

Rationale for the combination of venetoclax and ibrutinib in T-prolymphocytic leukemia

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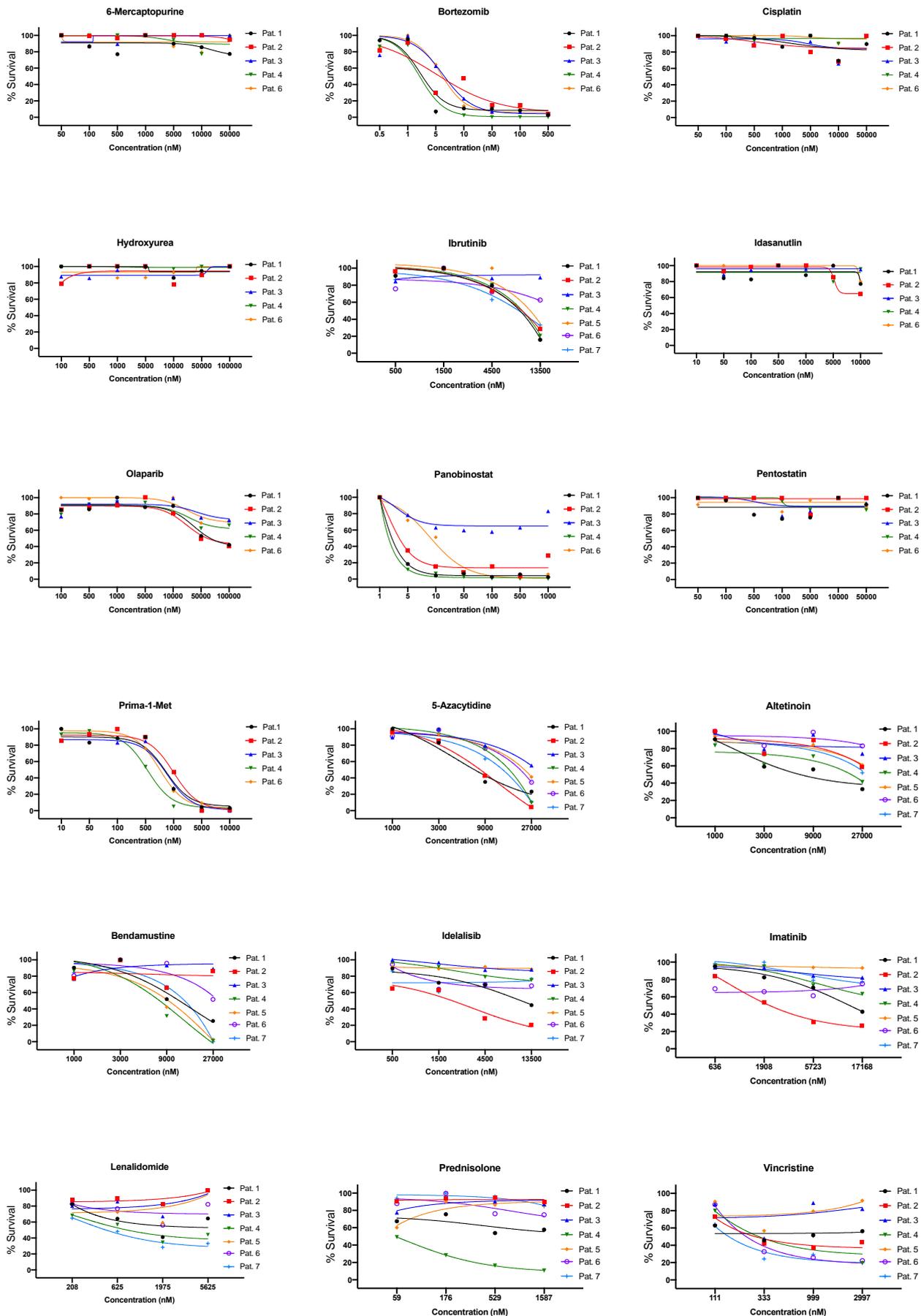
Supplementary Material

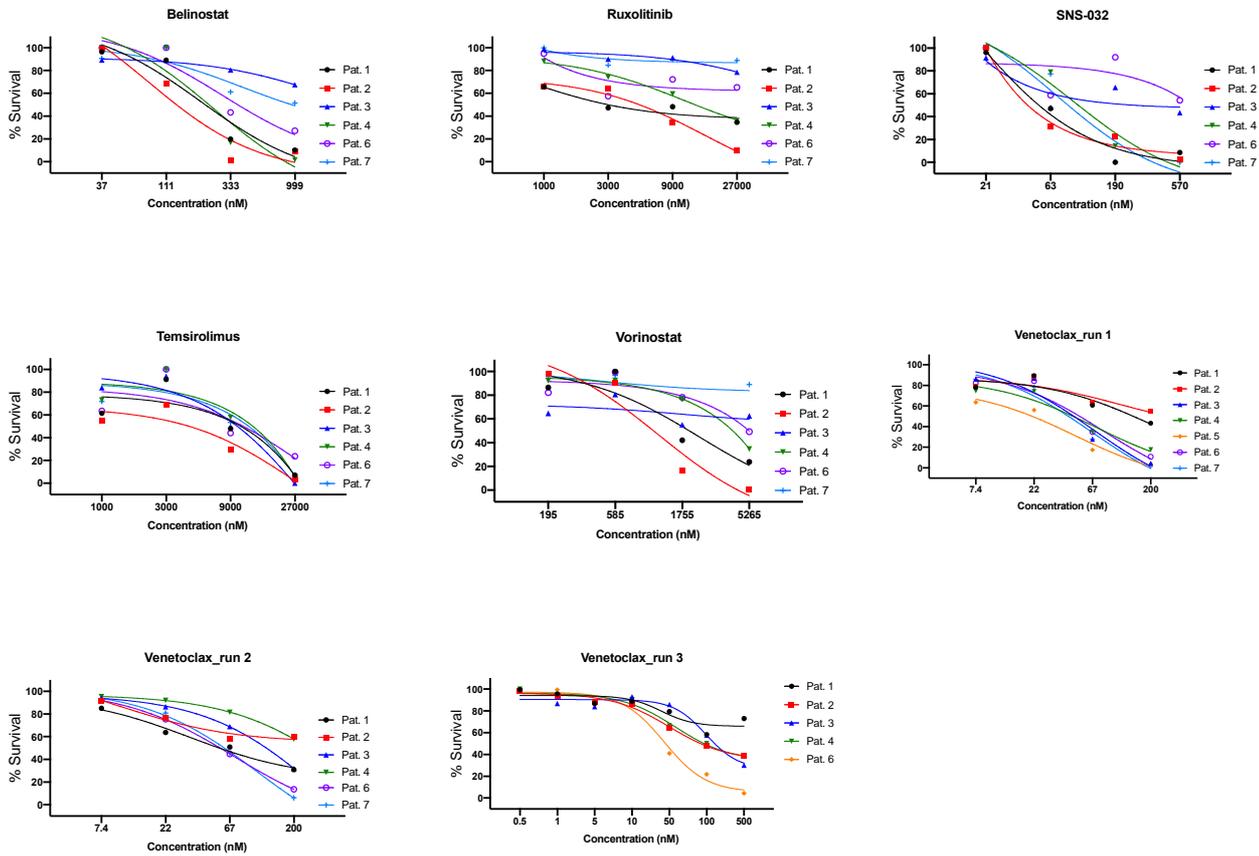
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Supplementary Fig. 1

