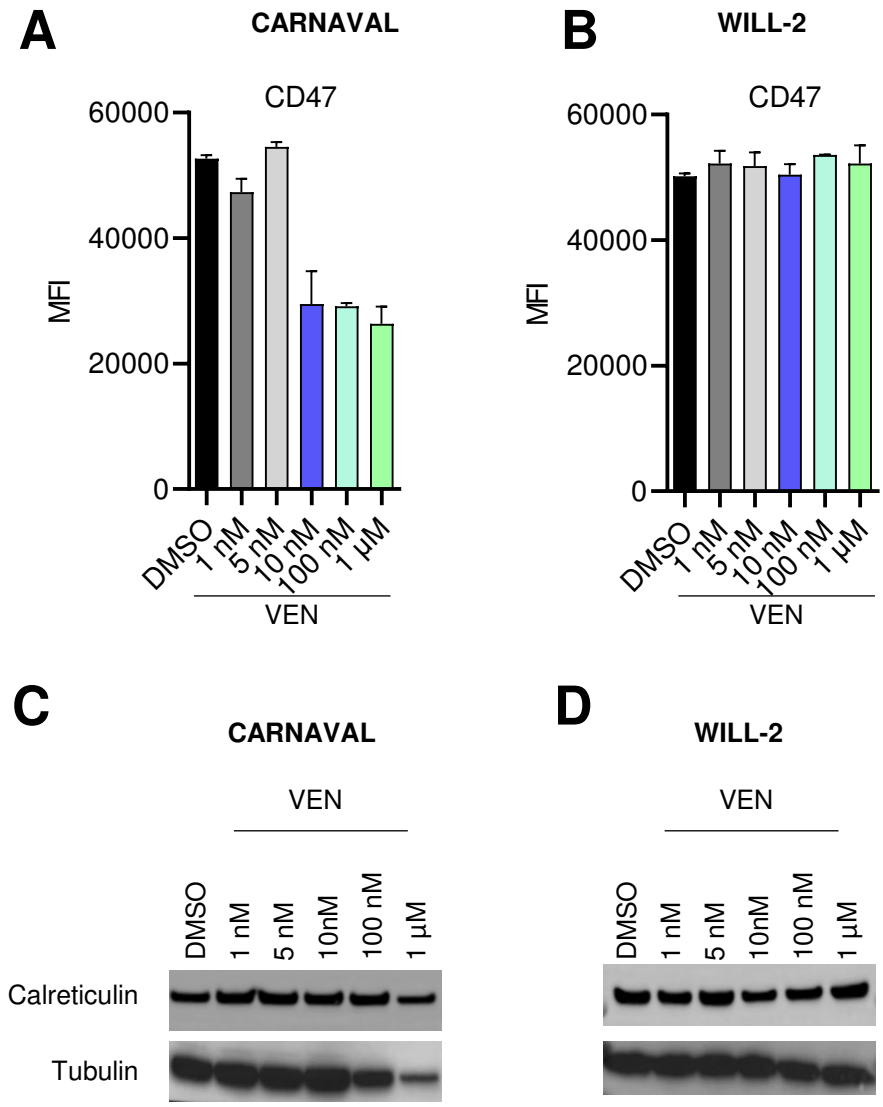

B
12 h
24 h
DMSO
1 nM
5 nM
100 nM
M
DMSO
1 nM
5 nM
100 nM
NTX
p21
cCaspase3
Caspase3
PARP
cPARP
Tubulin


C
CARNAVAL
Phagocytosis (%)
60
40
20
0
*
*
ns
n=4
w/o:DMSO
w/o:NTX
CTX:DMSO
CTX:NTX
RTX:DMSO
RTX:NTX

