European Review for Medical and Pharmacological Sciences 2019; 23: 413-425

Zyxin: a mechanotransductor to regulate gene expression

Y.-X. WANG[1,2], D.-Y. WANG[3], Y.-C. GUO[1], J. GUO[1]

[1]Academy of Medicine and Life Sciences, Nanjing University of Chinese Medicine, Nanjing, China
[2]The First School of Clinical Medicine, Nanjing University of Chinese Medicine, Nanjing, China
[3]Model Animal Research Center, Nanjing University, Nanjing, China

YuXuan Wang and *Dingyu Wang* contributed equally to this work

Abstract. – OBJECTIVE: Cells answer to biochemical, electrical and mechanical signals in the environment, which regulate their behavior. Mechanical signals can propagate through mechanically stiff structures like focal adhesions (FAs). Zyxin, a LIM domain protein, is localized primarily at focal adhesion plaques. Growing evidence suggests that zyxin is a vital mechanotransductor to regulate the gene expression. In this review, we summarize the features of zyxin and the molecular mechanism of how zyxin participate in the cellular activity.

MATERIALS AND METHODS: An English-language literature search is based on a keyword-based query of multiple databases (MEDLINE, Embase) and bibliographies from identified publications. The references in the selected paper are also considered as an additional source of data. The search was last updated in April 2018; no limitations are applied.

RESULTS: Zyxin enhances actin polymerization with the aid of Enabled (Ena)/vasodilator-stimulated phosphoprotein (VASP) proteins in response to mechanical tension, to perform its role in stress fibers (SFs) remodeling and repair. Zyxin can translocate from focal adhesions (FAs) to the nucleus responds to stretch, and regulate gene transcription by interaction with transcription factors like nuclear matrix protein 4 (NMP4). Misregulation of nuclear functions of zyxin appears to be associated with pathogenic effects and diseases, such as prostate cancer and non-small-cell lung cancer.

CONCLUSIONS: Zyxin is a crucial ingredient of the cellular mechanotransducing system and can modulate the gene expression. Given its clinical relevance, zyxin is also a promising target for the diagnosis and treatment of certain diseases. Understanding the role of zyxin in force sensing and gene expression regulating provides a compelling challenge for future biomechanics studies, and offers attractive evidence for zyxin as a potential diagnostic marker and therapeutic target for clinical diseases.

Key Words:
Zyxin, Focal adhesions, Nuclear-cytoplasmic communication, Gene expression, Mechanotransductor, Cytoskeleton, Clinical diagnosis and treatment.

Introduction

During their lifetimes, cells encounter a variety of stimuli that can dramatically affect their behavior, such as mechanical stimulation, which can raise different kinds of signal pathways and regulate physiologic activity[1]. Dysregulation of mechanical force is responsible for a variety of diseases, including neuronal and muscular degeneration[2], potential immune system disorders[3], hypertension[4], and polycystic kidney disease[5]. Cells respond to mechanical properties such as stiffness, contractility and tensile strength through punctually, appropriately graded adjustments to maintain tissue homeostasis[6]. Also, the gene expression in living cells can be regulated by mechanical stimuli[7]. Multiple studies[8-10] have highlighted the mechanosensitive features of the protein zyxin. Zyxin may enter the nucleus associated with other proteins in response to mechanical force stimulation and is exported from the nucleus through intrinsic leucine-rich nuclear export sequences (NES). Due to the ability to shuttle between cytoplasm and nucleus, zyxin may mediate cell function in a force-dependent manner, and its ability to detect mechanical force could be an integral part of the regulation of the gene expression[11,12]. Zyxin has also displayed force sensitive translocation to focal adhesions (FAs) and stress fibers (SFs) in fibroblasts and endothelial cells, helping the remodeling and repair of SFs[10]. Given the great importance of zyxin in