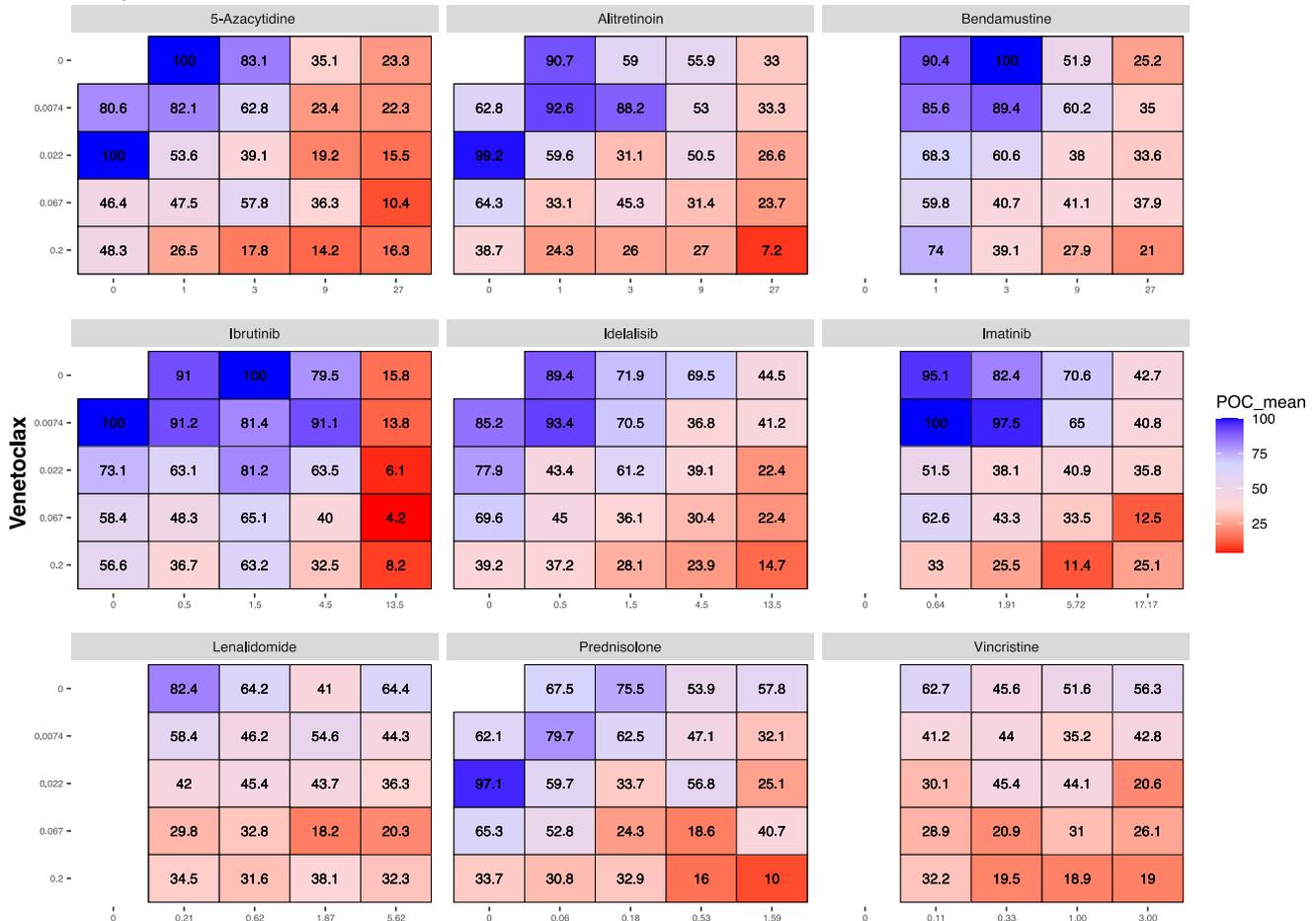
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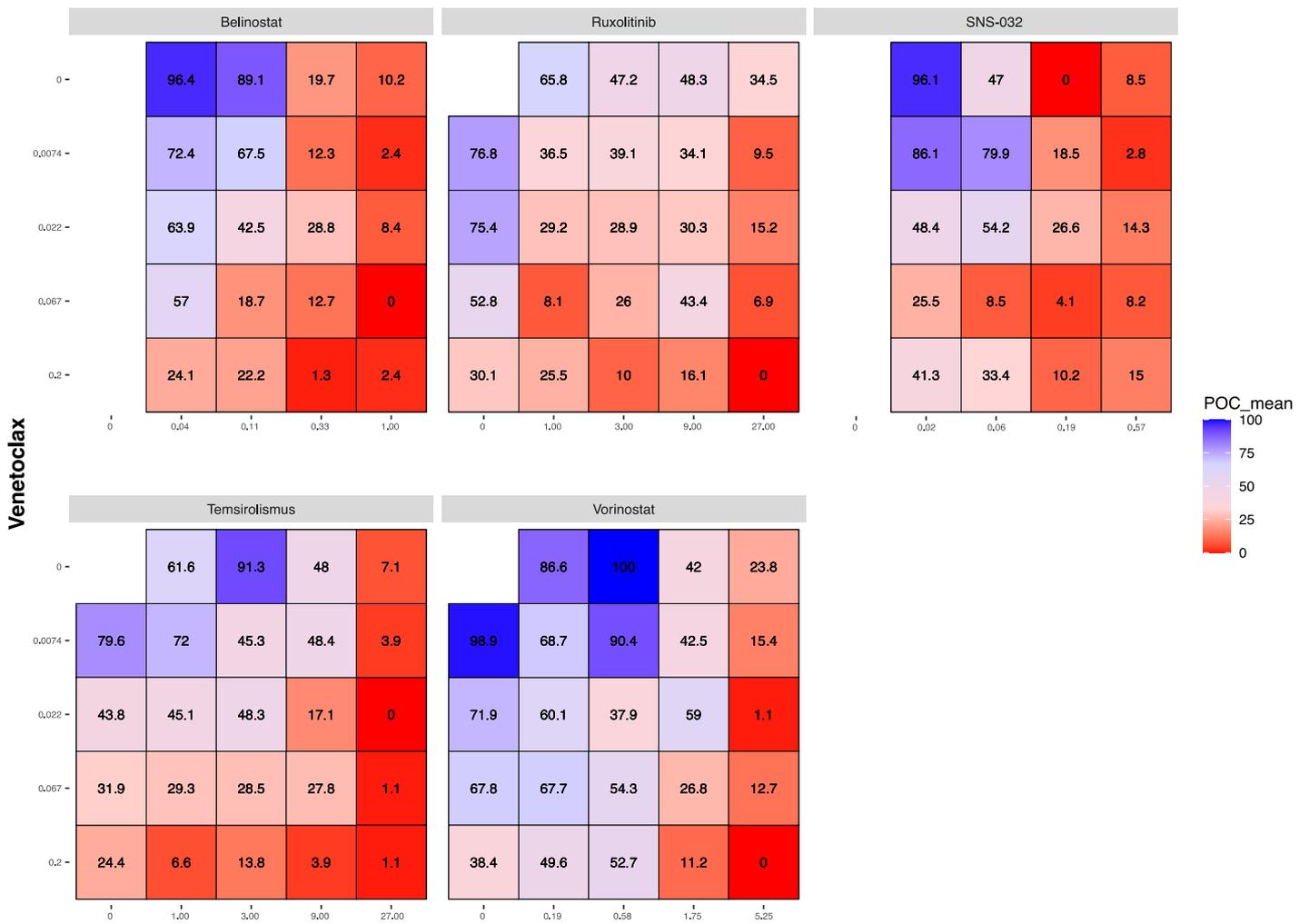


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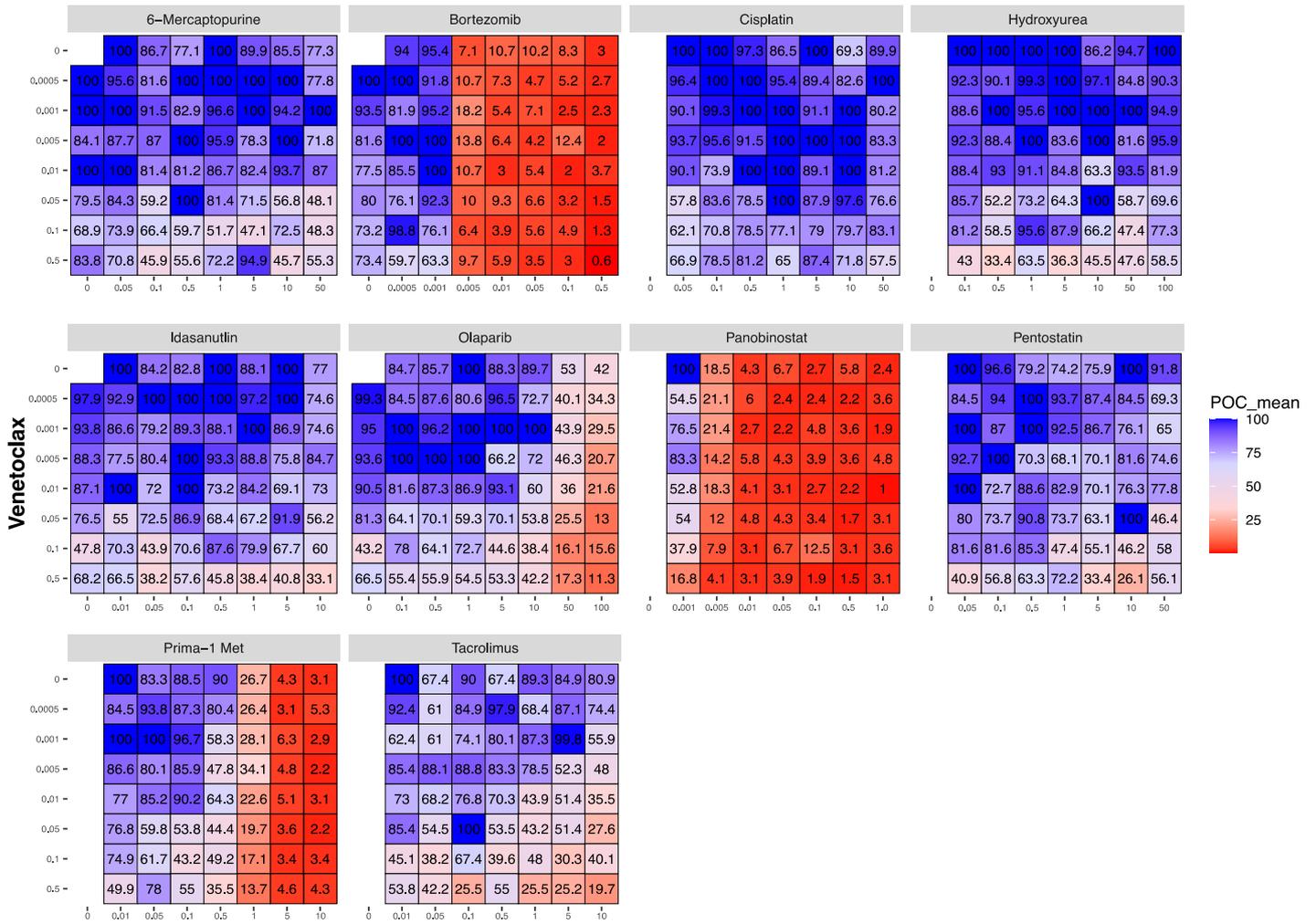
Patient 1, Run1