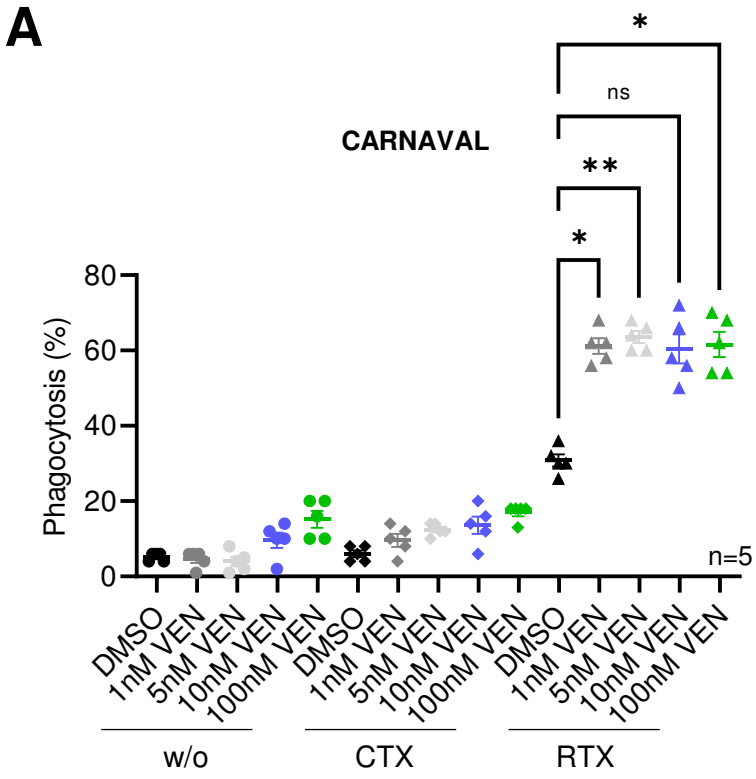

A
AZD5991
relative growth (%)
125
100
75
50
25
0
0.01
0.1
1
concentration [µM]
CARNAVAL
WILL-2
B
CARNAVAL
WILL-2
AZD5991
AZD5991
DMSO
10 nM
100 nM
250 nM
500 nM
1 µM
Mcl-1
cPARP
PARP
cCaspase3
Caspase3
Tubulin
C
CARNAVAL
ROI
800
600
400
200
0
*
*
n=4
w/o:DMSO
w/o:AZD5991
CTX:DMSO
CTX:AZD5991
RTX:DMSO
RTX:AZD5991
D
WILL-2
ROI
500
400
300
200
100
0
**
*
n=4
w/o:DMSO
w/o:AZD5991
RTX:DMSO
RTX:AZD5991
Dara:DMSO
Dara:AZD5991

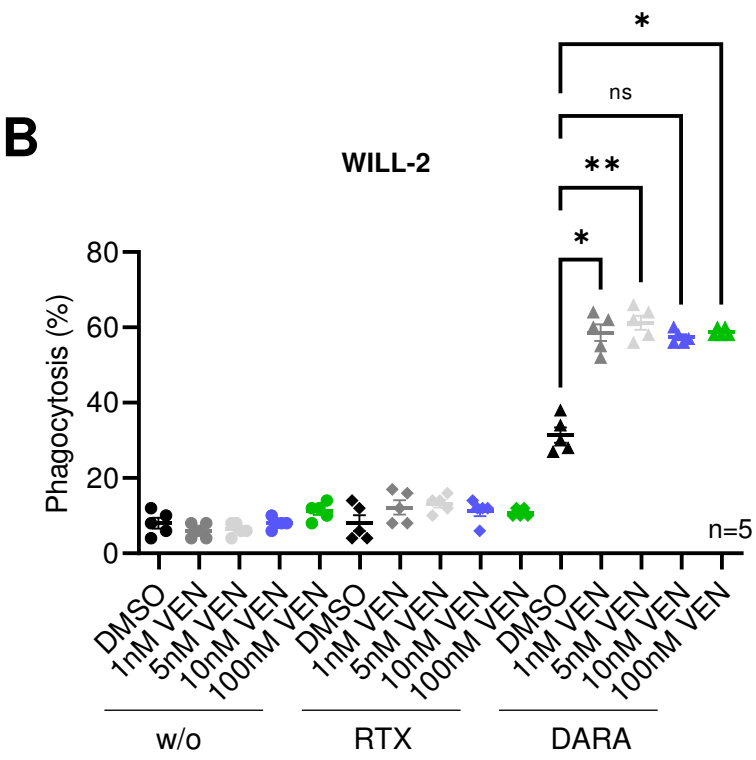
Vogiatzi et al., 2022 Supplementary Figure 8

