Supporting Information

Discovery of (2*S*)-8-[(3*R*)-3-Methylmorpholin-4-yl]-1-(3-methyl-2-oxobutyl)-2-(trifluoromethyl)-3,4-dihydro-2*H*-pyrimido[1,2-*a*]pyrimidin-6-one: A Novel Potent and Selective Inhibitor of Vps34 for the Treatment of Solid Tumors

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Figure S1. Line chart²¹ of an initial set of pyrimidinone derivatives. Activities on Vps34, GFP-FYVE and class I PI3Ks α , β , δ and γ , were measured and combined to establish “On target” (Vps34, GFP-FYVE) / “Off target” (class I PI3Ks) profiles (pIC₅₀ scale) complemented by in vitro eADME data ((%) lability in human and mouse liver microsomes, values divided per 10 to fit visually within the pIC₅₀ scale). Profile Correlation clustering (n=9) allowed identification of a sub-population that offer activity versus target and suitable level of selectivity versus off target while displaying a fair metabolism profile (cluster 7, with compound 2 and 3 highlighted in red). Compound 4, having a less favourable in vitro ADME profile, is highlighted in red and located in cluster 9. It was preferred to favor in vitro ADME positioning than selectivity ratio (cluster 6 or 1).

Figure S2. Impact of systematic exchange between non-substituted morpholine and 3(R)-methyl morpholine residues on the pyrimidinone scaffold is studied using Molecular Matching Pairs Analysis.²¹ Pairs shown here allow underlining the critical impact of the methyl morpholine group on the overall profile of the compounds. Pairs are materialized by solid lines, “On target” potency (pIC_{50} Vps34) is represented by dot size, cell potency (pIC_{50} GFP-FYVE) is represented along the Y axis while “Off target” potency (pIC_{50} PI3k γ) is color coded (red = no selectivity, green = selectivity). The figure clearly emphasize the competitive edge brought by the 3(R)-methyl morpholine on the selectivity versus “Off target”. Overall, increased pIC_{50} GFP-FYVE is observed, along with a global positive shift in selectivity versus class I PI3Ks.

Figure S3. Kintativ® profile of compound **31** (1 μ M) in Ramos cells.

Table S1. Crystallographic data

Compound	2	3	4	15	31
Data Collection					
Space Group	P 2 ₁ 2 ₁ 2	P 2 ₁ 2 ₁ 2	P 2 ₁ 2 ₁ 2	P 2 ₁ 2 ₁ 2	P 2 ₁ 2 ₁ 2
Cell dimensions (Å)	87.73	93.80	88.83	92.03	93.09
	154.87	147.01	145.46	146.93	146.36
	61.34	61.78	61.35	61.63	61.61
Resolution (Å)	87.74 – 2.99 (3.15 – 2.99)	49.00 – 2.70 (2.83 – 2.70)	75.91 – 1.93 (2.03 – 1.93)	146.93 – 2.83 (2.99 – 2.83)	146.36 – 2.80 (2.95 – 2.80)
Total reflections	118842	155155	247617	142029	63790
Unique reflections	16333	24128	60643	20552	17295
Completeness (%)	99.0 (96.7)	99.2 (94.5)	99.5 (97.8)	99.9 (100)	81.5 (84.7)
Rsym	14.7 (49.5)	9.2 (62.6)	6.2 (39.7)	10.5 (56.2)	10.8 (48.8)
I/σ(I)	9.4 (2.8)	14.2 (2.5)	13.8 (2.6)	15.5 (3.7)	10.6 (3.0)
cc1/2	Nc	99.8 (92.9)	Nc	Nc	Nc
Refinement					
Reflections used	16271	24071	60255	20509	17268
Rfree / R	24.6 / 21.2	22.1 / 18.0	20.8 / 18.4	22.8 / 17.6	25.5/18.9
Average B-values (Å ²)	34.1	62.5	28.5	56.1	46.4
Number of atoms					
Protein	4447	4415	4492	4423	4398
Heterogen atoms (not water)	113	95	32	30	38
Solvent	84	53	381	21	50
Rms deviations					
Bond lengths (Å)	0.008	0.010	0.008	0.010	0.009
Bond angles (°)	0.92	1.11	0.93	1.16	1.06
MolProbity score	1.85	1.97	1.37	2.22	1.92

Data in parentheses correspond to the highest resolution shells. To calculate Rfree, 5% of the reflections were excluded from the refinement. Rsym is defined as $R_{\text{sym}} = \sum hkl \sum i |I_i(hkl) - \langle I(hkl) \rangle| / \sum hkl \sum i I_i(hkl)$.