Corresponding Author: Jun Guo, MD; e-mail: Guoj@njucm.edu.cn

regulating cellular mechanical activities, understanding its role in force sensing and transduction is meaningful to biomechanics study. Focal adhesions and actin stress fibers are well-known structures that form and develop in a mechanical force-dependent manner[13,14]. Focal adhesions, also referred to as focal plaques or focal contacts, are an integral component of the transmission of mechanical stimuli[15-17]. FAs are complex multi-protein structures that form upon integrin engagement with the extracellular matrix (ECM) and link the ECM to the intracellular cytoskeleton[18]. Moreover, FAs serve as critical signaling hubs that transmit chemical (extracellular protein ligands) and physical (rigidity, composition) cues about the extracellular environment[13]. Exogenous mechanical forces in ECM are transduced through FAs to the actin cytoskeleton, which indicates that the FAs and SFs are mechanical mediums for maintaining the force balance in the changing mechanical environment and signal transmission[8,19]. It is intriguing to mention that, only partial focal complexes mature into large and stable focal adhesions, and then could recruit many more proteins like zyxin[20]. Zyxin dissociates from FAs by reducing mechanical loads on the FAs and regains accumulation at FAs by stretching the substratum[8]. In other words, the application of external forces can change the localization of the FAs protein zyxin. For instance, zyxin proteins would be recruited to actin stress fibers when adherent cells are stretched by pulling on the underlying flexible substrate. Meanwhile, actin assembly at FAs is enhanced[8,10]. By contrast, zyxin-deficient cells fail to respond to external strain[21]. Delocalization of zyxin from FAs or genetic ablation of zyxin leads to unusual and integrin-independent migration of cells[8]. Zyxin is also recruited to stress fiber strain sites for SFs repair and stabilization[22]. Consequently, zyxin plays as a crucial ingredient of the mechanotransducing system. The focus of this review is the protein zyxin, an adhesion plaque component that has been implicated in signaling events and mechanotransducing system at the adhesive membrane.

Molecular Structure of Zyxin

Zyxin is primarily localized at focal adhesion plaques, actin stress fibers, and cell-ECM and cell-cell junction areas, transiently exist in some nuclei[11,23]. FAs are structures located at the ends of actin fibers and serve as force transmission sites[24]. Zyxin is one of the FAs constituents (82KDa molecular weight), that possesses two distinct motifs: N-terminal proline-rich domain and C-terminal LIM domain[25,26]. The N-terminus of zyxin has been reported to bind some partners, including the actin filament cross-linker α-actinin[27,28], the actin assembly modulator Ena/VASP[29], the cytoskeletal proteins LIM and SH3 domain protein 1 (LASP-1) and the LIM-nebulette (LASP-2)[30]. Among which, the VASP family proteins form complexes with four proline-rich ActA repeats to facilitate actin-polymerization at FAs and SFs[31]. The overexpressed zyxin LIM domain substitutes the endogenous zyxin from FAs would induce the mislocalization of VASP and mammalian Ena[8,32]. The C-terminus LIM domains of zyxin that consist of three motifs (termed by the initials of LIN-11, Isl-1 and MEC-3) are essential for its force sensing function by accumulating at FAs or force-bearing sites[33]. LIM domains are dual zinc-finger protein-protein or protein-DNA binding interfaces. Some LIM proteins which localize to the nucleus have been proved to perform a transcriptional role[9,33]. Two leucine-rich NES lie in the central region of zyxin are believed to mediate nuclear export (Figure 1)[34]. Additionally, the zyxin-nectin interaction proves that amino acids 230-280 of zyxin are required for localization to cell-cell adhesions[35].

The functional diversity of LIM proteins suggests that the LIM domain plays a unique role in various cellular processes.

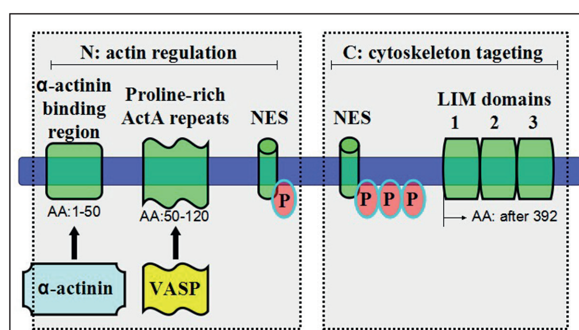

Figure 1. Molecule structure of zyxin and its domains. The N-terminus of zyxin has four proline-rich ActA repeats (amino acids 50-120) for the interaction with actin regulators VASP and Mena, which also has α-actinin binding sites (amino acids 1-50)[36]. C-terminal LIM domains (after amino acids 392) contain cysteine/histidine zinc-coordinating LIN-11, Isl-1 and MEC-3, which are essential for its location to FAs, force-induced targeting and protein interactions[33,37]. Two leucine-rich nuclear export sequences and relevant serine phosphorylation sites lie in the central region of zyxin[36].

