

Review

The LATS1 and LATS2 tumor suppressors: beyond the Hippo pathway

Noa Furth¹ and Yael Aylon*¹

Proper cellular functionality and homeostasis are maintained by the convergent integration of various signaling cascades, which enable cells to respond to internal and external changes. The Dbf2-related kinases LATS1 and LATS2 (LATS) have emerged as central regulators of cell fate, by modulating the functions of numerous oncogenic or tumor suppressive effectors, including the canonical Hippo effectors YAP/TAZ, the Aurora mitotic kinase family, estrogen signaling and the tumor suppressive transcription factor p53. While the basic functions of the LATS kinase module are strongly conserved over evolution, the genomic duplication event leading to the emergence of two closely related kinases in higher organisms has increased the complexity of this signaling network. Here, we review the LATS1 and LATS2 intrinsic features as well as their reported cellular activities, emphasizing unique characteristics of each kinase. While differential activities between the two paralogous kinases have been reported, many converge to similar pathways and outcomes. Interestingly, the regulatory networks controlling the mRNA expression pattern of *LATS1* and *LATS2* differ strongly, and may contribute to the differences in protein binding partners of each kinase and in the subcellular locations in which each kinase exerts its functions.

Cell Death and Differentiation (2017) 24, 1488–1501; doi:10.1038/cdd.2017.99; published online 23 June 2017

Facts

- LATS1 and LATS2 proteins show extensive sequence similarity and share similar modes of post-translational modifications.
- The *LATS* genes are differentially regulated at the transcriptional level.
- LATS kinases engage divergent binding partners, although these effectors often converge on similar cellular processes.
- Whole-body deletion, as well as tissue-specific deletion, of either *Lats1* or *Lats2*, reveals critical differences in the *in vivo* functions of the two kinases.

Open Questions

- Additional signaling pathways: what other functions do LATS kinases have beyond restricting YAP/TAZ activity?
- Redundant *versus* divergent function: what is the contribution of each kinase to distinct biological processes?
- Phosphorylation substrates of LATS: how is LATS kinase target recognition determined beyond simple amino-acid sequence motifs?
- Pro- *versus* anti-tumor effects: how does cellular context direct LATS toward apparently opposing functional outcomes?

In recent years, the LATS1 and LATS2 kinases have become the focus of intense research interest. They are gaining prominence due to their broad range of biological activities in

cell cycle regulation, differentiation and motility, as well as the diverse pathological outcomes of their deregulation. LATS kinases are critical for organism fitness, genome integrity and cancer prevention. The core kinase module is evolutionarily conserved from yeast through flies to humans, although effectors and biological impact have expanded over the course of evolution.

The yeast ortholog of LATS, Dbf2 is localized to the spindle pole body (yeast centrosome) and regulates mitotic exit.¹ Cdc15 (homolog of MST) is required for Dbf2 activation,^{2,3} and together they constitute a kinase module of the mitotic exit network.^{1,3} This module has been conserved in humans, manifested by LATS phosphorylation and activation by MST1/2 (MST) kinases.⁴ During evolution, this module recruited numerous different effectors, most notably the transcriptional coactivators YAP and TAZ, and extended its repertoire of biological functions. The *Caenorhabditis elegans* LATS, Ce-Wts-1, is associated with development, lifespan and body length control.⁵ Interestingly, nematodes lack YAP/TAZ,⁶ and Ce-Wts-1 exerts its function via effectors of the TGF-beta signaling pathway.⁵

Deletion of the *Drosophila* Warts (*Wts*, the fly ortholog of *LATS*) causes dramatic tissue overgrowth and abnormalities in cellular polarity.^{7,8} The fly Warts-Hippo (*Hpo*, MST ortholog) module exerts some of its functions via phosphorylation and inhibition of Yorkie (*Yki*, YAP and TAZ ortholog),⁹ while maintaining ancestral *Yki*-independent functions.¹⁰ The strong evolutionary conservation of the MST/LATS/YAP cascade (the Hippo pathway) is exemplified by the fact that human LATS

¹Department of Molecular Cell Biology, The Weizmann Institute of Science, POB 26, 234 Herzl St., Rehovot 7610001, Israel

*Corresponding author: Y Aylon, Department of Molecular Cell Biology, The Weizmann Institute of Science, POB 26, 234 Herzl St., Rehovot 7610001, Israel; Tel: +972 8 9342390; Fax: +972 8 9346004; E-mail: yael.aylon@weizmann.ac.il

Received 23.3.17; revised 30.4.17; accepted 15.5.17; Edited by G Melino; published online 23.6.17

proteins are able to rescue the loss of Wts functions in *Drosophila*.^{11,12}

As in other developmental pathways, complexity tends to increase over evolution. This is evidenced by the existence of additional components impacting the Hippo pathway, a diversity that might have been facilitated by duplication of the single ancestral *LATS* gene into two paralogs (coinciding with the duplication of other Hippo components, i.e., *MST*, *TEAD* and *YAP*) during deuterostome evolution.⁶ Genetic studies in mice have underscored the functional differences between the duplicated LATS kinases. Loss of *Lats2* is embryonic lethal on or before embryonic day E12.5, and this lethality is postulated to result from aberrant proliferation, mitotic defects and accumulated genomic instability.^{13,14} In contrast, *Lats1*-null mice are viable. However, they suffer from developmental defects such as infertility, growth retardation, pituitary dysfunction and lack of ductal structures in the mammary gland. In addition, *Lats1*^{-/-} mice are prone to spontaneous and oncogene-induced sarcomas.¹⁵

In this review, we examine the features of LATS1 and LATS2, some of which are redundant (presumably representing a common primordial LATS function), and others distinct (presumably acquired in the course of evolutionary diversification). The common ability of both LATS kinases to repress YAP/TAZ has been studied extensively (reviewed recently in Zanconato *et al.*¹⁶ and Meng *et al.*¹⁷). Therefore we will focus mainly on LATS utilization of effectors other than YAP/TAZ, and the impact of those interactions on cell fate.

Protein Structure and Post-Translational Modifications of LATS Kinases

Human LATS1 and LATS2 are Ser/Thr kinases of the AGC subfamily, most closely related to the nuclear Dbf2-related kinases (NDR1/2).¹⁸ While LATS1 and LATS2 share extensive sequence similarity within their kinase domain (85% similarity) located at the C terminus of the proteins, the N terminus portion displays significantly lower conservation (Figure 1 and detailed in Table 1).^{19,20} Immediately carboxyterminal to the catalytic domain of both kinases is a hydrophobic motif; this pattern is akin to other AGC kinases such as AKT, S6K1 and PKC.¹⁸ Within the lowly conserved amino (N) terminus, there are two stretches of conserved sequences (LCD1 and 2) that are required for proper LATS regulation and function.^{21,22} Also within the N terminus, both LATS1 and LATS2 harbor evolutionarily conserved ubiquitin-associated domains. Such domains are known to bind ubiquitinated proteins and may function in LATS activation.²³ Interestingly, each of the kinases possesses unique features, which may facilitate different protein–protein interactions; the N terminus of LATS1 contains a proline-rich domain,²⁴ while a unique PAPA repeat is found in LATS2.²⁰ Furthermore, *LATS2* encodes one, and *LATS1* encodes two, PPxY motifs; these are essential for interaction with the WW hydrophobic pockets of YAP, TAZ and other Hippo pathway components.²⁵

Superimposed on the amino-acid sequence is a combinatorial ‘code’ of post-translational modifications governing LATS activity (Figure 1; Table 1). Upstream signals such as cell cycle progression, cytoskeleton alterations and growth signals shape this code, defining different cellular outcomes.

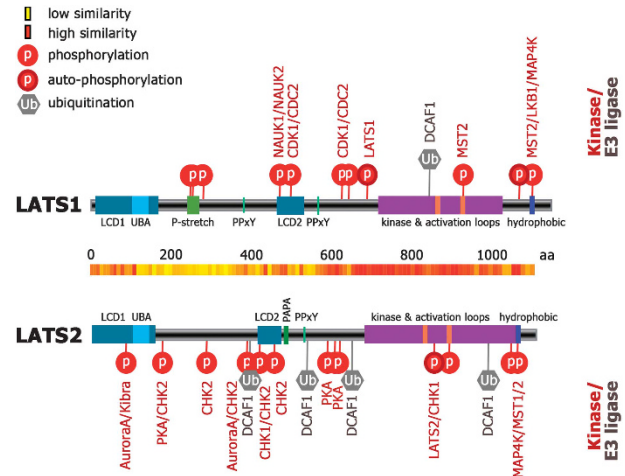


Figure 1 Schematic comparison of human LATS1 and LATS2 protein structures. Structural motifs, as defined by UniProtKB database, are represented as solid boxes on gray background. Reported phosphorylation sites are designated as red lollipops, with the phosphorylating kinase indicated above in dark red. Ubiquitination sites are gray denoted by hexagons, with the reported E3 ligase indicated above in dark gray. The heatmap between the LATS1 and LATS2 protein schemes represents the similarity of the aligned sequences, where dark orange represents high and yellow represents low amino-acid similarity. Similarity was calculated using the Waterman–Eggert local alignment application (EMBOSS explorer), comparing LATS1 (O95835-1) and LATS2 (Q9NRM7). Numbers above heatmap represent amino-acid position

LATS1 and LATS2 share dual phosphorylation–autophosphorylation mechanisms that are commonly employed by a subset of AGC kinases (including the aforementioned).²⁶ MST-dependent phosphorylations of LATS (S909/T1079 on LATS1 and T1041 on LATS2) increase its kinase activity.^{4,27} Subsequently, MOB1 binding to the LATS hydrophobic domain relieves LATS autoinhibition and facilitates activating autophosphorylation (LATS1 residues S674 and S1049; LATS2 residue S835).²⁸ In humans and flies, recruitment of LATS to the plasma membrane promotes MST-dependent phosphorylation and activation.²⁹ PP2A-mediated dephosphorylation of these sites may counter MST-mediated phosphorylation to quench LATS1 activation.^{28,30} The effect of PP2A on LATS2 phosphorylation status has not been examined.

Importantly, MST are neither obligatory nor the sole LATS activators. For instance, deletion of *Mst* in mouse liver results in YAP hyperactivity without reduction in LATS phosphorylation status.³¹ In line with this, MAP4Ks phosphorylate both LATS1 and LATS2 hydrophobic motifs resulting in their activation and YAP inhibition.³² Similarly, phosphorylation of LATS2 by PKA bypasses MST to augment LATS2 kinase activity toward YAP.³³ Other MST-independent phosphorylation of LATS may also result in cellular activities that are not related to YAP/TAZ regulation. For example, CHK1/2 phosphorylation of LATS2 S408 is associated with DNA damage-induced apoptosis.³⁴ Likewise, LATS2 is activated by CHK1 and ATR in response to oncogenic H-RAS.^{35,36}

Additional phosphorylation events of LATS1 and LATS2 relate to mitotic progression. A subset of molecules of both LATS1 and LATS2 is located at the centrosome,^{13,24} an organelle known for its crucial role in cell division.³⁷ In mitosis,

Table 1 Summary of reported LATS1/2 protein motifs and post-translational modifications

LATS1				LATS2					
Amino acid	Motif/modification type	Catalyzed by	Associated with	Ref	Amino acid	Motif/modification type	Catalyzed by	Associated with	Ref
13–167	LCD1			21,22	1–160	LCD1			21,22
103–143	UBA domain				101–141	UBA domain			
1075–1080	Hydrophobic motif				1037–1042	Hydrophobic motif			
236–266	Proline-rich domain (P-stretch)			24	403–463	LCD2			21,22
373–376	PPXY motif				467–480	Proline–alanine (PAPA) repeat			20
458–523	LCD2			21,22	515–518	PPXY motif			19,20
556–559	PPXY motif				666–1046	S-TKc (catalytic domain)			19,20
705–1010	S-TKc (catalytic domain)			19,20	668–973	S-TKc (catalytic domain)			19,20
845–857; 905–915	Activation loop				808–820; 868–878	Activation loop			
S244	Phos		Nuclear phosphoproteins Cancer; mitosis	194	S83	Phos	Aurora A, KIBRA	Mitosis	38,40,195
T246	Phos		Cancer; mitosis	196–198	S172	Phos	PKA, CHK2	YAP regulation	33,34
S278	Phos		Mitosis	198	T279	Phos	CHK2	UV radiation	198
S462	Phos			199,200	S380	Phos	Aurora A, CHK2	Mitosis	34,39,197
S464	Phos	NuaK1, NuaK2	Protein stability	41,198	S408	Phos	CHK1, CHK2	UV radiation	34
T490	Phos	CDK1/CDC2	Mitosis	197,201	S446	Phos	CHK2	Cancer	34
S613	Phos	CDK1/CDC2	Cancer; mitosis	4,197,201	S576	Phos	PKA	YAP regulation	197,198
S633	Phos		Autophosphorylation	4	S592	Phos	PKA	YAP regulation	33
S674	Phos	MST2	Activation	4,121	S598	Phos	Trans-auto-phosphorylation, CHK1	Apoptosis	33
S909	Phos		Autophosphorylation	4	S835	Phos			153
S1049	Phos	MST2, LKB1, MAP4K	Activation	4	S872	Phos		Activation	27
T1079	Phos		Activation	4,30,32,118,119,202–204	T1024	Phos		Mitosis	198
S1111	Phos	DCAF1	Protein stability	199	T1026	Phos	MST1, MST2, MAP4K	Mitosis	198
K830	Ub		Protein stability	51	T1041	Phos	DCAF1	Activation	4,27,30,32,118,119,203,204
Unknown	Ub	Itch	Protein stability	48,49	K383	Ub	DCAF1	Kinase inactivation	51
Unknown	Ub	WWP1	Protein stability	50	K527	Ub	DCAF1	Kinase inactivation	51
					K633	Ub	DCAF1	Kinase inactivation	51
					K968	Ub	DCAF1	Kinase inactivation	51
					Unknown	Ub	SIAH2	Protein stability	52

LATS1 (but not *LATS2*) is phosphorylated on T490 and S613 by CDK1/CDC2,¹² whereas *LATS2* (but not *LATS1*) is phosphorylated on S83 and S380 by Aurora A kinase.^{38–40} This may reflect a general divergent and complementary phosphorylation pattern, whose functional consequences remain to be explored.