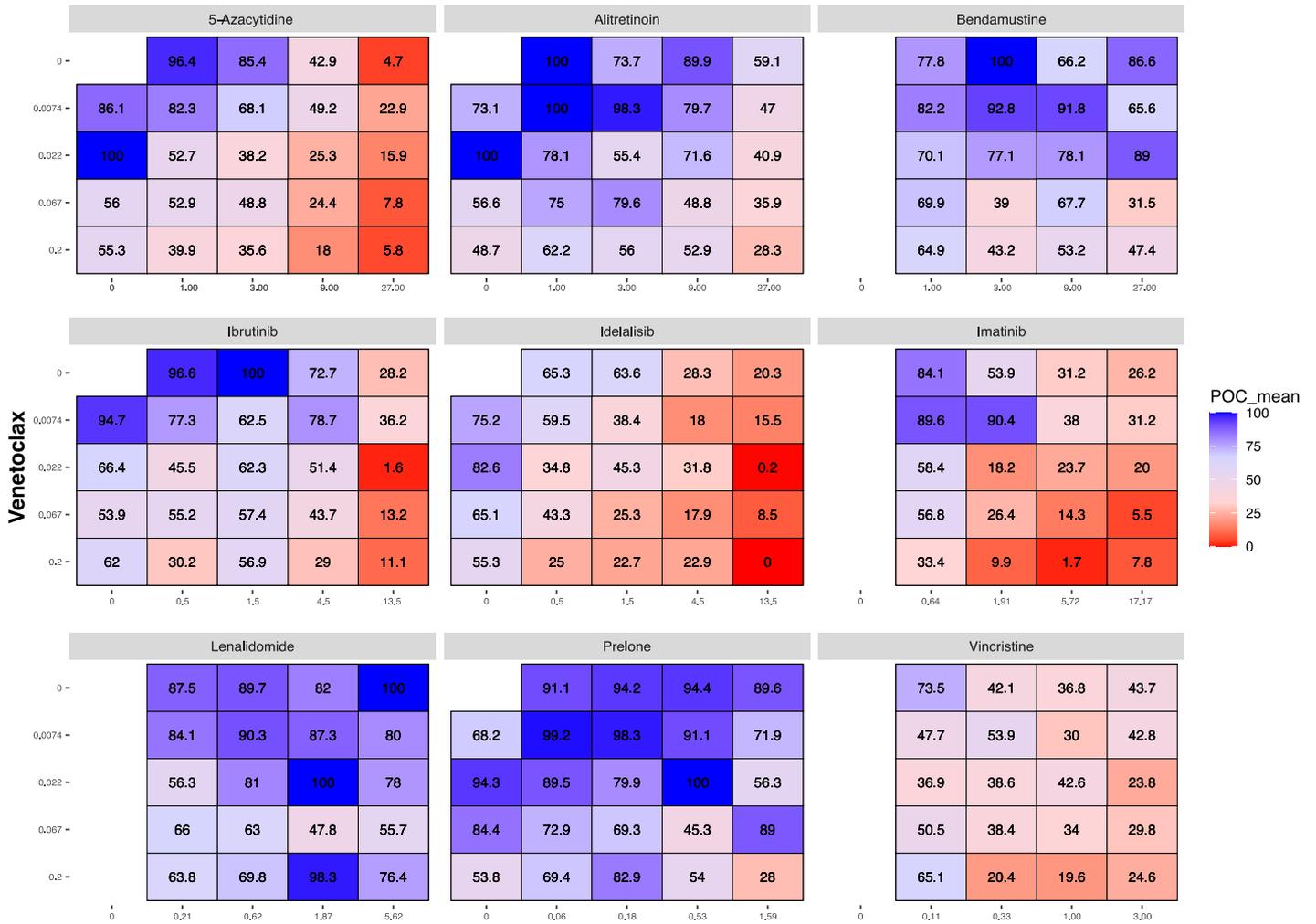
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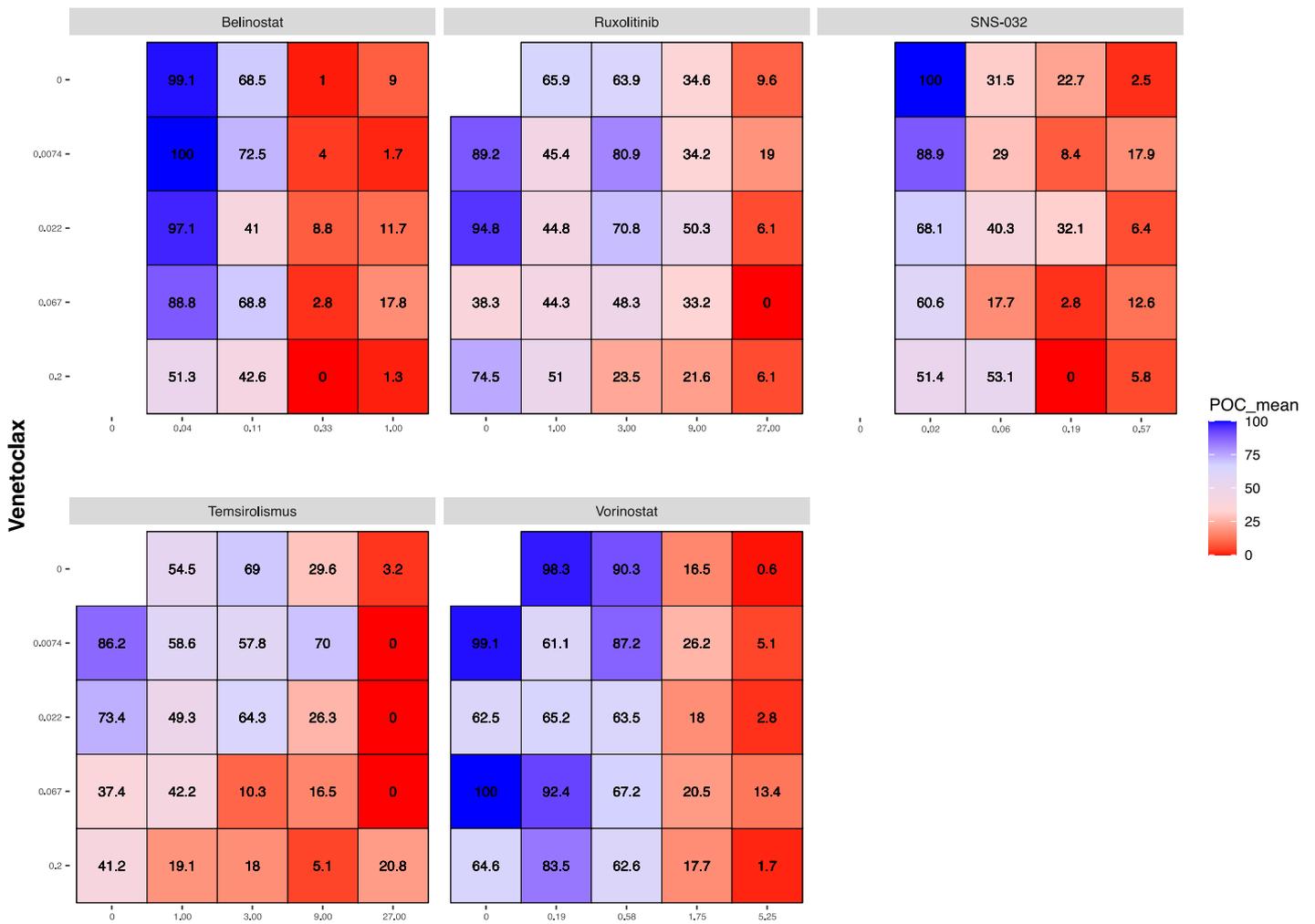
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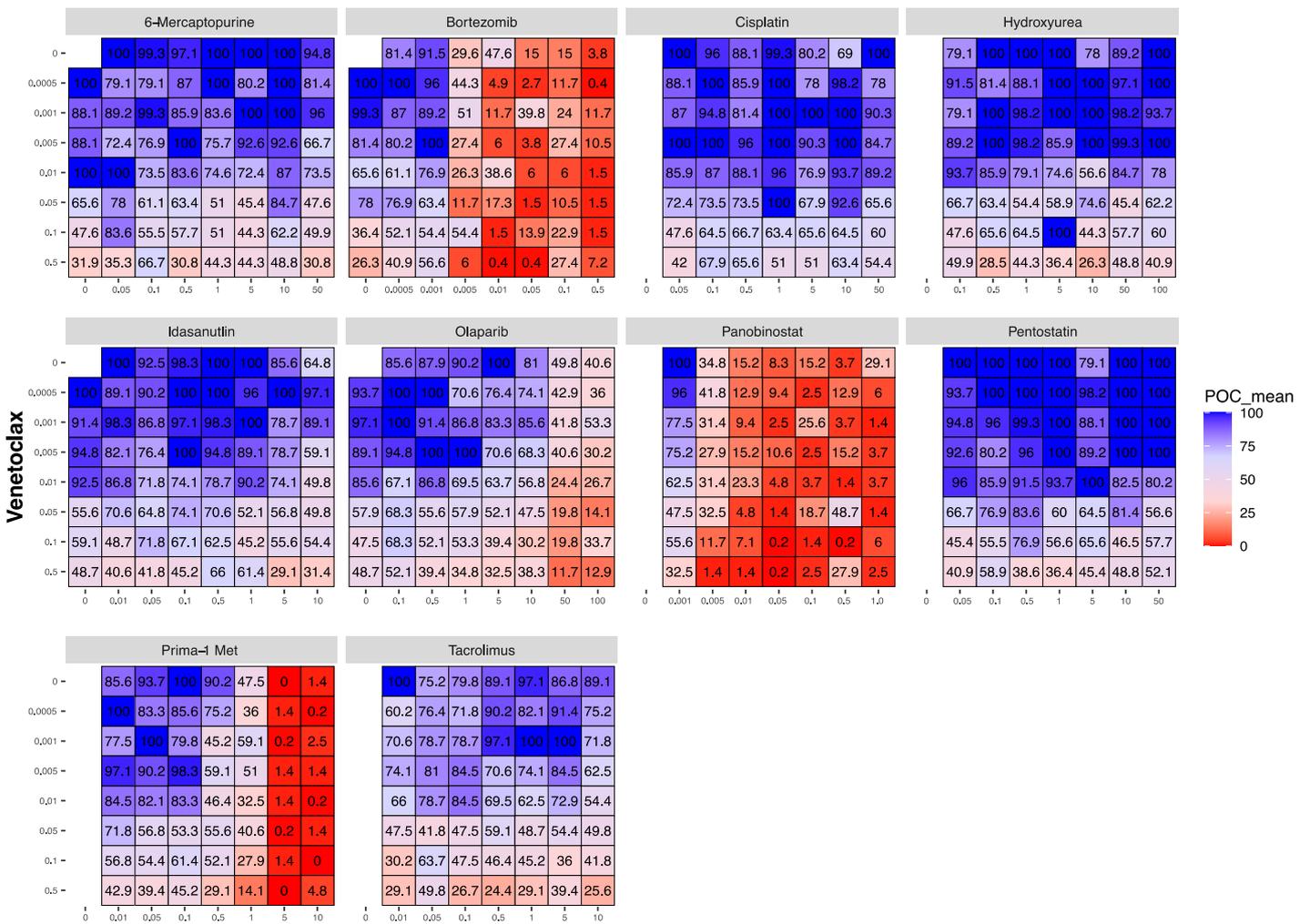
Patient 2 Run 1