Zyxin As a Mechanotransductor

Stress fibers, whose formation and development are mechanical force-dependent, are formed through a combination of actin *de novo* polymerization that occurs at FAs and the merging of previously formed fragments[38]. It has been illustrated that actin-regulatory proteins, Arp2/3 complex, mammalian Diaphanous (mDia)-related formins and Ena/VASP proteins are involved in the process of actin polymerization[39-42]. Compared to mDia and Arp2/3, Ena/VASP proteins are noticeably localized at FAs, which demonstrates a role of Ena/VASP in the local actin polymerization at FAs. The recruitment of Ena/VASP to FAs depends on the accumulation of the LIM protein zyxin at FAs[8].

Hirata et al[8] concluded previous experimental results from Rottner K, Lele TP and some other researchers, then put forward the role of zyxin as a crucial element of the mechanotransducing system at FAs. Proteomic studies[43,44] have indicated that LIM domain proteins such as Hic-5, paxillin, CRP2 and zyxin are sensitive to mechanical stress in the actin cytoskeleton. Among them, three LIM domain proteins have been shown to be recruited to SFs in response to stretch: Hic-5, CRP2 and zyxin[45,46]. Zyxin exhibits an expeditious and intensive mobilization from FAs to actin filaments in response to a unidirectional cyclic stretch of cells, while other FAs proteins remain concentrating at the substratum attachment sites[10]. Therefore, zyxin is the only LIM domain protein proved mechanoresponsive.

As mentioned above, zyxin is rapidly mobilized from FAs to the remodeling actin filaments responds to stretch. Zyxin flows away from FAs in synchrony with newly assembled actin bundles during SFs assembly[45]. In answer to a uniaxial cyclic stretch, zyxin-null cells fail to strengthen the actin SFs commonly, elucidating zyxin-independent and zyxin-dependent facets of the stretch response[47]. Live-cell imaging technology is applied to investigate zyxin dynamics in response to actin SFs thinning. Actin incorporation is high at zyxin-rich FAs but decreases when over-expressed LIM domains replace zyxin[36]. Moreover, after knocking down zyxin's expression with RNAi, a perinuclear actin cap structure induced by mild shear stress would be failed to form, and a reduction happened in the pulling force at cell-fibronectin bead contact sites[48]. Lastly, zyxin dissociates from SFs with relief of tension through laser severing and is reversibly recruited to SFs in response to atomic force microscopy (AFM) stylus-driven tension induction[49]. Zyxin enhances actin polymerization with the aid of Ena/VASP proteins in response to mechanical tension in FAs, to perform its role in SFs remodeling and repair. In other words, mechanical signals are transduced into the actin polymerization response via zyxin accumulation accompanied by Ena/VASP recruitment.

Externally Applied Force: Zyxin Mobilizes to FAs

Zyxin is recruited to Force-Bearing Sites

Researechers[49,50] showed that zyxin shuttles from FAs to tension zones within SFs and triggers local recruitment of α-actinin and VASP that thickens and reinforces SFs. In other words, zyxin accumulates in the sites of externally applied forces. However, Uemura et al[12] put forward their experimental results: zyxin only accumulates at the leading edge, it does not indiscriminately localize to force-bearing sites; instead, it is capable of distinguishing between these distinct adhesion sites. These results indicate that force-dependent zyxin accumulation occurs at the leading edge but not at the trailing edge of migrating cells.

Scholars[51,52] have reported the crucial role of the C-terminus LIM domain of zyxin in mediating zyxin binding to force-bearing sites. The LIM domain consists of three motifs. Studies have found that multiple zyxin LIM domains fail to increase force sensitivity while single zyxin LIM domain has the best force-sensitivity. These results demonstrate the essentiality of all three LIM motifs for linking binding partners with zyxin at force-bearing sites, while the impossible interaction occurs to multiple LIM domains[29]. The yet known partners have been already implied dropping out of the process that zyxin localized to force-bearing places, such as p130Cas, Cysteine-Rich Protein (CRP), and synemin[53-56]. It ought to be other zyxin binding partners participating in this process. However, Hoffman et al[47] prove that force-induced zyxin phosphorylation relies on the activation of the MAPK signaling pathways, rather than p130Cas, indicating that there is the possibility that zyxin would be recruited to force-bearing sites without binding any partners. The underlying mechanism of how zyxin is recruited to force-bearing sites remains to be clarified.

