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The LKB1 complex-AMPK pathway: the tree that hides the forest

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Abstract

Initially identified as the *Caenorhabditis elegans* PAR-4 homologue, the serine threonine kinase LKB1 is conserved throughout evolution and ubiquitously expressed. In humans, LKB1 is causally linked to the Peutz-Jeghers syndrome (PJS) and is one of the most commonly mutated genes in several cancers like lung and cervical carcinomas. These observations have led to classify *lkb1* as tumour suppressor gene. Although considerable dark zones remain, an impressive leap in the understanding of LKB1 functions has been done during the last decade. Role of LKB1 as a major actor of the AMPK/mTOR pathway connecting cellular metabolism, cell growth and tumorigenesis has been extensively studied probably to the detriment of other functions of equal importance. This review will discuss about LKB1 activity regulation, its effectors and clues on their involvement in cell polarity.

MESH Keywords Humans ; Neoplasms ; physiopathology ; Protein Kinases ; genetics ; metabolism ; Protein-Serine-Threonine Kinases ; genetics ; metabolism

Introduction

Initially identified as the *Caenorhabditis elegans* PAR-4 homologue involved in early asymmetric cell division [1], the vertebrate LKB1 gene *STK11* [2], also called XEEK1 in *Xenopus* [3], is conserved throughout evolution from worms to mammals. In humans, *STK11* gene locates on chromosome 9p13.3 and includes 11 exons [2] which encode for two LKB isoforms by alternative splicing [4, 5]. The long LKB1 (50kDa) form is detectable from 7 days old embryo and is ubiquitously expressed in adult [6] with a notable higher expression in pancreas, liver and skeletal muscle [6, 7]. The shorter form (48kDa), which lacks the last 29 C-terminus residues, appears restricted to testis [5]. Interest in LKB1 has dramatically increased when it was found to be causally linked to the Peutz-Jeghers syndrome [2, 8], a rare autosomal dominant disorder characterized by hamartomatous polyposis and mucocutaneous melanin pigmentation [9]. Indeed, in 70% of PJS cases [10], LKB1 kinase activity appears lost due to genomic deletions [11] or single mutations affecting either transcript splicing [12] or protein primary structure [11, 13]. Patients affected by this syndrome have a 18 times higher relative risk of developing cancers at multiple sites, mostly lung, pancreas, ovary, breast and colon than the general population [14, 15]. This had led to define whether sporadic mutations of *LKB1* gene could occur in tumours. At the exception of non-small-cell lung cancer where *LKB1* is found mutated in 33% of cases, somatic mutations of this gene are rather rare [16]. Nevertheless, its haploinsufficiency has been clearly involved in poor outcome for pancreas [17], breast [18], endometrial [19] and liver adenocarcinomas [20]. These observations have led to classify LKB1 as a tumour suppressor which is a rare feature for a kinase [16].

Generation of LKB1 knock out (KO) mouse models was the most informative to determine LKB1 functions. Thus, homozygous LKB1 KO mice die at 10.5 days of development mainly due to defect in neural tube closure and vascular abnormalities attributed, for this latter, to a VEGF level expression [21]. At the heterozygous state, mice recapitulate PJS with an average tumour free survival of 43 weeks [22]. In addition, conditional LKB1 KO mouse models targeting numerous organs confirmed LKB1 suppressor activity in breast [23], pancreas [24], prostate [25] and skin [26] where tumours appearance is correlated with an altered cell organization such as the nucleus positioning in Langerhans β -cells in the case of pancreas [27, 28]. In the same manner, LKB1 deficiency in the thymus prevents peripheral T cell maturation [29] while in the liver this leads to defects of glucose homeostasis [30]. At the cellular level, LKB1 activity has pleiotropic effects. It has been involved in cell cycle arrest [31, 32], apoptosis [33, 34], autophagia [35], energy metabolism [36] as well as in directional cell migration [37, 38] and epithelial apico-basal polarity [39]. Active LKB1 seems to exert these cellular effects mainly through phosphorylation, at a conserved N-terminal residue, of 14 members of the AMPK family kinase which thus become catalytically actives [40, 41] so driving this wide range of physiological processes.