Differential phosphorylation of *LATS1* and *LATS2* also affects their stability. Thus, *LATS1* phosphorylation on S464 by NUA1 reduces its protein levels,⁴¹ whereas KIBRA stabilizes *LATS2* by augmenting its phosphorylation and inhibiting its ubiquitination.⁴² Additionally, *LATS* protein stability and kinase activity can be bolstered by binding to heat-shock proteins. For instance, both *LATS* kinases are clients of the molecular chaperone HSP90.⁴³ Interestingly, MOB1 binding rescues *LATS* destabilization caused by HSP90 inhibition,⁴³ suggesting that MOB1 also functions to stabilize the *LATS* proteins. On the other hand, destabilizers of *LATS* include the LIM domain-containing proteins Ajuba, Dachous and Zyxin,^{44,45} which facilitate cell proliferation by reducing *LATS* protein levels and inhibition of *LATS* activity.⁴⁵

LATS protein stability is regulated also through ubiquitination by a number of E3 ligases. Thus, NEDD4 ubiquitinates and promotes the degradation of both kinases,^{46,47} whereas additional E3 ligases with WW domains, such as ITCH and WWP1, specifically bind and destabilize *LATS1*.^{48–50} The WW–PPxY interaction between these E3 ligases and *LATS1* might serve a dual purpose, by both decreasing *LATS1* levels and displacing YAP/TAZ from its PPxY-binding site. Interestingly, CRL4-DCAF1 performs inhibitory ubiquitination of both kinases.⁵¹ However, whereas *LATS1* is polyubiquitinated and directed to proteasomal degradation, *LATS2* is oligoubiquitinated at multiple sites, resulting in kinase inactivation without enhanced degradation. This might reflect a cellular mechanism to free YAP/TAZ from *LATS2* inhibition while retaining *LATS2* kinase-independent functions. Yet, *LATS2* is targeted for degradation by a distinct E3 ligase, SIAH2.⁵² Intriguingly, SIAH2 activity is associated with hypoxic response,⁵³ and a decrease in *LATS* protein levels is critical for ROS-induced senescence.⁵⁴

Regulation of *LATS* Gene Expression

Classically, tumor suppressors may undergo loss of function due to genomic deletions or mutations, or through epigenetic silencing. Loss of heterozygosity of *LATS1* was reported in ovarian,^{55,56} cervical⁵⁷ and breast^{58–60} cancer. Likewise, frequent copy number loss of *LATS2* also occurs in breast,⁶¹ ovarian,⁶² hepatocellular^{63,64} and lung⁶⁵ cancer, as well as in chronic lymphocytic leukemia.⁶⁶

On the other hand, mutations in the *LATS* genes are relatively rare. However, due to the growing popularity of large genomic sequencing projects, evidence of *LATS* mutations in cancer is gradually emerging.⁶⁷ In basal cell carcinoma of the skin, mutations occur specifically within the kinase domain of either *LATS1* or *LATS2* (16% or 12%, respectively), but rarely in both together.⁶⁸ Interestingly, in other tumor types only one of the kinases is significantly mutated. This is exemplified in esophageal and non-small-cell lung cancer, where tumor-specific mutations were found in *LATS2* but not *LATS1*.^{65,69} This further suggests that *LATS1* and *LATS2* may play distinct,

non-redundant roles in some tumors. Nevertheless, the low rates of mutations in *LATS* genes emphasize that other mechanisms are dominant in reducing *LATS* activity, and it remains to be shown whether these mutations are driver rather than passenger mutations during tumorigenesis.

Promoter hypermethylation is another mechanism by which tumor suppressors are often inactivated.⁷⁰ Such mode of inactivation has been documented for *LATS1*,^{71–73} *LATS2*,^{74,75} and in some cases for both, in various types of tumors.^{76–82} Importantly, downregulation of *LATS* expression has been associated with more aggressive cancer phenotypes.^{74,76–78,83} Promoter silencing can be mediated also by long non-coding RNAs (lncRNAs), which recruit the epigenetic machinery. Recently, it has been reported that the oncogenic lncRNAs PVT1, AGAP2-AS1 and LINC00673, whose elevated expression correlates with bad prognosis in non-small-cell lung and gastric cancers, tether polycomb repressive complexes to the *LATS2*, but not *LATS1*, promoter.^{84–86} Depletion of lncRNA reinstates *LATS2* expression and causes p53-dependent cell death.

More broadly, *LATS2* mRNA levels are exquisitely sensitive to tumor suppressive signaling, and are tightly regulated both transcriptionally and post-transcriptionally (Figure 2 and detailed in Table 2), while this seems to be less pertinent to *LATS1* expression. Induction of *LATS2* contributes to p53 tumor suppressive functions through a positive feedback mechanism, wherein the *LATS2* protein promotes p53 stabilization by binding and inactivating the major p53 inhibitor MDM2, while p53 directly positively regulates the transcription of the *LATS2* gene.^{87–89} In addition to regulating basal levels of *LATS2*, binding of p53 to the *LATS2* promoter augments transcription in response to genotoxic, developmental and metabolic stresses.^{35,87,89–91}

Like p53, FOXF3 also interacts directly with the *LATS2* promoter to induce *LATS2* expression.⁹² Interestingly, the levels of FOXF3 are positively regulated by MST,⁹³ representing an additional mechanism by which MST promotes *LATS2* activity. Intriguingly, also within the Hippo pathway, YAP/TAZ and their canonical partner transcription factor TEAD directly transactivate *LATS2* (but not *LATS1*) gene expression.^{94,95} Hence, YAP/TAZ positively regulates the expression of one of their key negative regulators. Similarly, in fly wing disks, *Wts* expression is upregulated upon expression of activated Yki, and this depends on the fly ortholog of TEAD, Scalloped.⁹⁴ It has been proposed that this negative feedback loop between *LATS2* and YAP/TAZ serves to dampen the duration of YAP activity,⁹⁵ maintain homeostasis⁹⁶ and render the Hippo pathway more robust, in order to resist the oncogenic effects of excessive YAP.⁹⁴

Nevertheless, positive regulation of *LATS* transcription does not always have a tumor suppressive outcome. For example, the *LATS1* promoter can be transactivated by CUX1, a transcription factor associated with acceleration of S-phase and tumorigenesis.^{97,98} *LATS2* overexpression in nasopharyngeal carcinomas was found to be associated with poor prognosis,⁹⁹ and in metastatic human breast cancer cells high levels of *LATS2* are associated with invasive and migratory capacities.¹⁰⁰ Furthermore, according to publicly available TCGA data, *LATS2* expression levels are elevated in glioblastoma, and the expression of both *LATS1* and *LATS2*

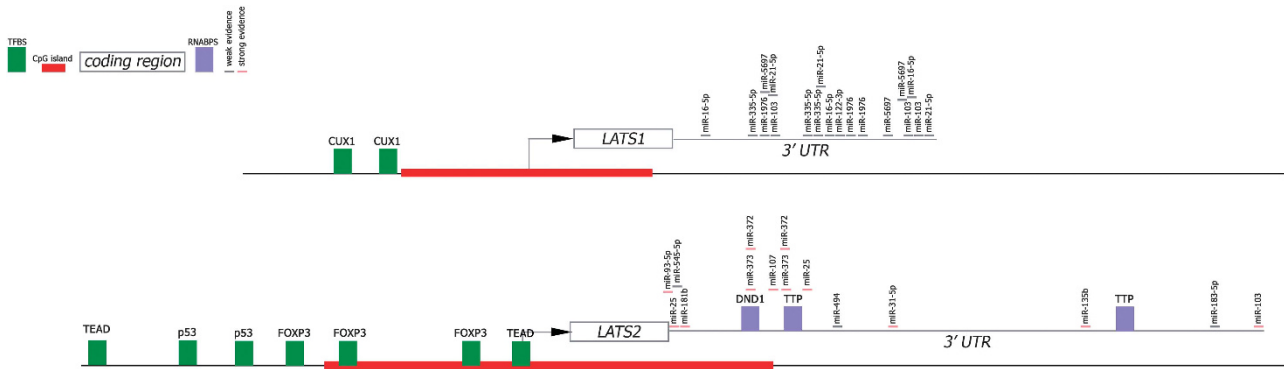


Figure 2 Scheme of human *LATS1* and *LATS2* genomic and mRNA structure. DNA is represented as a single black line. CpG islands (GC content > 50%), as defined by UCSC Genome Browser (GRCh37/hg19), are indicated by thick red lines, the length of which corresponds to the relative length of the *LATS1* and *LATS2* CpG stretch. Transcription factor binding sites (TFBS) are represented schematically as dark green boxes, whereas RNA-binding protein sites (RNABPS) are mauve colored. The coding regions of *LATS1* and *LATS2* are not drawn to scale, but the 3'UTR is drawn in the same scale as the CpG island designation. miR binding sites with strong experimental documentation are indicated by an orange underline, whereas putative miR binding sites, documented in broader screens with less conclusive direct evidence (miRTargetBase¹⁹³), are indicated by light gray lines

is significantly augmented in stomach cancer.¹⁰¹ This suggests that, contrary to the common assignment of *LATS1* and *LATS2* as tumor suppressors, retention of high *LATS* expression may actually sometimes be beneficial to the tumor, at least in some settings.

Post-Transcriptional Regulation of *LATS* mRNA

Several RNA-binding proteins have been shown to affect *LATS2* mRNA stability. Both Piwi-like 2 (PiwiL2), a protein which usually mediates gene silencing, and Deadend 1 (DND1) stabilize the *LATS2* transcript.^{102,103} DND1 binds to the 3'UTR to protect *LATS2* mRNA from microRNA (miR)-mediated repression.^{104,105} On the other hand, TTP, an AU-rich domain RNA-binding protein, promotes the degradation of *LATS2* mRNA by binding to its 3'UTR.¹⁰⁶ Interestingly, the DND1-binding site overlaps not only with miR target regions but also with one TTP binding site, consistent with the notion that multiple layers of RNA-binding proteins and miRs are in place to safeguard and modulate *LATS2* mRNA levels.

Strong evidence exists that at least four miRs directly bind the *LATS2* mRNA 3'UTR to repress *LATS2* expression (Figure 2 and detailed in Table 2). One miR, miR-135b, targets the mRNA of *LATS2*,¹⁰⁷ as well as of additional components within the Hippo pathway (for instance *MOB1* and *NDR2*¹⁰⁸), making it a 'Hippo-centric miR'. In contrast miR-31, an oncogenic miR overexpressed in numerous cancers,¹⁰⁹ specifically targets *LATS2* mRNA.¹¹⁰ Additionally, miR-372 and miR-373 have been shown to inhibit *LATS2* mRNA, causing reduction of *LATS2* expression and protein levels in testicular germ cell tumors,¹¹¹ and in cell lines derived from gastric cancer¹¹² and esophageal cancer.¹¹³

In contrast to *LATS2*, strong evidence of direct targeting of the *LATS1* mRNA 3'UTR by miRs is lacking. This may, in part, be due to the difference in length of the 3'UTRs (*LATS1* 814 nucleotides versus *LATS2* 1838 nucleotides, Figure 2), which might render the *LATS2* mRNA more vulnerable to miR-mediated inhibition. This supports the notion that, subsequent to the diversification of the ancestral *LATS* into two genes,

evolution has shaped each of these genes to receive inputs from different signaling modules, thereby expanding substantially the connectivity of the Hippo pathway and providing it with a broader portfolio of 'networking' opportunities. Indeed, consistent with such conjecture, although both *LATS1* and *LATS2* 3'UTRs are each highly conserved across different species, there is a very low similarity between them (only 3% similarity between the 3'UTRs of human *LATS1* and *LATS2*, according to the BLAST algorithm).

Interestingly, some miRs can have an indirect positive impact on *LATS1* expression. For instance, miR-106b targets *ITCH* mRNA, encoding an E3 ligase that promotes *LATS1* degradation, and in this way positively modulates *LATS1* protein levels.¹¹⁴ Likewise, miR-9 and miR-137 suppress the translation of *CUL4A*, a negative regulator of *LATS1*.¹¹⁵ Notably, miR-9-3p (processed from the complementary strand of miR-9) targets *TAZ* mRNA,¹¹⁶ thus, both strands of miR-9 function to reinforce the tumor suppressive potential of the Hippo pathway and quench the output of its oncogenic effectors.