Zyxin Recruits VASP to the Force-Bearing Sites

The Ena/VASP family is a growing collection of related proteins that have been implicated in the Abl signaling pathway and the assembly of the actin cytoskeleton[57]. VASP, the vasodilator-stimulated phosphoprotein, is a member of the Ena/VASP family. VASP is identified as a prominent 46 kDa substrate for cAMP- and cGMP-dependent kinases in platelets[58]. Immunolocalization works[34] have revealed that VASP is localized at integrin-rich adhesion plaque. Molecular cloning of VASP cDNAs established that human VASP is a protein of 380 aa with a central proline-rich core that is quite distinct from the proline sequence found in zyxin[59]. The proline region of VASP interacts directly with profilins, the small actin monomer binding proteins that have been demonstrated in the actin filament assembly regulation and signal transduction[60,61]. The direct interaction between VASP and ActA proteins of bacterium *Listeria* further confirms the role of VASP in actin filament dynamics[39]. As described above, ActA is required for the ability of the bacterium to assemble actin filaments on its surface. Interestingly, VASP has been shown to associate directly with zyxin[31]. VASP interacts with a proline repeat region of ActA that resembles the sequences found in zyxin[26,62]. The zyxin-deficient cells would not determine the position of VASP to FAs anymore[63], so does the mislocalization of zyxin[26]. Moreover, zyxin-dependent recruitment of VASP to sites of tension-induced cytoskeletal damage is found to regulate actin filament repair[64].

We can see the prevailing thought is that zyxin acts merely as a scaffold protein for VASP binding[26]. However, Grange et al[65] refute this view by identifying the LIM domain-VASP interaction. The series of four proline-rich (FPPPPP) motifs can bind zyxin with VASP[31,66,67], as well as through the LIM domain region[68]. It is intriguing that defects in SFs reinforcement following stretch stimuli are also observed in cells lacking zyxin[10], which highlights a critical role for VASP in organizing actin at FAs. They discover that zyxin-VASP binding through both the proline-rich motifs and the LIM domains alters specific VASP functions; neither individual interaction alters VASP's actin regulatory activities. Of interest, full-length zyxin dramatically reduces VASP-mediated actin bundling and actin assembly. These results suggest a model where zyxin-VASP complexes occur in complex organizations with suppressed actin regulatory activity[65].

The LIM domain of Zyxin is Sufficient for Force-Dependent Recruitment

Many studies tried to find out which molecule recruits zyxin to FAs in a force-dependent manner and which structure of zyxin play the core role in this process[8,12,29]. Yi et al[56] reported that protein p130Cas, which is the FAs-associated adapter, interacts with the LIM region of zyxin. Crawford[69] found the direct interaction between zyxin and α-actinin and soon after an α-actinin-binding site was identified in the N-terminal region of zyxin[26,28]. That leads to a long period in which zyxin N-terminal region was believed to be necessary for force-dependent localization of zyxin to FAs. However, Hirata et al[8] observed the accumulation of the separate LIM region of zyxin at FAs in a force-dependent manner. Also, they found that the force-induced accumulation of endogenous zyxin at FAs was inhibited by the expression of the LIM region[8]. These consequences suggest that the LIM region of zyxin is crucial for the force-dependent recruitment of zyxin to FAs, and raise the possibility that the LIM domain of zyxin is sufficient for force-dependent recruitment. Uemura et al[12] conducted the experiments and published their results in 2011. By analyzing the zyxin mutants with the truncated LIM-domain (ΔLIM-GFP) and with only the LIM domains (LIM-GFP), they concluded that the LIM domain of zyxin is sufficient for responding to the traction force generated by migrating cells. Furthermore, individual or truncated LIM motifs are not sufficient for force-dependent accumulation, and zyxin recruitment requires all three LIM motifs. To date, this finding has been acknowledged by most researchers in the field[17,29,70,71].