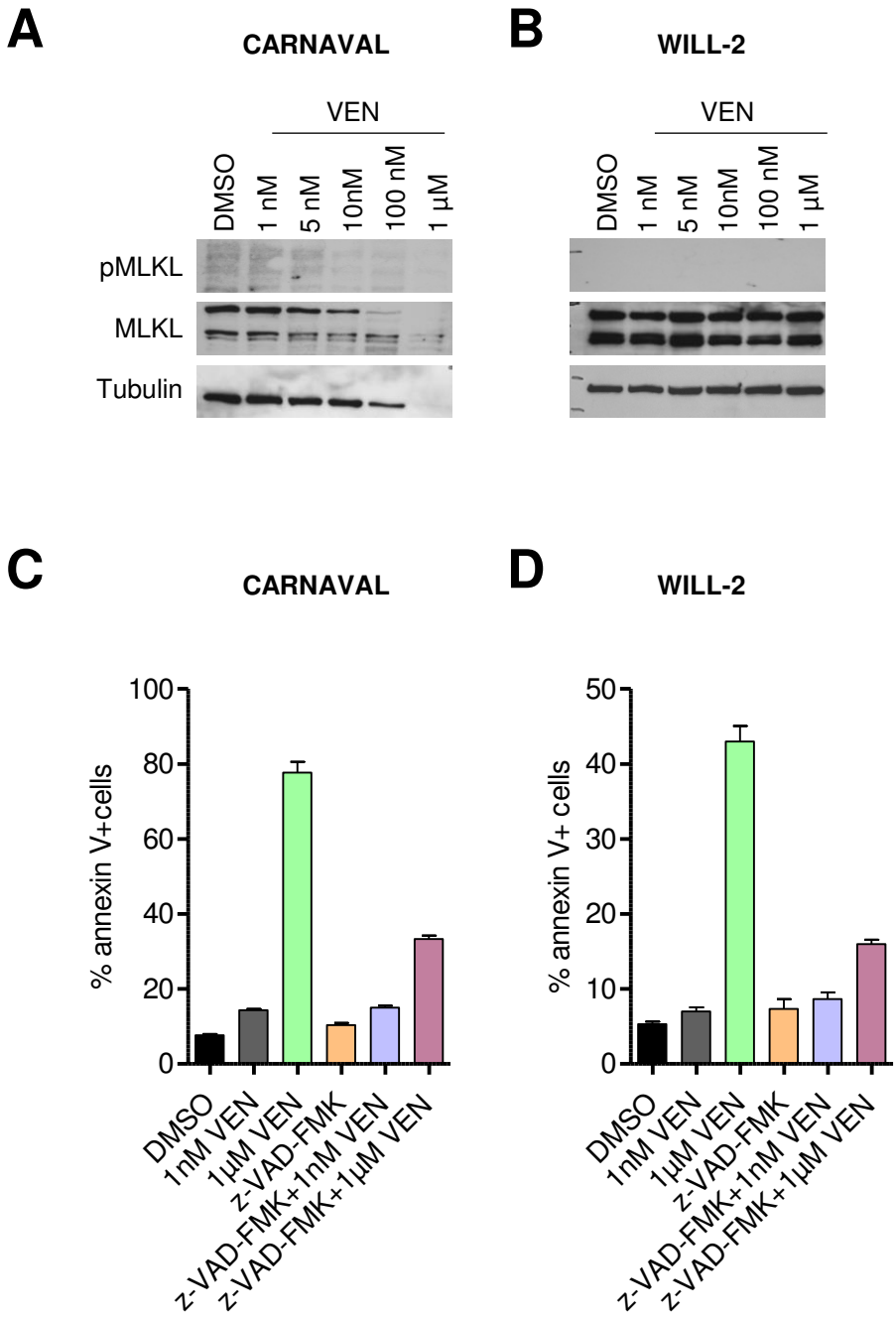



A
CARNAVAL
CD47
MFI
60000
40000
20000
0
DMSO
1 nM
5 nM
10 nM
100 nM
1 µM
VEN
B
WILL-2
CD47
MFI
60000
40000
20000
0
DMSO
1 nM
5 nM
10 nM
100 nM
1 µM
VEN

C
CARNAVAL
VEN
DMSO
1 nM
5 nM
10nM
100 nM
1 µM
Calreticulin
Tubulin
D
WILL-2
VEN
DMSO
1 nM
5 nM
10nM
100 nM
1 µM


A
CARNAVAL
*
ns
**
*
Phagocytosis (%)
80
60
40
20
0
n=5
DMSO
1nM VEN
5nM VEN
10nM VEN
100nM VEN
DMSO
1nM VEN
5nM VEN
10nM VEN
100nM VEN
DMSO
1nM VEN
5nM VEN
10nM VEN
100nM VEN
w/o
CTX
RTX


B
WILL-2
*
ns
**
*
Phagocytosis (%)
80
60
40
20
0
n=5
DMSO
1nM VEN
5nM VEN
10nM VEN
100nM VEN
DMSO
1nM VEN
5nM VEN
10nM VEN
100nM VEN
DMSO
1nM VEN
5nM VEN
10nM VEN
100nM VEN
w/o
RTX
DARA