LATS Protein Interactions and Cellular Localization

Pathway analysis of fly Warts binding partners (data from Kwon *et al.*¹¹⁷) revealed an enrichment of metabolic and DNA repair pathways. Some of these functions may be conserved in mammals, as energy stress and DNA damage have been shown to activate *LATS*.^{118–121} Two studies have provided comprehensive pictures of the mammalian Hippo signaling interactome (refs 122,123 and Figure 3). As expected, proteins binding to both *LATS1* and *LATS2* are enriched for 'Hippo signaling'. However, proteins binding exclusively to *LATS1* or *LATS2* are quite different in their pathway enrichment. Thus, proteins associated with *LATS1*, but not *LATS2*, are related to Estrogen signaling, whereas *LATS2* has Evolved a divergent interactome related to cell cycle, metabolism and p53. Of note, in both of these studies, the number of unique *LATS2* interacting proteins was higher than of unique *LATS1* interactors. Although this might have arisen from technical

Table 2 Summary of reported regulators of LATS1/2 mRNA levels

LATS1				LATS2			
Transcription factor	Binding position relative to TSS (approx.)	Ref	Transcription factor	Binding position relative to TSS (approx.)	Ref		
CUX1	-400 bp -600 bp	88	p53	-4700 bp -3000 bp -2500 bp -2000 bp -800 bp -600 bp -400 bp -10 000 bp 0 bp	89		
			FOXP3		92		
			YAP-TEAD		94,95		

LATS1				LATS2					
miRNA	3'UTR location	F function	Validation method	Refs	miRNA	3'UTR location	Function	Validation method	Ref
hsa-miR-103a-3p	717, 745, 301		NGS	205	hsa-miR-103	1833	Promotes invasion and proliferation of colorectal cancer cells	qPCR, luciferase assay, western blot	206
hsa-miR-16-5p	94, 480, 718		NGS	205	hsa-miR-6773-3p	NA		NGS	207
hsa-miR-4279	NA		NGS	208	hsa-miR-3153	NA		NGS	207
hsa-miR-1976	277, 538, 570		NGS	208	hsa-miR-25-3p	408 or 1	Proliferation and invasion of the gastric cancer cells	Reporter assay, western blot, qPCR	209
hsa-miR-6747-3p	NA		NGS	208	hsa-miR-545-5p	14		NGS	207
hsa-miR-6727-3p	NA		NGS	208	hsa-miR-4668-5p	NA		NGS	207
hsa-miR-4722-3p	NA		NGS	208	hsa-miR-4257	NA		NGS	207
hsa-miR-3691-3p	NA		NGS	208	hsa-miR-494-3p	507		NGS	207
hsa-miR-5697	292, 660, 720		NGS	208	hsa-miR-6847-5p	NA		NGS	207
hsa-miR-335-5p	231, 413, 428	Suppresses lung and bone metastasis of breast cancer cells	Microarray	210	hsa-miR-183-5p	1673		NGS	207
hsa-miR-122-3p	491		NGS	208	hsa-miR-135b-3p	1271	Lung cancer metastasis; MAPK/ERK signaling	Microarray, qPCR	107,108,211
hsa-miR-21-5p	441, 300, 775	Promotes tumor invasion	Microarray	212,213	hsa-miR-1182	NA		NGS	207
hsa-miR-5581-5p	NA		NGS	208	hsa-miR-93-5p	2	Enhances tumor cells survival	Reporter assay, western blot, qPCR	214
hsa-miR-4297	NA		NGS	208	hsa-miR-1178-5p	NA		NGS	207
hsa-miR-3614-3p	NA		NGS	208	hsa-miR-574-5p	NA		NGS	207
hsa-miR-5697	292, 660, 720		NGS	208	hsa-miR-562	NA		NGS	207
hsa-miR-3653-5p	NA		NGS	208	hsa-miR-4789-5p	NA		NGS	207
					hsa-miR-570-5p	NA		NGS	207
					hsa-miR-6867-5p	NA		NGS	215
					hsa-miR-548ba	NA		NGS	207
					hsa-miR-223-5p	NA		NGS	215
					hsa-miR-548ai	NA		NGS	207
					hsa-miR-223-3p	NA		NGS	207
					hsa-miR-548ag	NA		NGS	207
					hsa-miR-6801-5p	NA		NGS	207
					hsa-miR-548 m	NA		NGS	207
					hsa-miR-4697-3p	NA		NGS	207

Table 2 (Continued)

LATS1				LATS2					
miRNA	3'UTR location	F function	Validation method	Refs	miRNA	3'UTR location	Function	Validation method	Ref
					hsa-miR-510-3p	NA		NGS	207
					hsa-miR-373-3p	249, 346	Bypass of p53-dependent senescence	Reporter assay, western blot, qPCR, microarray	111,113,216
					hsa-miR-1256	NA		NGS	207
					hsa-miR-6739-5p	NA		NGS	207
					hsa-miR-181b-5p	2	Promotes proliferation and invasion	Reporter assay, western blot, qPCR	217
					hsa-miR-372-3p	247, 347	Bypass of p53-dependent senescence	Reporter assay, western blot, qPCR, microarray	104,111,216
					hsa-miR-202-3p	NA		NGS	207
					hsa-miR-215-3p	NA		NGS	207
					hsa-miR-6733-5p	NA		NGS	209
					hsa-miR-107	307	Proliferation and invasion of gastric cancer cells	Reporter assay, western blot, qPCR	
					hsa-miR-31-5p	678	Promotes lung cancer	Reporter assay, qPCR	109

reasons, it also suggests inherent differences between the kinases. Hence, as is also the case for regulation by miRs, evolution following the gene duplication event may have resulted in a broader spectrum of LATS2-binding partners, in order to increase its networking capabilities.

Among other things, choice of binding partners both is affected by, and affects, protein subcellular localization. Both LATS kinases have been detected on centrosomes,^{13,24} which are presumably associated with their role in regulation of mitosis. Both can also be tethered to the plasma membrane^{29,124} or localize to the cytoplasm.^{21,125} It is commonly assumed that LATS kinases phosphorylate YAP/TAZ in the cytoplasm. Yet, phosphorylation-dependent activation of LATS has been observed in the nucleus,^{51,126} while dephosphorylation of LATS1 and subsequent activation of YAP/TAZ can occur both in the nucleus and cytoplasm.¹²⁷ Furthermore, LATS1 was recently shown to localize to either the nucleus or the cytoplasm of mammary epithelial cells, depending on cell lineage.¹²⁸

Many of the functions unique to LATS2 have been attributed to its nuclear localization²⁰ and its interaction with nuclear proteins. Upon mitotic or oncogenic stress, nuclear LATS2 potentiates the activity of the tumor suppressor p53.^{35,36,89} In addition, nuclear LATS2 regulates chromatin dynamics by binding to polycomb repressive complex 2 (PRC2).¹²⁹ Nuclear LATS2 was shown to restrict oncogenic β -catenin signaling by disrupting the chromatin-bound β -catenin-BCL9 complex.¹³⁰ Accordingly, cardiac muscle-specific conditional knockout of *Lats2* generates an elevated Wnt signature,¹³¹ and LATS2 expression is inversely correlated with the levels of Wnt target genes in human colorectal cancer.¹³⁰ In contrast, in similar experiments LATS1 was not shown to bind chromatin or restrict β -catenin-induced transcription.¹³⁰

Also within the nucleus, LATS2 restrains steroid receptor transcriptional activity. In the prostate, LATS2 inhibits androgen receptor chromatin binding and transcriptional activity,¹³² while in breast tissue it modulates estrogen receptor (ER) activity.¹³³ More recently, LATS kinases have been shown to restrict the activity of ER by binding and promoting its degradation.¹²⁸ These studies implicate a nuclear function of LATS kinases in cell lineage commitment and in preventing the malignant progression of breast and prostate cancers.^{128,132}

Together, a spectrum of subcellular localizations enables LATS kinases to impact a variety of physiological functions.¹³⁴

Cell cycle Regulation and Apoptosis

Both LATS1 and LATS2 are involved in processes related to different stages of the cell cycle. Inhibition of CycE/CDK2 activity by LATS1 and LATS2 limits G1/S transition, under basal²¹ as well as potentially genotoxic conditions.^{120,121} In addition, LATS2 phosphorylation of DYRK1A promotes the assembly of the DREAM complex, which represses the expression of S-phase E2F target genes to promote senescence.¹³⁵

Multiple studies have linked LATS kinases to mitosis. Both LATS1 and LATS2 can bind to CDC25B¹³⁶ and phosphorylate CDC26,¹³⁷ master regulators of mitotic exit. Other studies suggest distinct modes of action for LATS1 and LATS2 during mitotic transition.¹³⁸⁻¹⁴⁰ In this scenario, LATS2 is

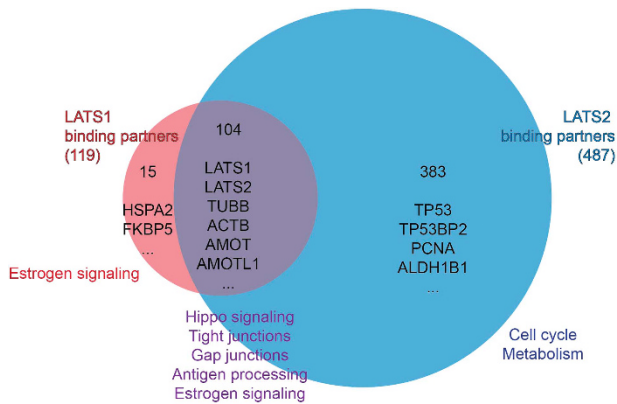


Figure 3 The protein interactome of LATS kinases. The Venn diagram depicts the overlap between putative LATS1 (119, light red circle) and LATS2 (487, light blue circle) binding partners in mammalian cells, as reported by Couzens *et al.*¹²³ and Wang *et al.*¹²² The numbers of common or exclusive binding partners and specific notable examples are indicated. Key-enriched biological processes (KEGG pathway database) are shown below the diagram

phosphorylated by Aurora A, and phospho-LATS2 translocates to the spindle along with LATS1, which phosphorylates Aurora B to ensure proper cytokinesis.³⁹ The above-described mitotic function of LATS kinases in mammalian cells is reminiscent of the role of yeast Dbf2,³ and may therefore represent an ancient dedication of the pathway to governing mitotic exit, which has been preserved in all metazoans.⁶

Since one of the functions of the mitotic exit pathway is to ensure that cytokinesis does not occur before proper partitioning of the genetic material, it may not be surprising that LATS1 and LATS2 are crucial in sensing mitotic stress that occurs in response to microtubule poisons such as nocodazole or during hyperproliferation owing to oncogene activation.^{35,89,141} These functions are strongly associated with the ability of LATS2 to promote activation of p53-dependent checkpoints, which may lead to either G1/S arrest or apoptosis.^{35,89} Indeed, extra chromosomes resulting from cytokinesis failure are sufficient to activate the Hippo pathway via the LATS2-p53 axis.¹⁴² Together with its ability to be transactivated by p53, this constitutes a LATS2-p53 tumor-suppressive positive feedback loop. In line with this, nuclear LATS2 can associate with p53 on the *p21* promoter to inhibit proliferation under stress conditions.¹⁴³ In this context, it is interesting to note that overexpression of kinase-dead LATS1 suppresses the ability of cells to induce p53 in response to mitotic stress.¹⁴⁴ Although this suggests that p53 is also sensitive to LATS1, it remains plausible that the effects of kinase-dead LATS1 might be due to dominant-negative inhibition of endogenous LATS2.

Alleviation of the MDM2-dependent inhibition of p53 can eliminate potentially transformed cells from the replicative pool.¹⁴⁵ Sustained K-RAS signaling promotes LATS1/MDM2/p53-dependent apoptosis.¹⁴⁶ Likewise, expression of oncogenic H-RAS facilitates LATS2-dependent phosphorylation of the pro-apoptotic protein ASPP1, and drives p53-dependent apoptosis.³⁶ Furthermore, ASPP1 can bind and inhibit LATS1-mediated phosphorylation of YAP, resulting in increased YAP activity.¹⁴⁷ This molecular wiring might exemplify another

mechanism by which LATS2 indirectly modulates LATS1 activity.