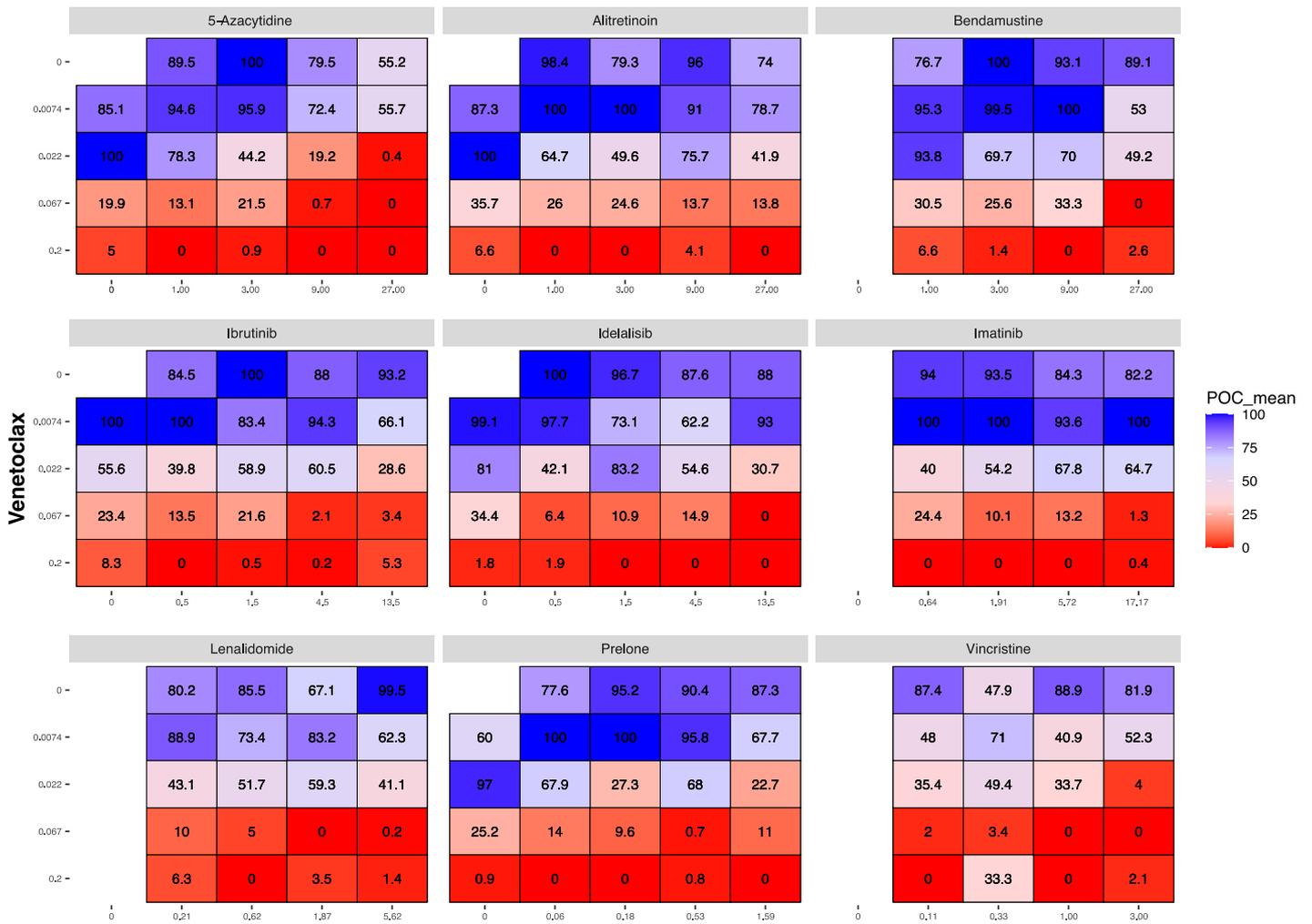
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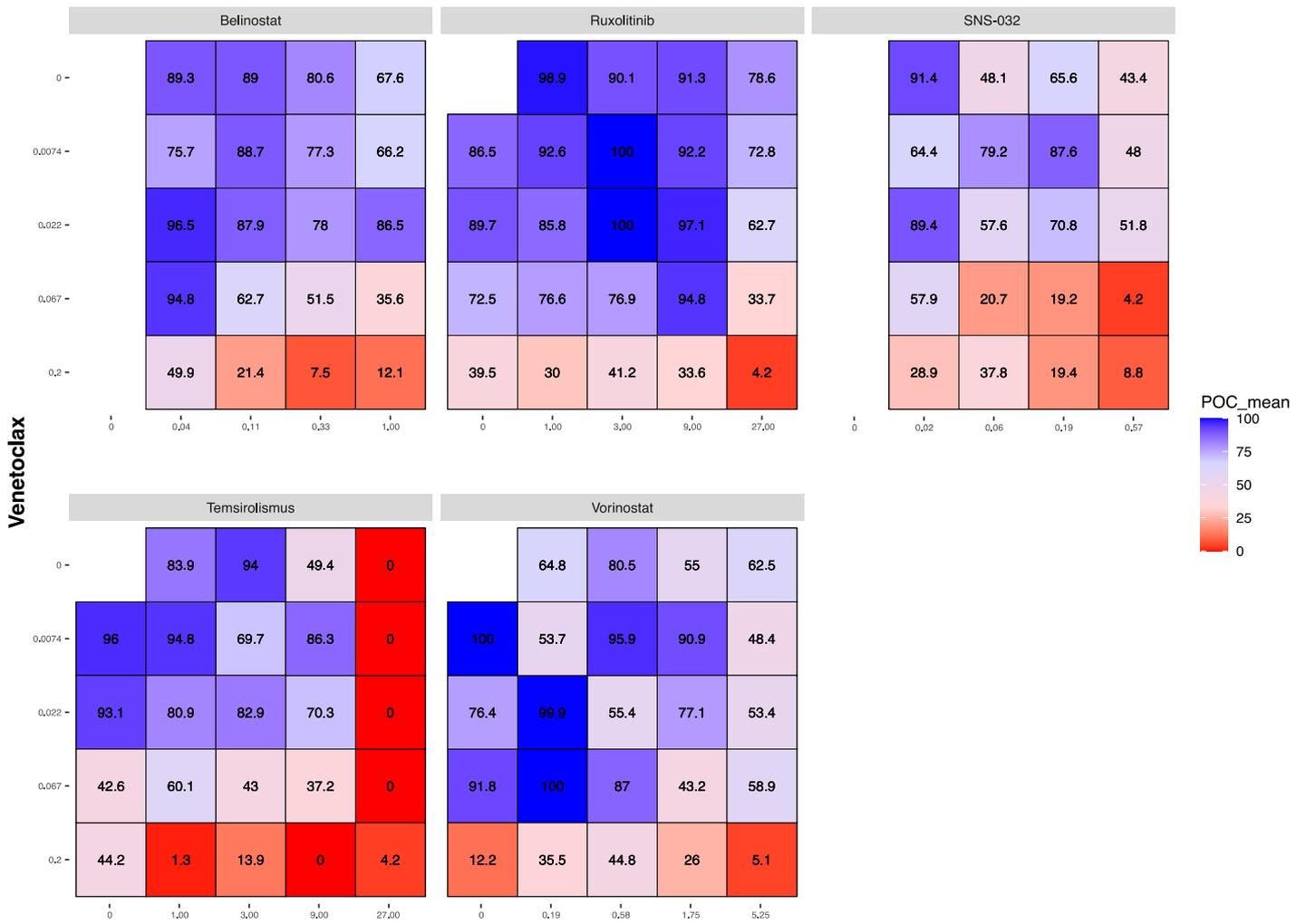
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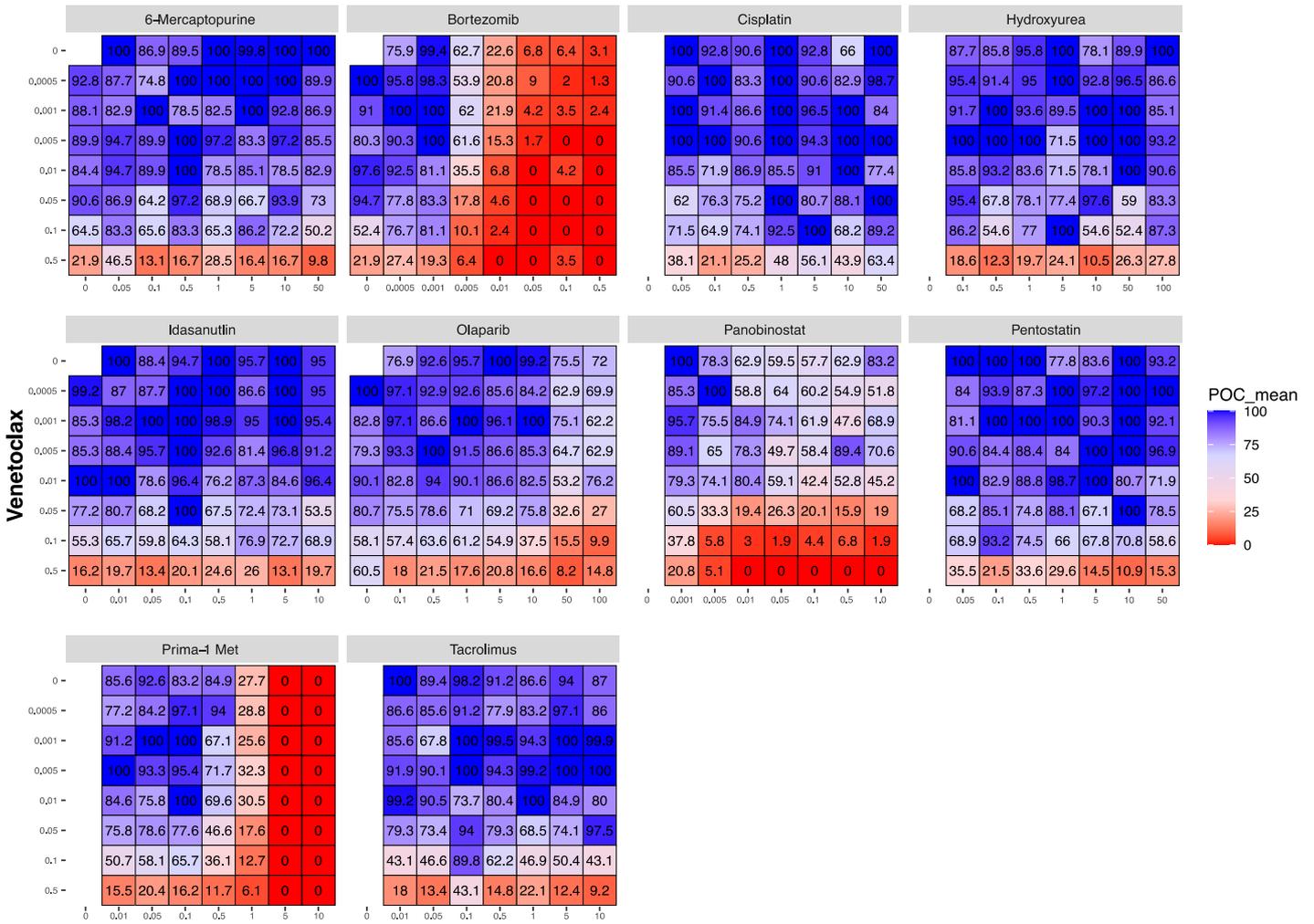
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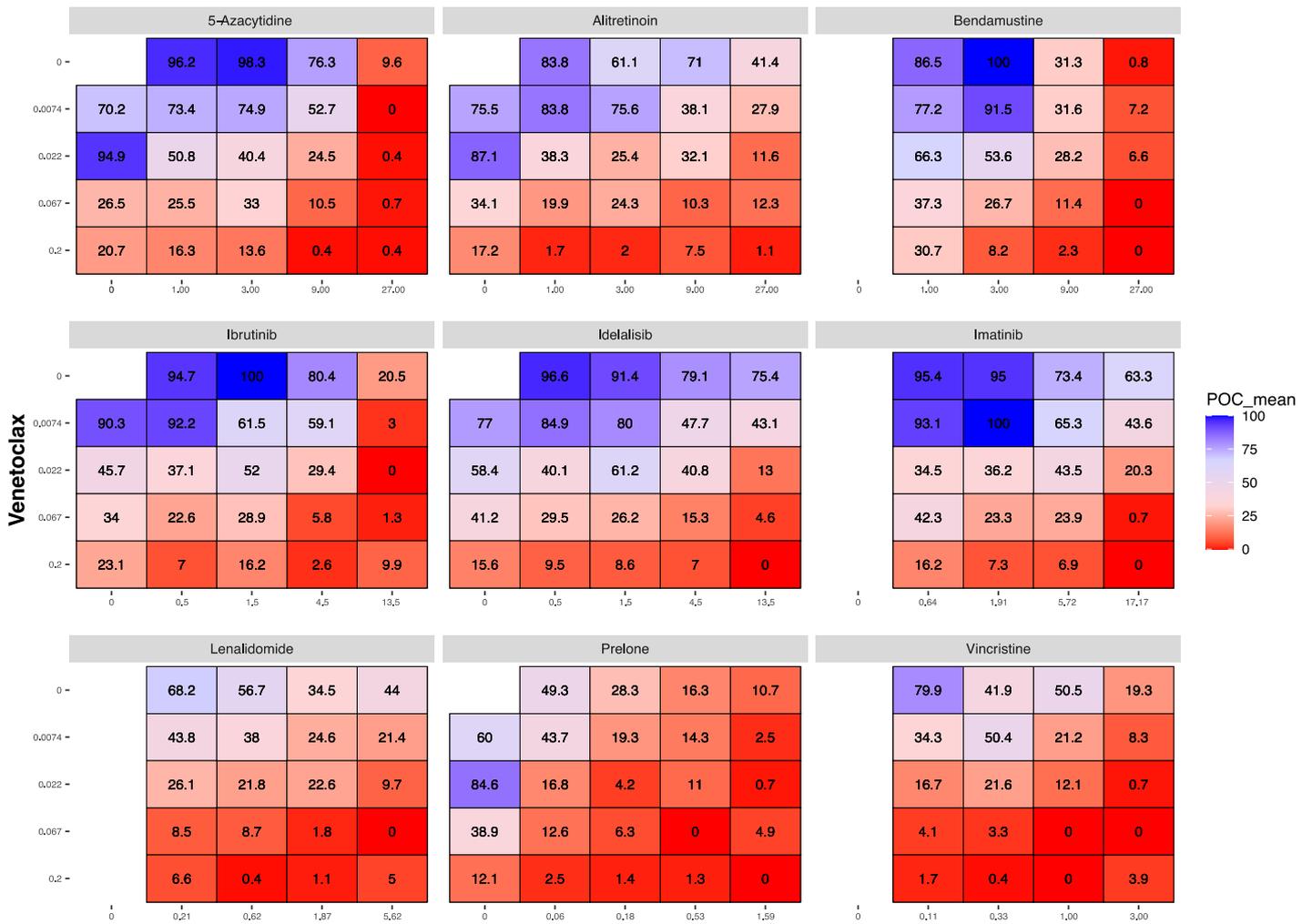