A
CARNAVAL
VEN
DMSO
1 nM
5 nM
10nM
100 nM
1 µM
pMLKL
MLKL
Tubulin
B
WILL-2
VEN
DMSO
1 nM
5 nM
10nM
100 nM
1 µM
C
CARNAVAL
% annexin V+cells
100
80
60
40
20
0
DMSO
1nM VEN
1µM VEN
z-VAD-FMK
z-VAD-FMK+1nM VEN
z-VAD-FMK+1µM VEN
D
WILL-2
% annexin V+ cells
50
40
30
20
10
0
DMSO
1nM VEN
1µM VEN
z-VAD-FMK
z-VAD-FMK+1nM VEN
z-VAD-FMK+1µM VEN

Supplementary Figure Legends

Supplementary Figure 1: Regulation of cell cycle upon VEN treatment in DHL cells

A-C, Determination of EC50 in CARNAVAL (A), WILL-2 (B) and Oci-Ly7 (C) cells using increasing concentrations of venetoclax (VEN). D-F, *Trypan blue* exclusion test of cell viability in CARNAVAL (D), WILL-2 (E) and Oci-Ly7 (F) cells treated with increasing concentrations of VEN for up to 48 h. Technical triplicates were counted per experiment. Graphs show results of three independent experiments (n=3, SD). G-H, Protein expression of pERK (Tyr204/Tyr187), ERK, pRb, Rb, p21, pAkt (Ser473) and AKT in CARNAVAL (G) and WILL-2 (H) cells after treatment with 1 nM, 5 nM, 100 nM VEN or DMSO (solvent control). I-J, Bcl-2 protein levels in 3 different Burkitt-Lymphoma (BL) (I) and t(17;19) BCP-ALL PDX (J) samples after treatment with 1 nM VEN for 12 h or DMSO analyzed by western blot. Tubulin served as a loading control.

Supplementary Figure 2: Interaction of Bcl-2/Bim in DHL cells upon VEN treatment

A-B, Interaction of Bcl-2 and Bim in CARNAVAL (A) and WILL-2 (B) cells after treatment with various concentrations of venetoclax (VEN) or DMSO (solvent control) for 12 h analysed by co-immunoprecipitation.

Supplementary Figure 3: Cell surface expression of CD19, CD20 and CD38 after exposure to VEN

Cell surface expression of CD19, CD20 and CD38 was analysed by flow cytometry after treatment with 1 nM venetoclax (VEN) for 12 h or DMSO (solvent control). (A) CD20 and CD38 expression in CARNAVAL cells, (B) CD20 and CD38 expression in WILL-2 cells, (C) CD20 and CD38 expression in Oci-Ly7 (D) CD20 expression in Burkitt-Lymphoma (BL) PDX, (E) CD19 expression in t(17;19) BCP-ALL PDX. Graphs show results of three independent experiments (n=3, SEM).

Supplementary Figure 4: Mononuclear cell and complement mediated cell killing in DHL cell lines upon VEN treatment

Antibody-dependent cellular cytotoxicity (ADCC) and complement dependent cytotoxicity (CDC) analysed by ^{51}Cr release assay in DHL cell lines treated with venetoclax (VEN). (A) CARNAVAL cells treated with 1 nM VEN or DMSO (solvent control) for 12 h, rituximab (RTX) or cetuximab (CTX; control antibody). (B) WILL-2 cells treated with 1 nM VEN or DMSO for 12 h, daratumumab (DARA) or rituximab (RTX; control antibody).

Supplementary Figure 5: ADCP using murine macrophages upon combination of VEN and therapeutic antibodies in DHL cell lines and PDX samples

ADCP of cells phagocytosed by murine macrophages with venetoclax (VEN)/antibody combinations as compared to VEN, antibody (rituximab, daratumumab, CD19-DE) alone or DMSO (solvent control) in independent experiments. (A) CARNAVAL cells were treated with 1 nM VEN for 12 h, rituximab (RTX) and cetuximab (CTX; control antibody). (B) WILL-2 cells subjected to 1 nM VEN for 12 h, daratumumab (DARA) and rituximab (RTX; control antibody). (C) Oci-Ly7 cell subjected to 1 nM VEN for 12 h, rituximab (RTX) and cetuximab (CTX; control antibody). (D) Burkitt-Lymphoma (BL) PDX sample treated with 1 nM VEN for 12 h, rituximab (RTX) and cetuximab (CTX; control antibody). (E) t(17;19) positive BCP-ALL PDX sample subjected to ADCP assays with 1 nM VEN for 12 h, CD19-DE and HER2-DE as the control antibody.

w/o: no antibody. Phagocytosis was determined as the percentage of macrophages with ingested CFSE green-positive cells per 100 macrophages by at least three independent observers. Each dot represents an independent experiment with different donors. Data are presented as mean ± SEM. Statistical analysis: ns= not significant; *p<0.05; **p<0.005; Mann-Whitney test. All antibodies used *in vitro* were applied to a final concentration of 10 µg/ml unless otherwise stated.

