Supplementary Table S1. Nf2/merlin head-tail interactions for comparison with Nf2/merlin binding to LATS1 using the PDBePISA server (https://www.ebi.ac.uk/msd-srv/prot_int/pistart.html).

Residues contributing to the Nf2/merlin head domain interface with Nf2/merlin mutant A585W tail domain. Black font is used for interactions with the F1 FERM Nf2/merlin subdomain and blue font for the F2 FERM Nf2/merlin subdomain.

A. Hydrogen bonds

Nf2/merlin	[Å]	Nf2/merlin
TYR 132 [OH]	2.70	ASP 559 [OD1]
ASN 175 [O]	2.94	ARG 554 [NH1]
GLN 178 [N]	2.79	LEU 549 [O]
LEU 176 [O]	2.67	LEU 551 [N]
ARG 187 [NH1]	2.83	GLU 545 [OE1]
TRP 191 [NE1]	3.19	GLN 538 [OE1]
GLU 194 [OE2]	3.64	HIS 534 [NE2]
GLU 194 [OE1]	2.96	GLN 538 [NE2]
GLU 194 [OE2]	2.80	GLN 538 [NE2]
LEU 214 [O]	2.93	LYS 573 [NZ]
GLU 215 [O]	3.73	LYS 573 [NZ]
TYR 217 [O]	3.03	LYS 573 [NZ]
ASN 226 [ND2]	3.13	LEU 595 [O]
ASN 226 [ND2]	2.94	PHE 592 [O]
ASN 226 [ND2]	3.70	GLU 593 [O]
LYS 227 [N]	2.98	GLU 593 [OE1]
LYS 228 [NZ]	3.27	GLU 594 [O]
GLU 260 [OE1]	2.10	ARG 588 [NH1]
GLU 260 [OE2]	2.75	ARG 588 [NH2]
ASP 281 [O]	2.80	TRP 585 [NE1]

B. electrostatic interactions

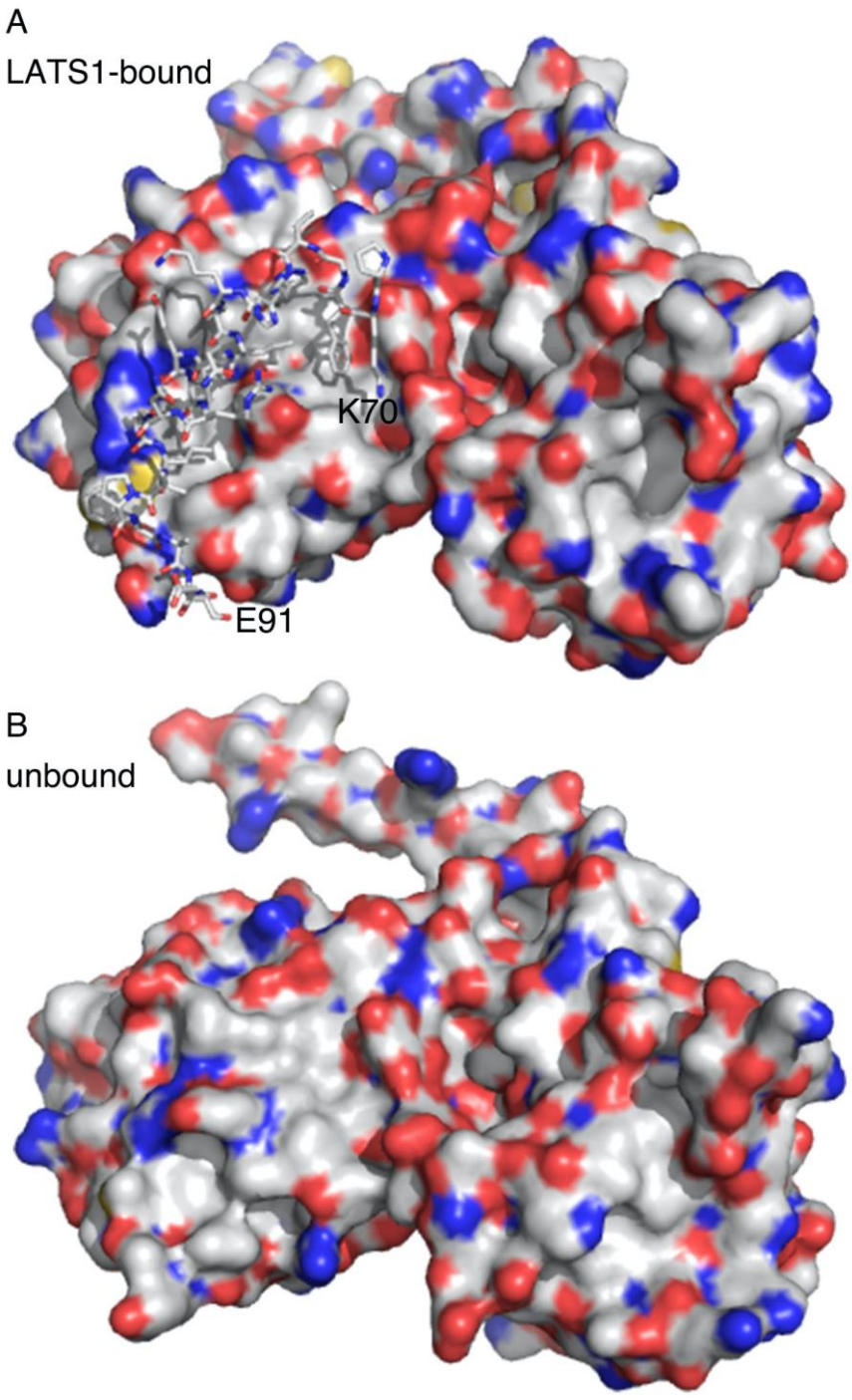
Nf2/merlin	[Å]	Nf2/merlin
ARG 187 [NH1]	2.83	GLU 545 [OE1]
GLU 194 [OE2]	3.64	HIS 534 [NE2]
GLU 260 [OE1]	2.10	ARG 588 [NH1]
GLU 260 [OE2]	3.01	ARG 588 [NH1]
GLU 260 [OE1]	3.52	ARG 588 [NH2]
GLU 260 [OE2]	2.75	ARG 588 [NH2]