In recent years, research on LKB1, especially on its regulation mode as well as on its driven functions at cellular level, has been greatly intensified in order to define how this kinase exerts its tumour suppressor properties. This review focuses on current knowledge about LKB1 activity regulation, its effectors and clues on their involvement in cell polarity.

Active LKB1 is a complex

Initial investigations on LKB1 functions and regulation mechanisms were difficult because of the poor LKB1 intrinsic kinase activity. However, identification of the pseudo kinase STRAD (STe-20 Related ADaptor) as a LKB1 partner was a real breakthrough since this association dramatically improves LKB1 kinase activity [42]. Interaction between STRAD and LKB1 appears to be stabilized by a third

protein, MO25 (mouse protein25) [43]. This has led to establish that, in fact, active LKB1 should be considered as a heterotrimeric complex (1:1:1) hereafter referred to as LKB1 complex (figure 1).

In lower organisms where the STRAD and MO25 homologues are expressed, even though this has not been formally established yet, the LKB1 complex formation should be conserved. Indeed in those organisms, MO25 and STRAD homologues contain the critical residues required for their activity such as the STRAD C-terminal WEF motif [44]. In mammals, an additional level of complexity occurs. Indeed, two human STRAD and MO25 paralogues, defined for both as α and β , have been characterised and appear ubiquitously expressed [43]. Furthermore, at least 4 STRAD α [45] and 2 STRAD β isoforms [46] all derived by alternative splicing have been defined. Therefore, cells may express more than one kind of LKB1 complex and in theory could reach up to 16 different ones. Interestingly, some C-terminus STRAD α splice variants, missing their MO25 binding motif, keep their ability to induce LKB1 kinase activity *in vitro* [45]. Nevertheless, this aspect remains unexplored, as LKB1 studies focus almost exclusively on the LKB1-STRAD α -MO25 α complex, but should be kept in mind since all these potential complexes could have specific functions or regulation modes.

The structure of the LKB1-STRAD α -MO25 α complex has been recently resolved and reveals that LKB1 kinase activity results from an allosteric modification induced by STRAD α [44]. Although unable to have an intrinsic kinase activity, due to absence of critical (Asp-Gly-Phe) residues to coordinate magnesium binding in its subdomain VII [42], STRAD α adopts a conformation close to an "active" kinase which associates with LKB1 as one of its substrates [44]. In addition, this reveals an unrecognized STRAD α role by promoting interaction between MO25 α and LKB1. Indeed, MO25 α plays a crucial role in stabilizing the conformation of the LKB1 activation loop required for the kinase activity induced by STRAD α binding [44]. Thus, this explains how lack of the LKB1 phosphorylation site in the activation loop, generally required to induce and stabilize the switch from inactive to active kinase, is dispensable. This feature leads to the conclusion that LKB1 needs to be associated with STRAD α and MO25 α to be active since no LKB1 autophosphorylation would be able to maintain this active conformation [44].

Regulation of LKB1 complex

Although few things have been clearly established on this point, the fact that LKB1 can phosphorylate and activate almost all known members of the AMPK family kinase implies that the LKB1 complex should be sharply regulated to trigger specific responses. Crystal structure had confirmed that LKB1 complex activity is regulated through the LKB1/STRAD interaction. Since expression level or stability of STRAD or LKB1 proteins does not appear to be involved in LKB1 complex regulation, the eventuality that post-translational modifications could be involved has been investigated.