Additional means by which LATS1 can impact apoptosis have been suggested. LATS1 is activated by death receptors downstream of RASSF1A and MST2.¹⁴⁸ In turn, LATS1 increases the expression of the pro-apoptotic protein BAX.¹⁴⁰ Furthermore, LATS1 binds and enhances the protease activity of Omi/HtrA2,¹⁴⁹ a mitochondrial protein that is released into the cytoplasm during apoptosis.¹⁵⁰ LATS1 also feedbacks to inhibit MST2 pro-apoptotic activities by phosphorylating RAF1 on Ser259.¹⁵¹ This phosphorylation promotes the inhibitory binding of RAF1 to MST2 and restricts RAF1 binding and activation of MEK signaling. Thus, by phosphorylating RAF1, LATS1 restricts both ERK-dependent cellular proliferation and MST2-dependent apoptosis.¹⁵¹

LATS2 can downregulate the expression of the anti-apoptotic proteins BCL-xL and BCL2 by a mechanism that requires its kinase activity.¹⁵² Interestingly, the LATS2-p53 functional axis can regulate apoptosis not only through the downstream activation of p53 transcriptional target genes, but also by non-transcriptional mechanisms. In particular, following UV irradiation, LATS2 phosphorylates the p21 protein, encoded by a major p53 transcriptional target gene, to induce its degradation.¹⁵³ In this way, cells bypass cell cycle arrest and are directed to die. Of note, the p53 family member p73 can act as potent inducer of apoptosis when bound to YAP.^{154,155} Interestingly, in leukemic cells, LATS2 promotes the pro-apoptotic activity of the p73-YAP complex.¹⁵⁶

Surprisingly, inhibition of the p73-YAP complex by the LATS kinases can also have an anti-apoptotic effect.¹⁵⁷ LATS2 can also inhibit DNA damage-induced apoptosis through inhibitory phosphorylation of c-Abl.¹⁵⁸ The tyrosine kinase c-Abl is a strong inducer of the YAP-p73 pro-apoptotic axis in response to DNA damage.^{159,160} Specifically, phosphorylation of YAP Tyr357 by c-Abl potentiates the binding to p73 and induction of pro-apoptotic genes.¹⁵⁹ Since, c-Abl and YAP can contribute or inhibit apoptosis,^{159,161-163} their inhibition by LATS kinases results in opposing outcomes. Overall, this highlights an interplay between LATS, YAP, p73 and c-Abl, whose eventual impact on apoptosis is highly cell context-dependent.

In sum, LATS kinases govern cell fate by manipulating both cell cycle and apoptosis. This becomes particularly important when cells are faced with replicative or oncogenic stress and must be removed from the proliferative pool in a cost-effective manner and with the least harm to the organism as a whole.

Migration and EMT

Epithelial to mesenchymal transition (EMT) and migration, two important features in development and oncogenic transformation, are both regulated by LATS kinases. Mechanistically, human LATS1 and *Drosophila* Warts can bind to actin and inhibit actin polymerization.^{164,165} In mammals, reduced LATS expression promotes cell migration by altering the functional state of p53¹⁰¹ and by increasing the activity of the YAP/TAZ transcriptional module.¹⁶⁶ It is noteworthy that YAP/TAZ sensitivity to cytoskeleton and cell motility dynamics is critical to their role in mechanosensing, some of which is LATS-independent.¹⁶⁷ Overall, the inhibitory effects of LATS kinases

on cell migration are in line with their assignment as tumor suppressors.

Surprisingly, LATS2 can also potentiate the activity of tumor-promoting factors and augment EMT. In fact, LATS2 was reported to increase the cell invasive capacities of metastatic breast cancer cell lines harboring mutant p53.¹⁰⁰ In that case, the underlying mechanism was proposed to be the phosphorylation of SNAIL1 by LATS2, leading to increased SNAIL1 stability, nuclear localization and transcriptional activity.¹⁰⁰

Embryogenesis and Stem Cells

The LATS-YAP/TAZ axis plays a key role in patterning of mammalian embryos and determining cell lineage and differentiation, as exemplified in mouse studies.^{168–170} In support of this, inhibition of *Lats1* and *Lats2* expression in early embryos results in irreversible lineage misspecification and aberrant polarization of the inner cell mass.¹⁷⁰ Specifically, LATS2 seems to play a critical role in early embryogenesis. The pluripotent transcription factors OCT4 and NANOG bind a region near the *Lats2* (but not *Lats1*) gene, and repress *Lats2* expression.¹⁷¹ Accordingly, deletion of *Lats2* (but not *Lats1*) is embryonic lethal.¹³ Mouse embryonic stem cells (mESCs) lacking *Lats2* display an altered chromatin landscape that retains H3K4me3/H3K27me3 bivalent histone marks;⁹¹ this may be related to the ability of LATS2 to associate with PRC2 to promote H3K27 tri-methylation.¹²⁹ In line with this, mESCs lacking *Lats2* are deficient in both sustaining pluripotency and responding to differentiation signals,⁹¹ suggesting a cellular mechanism for the embryonic lethality phenotype of *Lats2*–/– mice. Importantly, inhibition of Yap/Taz activity fails to rescue the transcriptional defect of *Lats2*–/– mESCs; rather, the ability of LATS2 to maintain mESC homeostasis is mediated by the LATS2-p53 functional axis.⁹¹

Members of the miR-290 family of microRNAs (mouse orthologues of human miR-372/373) are highly expressed in undifferentiated mESCs, and can promote their proliferation by potentiating G1 to S transition. Downregulation of *Lats2* by these miRs contributes to pluripotency by interfering with the ability of LATS2 to promote G1 arrest.¹⁷² Intriguingly, on the other hand, reprogramming to induced pluripotent stem cells has been shown to be inhibited by LATS2 via a p53-independent mechanism that does not accelerate cell proliferation.¹⁷³

In more advanced stages of development, such as lineage-specific differentiation, LATS2 was shown to contribute to the differentiation process. For example, LATS2 inhibits preadipocyte proliferation and promotes adipocyte differentiation by inducing a PPAR γ pro-adipogenic transcriptional program.^{174,175} Although this was shown to be mediated by cytoplasmic retention of TAZ, it still remains to be investigated whether this function is shared also with LATS1. Altogether, LATS2 plays a unique role in embryonic stem cells and in differentiation.

Tissue-Specific Roles of LATS Kinases

YAP/TAZ are key regulators of liver size and, when hyperactivated, can drive liver tumorigenesis.^{163,176} Thus, it is not

surprising that inactivation of both LATS kinases in liver cells leads to failure of proper differentiation and augments proliferation.^{177,178} Embryonic deletion of both kinases in the mouse liver results in neonate lethality.¹⁷⁷ In adult livers, acute deletion of *Lats1/2* results in dedifferentiation of hepatocytes into immature biliary epithelial cells, fibrosis and lethal liver impairment.¹⁷⁸ LATS2 also has additional hepatic functions, which are not mediated by YAP/TAZ activity and are not shared with LATS1. For example LATS2, but not LATS1, inhibits hepatic cholesterol accumulation by binding and quenching the transcriptional activity of SREBP1 and SREBP2, transcription factors that are master regulators of lipid and cholesterol homeostasis.⁹⁰ Consequently, mice lacking *Lats2* in the liver have deregulated cholesterol metabolism and are prone to fatty liver disease, suggesting that LATS2 plays a role in metabolic homeostasis.⁸⁵

As in the liver, *Lats1* and *Lats2* are essential also in the kidney ureteric bud lineage: deletion of both *Lats* genes results in severe defects in branching morphogenesis, deregulated cell polarity and hyperactivation of YAP and TAZ.¹⁷⁹

In the heart, inactivation of both LATS kinases reflects a role for LATS in restricting cardiomyocyte renewal and regeneration.¹⁸⁰ Interestingly, the individual functions of each kinase in cardiomyocytes may not be fully redundant, since inactivation of *Lats2* is sufficient to cause myocardial expansion¹³¹ and *Lats2* overexpression negatively regulates ventricular mass in the heart.¹⁸¹ Furthermore, the kinase activity of LATS2 is required for YAP's ability to regulate coronary vascular formation.¹⁸² In line with these observations, expression of *Lats2*, but not *Lats1*, promotes apoptosis in cultured cardiomyocytes.¹⁸¹

Both LATS kinases are expressed ubiquitously throughout different human tissues,¹³⁴ except the spleen in which neither kinase is detected (Human Protein Atlas available at: www.proteinatlas.org).¹⁸³ Protein levels differ, with very few tissues showing similar trends of expression between LATS1 and LATS2. While LATS1 protein is detected in high levels throughout most tissues, LATS2 protein levels seem to vary, with highest expression in the gastrointestinal tract and the brain.¹⁸³ The functional impact of each kinase in different tissues remains to be further examined.

Conclusion

The great interest in the Hippo pathway components has generated a wealth of new information. Yet, many of the studies have focused exclusively on the pathway downstream effectors YAP and TAZ, and LATS kinases function, if addressed at all, has been examined merely in the light of their effect on YAP/TAZ. Furthermore, most studies employ only one *LATS* gene or protein, making it difficult to identify true differences between LATS1 and LATS2. In this review, we have tried to tease out and analyze discrete characteristics of LATS1 and LATS2, as recorded in the literature to date. We show that although, as expected, there does exist substantial functional overlap between these two paralogs, many of their features are nevertheless distinct.

The *LATS* duplication event set the stage for evolution to 'teach' us about LATS function. Gene duplication establishes a platform for exploring genetic novelty, while augmenting

genomic robustness by buffering paralogs.¹⁸⁴ Actually, evolution pushes the duplicated genes toward diversification, as total redundancy among duplicates is both genetically unfavorable and potentially disruptive to biochemical pathways due to dosage sensitivity.¹⁸⁵ Together, this suggests that the second copy is liberated from selective pressure and can evolve novel functions, as long as any ensuing functional losses can be complemented by the other copy.¹⁸⁶ Interestingly, alterations in gene expression often precede functional changes in paralog evolution.¹⁸⁷

LATS1 and LATS2 embody this evolutionary format. The most striking differences between LATS1 and LATS2 occur on the transcript level. The difference in transcription factors regulating *LATS1* versus *LATS2* may represent the necessity to keep tight reins on the 'brakes' and 'gas' of proliferation signals by maintaining proficient levels of LATS in both conditions. Further indication of tight regulation on the transcriptional level is evident in their 3'UTRs: *LATS2* contains a long, highly regulated 3'UTR, whereas the shorter *LATS1* UTR may evade, at least to some extent, negative (miR) or positive (RNA-binding proteins) regulation. Interestingly, lengthening of 3'UTRs has been associated with increased morphological complexity over evolution¹⁸⁸ and might be linked to observations that regulatory motifs in UTRs are often conserved in genes within similar functional pathways.¹⁸⁹ It will be interesting to examine the possibility that *LATS2* has evolved functions that enable it to be co-regulated within the context of a larger functional gene family; this concept is illustrated by the observation that miR-372/3 commonly targets *LATS2* as well as other factors that are critical in stem cell differentiation.¹⁹⁰

The divergent expression patterns of *LATS1* and *LATS2* might contribute to their likelihood of encountering distinct binding partners that, in turn, might tether the two LATS proteins to different cellular localizations and facilitate their distinct functions. This is illustrated by the specialized connection of LATS1 to estrogen signaling, and of LATS2 to stem cell differentiation and to the p53 network. It is important to note, however, that even in these 'dedicated' interactions, there is substantial redundancy between LATS1 and LATS2, which probably underpins their ability to serve as partial backups for each other. Thus, LATS2-specific interacting partners are not enriched in estrogen signaling,^{122,123} yet both LATS1 and LATS2 have been shown to regulate the stability of the ER.¹²⁸ Likewise, p53 exclusively binds LATS2 but not LATS1¹²² and transcriptionally activates the *LATS2* but not *LATS1* promoter,⁸⁹ but LATS1 can nevertheless modulate p53-dependent apoptosis.¹⁴⁶ Similarly, OCT4 and NANOG repress *Lats2* but not *Lats1* expression,¹⁷¹ which is essential for proper embryonic development, but *Lats1* is also important for embryogenesis, since re-expression can rescue *Lats* depletion phenotype in early embryogenesis.¹⁷⁰

In fact, a considerable proportion of LATS functions intersect on different elements of the same pathway. For instance, LATS2 is phosphorylated by Aurora A and LATS1 phosphorylates Aurora B.³⁹ Both Aurora kinases impact mitotic progression, however Aurora A associates with the spindle poles to regulate entry into mitosis and spindle assembly, whereas Aurora B regulates chromosome cohesion and cytokinesis.¹⁹¹ Therefore, although the LATS1/2-specific

mechanisms may have diverged, in most cases the broader physiological 'agenda' of the LATS kinases has been retained. Probably for these reasons, both LATS genes undergo selective silencing in cancer.

The LATS kinases restrict the 'canonical' Hippo effectors YAP and TAZ, and also control 'non-canonical' novel signaling pathways to integrate critical cellular processes. However, the distinction between 'canonical' and emergent LATS functions quickly becomes blurry. Some novel activities of LATS indirectly impinge on YAP/TAZ functions.^{36,90,147} Additionally, due to YAP-LATS2 feedback, hyperactivation of YAP is expected to also inherently affect LATS2 non-canonical functions. Furthermore, LATS2 has been shown to act upstream to LATS1 and enhance its kinase activity toward non-canonical effectors.³⁹ Many non-canonical LATS kinase-regulated events are not associated with the HXRXXS/T consensus LATS phosphorylation motif,¹⁹² suggesting that in these cases LATS substrate selection is shaped by factors other than just amino-acid sequence. Thus, complicated and multi-directional mechanisms are in place, even within the Hippo module itself.

