

**Supplemental Table 2: HDX-MS protection summary of key AXP binding residue-containing peptides.** Numbers indicate % $\Delta$ HDX upon incubation of purified AMPK with indicated adenine nucleotide(s). Listed are MS-resolved peptides that contain the indicated AXP-binding residues.

		Wildtype holo-AMPK ( $\alpha_1\beta_2\gamma_1$ )				$\alpha_1$ -K47G, $\gamma_1$ -L129D/V130D/V276G/L277G (CBS4 only)	$\alpha_1$ -K47G, $\gamma_1$ -L129D/V130D/I312D (CBS3 only)	$\alpha_1$ -K47G, $\gamma_1$ -V276G/L277G/I312D (CBS1 only)
CBS	Sub-site	2.5 mM ATP	2.5 mM AMP	40 $\mu$ M AMP 400 $\mu$ M ADP 4.5 mM ATP	300 $\mu$ M AMP 1.0 mM ADP 3.8 mM ATP	2.5 mM AMP		
CBS-1	$\beta$ -P: T87, T89 Ribose: D90	-9	+4	-13	-15	+16	+14	-3
	Adenine: K127, L129, V130	-6	0	-3	-4	N/A	N/A	-2
	$\alpha$ -P: H151 $\gamma$ -P: R152	N/A	N/A	N/A	N/A	N/A	N/A	N/A
CBS-3	Ribose: D245	-2	-16	-6	-7	-18	-7	-13
	Adenine: I240	-7	-17	-11	-11	-21	-11	-19
	Adenine: V297	-6	-9	-7	-9	2	-1	1
	Adenine: V276+L277	N/A	N/A	N/A	N/A	-20	N/A	-17
	$\alpha$ -P: R299	-6	-9	-7	-9	2	-1	1
	$\alpha$ -P: K170	-1	0	-1	-1	N/A	1	0
CBS-4	$\beta$ -P: L315+S316 Ribose: D317	1	-1	0	0	-1	-1	-2
	Ribose: T200,	-8	N/A	-10	-8	-31	-15	-24
	Adenine: A205	-7	-16.5	-10.5	-10.5	-20.5	-4	-12
	Adenine: I312;	-3	-8	-4	-3	-9	N/A	N/A
	Adenine: V225 $\alpha$ -P: S226	-13	-33.5	-14	-16	-29	-6	-15
	$\alpha$ -P: H151	N/A	N/A	N/A	N/A	N/A	N/A	N/A
	$\gamma$ -P: R299	-6	-9	-7	-9	2	-1	1