Post-translational modifications

Several phosphorylation sites have been defined on LKB1. Four threonines phosphorylated through LKB1-STRAD interaction are considered as autophosphorylation sites revealing LKB1 complex activity [42]. In the same conditions, STRAD α phosphorylation sites have also been identified, though these sites are not conserved in the STRAD β paralogue [44]. LKB1 Thr363 is phosphorylated after DNA damage in an ATM kinase-dependent manner [47, 48]. This phosphorylation does not modify LKB1 complex catalytic activity, but has been recently demonstrated to be required for inactivation of CRTC2, a transcriptional coactivator of CREB, a critical component of germinal center for B cell proliferation [48]. Even if LKB1 Ser325 and 428, respectively phosphorylated by at least ERK and p90RSK, do not affect LKB1 complex kinase activity [49, 50] they have interestingly been described to impede LKB1 inhibitory effect in anchorage-independent growth, concomitantly to a reduction of AMPK activation [49–51]. Phosphorylation on several other LKB1 sites like Ser31 or Thr189 has also been observed, but kinases involved and functional consequences are still unknown [52]. However, these sites do not appear to be involved in LKB1 complex activity since their substitution by alanine does not significantly affect the LKB1 kinase activity measured *in vitro* [52]. LKB1 can also be farnesylated at a C-terminal CAAX sequence only present in the LKB1 long form. This modification gives rise to LKB1 higher affinity for membrane localization, as alanine replacement of the critical cysteine in CAAX sequence abolishes it [47, 53, 54]. Nevertheless, farnesylation does not affect LKB1 complex activity [49, 51]. The only LKB1 post translational modification referred to modulate kinase activity of the complex is acetylation. Indeed, it has been reported that LKB1 could be acetylated on nine different lysines. Among those, acetylation of the LKB1 Lys48 reduces its affinity for STRAD α and then decreases activity of the complex [55]. Although the acetyl transferase responsible for this modification has not been identified yet, SIRT1 appears to be as the deacetylase able to remove this acetyl group and then favour LKB1 complex formation and activity [55]. These observations suggest that LKB1 complex activity is not mainly regulated by a post translational modification, even though some of them have functional consequences [50]. Furthermore, to our knowledge, no studies have still observed LKB1 complex activation under any stimulating cues, leading to the hypothesis that LKB1 complex could be constitutively active [51, 53] and that its regulation may dependent on its intracellular localization [53].

Subcellular localisations

LKB1 complex localization in cells was initially investigated in over-expressing systems using tagged constructs [42, 56–58]. It was then observed that LKB1 predominantly localizes in the nucleus and to lesser extent in the cytosol. A nuclear localization signal (NLS) at

the N-amino terminal of LKB1 was found to be critical for LKB1 nuclearization [56–58]. Interestingly, overexpressed STRAD α is clearly able to displace LKB1 from the nucleus to the cytoplasm compartment [42, 43, 56] whereas STRAD β was unable to do so [56]. Thus, it was proposed that active LKB1/STRAD α was cytosolic whereas the nuclear LKB1 was active through its interaction with STRAD β [56]. Nevertheless, recent results challenge this model. Indeed, improvement of LKB1 serological tools allowed to immunostain endogenous LKB1. Results from those experiments, confirmed by cell fractionation assays, show that endogenous LKB1 localizes mainly in the cytosolic and membrane fractions and appears to be absent from the nucleus [4, 53]. Failure to detect endogenous nuclear LKB1 has been reported in different cell types, leading to reconsider the functional relevance of the identified NLS sequence. In addition, while LKB1 functions appear to be largely conserved throughout evolution, its *C. elegans* or *Drosophila* homologues do not have NLS sequence and are not significantly found in the nucleus [54].

Endogenous LKB1 immunostaining in migrating cells shows LKB1 concentration at the leading edge [38]. In the same way, in fully polarized epithelial cells LKB1 localizes in a E-cadherin dependent manner at the basolateral domain [38, 53] overlapping with adherens junctions [38, 53]. Farnesylation is required for this membranous localization and, more importantly, membranous LKB1 colocalizes and interacts with STRAD α thus supporting complex activity at basolateral domain [53]. This endogenous LKB1 complex localization at the plasma membrane has also been observed in *C. elegans* [59]. In *Drosophila*, although STRAD has not been localized yet, LKB1 was observed at the cell cortex [54, 60]. Functionally, the LKB1 complex presence at the basolateral domain is correlated to its ability to activate one of its substrate, AMPK [53], suggesting that LKB1 complex regulation could be governed through its intracellular localization allowing proximity with its substrates. This eventuality is strengthened by sparse observations in which LKB1 Ser431 phosphorylation reduces its affinity for the membrane [4] as well as its ability to properly activates AMPK [50].