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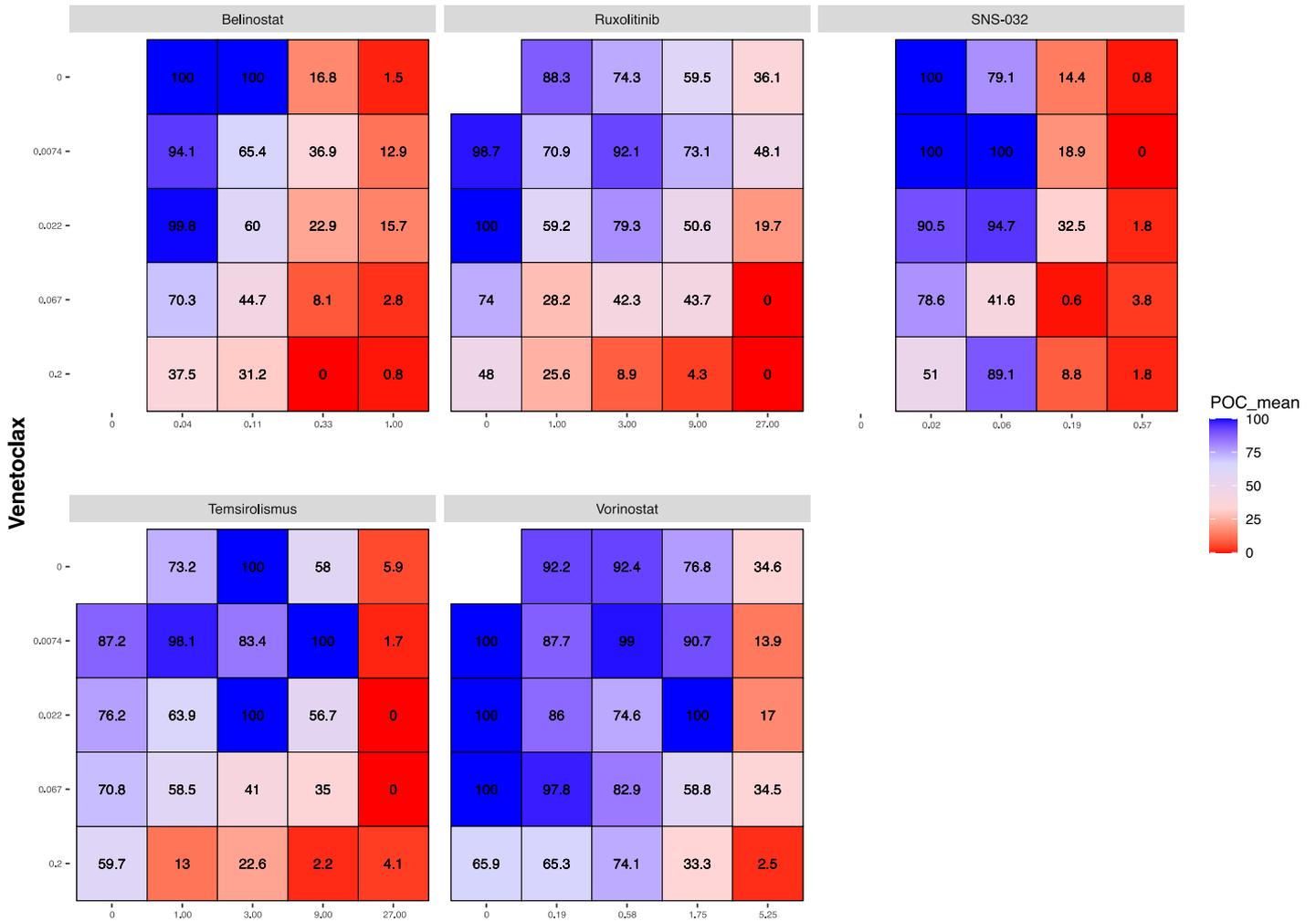
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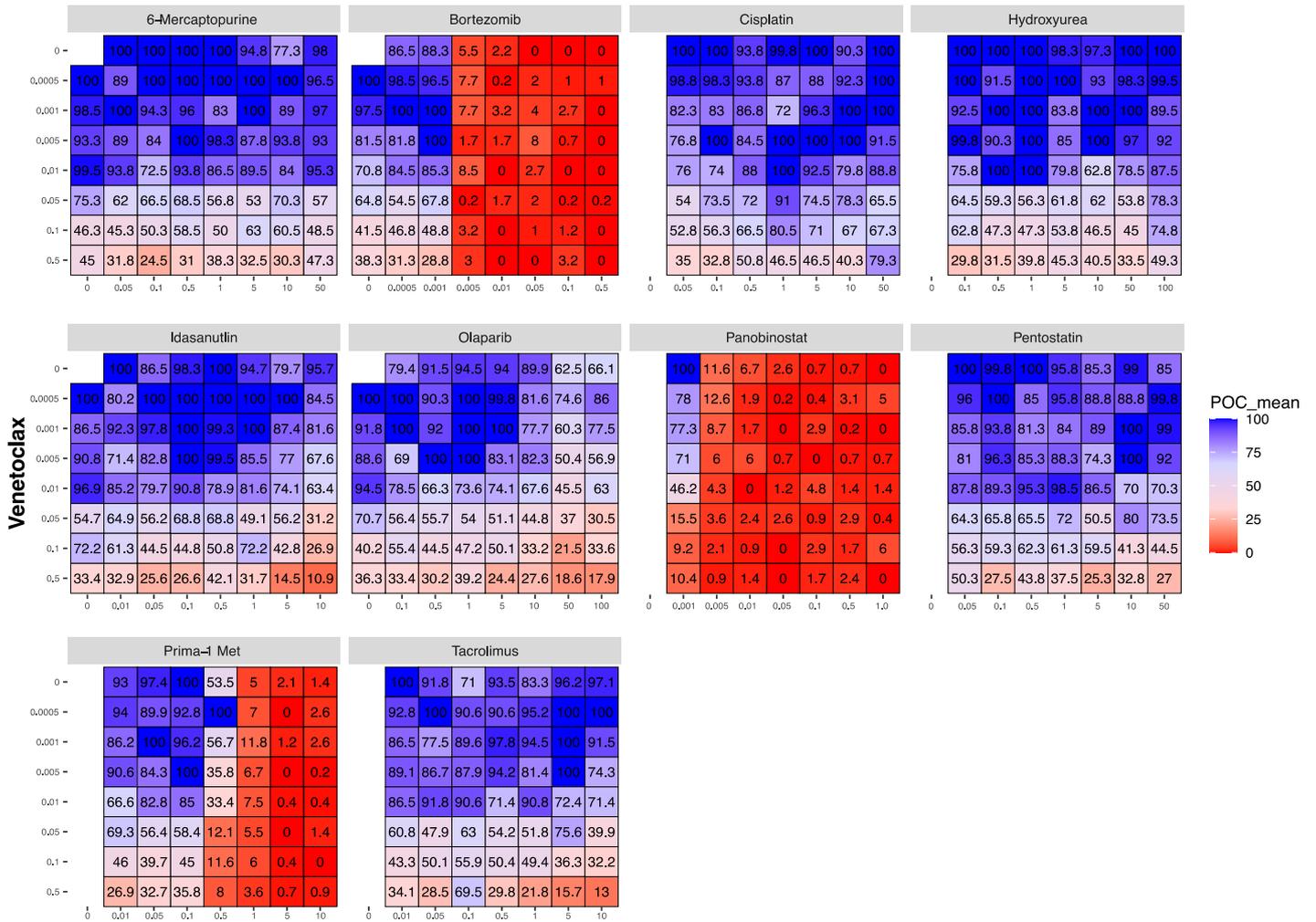
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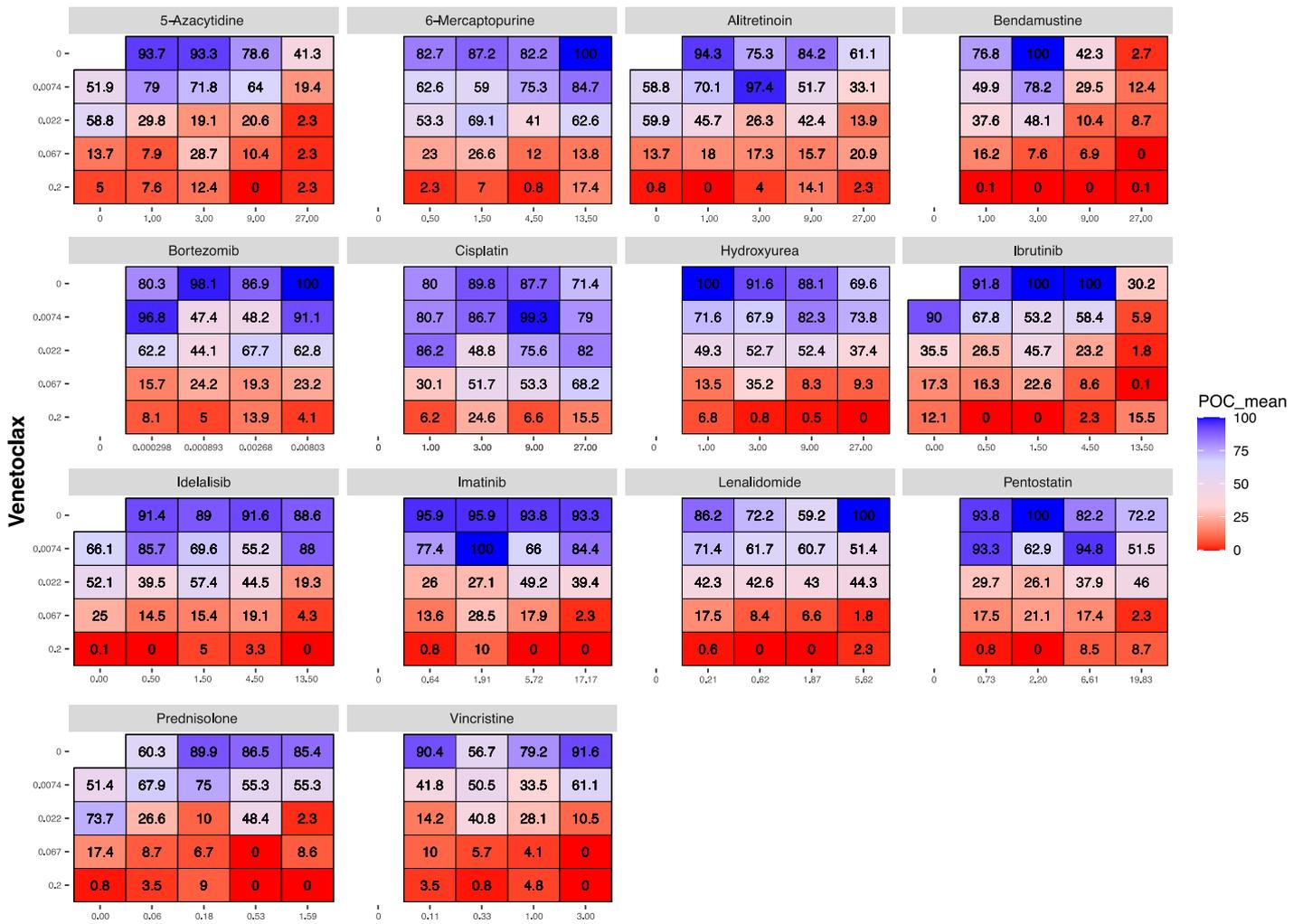
Patient 4 Run 2