Consequently, LATS-dependent cell fate decisions are the sum total of innumerable signaling inputs and outputs, the weight of each signal being determined (among many other factors) by cell density, cell type, developmental stage, neighboring cells and whether the cells are normal or transformed. Together, these complex signals lead to a vast and sometimes contradictory spectrum of LATS functions and activities. Some of the most striking examples are illustrated by the ability of LATS1 and LATS2 to both promote and inhibit apoptosis,^{139,152,153,157,158} and the ability of LATS2 to both augment and inhibit differentiation^{91,129} or cellular migration.^{100,101}

Many open questions still remain to be answered. More meticulous studies need to be performed to accurately define LATS1 and LATS2 shared versus distinct functions. Advances in understanding LATS signaling may aid to resolve basic scientific enigmas such as how kinases choose phosphorylation substrates, how signaling pathways balance cell division, differentiation and proliferation, and how these pathways are skewed during cancerous transformation. Moreover, deciphering the details of LATS-mediated tumor suppression will hopefully elucidate opportunities for improved early detection, prognostication and treatment of cancer.

Conflict of Interest

The authors declare no conflict of interest.

1. Visintin R, Amon A. Regulation of the mitotic exit protein kinases Cdc15 and Dbf2. *Mol Biol Cell* 2001; **12**: 2961–2974.
2. Rock JM, Lim D, Stach L, Ogradowicz RW, Keck JM, Jones MH *et al*. Activation of the yeast Hippo pathway by phosphorylation-dependent assembly of signaling complexes. *Science* 2013; **340**: 871–875.
3. Bardin AJ, Amon A. Men and sin: what's the difference? *Nat Rev Mol Cell Biol* 2001; **2**: 815–826.
4. Chan EHY, Nousiainen M, Chalamalasetty RB, Schäfer A, Nigg EA, Silljé HHW. The Ste20-like kinase Mst2 activates the human large tumor suppressor kinase Lats1. *Oncogene* 2005; **24**: 2076–2086.
5. Cai Q, Wang W, Gao Y, Yang Y, Zhu Z, Fan Q. Ce-wts-1 plays important roles in *Caenorhabditis elegans* development. *FEBS Lett* 2009; **583**: 3158–3164.
6. Hilman D, Gat U. The evolutionary history of YAP and the Hippo/YAP pathway. *Mol Biol Evol* 2011; **28**: 2403–2417.

7. Bryant PJ, Watson KL, Justice RW, Woods DF. Tumor suppressor genes encoding proteins required for cell interactions and signal transduction in *Drosophila*. *Development* 1993; **239**: 239–249.
8. Justice RW, Zilian O, Woods DF, Noll M, Bryant PJ. The *Drosophila* tumor suppressor gene warts encodes a homolog of human myotonic dystrophy kinase and is required for the control of cell shape and proliferation. *Genes Dev* 1995; **9**: 534–546.
9. Zhao B, Li L, Lei Q, Guan K-L. The Hippo-YAP pathway in organ size control and tumorigenesis: an updated version. *Genes Dev* 2010; **24**: 862–874.
10. Dewey EB, Sanchez D, Johnston CA. Warts phosphorylates mud to promote pins-mediated mitotic spindle orientation in *Drosophila*, independent of Yorkie. *Curr Biol* 2015; **25**: 2751–2762.
11. Staley BK, Irvine KD. Hippo signaling in *Drosophila*: recent advances and insights. *Dev Dyn* 2012; **241**: 3–15.
12. Tao W, Zhang S, Turenchalk GS, Stewart RA St, John MA, Chen W *et al*. Human homologue of the *Drosophila melanogaster* lats tumour suppressor modulates CDC2 activity. *Nat Genet* 1999; **21**: 177–181.
13. McPherson JP, Tambllyn L, Elia A, Migon E, Shehabeldin A, Matysiak-Zablocki E *et al*. Lats2/Kpm is required for embryonic development, proliferation control and genomic integrity. *EMBO J* 2004; **23**: 3677–3688.
14. Yabuta N, Okada N, Ito A, Hosomi T, Nishihara S, Sasayama Y *et al*. Lats2 is an essential mitotic regulator required for the coordination of cell division. *J Biol Chem* 2007; **282**: 19259–19271.
15. St John MA, Tao W, Fei X, Fukumoto R, Carcangiu ML, Brownstein DG *et al*. Mice deficient of Lats1 develop soft-tissue sarcomas, ovarian tumours and pituitary dysfunction. *Nat Genet* 1999; **21**: 182–186.
16. Zancanato F, Cordenonsi M, Piccolo S. YAP/TAZ at the roots of cancer. *Cancer Cell* 2016; **29**: 783–803.
17. Meng Z, Moroishi T, Guan K-L. Mechanisms of Hippo pathway regulation. *Genes Dev* 2016; **30**: 1–17.
18. Avruch J, Zhou D, Fitamant J, Bardeesy N, Mou F, Barrufet LR. Protein kinases of the Hippo pathway: regulation and substrates. *Semin Cell Dev Biol* 2012; **23**: 770–784.
19. Hori T, Takaori-Kondo A, Kamikubo Y, Uchiyama T. Molecular cloning of a novel human protein kinase, kpm, that is homologous to warts/lats, a *Drosophila* tumor suppressor. *Oncogene* 2000; **19**: 3101–3109.
20. Yabuta N, Fujii T, Copeland NG, Gilbert DJ, Jenkins NA, Nishiguchi H *et al*. Structure, expression, and chromosome mapping of LATS2, a mammalian homologue of the *Drosophila* tumor suppressor gene lats/warts. *Genomics* 2000; **63**: 263–270.
21. Li Y, Pei J, Xia H, Ke H, Wang H, Tao W. Lats2, a putative tumor suppressor, inhibits G1/S transition. *Oncogene* 2003; **22**: 4398–4405.
22. Yabuta N, Mukai S, Okamoto A, Okuzaki D, Suzuki H, Torigata K *et al*. N-terminal truncation of Lats1 causes abnormal cell growth control and chromosomal instability. *J Cell Sci* 2013; **126**: 508–520.
23. Kim M, Kim M, Park S-J, Lee C, Lim D-S. Role of Angiomotin-like 2 mono-ubiquitination on YAP inhibition. *EMBO Rep* 2016; **17**: 64–78.
24. Nishiyama Y, Hirota T, Morisaki T, Hara T, Marumoto T, Iida S *et al*. A human homologue of *Drosophila* warts tumor suppressor, h-warts, localized to mitotic apparatus and specifically phosphorylated during mitosis. *FEBS Lett* 1999; **459**: 159–165.
25. Sudol M. Newcomers to the WW domain-mediated network of the Hippo tumor suppressor pathway. *Genes Cancer* 2010; **1**: 1115–1118.
26. Pearce LR, Komander D, Alessi DR. The nuts and bolts of AGC protein kinases. *Nat Rev Mol Cell Biol* 2010; **11**: 9–22.
27. Hoa L, Kulaberoglu Y, Gundogdu R, Cook D, Mavis M, Gomez M *et al*. The characterization of LATS2 kinase regulation in Hippo-YAP signalling. *Cell Signal* 2016; **28**: 488–497.
28. Hergovich A, Schmitz D, Hemmings BA. The human tumour suppressor LATS1 is activated by human MOB1 at the membrane. *Biochem Biophys Res Commun* 2006; **345**: 50–58.
29. Yin F, Yu J, Zheng Y, Chen Q, Zhang N, Pan D. Spatial organization of Hippo signaling at the plasma membrane mediated by the tumor suppressor Merlin/NF2. *Cell* 2013; **154**: 1342–1355.
30. Praskova M, Xia F, Avruch J. MOBKL1A/MOBKL1B phosphorylation by MST1 and MST2 inhibits cell proliferation. *Curr Biol* 2008; **18**: 311–321.
31. Zhou D, Conrad C, Xia F, Park J-S, Payer B, Yin Y *et al*. Mst1 and Mst2 maintain hepatocyte quiescence and suppress hepatocellular carcinoma development through inactivation of the Yap1 oncogene. *Cancer Cell* 2009; **16**: 425–438.
32. Meng Z, Moroishi T, Mottier-Pavie V, Plouffe SW, Hansen CG, Hong AW *et al*. MAP4K family kinases act in parallel to MST1/2 to activate LATS1/2 in the Hippo pathway. *Nat Commun* 2015; **6**: 8357.
33. Kim M, Kim M, Lee S, Kuninaka S, Saya H, Lee H *et al*. cAMP/PKA signalling reinforces the LATS-YAP pathway to fully suppress YAP in response to actin cytoskeletal changes. *EMBO J* 2013; **32**: 1543–1555.
34. Okada N, Yabuta N, Suzuki H, Aylon Y, Oren M, Nojima H. A novel Chk1/2-Lats2-14-3-3 signaling pathway regulates P-body formation in response to UV damage. *J Cell Sci* 2011; **124**: 57–67.
35. Aylon Y, Yabuta N, Besserglick H, Buganim Y, Rotter V, Nojima H *et al*. Silencing of the Lats2 tumor suppressor overrides a p53-dependent oncogenic stress checkpoint and enables mutant H-Ras-driven cell transformation. *Oncogene* 2009; **28**: 4469–4479.
36. Aylon Y, Ofir-Rosenfeld Y, Yabuta N, Lapi E, Nojima H, Lu X *et al*. The Lats2 tumor suppressor augments p53-mediated apoptosis by promoting the nuclear proapoptotic function of ASPP1. *Genes Dev* 2010; **24**: 2420–2429.
37. Wang G, Jiang Q, Zhang C. The role of mitotic kinases in coupling the centrosome cycle with the assembly of the mitotic spindle. *J Cell Sci* 2014; **127**: 4111–4122.
38. Toji S, Yabuta N, Hosomi T, Nishihara S, Kobayashi T, Suzuki S *et al*. The centrosomal protein Lats2 is a phosphorylation target of Aurora-A kinase. *Genes Cells* 2004; **9**: 383–397.
39. Yabuta N, Mukai S, Okada N, Aylon Y, Nojima H. The tumor suppressor Lats2 is pivotal in Aurora A and Aurora B signaling during mitosis. *Cell Cycle* 2011; **10**: 2724–2736.
40. Zhang L, Iyer J, Chowdhury A, Ji M, Xiao L, Yang S *et al*. KIBRA regulates aurora kinase activity and is required for precise chromosome alignment during mitosis. *J Biol Chem* 2012; **287**: 34069–34077.
41. Humbert N, Navaratnam N, Augert A, Da Costa M, Martien S, Wang J *et al*. Regulation of ploidy and senescence by the AMPK-related kinase NUAK1. *EMBO J* 2010; **29**: 376–386.
42. Xiao L, Chen Y, Ji M, Dong J. KIBRA regulates Hippo signaling activity via interactions with large tumor suppressor kinases. *J Biol Chem* 2011; **286**: 7788–7796.
43. Huntoon CJ, Nye MD, Geng L, Peterson KL, Flatten KS, Haluska P *et al*. Heat shock protein 90 inhibition depletes LATS1 and LATS2, two regulators of the mammalian Hippo tumor suppressor pathway. *Cancer Res* 2010; **70**: 8642–8650.
44. Das Thakur M, Feng Y, Jagannathan R, Seppa MJ, Skeath JB, Longmore GD. Ajuba LIM proteins are negative regulators of the Hippo signaling pathway. *Curr Biol* 2010; **20**: 657–662.
45. Rauskolb C, Pan G, Reddy BVVG, Oh H, Irvine KD. Zyxin links fat signaling to the Hippo pathway. *PLoS Biol* 2011; **9**: e1000624.
46. Bae SJ, Kim M, Kim S-H, Kwon YE, Lee J-H, Kim J *et al*. NEDD4 controls intestinal stem cell homeostasis by regulating the Hippo signalling pathway. *Nat Commun* 2015; **6**: 6314.
47. Salah Z, Cohen S, Itzhaki E, Aqeilan RI. NEDD4 E3 ligase inhibits the activity of the Hippo pathway by targeting LATS1 for degradation. *Cell Cycle* 2013; **12**: 3817–3823.
48. Ho KC, Zhou Z, She Y-M, Chun A, Cyr TD, Yang X. Itch E3 ubiquitin ligase regulates large tumor suppressor 1 stability [corrected]. *Proc Natl Acad Sci USA* 2011; **108**: 4870–4875.
49. Salah Z, Melino G, Aqeilan RI. Negative regulation of the Hippo pathway by E3 ubiquitin ligase ITCH is sufficient to promote tumorigenicity. *Cancer Res* 2011; **71**: 2010–2020.
50. Yeung B, Ho K-C, Yang X. WWP1 E3 ligase targets LATS1 for ubiquitin-mediated degradation in breast cancer cells. *PLoS ONE* 2013; **8**: e61027.
51. Li W, Cooper J, Zhou L, Yang C, Erdjument-Bromage H, Zagzag D *et al*. Merlin/NF2 loss-driven tumorigenesis linked to CRL4(DCAF1)-mediated inhibition of the Hippo pathway kinases Lats1 and 2 in the nucleus. *Cancer Cell* 2014; **26**: 48–60.
52. Ma B, Chen Y, Chen L, Cheng H, Mu C, Li J *et al*. Hypoxia regulates Hippo signalling through the SIAH2 ubiquitin E3 ligase. *Nat Cell Biol* 2015; **17**: 95–103.
53. Nakayama K, Qi J, Ronai Z. The ubiquitin ligase Siah2 and the hypoxia response. *Mol Cancer Res* 2009; **7**: 443–451.
54. Takahashi A, Ohtani N, Yamakoshi K, Iida S, Tahara H, Nakayama K *et al*. Mitogenic signalling and the p16INK4a-Rb pathway cooperate to enforce irreversible cellular senescence. *Nat Cell Biol* 2006; **8**: 1291–1297.
55. Cooke IE, Shelling AN, Le Meuth VG, Chernack ML, Ganesan TS. Allele loss on chromosome arm 6q and fine mapping of the region at 6q27 in epithelial ovarian cancer. *Genes Chromosomes Cancer* 1996; **15**: 223–233.
56. Lee JH, Kavanagh JJ, Wildrick DM, Wharton JT, Blick M. Frequent loss of heterozygosity on chromosomes 6q, 11, and 17 in human ovarian carcinomas. *Cancer Res* 1990; **50**: 2724–2728.
57. Mazurenko N, Attaleb M, Gritsko T, Semjonova L, Pavlova L, Sakharova O *et al*. High resolution mapping of chromosome 6 deletions in cervical cancer. *Oncol Rep* 1999; **6**: 859–863.
58. Fujii H, Zhou W, Gabrielson E. Detection of frequent allelic loss of 6q23-q25.2 in microdissected human breast cancer tissues. *Genes Chromosomes Cancer* 1996; **16**: 35–39.
59. Theille M, Seitz S, Arnold W, Jandrig B, Frege R, Schlag PM *et al*. A defined chromosome 6q fragment (at D6S310) harbors a putative tumor suppressor gene for breast cancer. *Oncogene* 1996; **13**: 677–685.
60. Noviello C, Courjal F, Theillet C. Loss of heterozygosity on the long arm of chromosome 6 in breast cancer: possibly four regions of deletion. *Clin Cancer Res* 1996; **2**: 1601–1606.
61. Lee EY, To H, Shew JY, Bookstein R, Scully P, Lee WH. Inactivation of the retinoblastoma susceptibility gene in human breast cancers. *Science* 1988; **241**: 218–221.
62. Sato T, Saito H, Morita R, Koi S, Lee JH, Nakamura Y. Allelotyping of human ovarian cancer. *Cancer Res* 1991; **51**: 5118–5122.
63. Wang HP, Rogler CE. Deletions in human chromosome arms 11p and 13q in primary hepatocellular carcinomas. *Cytogenet Cell Genet* 1988; **48**: 72–78.
64. Chen C-F, Yeh S-H, Chen D-S, Chen P-J, Jou Y-S. Molecular genetic evidence supporting a novel human hepatocellular carcinoma tumor suppressor locus at 13q12.11. *Genes Chromosomes Cancer* 2005; **44**: 320–328.
65. Strazisar M, Mlakar V, Glavac D. LATS2 tumour specific mutations and down-regulation of the gene in non-small cell carcinoma. *Lung Cancer* 2009; **64**: 257–262.
66. Ouillet P, Erba H, Kujawski L, Kaminski M, Shedden K, Malek SN. Integrated genomic profiling of chronic lymphocytic leukemia identifies subtypes of deletion 13q14. *Cancer Res* 2008; **68**: 1012–1021.
67. Yu T, Bachman J, Lai Z-C. Mutation analysis of large tumor suppressor genes LATS1 and LATS2 supports a tumor suppressor role in human cancer. *Protein Cell* 2015; **6**: 6–11.
68. Bonilla X, Parmentier L, King B, Bezrukov F, Kaya G, Zoete V *et al*. Genomic analysis identifies new drivers and progression pathways in skin basal cell carcinoma. *Nat Genet* 2016; **48**: 398–406.