Altogether, this gives rise to the idea that the LKB1 complex is constitutively active and regulation of cellular processes in which it is involved appears dependent of its subcellular localizations. Very recently, a second LKB1 localization identified in primary cilium of polarized epithelial cells [61] strengthen this hypothesis, nevertheless mechanism(s) regulating these spatial segregations remain to be defined.

LKB1 complex effectors and substrates

Effectors

LKB1 effectors have been identified mainly by mass spectrometry and yeast two hybrid assays. Thus, LKB1 was found to interact with the Hsp90/Cdc37 protein complex contributing likely to its cellular stability [62, 63]. LKB1 was also reported to interact with LIP1 (LKB1 interacting protein 1) which contains 6 Leucine rich repeats (LRR) [64]. Function of LIP1 and consequences of its interaction with LKB1 remain elusive even though effects on TGF β signalling pathway have been suggested [64]. In the same manner, LKB1 interaction with PTEN [65] was observed without clear evidence of functional consequences. Although poorly investigated in the LKB1 context, potentially through LIP1, TGF β and PTEN have been described to be involved in several syndromes like juvenile polyposis which leads to formation of gastrointestinal hamartomas bearing different histological features [66]. Nevertheless, since mechanisms responsible for the formation of those benign polyps are still undefined, deregulation of the cross talk between TGF β , through SMAD4, and mTOR pathway negatively regulated by PTEN and LKB1 [36], could play a role in these pathologies. In addition, LKB1 was also described to interact with Estrogen receptor- α (ER α) [67] and with Brahma related gene-1 (BRG1) [68], both involved in transcriptional regulation. Nevertheless, LKB1 does not appear to phosphorylate these latest partners suggesting perhaps potential indirect LKB1 effect [67, 68].

LKB1 complex substrates, the AMPK related kinase family

AMPK (AMP kinase) was the first LKB1 substrate identified and it is, by far, the one to have been better characterised [69, 70]. This kinase is a heterotrimer composed of a catalytic (AMPK α) and two regulatory (AMPK β and AMPK γ) subunits. AMPK is activated when the intracellular AMP/ATP ratio increases, for instance during hypoxia or nutrient deprivation. In these conditions, LKB1 complex kinase activity does not increase but cytosolic accumulated AMP binds to AMPK γ subunit [71] allowing, by allosteric modification, the accessibility to the critical threonine of the AMPK T-loop kinase domain targeted by LKB1 complex, so leading to its phosphorylation and then to AMPK activation. Main interest in this LKB1 substrate comes from the indication that AMPK modulates the activity of multiple downstream targets such as Raptor (Regulatory associated protein of mTOR) which, in turn, repress the mTORC1 pathway [36]. mTORC1 integrates growth factors and nutriment inputs and controls eukaryote cell growth deregulated in most human tumours [72]. Thus, LKB1 could be the link between energy metabolism and tumour growth suspected since more than 50 years [51]. This hypothesis is particularly attractive to explain the mechanism by which LKB1 exerts its tumours suppressor activity. However, AMPK activation could occur by multiples alternative ways than through the LKB1 complex. Indeed, AMPK can be activated by alternative kinases such as CAMKK β (Ca²⁺/calmodulin-dependent protein kinase kinase β) [73] or TGF β -activated kinase-1 (TAK1) [74] which probably contribute to some consequences attributed to LKB1 complex. Details about AMPK regulated pathways, especially its involvement in mTOR pathway and cell contractility regulation, are reviewed by Tomi Makela and al. in this issue. However and even though AMPK involvement in LKB1

functions appears until now preponderant, the LKB1 complex is able to activate 13 other kinases [47] belonging to the AMPK family which are poorly studied compare to AMPK (figure 2). Most of these kinases have been confirmed to be activated by LKB1 in physiological conditions since their activity is significantly reduced in Lkb1-deficient models.

