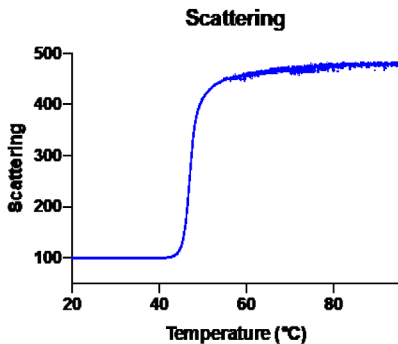
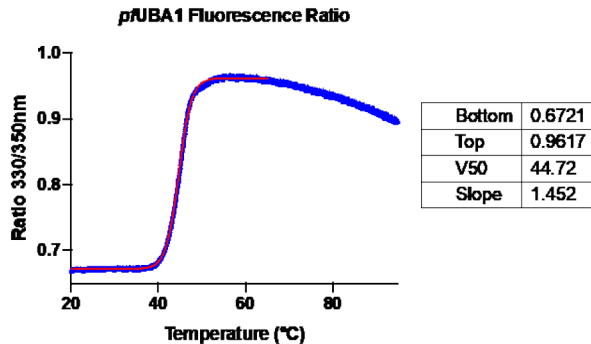


Scattering



Ratio 330/350nm

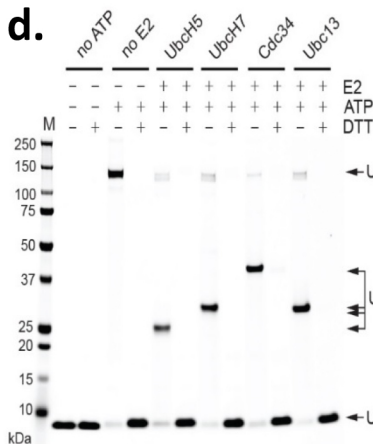


Bottom	0.6721
Top	0.9617
V50	44.72
Slope	1.452

d.

	no ATP			no E2			UbcH5			UbcH7			Cdc34			Ubc13			E2	ATP	DTT
	-	-	-	-	+	+	-	+	+	-	+	-	+	-	+	-	+	-	-	+	+
M																					
250																					
150																					
100																					
75																					
50																					
37																					
25																					
20																					
15																					
10																					
kDa																					

← U
← U
← U
← U
← U

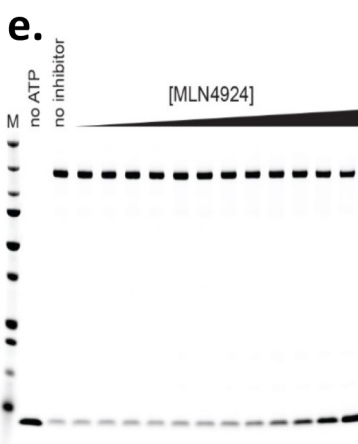


e.

no ATP
no inhibitor

[MLN4924]

This Western blot shows the phosphorylation of p38 in the absence of ATP. The top row of bands, labeled 'no ATP', shows a strong, consistent band across all lanes, indicating that p38 is constitutively phosphorylated when ATP is absent. The bottom row of bands, labeled 'no inhibitor', shows a very faint band in the first lane (no MLN4924) that gradually increases in intensity across the subsequent lanes as the concentration of MLN4924 increases. This indicates that MLN4924 promotes p38 phosphorylation even in the absence of ATP, but the effect is much weaker than when ATP is present.



f.

100
75
50
25
0

% U-E1 thioester

IC₅₀ = 47 ± 5 μM

0.01 0.1 1 10 100

MLN4924 (μM)