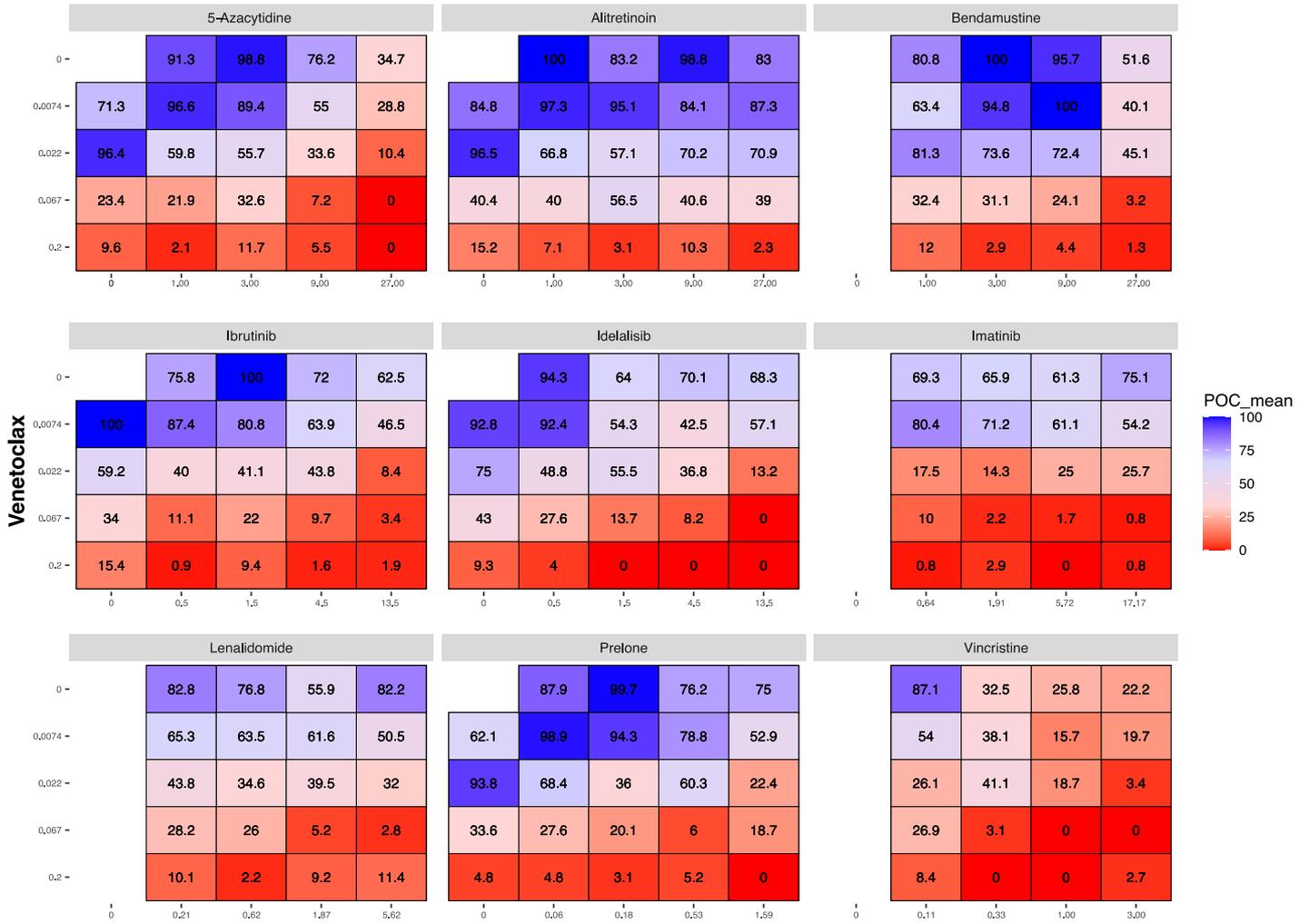
Patient 4 Run 3



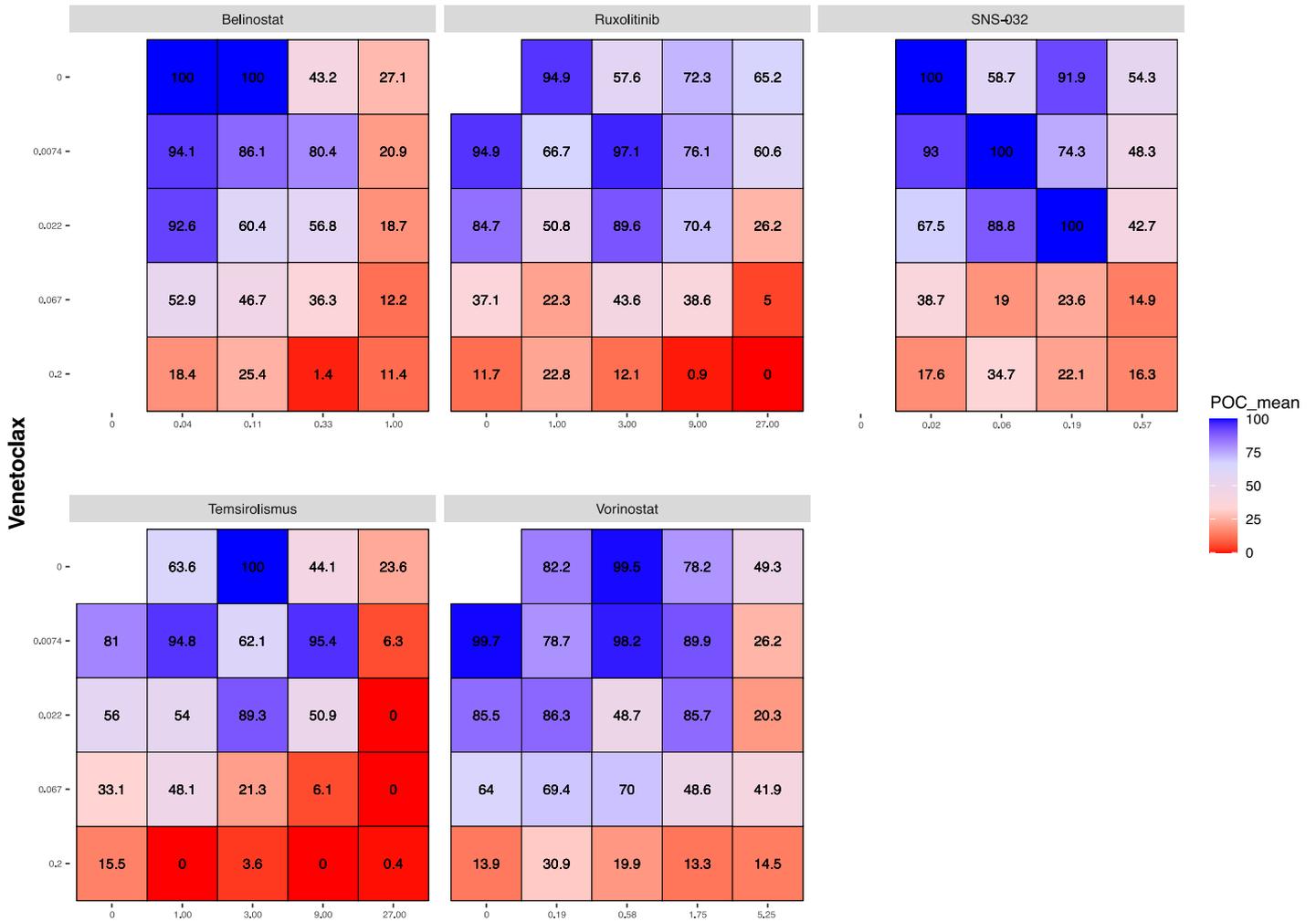
Patient 5 Run 1



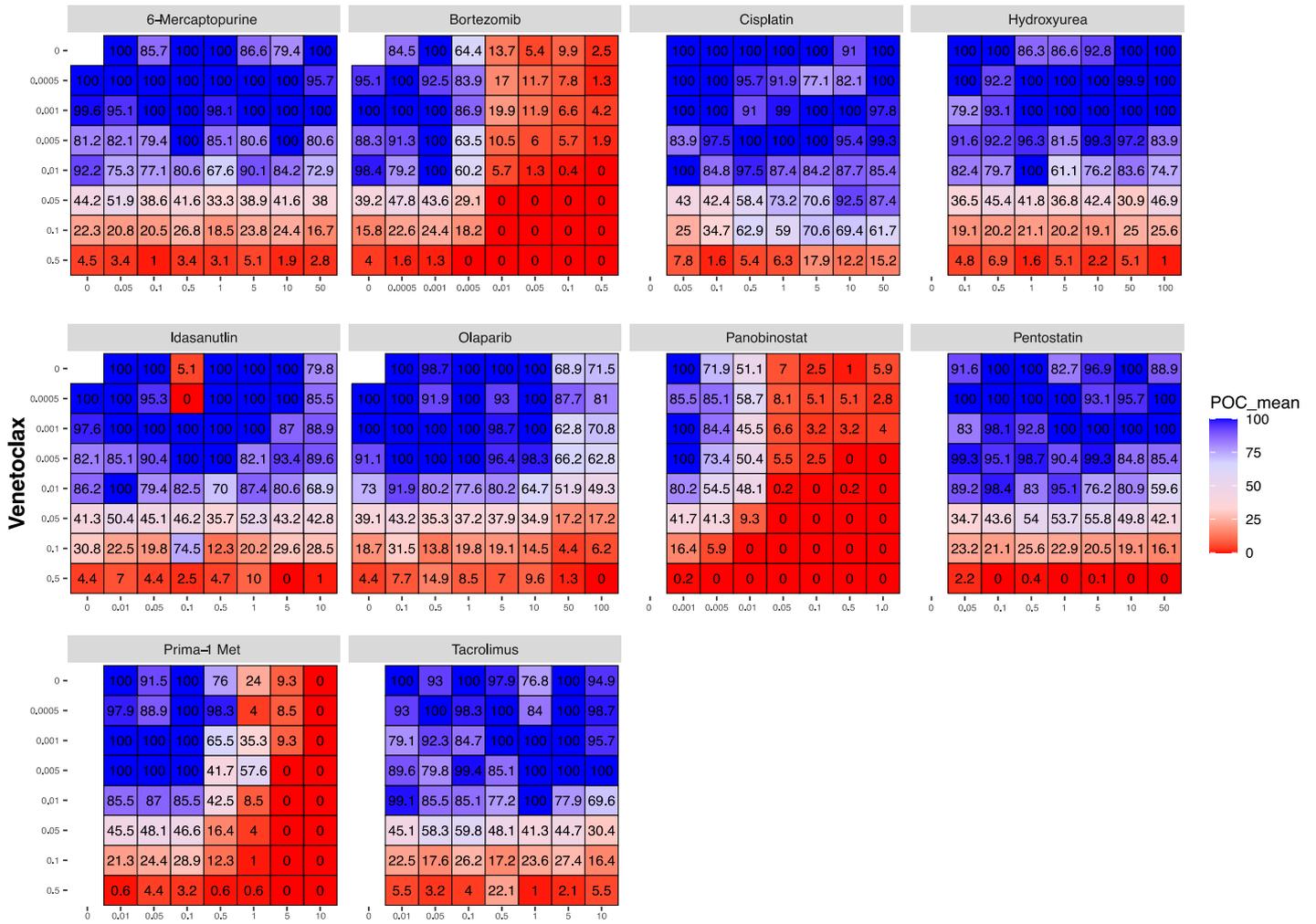
Patient 6 Run 1



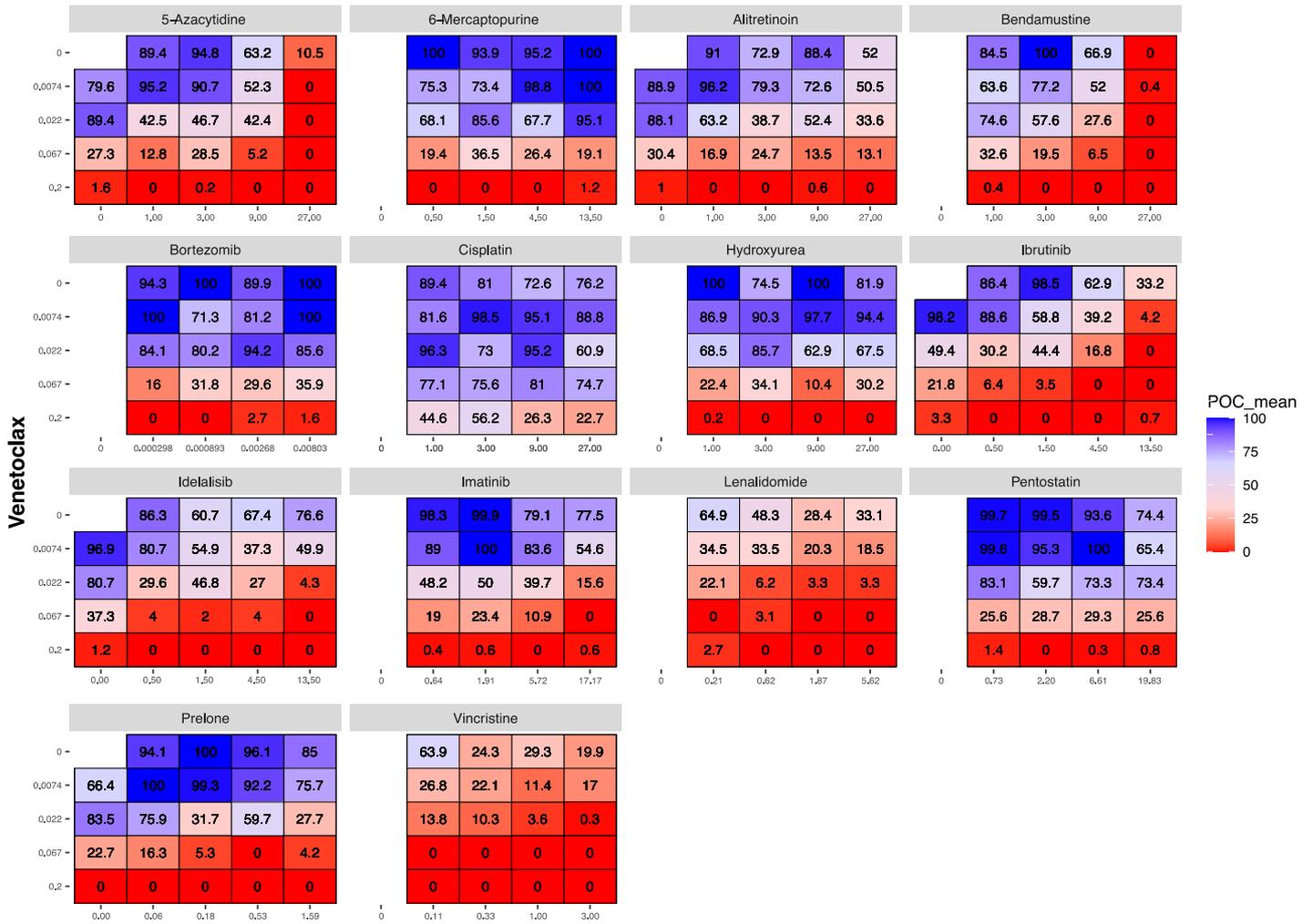
Patient 6 Run 2



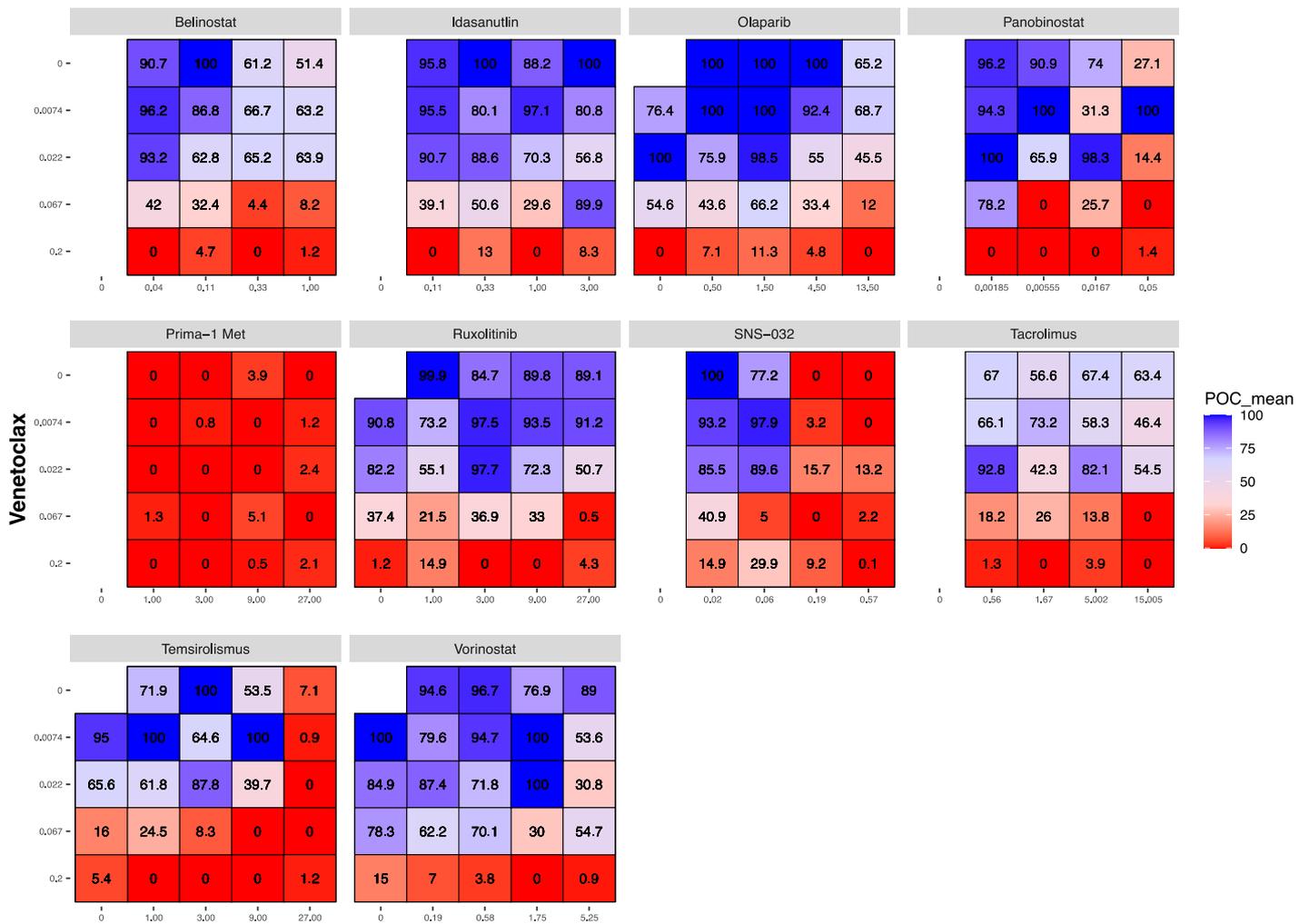
Patient 6 Run 3



Patient 7 Run 1

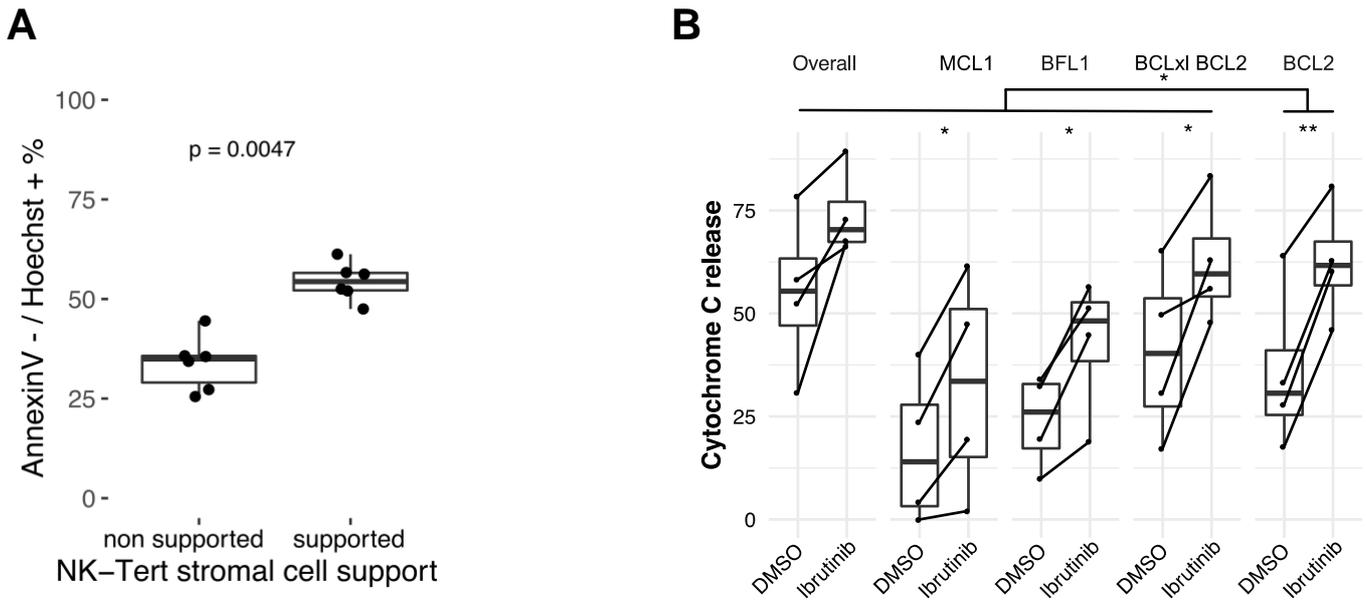


Patient 7 Run 2



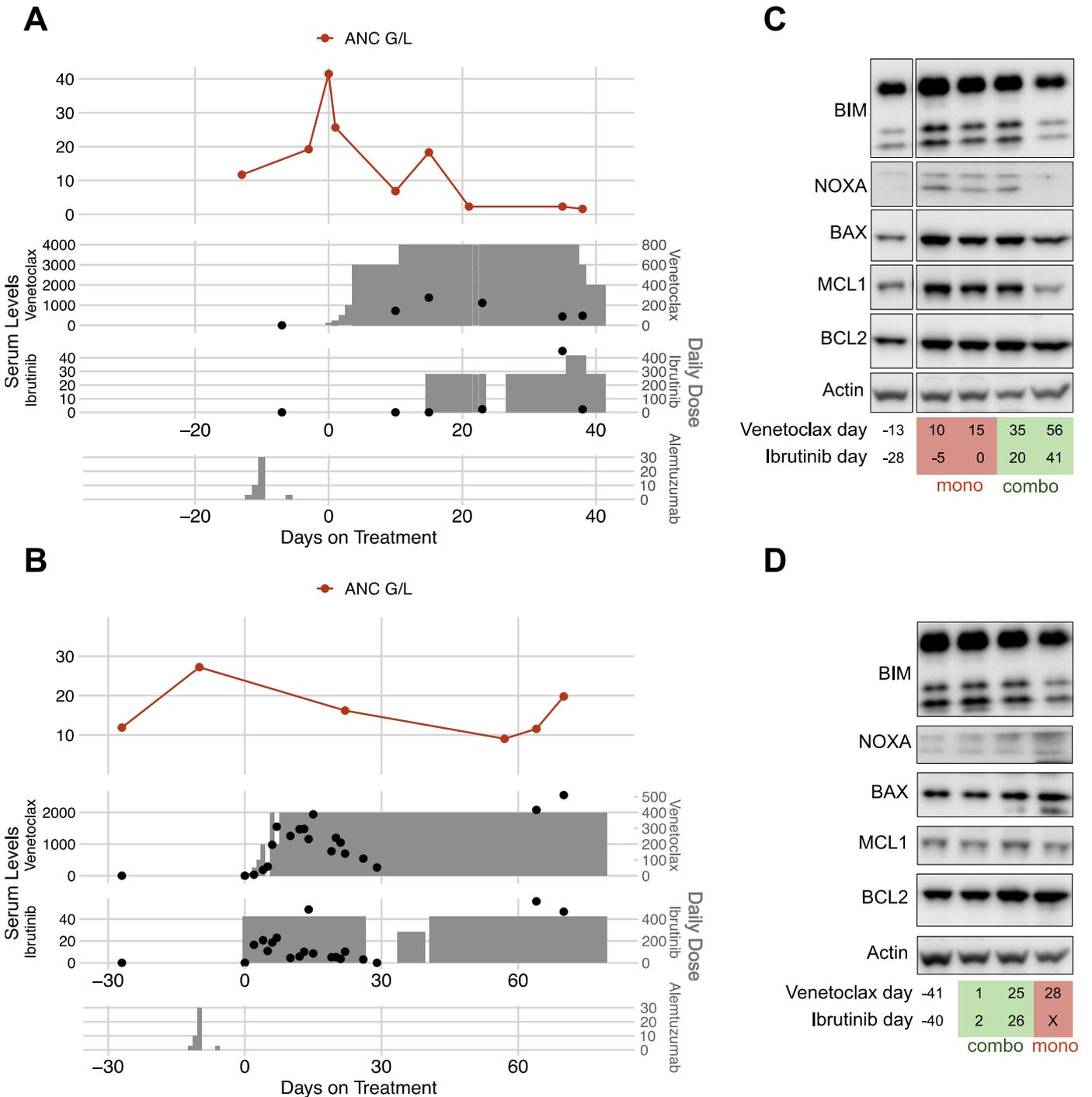
Supplementary Fig. 1.: Single Dose Response Curves and individual combinatorial heatmaps for drug screening A) Single dose response curves: Single dose response curves separated by patient for the indicated drugs. B) Heatmaps depicting the combinatorial response for each patient. Each tile represents percentage of control (mean in Run1 and 2). Drug concentrations are given as μM .