69. Ishizaki K, Fujimoto J, Kumimoto H, Nishimoto Y, Shimada Y, Shinoda M *et al.* Frequent polymorphic changes but rare tumor specific mutations of the *LATS2* gene on 13q11-12 in esophageal squamous cell carcinoma. *Int J Oncol* 2002; **21**: 1053–1057.
70. Esteller M. Epigenetic gene silencing in cancer: the DNA hypermethylome. *Hum Mol Genet* 2007; **16**(Spec No 1): R50–R59.
71. Reddy VR, Annamalai T, Narayanan V, Ramanathan A. Hypermethylation of promoter region of LATS1—a CDK interacting protein in oral squamous cell carcinomas—a pilot study in India. *Asian Pac J Cancer Prev* 2015; **16**: 1599–1603.
72. Wierzbicki PM, Adrych K, Kartanowicz D, Stanislawowski M, Kowalczyk A, Godlewski J *et al.* Underexpression of LATS1 TSG in colorectal cancer is associated with promoter hypermethylation. *World J Gastroenterol* 2013; **19**: 4363–4373.
73. Chen K-H, He J, Wang D-L, Cao J-J, Li M-C, Zhao X-M *et al.* Methylation-associated inactivation of LATS1 and its effect on demethylation or overexpression on YAP and cell biological function in human renal cell carcinoma. *Int J Oncol* 2014; **45**: 2511–2521.
74. Yao F, Liu H, Li Z, Zhong C, Fang W. Down-regulation of LATS2 in non-small cell lung cancer promoted the growth and motility of cancer cells. *Tumour Biol* 2015; **36**: 2049–2057.
75. Jiménez-Velasco A, Román-Gómez J, Agirre X, Barrios M, Navarro G, Vázquez I *et al.* Downregulation of the large tumor suppressor 2 (LATS2/KPM) gene is associated with poor prognosis in acute lymphoblastic leukemia. *Leukemia* 2005; **19**: 2347–2350.
76. Takahashi Y, Miyoshi Y, Takahata C, Irahara N, Taguchi T, Tamaki Y *et al.* Down-regulation of LATS1 and LATS2 mRNA expression by promoter hypermethylation and its association with biologically aggressive phenotype in human breast cancers. *Clin Cancer Res* 2005; **11**: 1380–1385.
77. Steinmann K, Sandner A, Schagdarsurengin U, Dammann RH. Frequent promoter hypermethylation of tumor-related genes in head and neck squamous cell carcinoma. *Oncol Rep* 2009; **22**: 1519–1526.
78. Jiang Z, Li X, Hu J, Zhou W, Jiang Y, Li G *et al.* Promoter hypermethylation-mediated down-regulation of LATS1 and LATS2 in human astrocytoma. *Neurosci Res* 2006; **56**: 450–458.
79. Sasaki H, Hikosaka Y, Kawano O, Yano M, Fujii Y. Hypermethylation of the large tumor suppressor genes in Japanese lung cancer. *Oncol Lett* 2010; **1**: 303–307.
80. Ladiz MAR, Najafi M, Kordi-Tamandani DM. Contribution of LATS1 and LATS2 promoter methylation in OSCC development. *J Cell Commun Signal* 2017; **11**: 49–55.
81. Oh J-E, Ohta T, Satomi K, Foll M, Durand G, McKay J *et al.* Alterations in the NF2/LATS1/LATS2/YAP pathway in schwannomas. *J Neuropathol Exp Neurol* 2015; **74**: 952–959.
82. Malik SA, Khan MS, Dar M, Hussain MU, Shah MA, Shafi SM *et al.* Molecular alterations and expression dynamics of LATS1 and LATS2 genes in non-small-cell lung carcinoma. *Pathol Oncol Res* 2017.
83. Liang R, Lin Y, Yuan C-L, Liu Z-H, Li Y-Q, Luo X-L *et al.* The clinical significance and biological function of large tumour suppressor 2 in hepatocellular carcinoma. *Cell Prolif* 2017; **50**.
84. Wan L, Sun M, Liu G-J, Wei C-C, Zhang E-B, Kong R *et al.* Long noncoding RNA PVT1 promotes non-small cell lung cancer cell proliferation through epigenetically regulating LATS2 expression. *Mol Cancer Ther* 2016; **15**: 1082–1094.
85. Huang M, Hou J, Wang Y, Xie M, Wei C, Nie F *et al.* Long noncoding RNA LINC00673 is activated by SP1 and exerts oncogenic properties by interacting with LSD1 and EZH2 in gastric cancer. *Mol Ther* 2017; **25**: 1014–1026.
86. Li W, Sun M, Zang C, Ma P, He J, Zhang M *et al.* Upregulated long non-coding RNA AGAP2-AS1 represses LATS2 and KLF2 expression through interacting with EZH2 and LSD1 in non-small-cell lung cancer cells. *Cell Death Dis* 2016; **7**: e2225.
87. Kostic C, Shaw PH. Isolation and characterization of sixteen novel p53 response genes. *Oncogene* 2000; **19**: 3978–3987.
88. Aylon Y, Oren M. The Paradox of p53: what, how, and why? *Cold Spring Harb Perspect Med* 2016; **6**: a026328.
89. Aylon Y, Michael D, Shmueli A, Yabuta N, Nojima H, Oren M. A positive feedback loop between the p53 and Lats2 tumor suppressors prevents tetraploidization. *Genes Dev* 2006; **20**: 2687–2700.
90. Aylon Y, Gershoni A, Rotkopf R, Biton IE, Porat Z, Koh AP *et al.* The LATS2 tumor suppressor inhibits SREBP and suppresses hepatic cholesterol accumulation. *Genes Dev* 2016; **30**: 786–797.
91. Aylon Y, Sarver A, Tovy A, Ainbinder E, Oren M. Lats2 is critical for the pluripotency and proper differentiation of stem cells. *Cell Death Differ* 2014; **21**: 624–633.
92. Li W, Wang L, Katoh H, Liu R, Zheng P, Liu Y. Identification of a tumor suppressor relay between the FOXP3 and the Hippo pathways in breast and prostate cancers. *Cancer Res* 2011; **71**: 2162–2171.
93. Du X, Shi H, Li J, Dong Y, Liang J, Ye J *et al.* Mst1/Mst2 regulate development and function of regulatory T cells through modulation of Foxo1/Foxo3 stability in autoimmune disease. *J Immunol* 2014; **192**: 1525–1535.
94. Park G-S, Oh H, Kim M, Kim T, Johnson RL, Irvine KD *et al.* An evolutionarily conserved negative feedback mechanism in the Hippo pathway reflects functional difference between LATS1 and LATS2. *Oncotarget* 2016; **7**: 24063–24075.
95. Moroiishi T, Park HW, Qin B, Chen Q, Meng Z, Plouffe SW *et al.* A YAP/TAZ-induced feedback mechanism regulates Hippo pathway homeostasis. *Genes Dev* 2015; **29**: 1271–1284.
96. Chen Q, Zhang N, Xie R, Wang W, Cai J, Choi K-S *et al.* Homeostatic control of Hippo signaling activity revealed by an endogenous activating mutation in YAP. *Genes Dev* 2015; **29**: 1285–1297.
97. Sansregret L, Nepveu A. The multiple roles of CUX1: insights from mouse models and cell-based assays. *Gene* 2008; **412**: 84–94.
98. Siam R, Harada R, Cadieux C, Battat R, Vadnais C, Nepveu A. Transcriptional activation of the Lats1 tumor suppressor gene in tumors of CUX1 transgenic mice. *Mol Cancer* 2009; **8**: 60.
99. Zhang Y, Hu C-F, Chen J, Yan L-X, Zeng Y-X, Shao J-Y. LATS2 is de-methylated and overexpressed in nasopharyngeal carcinoma and predicts poor prognosis. *BMC Cancer* 2010; **10**: 538.
100. Zhang K, Rodriguez-Aznar E, Yabuta N, Owen RJ, Mingot JM, Nojima H *et al.* Lats2 kinase potentiates Snail1 activity by promoting nuclear retention upon phosphorylation. *EMBO J* 2012; **31**: 29–43.
101. Furth N, Bossel Ben-Moshe N, Pozniak Y, Porat Z, Geiger T, Domany E *et al.* Down-regulation of LATS kinases alters p53 to promote cell migration. *Genes Dev* 2015; **29**: 2325–2330.
102. Zhu R, Lacovino M, Mahen E, Kyba M, Matin A. Transcripts that associate with the RNA binding protein, DEAD-END (DND1), in embryonic stem (ES) cells. *BMC Mol Biol* 2011; **12**: 37.
103. Wu Q, Ma Q, Shehadeh LA, Wilson A, Xia L, Yu H *et al.* Expression of the Argonaute protein PiwiL2 and piRNAs in adult mouse mesenchymal stem cells. *Biochem Biophys Res Commun* 2010; **396**: 915–920.
104. Kedde M, Strasser MJ, Boldajipour B, Oude Vrielink JAF, Slanchev K, le Sage C *et al.* RNA-binding protein Dnd1 inhibits microRNA access to target mRNA. *Cell* 2007; **131**: 1273–1286.
105. Ali S, Karki N, Bhattacharya C, Zhu R, MacDuff DA, Stenglein MD *et al.* APOBEC3 inhibits DEAD-END function to regulate microRNA activity. *BMC Mol Biol* 2013; **14**: 16.
106. Lee HH, Vo M-T, Kim HJ, Lee UH, Kim CW, Kim HK *et al.* Stability of the LATS2 tumor suppressor gene is regulated by tristetraprolin. *J Biol Chem* 2010; **285**: 17329–17337.
107. Hua K, Jin J, Zhao J, Song J, Song H, Li D *et al.* miR-135b, upregulated in breast cancer, promotes cell growth and disrupts the cell cycle by regulating LATS2. *Int J Oncol* 2016; **48**: 1997–2006.
108. Lin C-W, Chang Y-L, Chang Y-C, Lin J-C, Chen C-C, Pan S-H *et al.* MicroRNA-135b promotes lung cancer metastasis by regulating multiple targets in the Hippo pathway and LZTS1. *Nat Commun* 2013; **4**: 1877.
109. Liu X, Sempere LF, Ouyang H, Memoli VA, Andrew AS, Luo Y *et al.* MicroRNA-31 functions as an oncogenic microRNA in mouse and human lung cancer cells by repressing specific tumor suppressors. *J Clin Invest* 2010; **120**: 1298–1309.
110. Mitamura T, Watari H, Wang L, Kanno H, Kitagawa M, Hassan MK *et al.* microRNA 31 functions as an endometrial cancer oncogene by suppressing Hippo tumor suppressor pathway. *Mol Cancer* 2014; **13**: 97.
111. Voorhoeve PM, le Sage C, Schrier M, Gillis AJM, Stoop H, Nagel R *et al.* A genetic screen implicates miRNA-372 and miRNA-373 as oncogenes in testicular germ cell tumors. *Cell* 2006; **124**: 1169–1181.
112. Cho WJ, Shin JM, Kim JS, Lee MR, Hong KS, Lee J-H *et al.* miR-372 regulates cell cycle and apoptosis of ags human gastric cancer cell line through direct regulation of LATS2. *Mol Cells* 2009; **28**: 521–527.
113. Lee K-H, Goan Y-G, Hsiao M, Lee C-H, Jian S-H, Lin J-T *et al.* MicroRNA-373 (miR-373) post-transcriptionally regulates large tumor suppressor, homolog 2 (LATS2) and stimulates proliferation in human esophageal cancer. *Exp Cell Res* 2009; **315**: 2529–2538.
114. Luo Z-L, Luo H-J, Fang C, Cheng L, Huang Z, Dai R *et al.* Negative correlation of ITCH E3 ubiquitin ligase and miRNA-106b dictates metastatic progression in pancreatic cancer. *Oncotarget* 2016; **7**: 1477–1485.
115. Deng J, Lei W, Xiang X, Zhang L, Lei J, Gong Y *et al.* Cullin 4A (CUL4A), a direct target of miR-9 and miR-137, promotes gastric cancer proliferation and invasion by regulating the Hippo signaling pathway. *Oncotarget* 2016; **7**: 10037–10050.
116. Higashi T, Hayashi H, Ishimoto T, Takeyama H, Kaida T, Arima K *et al.* miR-9-3p plays a tumour-suppressor role by targeting TAZ (WWTR1) in hepatocellular carcinoma cells. *Br J Cancer* 2015; **113**: 252–258.
117. Kwon Y, Vinayagam A, Sun X, Dephoure N, Gygi SP, Hong P *et al.* The Hippo signaling pathway interactome. *Science* 2013; **342**: 737–740.
118. Wang W, Xiao Z-D, Li X, Aziz KE, Gan B, Johnson RL *et al.* AMPK modulates Hippo pathway activity to regulate energy homeostasis. *Nat Cell Biol* 2015; **17**: 490–499.
119. Mo J-S, Meng Z, Kim YC, Park HW, Hansen CG, Kim S *et al.* Cellular energy stress induces AMPK-mediated regulation of YAP and the Hippo pathway. *Nat Cell Biol* 2015; **17**: 500–510.
120. Pefani D-E, Latusek R, Pires I, Grawenda AM, Yee KS, Hamilton G *et al.* RASSF1A-LATS1 signalling stabilizes replication forks by restricting CDK2-mediated phosphorylation of BRCA2. *Nat Cell Biol* 2014; **16**: 962–971.
121. Matsuoka S, Ballif BA, Smogorzewska A, McDonald ER, Hurov KE, Luo J *et al.* ATM and ATR substrate analysis reveals extensive protein networks responsive to DNA damage. *Science* 2007; **316**: 1160–1166.
122. Wang W, Li X, Huang J, Feng L, Dolint KG, Chen J. Defining the protein-protein interaction network of the human Hippo pathway. *Mol Cell Proteomics* 2014; **13**: 119–131.