Supplementary Figure 6: ADCP in CARNAVAL cells upon NTX and RTX treatment

(A) Determination of EC50 in CARNAVAL using increasing concentrations of navitoclax (NTX). (B) Protein levels of p21, cCaspase3, Caspase3, cPARP and PARP in CARNAVAL after treatment with various concentrations of NTX for 12 or 24 h or DMSO (solvent control). Tubulin served as a loading control. (C) *In vitro* phagocytosis assay with human macrophages in CARNAVAL cells treated with 50 nM NTX for 12 h or DMSO, rituximab (RTX) and cetuximab (CTX; control antibody). Each dot represents an independent experiment with different human donors (n=4, SEM). Phagocytosis was analyzed as described in the legend to Figure 2. Statistical analysis: *p<0.05; Mann-Whitney test.

Supplementary Figure 7: ADCP in CARNAVAL and WILL-2 after co-treatment with AZD5991 and antibodies

(A) Determination of EC50 in CARNAVAL and WILL-2 cells using escalating concentrations of AZD5991. (B) Protein expression of Mcl-1 and pro-apoptotic markers cCaspase3, Caspase3, cPARP and PARP in CARNAVAL and WILL-2 cells with increasing AZD5991 concentrations. (C) ADCP by using the IncuCyte technology in CARNAVAL and WILL-2 pre-treated with 500 nM AZD5991 or DMSO al solvent control 24 h prior subjection to rituximab (RTX), cetuximab (CTX) or no antibody (w/o). Experiments were performed four times with different human donor macrophages (n=4, SEM); Statistical analysis: *p<0.05; **p<0.005; Mann-Whitney test. ROI: red objects count; Tubulin served as a loading control.

Supplementary Figure 8: Analysis of murine macrophage markers in LC-treated mice

Murine (m) F4-80 and mCD11b determined by flow analysis in bone marrow isolated from mice sacrificed 5 days after treatment intraperitoneally with 100 µl liposomal clodronate (LC) or left untreated (control).

Supplementary Figure 9: Calreticulin and CD47 expression in CARNAVAL and WILL-2 cells with escalating concentrations of VEN

A-B, Surface expression of CD47 analyzed by flow cytometry in CARNAVAL (A) and WILL-2 (B) cells with increasing concentrations of venetoclax (1 nM - 1 µM / 12h). C-D, Protein levels of Calreticulin in CARNAVAL (C) and WILL-2 (D) cells with escalating concentrations of venetoclax (VEN, 1 nM - 1 µM / 12h). MFI: mean fluorescence intensity; Experiments were performed three times (n=3, SEM). Tubulin served as a loading control.

Supplementary Figure 10: ADCP in CARNAVAL and WILL-2 with escalating concentrations of VEN

ADCP of cells phagocytosed by human macrophages with increasing concentrations of venetoclax (VEN) /antibody combinations as compared to VEN or antibody (rituximab, daratumumab) alone in independent experiments. CARNAVAL cells after treatment with 1-100 nM VEN for 12 h and DMSO (solvent control), rituximab (RTX) and the control antibody cetuximab (CTX) (B) WILL-2 cells treated with 1-100 nM VEN for 12 h and DMSO (solvent control), daratumumab (DARA) and the control antibody rituximab (RTX). Phagocytosis was analyzed as described in the legend to Figure 2. Experiments were performed five times with different human donor macrophages (n=5, SEM). Statistical analysis: ns= not significant; *p<0.05; **p<0.005; Mann-Whitney test.

Supplementary Figure 11: A-B, Protein levels of necroptotic markers pMLKL and MLKL in CARNAVAL (A) and WILL-2 (B) cells with escalating concentrations of venetoclax (VEN, 1 nM-1 µM / 12h). C-D, Annexin V staining in CARNAVAL (C) and WILL-2 (D) cells pre-treated with 50 µM Z-VAD FMK for 1h followed by 12h with 1 nM or 1µM venetoclax (VEN); (n=3, SEM)