Supplementary Fig. 2



Supplementary Figure 2: Influence of stromal support on viability of T-PLL samples and BH3 profiling during ex-vivo ibrutinib treatment A) Comparison of AnnexinV positivity in primary T-PLL samples with and without NK-Tert stromal support: Percentage of annexin V negative / Hoechst positive cells of 6 T-PLL samples after 24 hours in vitro in R10 with or without stromal support by co-culture with NKT, paired T-Test. B) BH3 profiling for primary TPLL samples treated with ibrutinib: cytochrome C release is shown for samples treated either with DMSO or ibrutinib 10 μ M. For statistics, the difference between DMSO- and ibrutinib treated samples was compared between BCL2 dependent cytochrome C release and all other groups using a one-sided T-test ($* = p \leq 0.05$) and for DMSO vs. ibrutinib-treated samples in each group with a two-sided T-test ($* = p \leq 0.05$, $** = p \leq 0.01$).

Supplementary Fig. 3



Supplementary Figure 3: Absolute Neutrophil Counts and BH3-family member expression in two treated patients. A) and B) Absolute Neutrophil Counts (ANC G/L) are shown for the two treated patients (A = Patient A, B = Patient B). **C) and D)** Expression of BH3 family members of the two treated patients during combinatorial treatment with venetoclax and ibrutinib (C = Patient A, D = Patient B). Primary antibodies used were directed against Bcl-2 (Biologend, #658702), Mcl-1 (BD Biosciences, #559027), Bim (Cell Signaling, #2933), Noxa (Cat: 14766, Cell Signaling), Bax (Cat: MA5-14003, Invitrogen), beta-actin (Santa Cruz Biotechnology, SC-47778)