Supplementary Figure S1. Comparison of the LATS1-bound Nf2/merlin structure with the unbound apo Nf2/merlin structure

Surface representation of Nf2/merlin (red, oxygen; blue, nitrogen; white, carbon; yellow, sulfur) with LATS1 shown in stick representation.

A. Upon binding to LATS1, rearrangement occurs and LATS1 fits well in the groove. *N*-(K70) and *C*-terminal residues (E91) are indicated.

B. In the apo form, the LATS1 binding groove is not well-formed and blocked by Glu-136 (not shown).



Supplementary Figure S2. The extended Nf2/merlin *C*-terminal α-helix is necessary to understand the allosteric binding mechanism

Superposition of our 1.6 Å structure (residues 21-338; F1, orange; F2, yellow; F3, green) onto the truncated 2.3 Å or 2.7 Å Nf2/merlin structures (residues 21-311, gray) bound to LATS1 (cyan; PDB entry 4zrk) (40) or LATS2 (gray; PDB entry 4zri) (40), confirms our observed allosteric mechanism: the F1-F2 subdomains (residues 21-215; F1, orange; F2, yellow) superimpose with root means squares deviations of 0.668 Å for LATS1 (or 0.581 Å for LATS2) for 1,363 atoms (or 1,379 atoms for LATS2, cyan) while including F3 (residues 21-311, green) superimposes much poorer with root means squares deviations of 0.996 Å for LATS1 (or 0.806 Å for LATS2) for 2,028 atoms (or 1,913 atoms for LATS2). The movements of over 3 Å for example seen at residue Lys-279 of the Nf2/merlin F3 subdomain upon binding of LATS to the Nf2/merlin F2 sub-domain is indicated by a double arrow. Furthermore, in the 2.3 Å truncated Nf2/merlin structure bound to LATS1, its *N*-terminus (LATS1 residues 69-73, cyan) points in the opposite direction compared to LATS1 seen in our 1.6 Å Nf2/merlin (residues 21-338) and LATS2 in the LATS2-bound structure (PDB entry 4zri) (40).