Internally Generated Force: Zyxin Flows Away from FAs to Actin SFs

Forces applied to cell-ECM adhesions are transmitted across the transmembrane integrin receptors to the cytoskeleton via molecular linkages with the FAs[30,72]. Within the FAs anchoring complex, integrin physically associates with multiple adaptor proteins involved in signal transduction, such as focal adhesion kinase (FAK), vinculin, talin, p130Cas, and paxillin[73-76]. Moreover, the internal movement of actin filaments can also be sensed by FAs through a slippage-clutch mechanism inside cells[77]. By means

of conformational changes, some molecules of these complexes, such as talin and paxillin, can ‘‘perceive’’ mechanical stimuli and transduce them into electrical or biochemical signals, subsequently triggering downstream signaling pathways to modulate cellular physiologic activities[78-81]. Thus, we can conclude that not only external forces transmitted across ECM adhesions focus on these FAs sites as already shown, but also internal forces generated within the actin cytoskeleton. To give an example, the recruitment of zyxin at force-bearing sites relies on myosin II and Rho-kinase activation, suggesting that zyxin not only responds to the externally applied force according to previous data, but also responds to the internally generated actin-myosin force[12]. Although this viewpoint has been accepted by some researchers[70], it still needs further research to explain how zyxin plays its role in sensing internally generated force.

Zyxin Mediates SFs Repair

Cells will recognize and respond to changes in cytoskeletal integrity to maintain mechanical homeostasis[64]. Actin stress fibers come through bounded, intense, force-mediated elongation and decreasing events that settle their function of stress transmission, followed by SFs repair that revives this capability[64]. SF strain sites recruit at least four different proteins found at FAs: zyxin, paxillin, α-actinin and VASP[19,71]. Paxillin, a 68 kDa FAs protein, has been confirmed the recruitment to SF strain sites, so does the zyxin[19,82]. Zyxin swiftly accumulates at the damage sites of strain-induced SFs, and paxillin recruitment even precedes zyxin recruitment. The recruitment and repair process of paxillin is parallel to, but independent of, the zyxin repair system[19]. The repair functions of zyxin are executed by the actin crosslinker α-actinin and the actin regulator VASP, which are recruited to SF strain sites in a zyxin dependent manner. Zyxin is recruited first dependently, in synchrony with VASP, and then recruits α-actinin bind the N-terminal region of zyxin[36]. Zyxin binding to VASP is required for VASP recruitment to either cyclically stretched SFs or to SF strains sites. Mutation of the proline-rich ActA repeats in the N-terminal region of zyxin eliminates VASP binding to zyxin, which may indicate the binding region on zyxin[36]. The mechanism of strain recognition and repair demonstrates the cellular machinery for quick modification of cytoskeletal tension respond to changes in cell contractility or external forces.

Force/Stretch Induced Nuclear Translocation and Changes of Zyxin Nuclear Activities

FAs proteins combine the actin filaments with integrins and regulate transmembrane mechanical force transmission. Zyxin, which acts as the mechanotransducer, is partly mediated by cytoskeletal tension[83,84]. Part of this response is mediated by regulating the physical strength of the FAs that resists cell traction forces to sustain cytoskeletal prestress[85]. These FAs proteins could facilitate to transfer mechanical to chemical signals by the Rho pathway, which induces myosin II phosphorylation by feedback and produces cytoskeletal forces[86,87]. Zyxin, known as FAs protein, alter its binding kinetics in a force-dependent manner which enables it to shuttle between the cytoplasm and nucleus, and in this manner, zyxin can serve as a transcription factor to regulate gene expression[9,88]. Three lines of evidence have demonstrated that zyxin shuttles between nuclear compartments and cytoplasm. First, in cells that are treated with leptomycin B, an inhibitor of Crm1-dependent nuclear export, zyxin accumulates in the nucleus[85]. Second, a leucine-rich nuclear export signal (NES) has been well-characterized within the central region of most zyxin proteins, and deletion or mutation of this NES also results in nuclear accumulation of zyxin[89]. Third, nuclear accumulation of endogenous zyxin is also observed after infection of cells with a vaccinia virus[90].

Growing studies[9,91,92] have reported the role of zyxin in transcriptional responses. Zyxin is transported into the nucleus respond to applied forces and antisense oligonucleotides against zyxin altered stretch-induced changes in gene expression in smooth muscle cells[93]. However, less is known about the underlying mechanism. Apart from the proline-rich region, LIM domains of zyxin may also contribute to the process of nuclear import[94]. However, neither is basic. Accumulation within nuclei is likely to occur via a particular mechanism[87]. There are some hypotheses so far. The hydrodynamic characteristics of chicken zyxin inform that zyxin performs as an elongate monomer of 69 kDa, which is too large to diffuse passively through the nuclear pore complex[87]. Furthermore, zyxin has no traditional (primary) nuclear localization sequence (NLS). Thus, it is possible that zyxin enters the nucleus in association with other NLS-containing proteins or alternative mechanisms[9]. Zyxin may use a unique nuclear import mechanism similar