Among them, the two brain-specific kinases (**BRSK-1 and -2**) also called SAD kinases (SAD-B and -A), mainly expressed in brain and testis, play a role in neuronal cell polarity. BRSK deficient mice exhibits drastic reduced axon growth as well as mislocalized axons and dendritic markers [75, 76]. In addition, BRSK-2 has been demonstrated to regulate centrosome duplication. Indeed, BRSK-2 localizes at centrosome through its association with γ -Tubulin and phosphorylates it at serine 131 which is required to centrosome duplication [77]. Thus, BRSK-2 could regulate polarity and neuronal proliferation as AMPK does for epithelial cells (see below). Moreover, like AMPK, BRSK-2 can be also phosphorylated and activated, independently from LKB1, notably by CAMKK [78].

The LKB1 complex had been described to phosphorylate the four human **MARK** kinases (microtubule affinity regulating kinase), homologous to the *C. elegans Par-1* gene product [40]. Even though, MARK kinases appear involved in epithelial cell polarity [79, 80], the relationship between LKB1 and MARK kinases remain unclear in mammals. Indeed, LKB1 deficiency either in primary fibroblasts or in cardiac muscle cells does not lead to significant modifications of MARK3 phosphorylation and activation [40]. This could be explained by the fact that TAO1 kinase (Thousand and one amino acid protein kinase) is able to phosphorylate the critical T-loop threonine and thereby activate MARK kinases [81]. Nevertheless, forced expression of active LKB1 triggers MARK2 function by suppressing tubulin polymerization through Tau phosphorylation and affecting cell migration and polarity [82]. Thus, in physiological conditions, MARK kinase activation by LKB1 could be restricted to some isoforms.

Like for other related AMPK kinases, activation of **QSK** and **SIK** kinases is due to their phosphorylation on the T-loop threonine by LKB1. For these kinases, this motif is a docking site for the 14-3-3 family members which participates to their full activation and concomitantly affects their subcellular localization [41]. Indeed, an active form of QSK localizes in punctate structures of unknown nature within the cytosol through 14-3-3 binding [41]. While QSK functions are not well established, QSK knockdown in *Drosophila* triggers mitotic defects like spindle and chromosome alignment abnormalities indicating that this protein might be involved also in cell proliferation [83]. In addition to the indication that both *Drosophila* AMPK and LKB1 deficiencies lead to similar defects [84, 85], it is tempting to speculate that those three kinases may cooperate in a same pathway during *Drosophila* cell division, potentially linked to energetic stress.

SIK1 (salt-induced kinase 1) has been described to play a role in steroidogenesis and TGF β signalling, for this latter by inhibiting type I TGF β receptor kinase [84]. More recently, SIK1 was also found to have a crucial role in p53 dependent apoptosis induced by cell detachment, called anoikis [33]. This study shows that following cell detachment, SIK is activated in a LKB1 dependent pathway triggering p53 phosphorylation and apoptosis through proapoptotic Bax and Puma induced expression. Thus, loss of LKB1, SIK1 or p53 interferes in anoikis induction so favouring metastasis development [33]. **SIK2**, also called QIK, appears to be highly expressed in adipose tissues and is able to phosphorylate insulin receptor substrate 1 (IRS1) at the same site than AMPK [85]. Similarly, SIK2 phosphorylates the CREB coactivator TORC2 at the same site than AMPK, leading to TORC2 capture by 14-3-3 proteins. Result of this binding is TORC2 retention in the cytosol preventing its transcriptional induction of gluconeogenic genes expression [85].

