

Category	Parameter	Description
Assay	Type of assay	Enzymatic, endpoint
	Target	SARS-CoV-2 Papain-like Protease
	Primary measurement	Fluorescence intensity
	Key reagents	Ub-Rhodamine
Library	Library size	5,576 compounds
	Library composition	Small molecule, including FDA-approved drugs, advanced preclinical compounds and focused sets.
	Source	Commercial and in-house curated collections (see Methods)
	Additional comments	
Screen	Format	1536 well plate
	Concentration tested	4.2 μ M in 2% DMSO
	Plate controls	Negative control; 2% DMSO, positive control; compound rac5c (see manuscript, Figure 4)
	Reagent/ compound dispensing system	See Methods
	Detection instrument and software	PHERASTAR FSX (BMG)
	Assay validation/QC	Robust Z' > 0.5
	Hit criteria	>4*MAD above negative control
	Hit rate	0.45%
Post-HTS analysis	Counter screen	Human USP21, Ub-Rhodamine
	Additional assay(s)	
	Confirmation of hit purity and structure	LC/MS