to that described for the cell adhesion and cell signaling protein β-catenin that interacts directly with the nuclear pore complex[87]. The zyxin proteins can enter the nucleus by reinforced-association with a mutant form of cell adhesion kinase β/proline-rich tyrosine kinase 2 (CAKβ/PYK2), which abnormally localizes to the nucleus[95]. Suresh Babu et al[96] exhibit a multiple-stage signaling pathway through the stretch-induced release of endothelial vasoconstrictor peptide endothelin-1 (ET-1), mediated by the transient receptor potential channel. Protein kinase G would mediate the phosphorylation of zyxin at serine 142, sequentially triggering the translocation of zyxin to the nucleus. Furthermore, zyxin acts as a transducer of transducing the mechanical signal into the nucleus in endothelial cells, where it orchestrates the expression of a prominent subset of stretch-sensitive genes through a novel DNA-response element[97]. In vascular smooth muscle cells (VSMCs), within minutes, zyxin translocates from FAs to the nucleus of VSMCs when exposes to a cyclic strain, and changes the expression of the mechanical-sensitive gene[98]. ChIP assays revealed that zyxin actually interacts with the promoter region of zyxin-dependent genes, such as interleukin-8, VCAM-1, HMNC1, Hey-1, HMGCR, and ICAM-1[93].

As mentioned above, zyxin not only contributes to organizing the actin cytoskeleton but also to the changes in the gene expression occurring as an adaptive response to enduring mechanical strain. The LIM-domains and the similar LIM-domain present in the zyxin homolog, lipoma-preferred protein, can directly induce gene expression in an artificial assay system. The phenomenon suggests that zyxin may act as a transcription factor[87]. Otherwise, zyxin may affect the gene expression exclusively through protein-protein interactions, as described for related zinc finger proteins like the GATA family of transcription[87].

Here is another supporting proof: long-drawn exposure to enhanced stretch, such as hypertension, can trigger endothelial dysfunction, a hallmark of pathological vascular remodeling processes[93]. DNA microarray pathway analyses of stretch-induced changes in endothelial cell gene expression revealed that zyxin mainly regulates proinflammatory pathways, suggesting a role for zyxin in vascular remodeling processes. Testing results of three stretch-sensitive genes revealed that zyxin controls the interleukin-8 and CXCL1 instead of the B-type endothelin receptor (ETB-R). In practice, zyxin interacts with the promoter region of these genes[93]. Furthermore, in human cultured endothelial cells that exposed to cyclic stretch, a nuclear protein-DNA complex forms that, according to supershift analysis, contains zyxin, indicating the significant role of zyxin in stretch-induced endothelial gene expression[93].

LIM domains have structures related to certain zinc fingers, which are known to mediate DNA binding in several transcription factors. Zyxin interacts with a variety of nuclear proteins including transcription factors or induces regulation of cytoplasmic proteins in the nucleus[88,99,100]. Zyxin acts as coactivators of transcription to regulate gene expression[85]. Proteins regulated by the stretch-induced accumulation of zyxin in nuclei are as followed:

1. **6E6:** A yeast two-hybrid library screening determines that zyxin acts as a protein partner for E6, from Human Papillomavirus (HPV) Type 6 and results in E6's nuclear translocation. Cotransfection of E6 from HPV (6E6) and zyxin leads to the aggregation of zyxin in the nucleus, where it can work as an activating transcription factor. 6E6 can also mobilize endogenous zyxin to the nucleus. Moreover, when zyxin binds to Gal4-BD for exogenously expressing, the results show that it has inscriptional activation potential and this activity is synergistically enhanced by 6E6 only when the interacting C-terminal LIM domain is present in the zyxin construct[101].
2. **Akt:** Kato et al[102] found that atrial natriuretic peptide (ANP) promotes cardiomyocyte survival by cGMP-dependent nuclear accumulation of zyxin and Akt. Nuclear translocation of zyxin also induces nuclear accumulation of activated Akt kinase. Zyxin and activated Akt participate in a cGMP-dependent signaling cascade leading from ANP receptors to nuclear accumulation of both molecules. Collectively, nuclear accumulation of zyxin and activated Akt may represent a fundamental mechanism that facilitates nuclear-signal transduction and potentiates cell survival[33]. Additionally, another research shows that zyxin binds to acinus-S, a nuclear speckle protein inducing apoptotic, chromatin condensation after cleavage by caspases, and restraints its apoptotic action, which is regulated by Akt[103].
3. **HNF-1β:** Hepatocyte nuclear factor-1β (HNF-1β), an epithelial tissue-specific transcription