About **NUAK2**, also referenced as ARK5, few things are known, except that mice deficient for this kinase exhibit an embryonic lethality after 16.5 days of development due to an exencephaly which results in a brain protrusion outside the skull [86]. Moreover, NUAK2 has been described to be activated through TNF α , leading to inhibition of the actomyosin contractile network through phosphorylation of threonine 696 and 853 of MYPT1 regulatory subunit of the myosin light chain phosphatase. Thereby, this inhibition induces increased phosphorylation of myosin light chain and cellular contractility [87]. Unfortunately, this study does not link this TNF α -dependent NUAK2 activity with the LKB1 complex. Nevertheless, **NUAK1** activated by LKB1 complex has been found recently to lead to similar effect on MYPT1 [88]. This suggests that, through NUAK kinases, LKB1 could play an important role in cell contractility regulation independently of Rho GTPases family effector such as Rho kinase [87]. Besides, NUAK1 has been reported to be increased in colorectal tumours where it promotes invasiveness [89]. NUAK1 is also involved in cell senescence [90] and apoptosis inhibition [91]. Beyond an already established connection between cell contractility and apoptosis [92], contractility driven by NUAK1 could have a major role in tumorigenesis progress in addition to these well established roles in invasion and metastatic processes.

Finally, **SNRK**, the less studied member of AMPK related kinases, is mainly expressed in testis and might be involved in spermatogenesis or spermatozoid motility [93]. This idea is coherent with the fact that male mice which express low levels of LKB1 are sterile [94].

LKB1 and cell polarity

From the seminal studies in *C.elegans*, that characterized PAR4, the LKB1 homologue and its involvement in asymmetric cell division, implication of this kinase in cell polarity and cell division had been suspected [1]. LKB1 contribution in cell cycle regulation was

confirmed by ectopic expression in various cellular models leading to induction of p21^{waf} Cdk inhibitor in a p53 dependant manner [31 , 32]. Beyond this effect on cell cycle regulation, the LKB1 complex could be directly involved in cell division process, as its knock down in *Drosophila* S2 cells affects chromosome alignment, spindle formation and centrosome number [83]. These effects could be the result of loss of either QSK, MARK-3 or AMPK activities which are individually important in cell division [83]. Nevertheless, those findings remain fragmentary and need to be confirmed in mammalian models.

In addition to PAR4, Kemphues and colleagues had characterized other genes involved as partitioning defective mutants that include mammalian PAR3 and PAR6 co-homologues. These proteins are well established cell polarity proteins and act together with PAR1 and PAR5, homologues of MARK kinases and 14-3-3 respectively. Thus, the role of LKB1 in cell polarity establishment was initially investigated and observed in *Drosophila* oocyte polarization [54]. In mammals, LKB1 complex contribution to apico-basal cell polarity was revealed by co-expressing STRAD α and LKB1 in colorectal cancer cell lines [39]. Indeed, in those cells, forced LKB1 activity induced leads to formation of brush border-like structures and an actin cap resembling to the apical domain of polarized epithelial cells [39]. Moreover, this apical-like domain was concomitantly associated with directed endosomal transport of basolateral markers, such as the transferrin receptor, excluded from the apical structure [39]. However, LKB1 involvement in epithelial cell polarity is less obvious than usually considered. Indeed, several reports describe that LKB1 knock-down in MDCK [53 , 61] or CaCo2 [53] cells derived respectively from canine kidney or human colon, only exhibits a slight delay in cell polarity. Moreover, during embryonic development of LKB1 deficient mice, no cell polarity defect is observed until 7 days of development while important processes of cell polarization have already taken place before this stage [21]. Studies in *Drosophila* can potentially explain this discrepancy. Indeed, it has been shown that AMPK or LKB1 loss leads to defects in *Drosophila* epithelial cell polarity only under energetic stress [60]. These results suggest that LKB1-AMPK is required to allow cell adaptation to unfavourable growth conditions, probably because epithelial cell polarity maintenance is an energetic consuming process. It would be interesting to define whether, in mammalian cells, a similar mechanism is conserved. Indeed, during epithelial derived tumours formation loss of polarity could be due not only to loss of structural protein functions but result also to limiting nutriment availability leading to an energetic stress that tumour cells have difficulties to bypass. Thus, observations that E-cadherin is partly involved in LKB1 dependent AMPK activation [53] and often lost during tumour progression [95], could reflect another way for it to affect cell polarity in addition to its well established structural functions [96]. This eventuality is strengthened by the fact that AMPK activity, involved in tight junction formation [97 , 98], is increased in epithelial cells through calcium switch [97 , 98] considered in part to act on E-cadherin and adherens junctions formation. Main role of the tight junctions is to seal the apical membranes of adjacent epithelial cells precluding the free diffusion of solutes toward the basolateral membranes [99]. However, they also restrict the lateral diffusion of some lipids and plasma membrane-associated proteins between apical and basolateral domains, which is thought to ensure that the two membrane domains do not mix [99]. How AMPK activity controls these events is unclear. It seems that its inhibitory properties on the mTOR pathway may play a role, since the effect of dominant negative AMPK construct on tight junctions formation can be partially rescued by mTOR inhibitors such as Rapamycin [97]. This is consistent with the fact that mTOR activity is opposed to apico-basal polarity by contributing to the epithelio-mesenchymal transition [100]. All of this underlines that outside of energetic stress conditions where LKB1-AMPK appears to be dispensable in epithelial cell polarity, the mTOR pathway activity should be maintained low by an alternative mechanism. Moreover, MARK kinase activities, well known to be required in epithelial cell polarity [79 , 80] would be also activated through a LKB1 independent manner potentially by TAO1 kinase as mentioned above [81].