123. Couzens AL, Knight JDR, Kean MJ, Teo G, Weiss A, Dunham WH *et al.* Protein interaction network of the mammalian Hippo pathway reveals mechanisms of kinase-phosphatase interactions. *Sci Signal* 2013; **6**: rs15.
124. Hirate Y, Hirahara S, Inoue K-I, Suzuki A, Alarcon VB, Akimoto K *et al.* Polarity-dependent distribution of angiotensin II localizes Hippo signaling in preimplantation embryos. *Curr Biol* 2013; **23**: 1181–1194.
125. Yang X, Yu K, Hao Y, Li D, Stewart R, Insogna KL *et al.* LATS1 tumour suppressor affects cytokinesis by inhibiting LIMK1. *Nat Cell Biol* 2004; **6**: 609–617.
126. Lee J-H, Kim T-S, Yang T-H, Koo B-K, Oh S-P, Lee K-P *et al.* A crucial role of WW45 in developing epithelial tissues in the mouse. *EMBO J* 2008; **27**: 1231–1242.
127. Zhang P, Wang S, Wang S, Qiao J, Zhang L, Zhang Z *et al.* Dual function of partitioning-defective 3 in the regulation of YAP phosphorylation and activation. *Cell Discov* 2016; **2**: 16021.
128. Britschgi A, Duss S, Kim S, Couto JP, Brinkhaus H, Koren S *et al.* The Hippo kinases LATS1 and 2 control human breast cell fate via crosstalk with ER α . *Nature* 2017; **541**: 541–545.
129. Torigata K, Daisuke O, Mukai S, Hatanaka A, Ohka F, Motooka D *et al.* LATS2 positively regulates polycomb repressive complex 2. *PLoS ONE* 2016; **11**: e0158562.
130. Li J, Chen X, Ding X, Cheng Y, Zhao B, Lai Z-C *et al.* LATS2 suppresses oncogenic Wnt signaling by disrupting β -catenin/BCL9 interaction. *Cell Rep* 2013; **5**: 1650–1663.
131. Heallen T, Zhang M, Wang J, Bonilla-Claudio M, Klysis E, Johnson RL *et al.* Hippo pathway inhibits Wnt signaling to restrain cardiomyocyte proliferation and heart size. *Science* 2011; **332**: 458–461.
132. Powzaniuk M, McElwee-Witmer S, Vogel RL, Hayami T, Rutledge SJ, Chen F *et al.* The LATS2/KPM tumor suppressor is a negative regulator of the androgen receptor. *Mol Endocrinol* 2004; **18**: 2011–2023.
133. Lit LC, Scott S, Zhang H, Stebbing J, Photiou A, Giamas G. LATS2 is a modulator of estrogen receptor alpha. *Anticancer Res* 2013; **33**: 53–63.
134. Visser S, Yang X. LATS tumor suppressor: a new governor of cellular homeostasis. *Cell Cycle* 2010; **9**: 3892–3903.
135. Tschöp K, Conery AR, Litovchick L, Decaprio JA, Settleman J, Harlow E *et al.* A kinase shRNA screen links LATS2 to the pRB tumor suppressor. *Genes Dev* 2011; **25**: 814–830.
136. Mukai S, Yabuta N, Yoshida K, Okamoto A, Miura D, Furuta Y *et al.* Lats1 suppresses centrosome overduplication by modulating the stability of Cdc25B. *Sci Rep* 2015; **5**: 16173.
137. Masuda K, Chiyoda T, Sugiyama N, Segura-Cabrera A, Kabe Y, Ueki A *et al.* LATS1 and LATS2 phosphorylate CDC26 to modulate assembly of the tetratricopeptide repeat subcomplex of APC/C. *PLoS ONE* 2015; **10**: e0118662.
138. Kamikubo Y, Takaori-Kondo A, Uchiyama T, Hori T. Inhibition of cell growth by conditional expression of kpm, a human homologue of *Drosophila* warts/lats tumor suppressor. *J Biol Chem* 2003; **278**: 17609–17614.
139. Yang X, Li DM, Chen W, Xu T. Human homologue of *Drosophila* lats, LATS1, negatively regulate growth by inducing G(2)/M arrest or apoptosis. *Oncogene* 2001; **20**: 6516–6523.
140. Xia H, Qi H, Li Y, Pei J, Barton J, Blackstad M *et al.* LATS1 tumor suppressor regulates G2/M transition and apoptosis. *Oncogene* 2002; **21**: 1233–1241.
141. Zhao B, Li L, Wang L, Wang C-Y, Yu J, Guan K-L. Cell detachment activates the Hippo pathway via cytoskeleton reorganization to induce anoikis. *Genes Dev* 2012; **26**: 54–68.
142. Ganem NJ, Cornils H, Chiu S-Y, O'Rourke KP, Arnaud J, Yimlamai D *et al.* Cytokinesis failure triggers Hippo tumor suppressor pathway activation. *Cell* 2014; **158**: 833–848.
143. Vigneron AM, Vousden KH. An indirect role for ASPP1 in limiting p53-dependent p21 expression and cellular senescence. *EMBO J* 2012; **31**: 471–480.
144. Iida S-I, Hirota T, Morisaki T, Marumoto T, Hara T, Kuninaka S *et al.* Tumor suppressor WARTS ensures genomic integrity by regulating both mitotic progression and G1 tetraploidy checkpoint function. *Oncogene* 2004; **23**: 5266–5274.
145. Haupt Y, Maya R, Kazaz A, Oren M. Mdm2 promotes the rapid degradation of p53. *Nature* 1997; **387**: 296–299.
146. Matallanas D, Romano D, Al-Mulla F, O'Neill E, Al-Ali W, Crespo P *et al.* Mutant K-Ras activation of the proapoptotic MST2 pathway is antagonized by wild-type K-Ras. *Mol Cell* 2011; **44**: 893–906.
147. Vigneron AM, Ludwig RL, Vousden KH. Cytoplasmic ASPP1 inhibits apoptosis through the control of YAP. *Genes Dev* 2010; **24**: 2430–2439.
148. Matallanas D, Romano D, Yee K, Meissl K, Kucerova L, Piazzolla D *et al.* RASSF1A elicits apoptosis through an MST2 pathway directing proapoptotic transcription by the p73 tumor suppressor protein. *Mol Cell* 2007; **27**: 962–975.
149. Kuninaka S, Nomura M, Hirota T, Iida S-I, Hara T, Honda S *et al.* The tumor suppressor WARTS activates the Omi/HtrA2-dependent pathway of cell death. *Oncogene* 2005; **24**: 5287–5298.
150. Hegde R, Srinivasula SM, Zhang Z, Wassell R, Mukattash R, Cilenti L *et al.* Identification of Omi/HtrA2 as a mitochondrial apoptotic serine protease that disrupts inhibitor of apoptosis protein-caspase interaction. *J Biol Chem* 2002; **277**: 432–438.
151. Romano D, Nguyen LK, Matallanas D, Halasz M, Doherty C, Kholodenko BN *et al.* Protein interaction switches coordinate Raf-1 and MST2/Hippo signalling. *Nat Cell Biol* 2014; **16**: 673–684.
152. Ke H, Pei J, Ni Z, Xia H, Qi H, Woods T *et al.* Putative tumor suppressor Lats2 induces apoptosis through downregulation of Bcl-2 and Bcl-x(L). *Exp Cell Res* 2004; **298**: 329–338.
153. Suzuki H, Yabuta N, Okada N, Torigata K, Aylon Y, Oren M *et al.* Lats2 phosphorylates p21/CDKN1A after UV irradiation and regulates apoptosis. *J Cell Sci* 2013; **126**: 4358–4368.
154. Strano S, Monti O, Pediconi N, Baccarini A, Fontemaggi G, Lapi E *et al.* The transcriptional coactivator Yes-associated protein drives p73 gene-target specificity in response to DNA damage. *Mol Cell* 2005; **18**: 447–459.
155. Basu S, Totty NF, Irwin MS, Sudol M, Downward J. Akt phosphorylates the Yes-associated protein, YAP, to induce interaction with 14-3-3 and attenuation of p73-mediated apoptosis. *Mol Cell* 2003; **11**: 11–23.
156. Kawahara M, Hori T, Chonabayashi K, Oka T, Sudol M, Uchiyama T. Kpm/Lats2 is linked to chemosensitivity of leukemic cells through the stabilization of p73. *Blood* 2008; **112**: 3856–3866.
157. Oka T, Mazack V, Sudol M. Mst2 and Lats kinases regulate apoptotic function of Yes kinase-associated protein (YAP). *J Biol Chem* 2008; **283**: 27534–27546.
158. Reuven N, Adler J, Meltser V, Shaul Y. The Hippo pathway kinase Lats2 prevents DNA damage-induced apoptosis through inhibition of the tyrosine kinase c-Abl. *Cell Death Differ* 2013; **20**: 1330–1340.
159. Levy D, Adamovich Y, Reuven N, Shaul Y. Yap1 phosphorylation by c-Abl is a critical step in selective activation of proapoptotic genes in response to DNA damage. *Mol Cell* 2008; **29**: 350–361.
160. Agami R, Blandino G, Oren M, Shaul Y. Interaction of c-Abl and p73alpha and their collaboration to induce apoptosis. *Nature* 1999; **399**: 809–813.
161. Strano S, Munarriz E, Rossi M, Castagnoli L, Shaul Y, Sacchi A *et al.* Physical interaction with Yes-associated protein enhances p73 transcriptional activity. *J Biol Chem* 2001; **276**: 15164–15173.
162. Sirvent A, Benistant C, Roche S. Cytoplasmic signalling by the c-Abl tyrosine kinase in normal and cancer cells. *Biol Cell* 2008; **100**: 617–631.
163. Dong J, Feldmann G, Huang J, Wu S, Zhang N, Comerford SA *et al.* Elucidation of a universal size-control mechanism in *Drosophila* and mammals. *Cell* 2007; **130**: 1120–1133.
164. Visser-Grievé S, Zhou Z, She Y-M, Huang H, Cyr TD, Xu T *et al.* LATS1 tumor suppressor is a novel actin-binding protein and negative regulator of actin polymerization. *Cell Res* 2011; **21**: 1513–1516.
165. Fang X, Adler PN. Regulation of cell shape, wing hair initiation and the actin cytoskeleton by Trc/Fry and Wts/Mats complexes. *Dev Biol* 2010; **341**: 360–374.
166. Moroishi T, Hansen CG, Guan K-L. The emerging roles of YAP and TAZ in cancer. *Nat Rev Cancer* 2015; **15**: 73–79.
167. Dupont S, Morsut L, Aragona M, Enzo E, Giulitti S, Cordenonsi M *et al.* Role of YAP/TAZ in mechanotransduction. *Nature* 2011; **474**: 179–183.
168. Varelas X, Samavarchi-Tehrani P, Narimatsu M, Weiss A, Cockburn K, Larsen BG *et al.* The Crumbs complex couples cell density sensing to Hippo-dependent control of the TGF- β -SMAD pathway. *Dev Cell* 2010; **19**: 831–844.
169. Nishioka N, Inoue K, Adachi K, Kiyonari H, Ota M, Ralston A *et al.* The Hippo signaling pathway components Lats and Yap pattern Tead4 activity to distinguish mouse trophectoderm from inner cell mass. *Dev Cell* 2009; **16**: 398–410.
170. Lorthongpanich C, Messerschmidt DM, Chan SW, Hong W, Knowles BB, Solter D. Temporal reduction of LATS kinases in the early preimplantation embryo prevents ICM lineage differentiation. *Genes Dev* 2013; **27**: 1441–1446.
171. Loh Y-H, Wu Q, Chew J-L, Vega VB, Zhang W, Chen X *et al.* The Oct4 and Nanog transcription network regulates pluripotency in mouse embryonic stem cells. *Nat Genet* 2006; **38**: 431–440.
172. Wang Y, Baskerville S, Shenoy A, Babiarz JE, Baehner L, Blleloch R. Embryonic stem cell-specific microRNAs regulate the G1-S transition and promote rapid proliferation. *Nat Cell Biol* 2008; **40**: 1478–1483.
173. Qin H, Blaschke K, Wei G, Ohi Y, Blouin L, Qi Z *et al.* Transcriptional analysis of pluripotency reveals the Hippo pathway as a barrier to reprogramming. *Hum Mol Genet* 2012; **21**: 2054–2067.
174. An Y, Kang Q, Zhao Y, Hu X, Li N. Lats2 modulates adipocyte proliferation and differentiation via Hippo signaling. *PLoS ONE* 2013; **8**: e72042.
175. Liu Q, Gu X, Zhao Y, Zhang J, Zhao Y, Meng Q *et al.* Pig large tumor suppressor 2 (Lats2), a novel gene that may regulate the fat reduction in adipocyte. *BMB Rep* 2010; **43**: 97–102.
176. Yimlamai D, Christodoulou C, Galli GG, Yanger K, Pepe-Mooney B, Gurung B *et al.* Hippo pathway activity influences liver cell fate. *Cell* 2014; **157**: 1324–1338.
177. Yi J, Lu L, Yanger K, Wang W, Sohn BH, Stanger BZ *et al.* Large tumor suppressor homologs 1 and 2 regulate mouse progenitor cell proliferation and maturation through antagonism of the coactivators YAP and TAZ. *Hepatology* 2016; **64**: 1757–1772.
178. Lee D-H, Park JO, Kim T-S, Kim S-K, Kim T-H, Kim M-C *et al.* LATS-YAP/TAZ controls lineage specification by regulating TGF β signaling and Hnf4 α expression during liver development. *Nat Commun* 2016; **7**: 11961.
179. Reginensi A, Enderle L, Gregorieff A, Johnson RL, Wrana JL, McNeill H. A critical role for NF2 and the Hippo pathway in branching morphogenesis. *Nat Commun* 2016; **7**: 12309.
180. Heallen T, Morikawa Y, Leach J, Tao G, Willerson JT, Johnson RL *et al.* Hippo signaling impedes adult heart regeneration. *Development* 2013; **140**: 4683–4690.
181. Matsui Y, Nakano N, Shao D, Gao S, Luo W, Hong C *et al.* Lats2 is a negative regulator of myocyte size in the heart. *Circ Res* 2008; **103**: 1309–1318.
182. Singh A, Ramesh S, Cibi DM, Yun LS, Li J, Li L *et al.* Hippo signaling mediators yap and taz are required in the epicardium for coronary vasculature development. *Cell Rep* 2016; **15**: 1384–1393.
183. Uhlén M, Fagerberg L, Hallström BM, Lindskog C, Oksvold P, Mardinoglu A *et al.* Proteomics. tissue-based map of the human proteome. *Science* 2015; **347**: 1260419.