Conclusions

Currently, there is no doubt that the LKB1 complex is at the centre of an important signalling node affecting numerous cellular processes whose deregulation contributes to pathologies such as Peutz-Jeghers syndrome and cancer. Although studies by many labs have focused on the role of LKB1 complex in the regulation of the AMPK-mTOR pathway, the 13 AMPK related LKB1 effectors remain currently poorly investigated. Thus, studies using LKB1 knock down models and relying consequence almost exclusively on the AMPK activity should be interpreted with caution since contribution of other kinases of this family might participate to consequences observed. Besides and although downstream signalling events of this complex begin to be defined, upstream signals that regulate LKB1 functions remain almost completely uncharacterized. Expressions of MO25 and STRAD paralogues as well as splice variants have probably some importance in specific LKB1 functions. The emerging notion that the complex is constitutively active in cells led to think that the subcellular localizations of LKB1 are probably a major feature to specify the functions of the kinase. How these localizations are achieved and which effectors and substrates are targeted are pending questions that should be addressed in a near future to understand all aspects of LKB1 complex involvement and disorders in physiopathology.

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Fig. 1

(A) Scheme of human LKB1 spliced variants, STRAD and Mo25 paralogues. Tilted and underline residues considered as autophosphorylation sites. Phosphorylated residues targeted by mentioned kinases (arrow). NLS for nuclear localization sequence. CAAX for the site of farnesylation. (B) Schematic representation of active and functional complex.

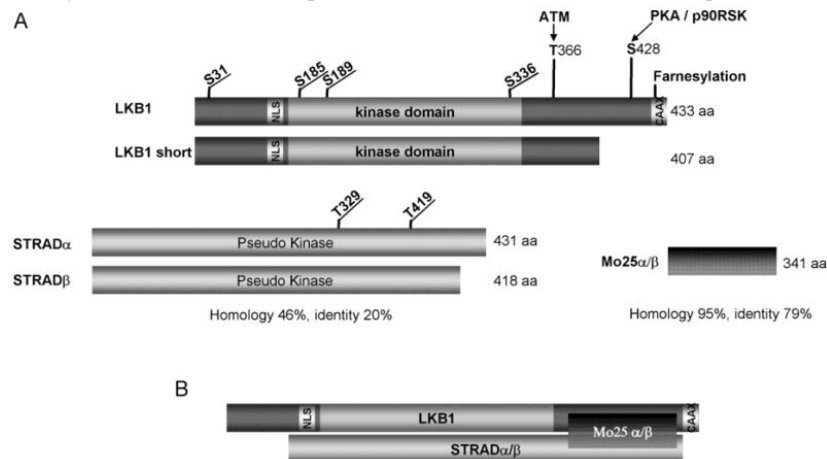


Fig. 2

Domain organization of AMPK-related kinases activated by LKB1 complexes. Kinase domain in light gray, ubiquitin associated domain in checked motif. Usual and alternative names, molecular weight and main identified functions, respectively from left to right.