184. Gu Z, Steinmetz LM, Gu X, Scharfe C, Davis RW, Li W-H. Role of duplicate genes in genetic robustness against null mutations. *Nature* 2003; **421**: 63–66.
185. Li J, Musso G, Zhang Z. Preferential regulation of duplicated genes by microRNAs in mammals. *Genome Biol* 2008; **9**: R132.
186. Ohno S. *Evolution by Gene Duplication*. Springer Berlin Heidelberg: Berlin, Germany, 1970.
187. Li W-H, Yang J, Gu X. Expression divergence between duplicate genes. *Trends Genet* 2005; **21**: 602–607.
188. Chen C-Y, Chen S-T, Juan H-F, Huang H-C. Lengthening of 3'UTR increases with morphological complexity in animal evolution. *Bioinformatics* 2012; **28**: 3178–3181.
189. Hausser J, Zavolan M. Identification and consequences of miRNA-target interactions—beyond repression of gene expression. *Nat Rev Genet* 2014; **15**: 599–612.
190. Subramanyam D, Lamouille S, Judson RL, Liu JY, Bucay N, Derynck R *et al*. Multiple targets of miR-302 and miR-372 promote reprogramming of human fibroblasts to induced pluripotent stem cells. *Nat Biotechnol* 2011; **29**: 443–448.
191. Carmena M, Ruchaud S, Earnshaw WC. Making the Auroras glow: regulation of Aurora A and B kinase function by interacting proteins. *Curr Opin Cell Biol* 2009; **21**: 796–805.
192. Hao Y, Chun A, Cheung K, Rashidi B, Yang X. Tumor suppressor LATS1 is a negative regulator of oncogene YAP. *J Biol Chem* 2008; **283**: 5496–5509.
193. Chou C-H, Chang N-W, Shrestha S, Hsu S-D, Lin Y-L, Lee W-H *et al*. miRTarBase 2016: updates to the experimentally validated miRNA-target interactions database. *Nucleic Acids Res* 2016; **44**: D239–D247.
194. Beausoleil SA, Jedrychowski M, Schwartz D, Elias JE, Villén J, Li J *et al*. Large-scale characterization of HeLa cell nuclear phosphoproteins. *Proc Natl Acad Sci USA* 2004; **101**: 12130–12135.
195. Emery A, Sorrell DA, Lawrence S, Easthope E, Stockdale M, Jones DO *et al*. A novel cell-based, high-content assay for phosphorylation of Lats2 by Aurora A. *J Biomol Screen* 2011; **16**: 925–931.
196. Van Hoof D, Muñoz J, Braam SR, Pinkse MWH, Linding R, Heck AJR *et al*. Phosphorylation dynamics during early differentiation of human embryonic stem cells. *Cell Stem Cell* 2009; **5**: 214–226.
197. Zhou H, Di Palma S, Preisinger C, Peng M, Polat AN, Heck AJR *et al*. Toward a comprehensive characterization of a human cancer cell phosphoproteome. *J Proteome Res* 2013; **12**: 260–271.
198. Dephoure N, Zhou C, Villén J, Beausoleil SA, Bakalarski CE, Elledge SJ *et al*. A quantitative atlas of mitotic phosphorylation. *Proc Natl Acad Sci USA* 2008; **105**: 10762–10767.
199. Daub H, Olsen JV, Bairlein M, Gnad F, Oppermann FS, Körner R *et al*. Kinase-selective enrichment enables quantitative phosphoproteomics of the kinome across the cell cycle. *Mol Cell* 2008; **31**: 438–448.
200. Pan C, Olsen JV, Daub H, Mann M. Global effects of kinase inhibitors on signaling networks revealed by quantitative phosphoproteomics. *Mol Cell Proteomics* 2009; **8**: 2796–2808.
201. Morisaki T, Hirota T, Iida S, Marumoto T, Hara T, Nishiyama Y *et al*. WARTS tumor suppressor is phosphorylated by Cdc2/cyclin B at spindle poles during mitosis. *FEBS Lett* 2002; **529**: 319–324.
202. Ni L, Zheng Y, Hara M, Pan D, Luo X. Structural basis for Mob1-dependent activation of the core Mst-Lats kinase cascade in Hippo signaling. *Genes Dev* 2015; **29**: 1416–1431.
203. Liu G, Yu F-X, Kim YC, Meng Z, Naipauer J, Looney DJ *et al*. Kaposi sarcoma-associated herpesvirus promotes tumorigenesis by modulating the Hippo pathway. *Oncogene* 2015; **34**: 3536–3546.
204. Mohseni M, Sun J, Lau A, Curtis S, Goldsmith J, Fox VL *et al*. A genetic screen identifies an LKB1-MARK signalling axis controlling the Hippo-YAP pathway. *Nat Cell Biol* 2014; **16**: 108–117.
205. Hafner M, Landthaler M, Burger L, Khorshid M, Hausser J, Berninger P *et al*. Transcriptome-wide identification of RNA-binding protein and microRNA target sites by PAR-CLIP. *Cell* 2010; **141**: 129–141.
206. Zheng Y-B, Xiao K, Xiao G-C, Tong S-L, Ding Y, Wang Q-S *et al*. MicroRNA-103 promotes tumor growth and metastasis in colorectal cancer by directly targeting LATS2. *Oncol Lett* 2016; **12**: 2194–2200.
207. Whisnant AW, Bogerd HP, Flores O, Ho P, Powers JG, Sharova N *et al*. In-depth analysis of the interaction of HIV-1 with cellular microRNA biogenesis and effector mechanisms. *MBio* 2013; **4**: e000193.
208. Karginov FV, Hannon GJ. Remodeling of Ago2-mRNA interactions upon cellular stress reflects miRNA complementarity and correlates with altered translation rates. *Genes Dev* 2013; **27**: 1624–1632.
209. Zhang M, Wang X, Li W, Cui Y. miR-107 and miR-25 simultaneously target LATS2 and regulate proliferation and invasion of gastric adenocarcinoma (GAC) cells. *Biochem Biophys Res Commun* 2015; **460**: 806–812.
210. Tavazoie SF, Alarcón C, Oskarsson T, Padua D, Wang Q, Bos PD *et al*. Endogenous human microRNAs that suppress breast cancer metastasis. *Nature* 2008; **451**: 147–152.
211. Ragusa M, Statello L, Maugeri M, Majorana A, Barbagallo D, Salito L *et al*. Specific alterations of the microRNA transcriptome and global network structure in colorectal cancer after treatment with MAPK/ERK inhibitors. *J Mol Med* 2012; **90**: 1421–1438.
212. Gabriely G, Wurdinger T, Kesari S, Esau CC, Burchard J, Linsley PS *et al*. MicroRNA 21 promotes glioma invasion by targeting matrix metalloproteinase regulators. *Mol Cell Biol* 2008; **28**: 5369–5380.
213. Jiang L, Li W, Wu M, Cao S. Circulating miRNA-21 as a biomarker predicts polycystic ovary syndrome (PCOS) in patients. *Clin Lab* 2015; **61**: 1009–1015.
214. Fang L, Du WW, Yang W, Rutnam ZJ, Peng C, Li H *et al*. MiR-93 enhances angiogenesis and metastasis by targeting LATS2. *Cell Cycle* 2012; **11**: 4352–4365.
215. Kishore S, Jaskiewicz L, Burger L, Hausser J, Khorshid M, Zavolan M. A quantitative analysis of CLIP methods for identifying binding sites of RNA-binding proteins. *Nat Methods* 2011; **8**: 559–564.
216. Belair C, Baud J, Chabas S, Sharma CM, Vogel J, Staedel C *et al*. *Helicobacter pylori* interferes with an embryonic stem cell micro RNA cluster to block cell cycle progression. *Silence* 2011; **2**: 7.
217. Xia Y, Gao Y. MicroRNA-181b promotes ovarian cancer cell growth and invasion by targeting LATS2. *Biochem Biophys Res Commun* 2014; **447**: 446–451.