factor, could regulate the gene expression in the kidney, liver, intestine, and other organs[104]. The LIM-domain protein zyxin is identified as a new binding partner of HNF-1β in renal epithelial cells. Zyxin shuttles to the nucleus with the co-localization of HNF-1β[91]. The interaction of the two proteins requires the participation of the second LIM domain of zyxin and the two particular domains of HNF-1β. The overexpression of zyxin motivates the transcriptional activity of HNF-1β, while small interfering RNA silencing of zyxin inhibits HNF-1β-dependent transcription[101].

4. **CBP:** Retinoids including all-trans retinoic acid (RA) have been widely used for cancer therapy. However, the acquired resistance remains the main obstacle to RA treatment. Former studies informed that zyxin mediates retinoic acid receptors (RARs) repression by forming a ternary complex with PTOV1 and the RAR coactivator CBP through translocating to the nucleus in response to RA. Accordingly, it promotes the dissociation of CBP from RAR at the RA-responsive promoter. Consistently, RA-induced cancer cell cytotoxicity is significantly impaired by Zyxin or PTOV1[105].
5. **CARP-1:** Zyxin contributes to UV-induced apoptosis. Cell cycle and apoptosis regulator protein-1 (CARP-1), a 130-kDa nuclear protein which is co-isolated with zyxin, are identified by microsequencer analysis. Zyxin connects with CARP-1 through its LIM region. Zyxin lacking the CARP-1 binding region presents lessened proapoptotic activity in response to UV-C irradiation[106].
6. **SIRT1:** After treatment with leptomycin B, zyxin accumulates in the nucleus co-localized with SIRT1 in COS-7 cells. Moreover, the SIRT1 deacetylates zyxin suggests that SIRT1 could interact with nuclear-accumulated zyxin and regulate its function through deacetylation. These consequences raise the possibility that SIRT1 regulates signal transmission from ECM to the nucleus by modulating the functions of zyxin via deacetylation[107].
7. **Xanf1:** By using a yeast two-hybrid system, experiments are designed to seek candidature partner protein of the homeodomain transcription repressor Xanf1, a crucial transcriptional regulator of the early stage of the forebrain growth. The LIM domain protein zyxin is identified from the African clawed frog Xenopuslaevis primarily. In the lysate of X. laevis embryos, the interaction of zyxin with Xanf1 is confirmed by the immunoprecipitation of an endogenous-zyxin-complex with the hybrid myc-Xanf1 protein. By using a set of deletion mutants of both proteins, it has been demonstrated that the combination of the LIM2 domain of zyxin and the Engrailed Homology 1 repressor domain of Xanf1 contributes to the interaction of these proteins[108].
8. **ZNF384:** Zyxin interacts *in vitro* with ZNF384 (zinc finger protein 384, also called the Cas interacting Zn-finger protein, CIZ, and NMP4), a transcription factor which shuttles between the nucleus and adhesion sites and is involved in osteoblast differentiation. Zyxin interacts directly with p130Cas and is postulated to link p130Cas to ZNF384. It is intriguing that zyxin or its binding partners have been implicated in the control of the gene expression in two tissues, bone, and smooth muscle, both of which are exquisitely responsive to mechanical stress[109].

Conclusions

In this review, we discuss the FAs protein zyxin, also a member of LIM domain proteins, which shows mechanosensitive features that indicate its role as a crucial ingredient of the mechanotransducing system. In response to mechanical force stimulation, zyxin flows away from FAs to actin stress fibers for SFs remolding and repair, enters the nucleus by association with other proteins to regulate gene expression. Mechanical signals are transduced into the actin polymerizing response via zyxin accumulation accompanied by Ena/VASP recruitment. Moreover, zyxin is recruited with VASP and α-actinin, which bind the N-terminal region of zyxin, to SF strain sites and mediate SFs repair. However, there are still many unsolved questions. Although previous results suggested a model where zyxin-VASP complexes occur in complex organizations with suppressed actin regulatory activity, it remains to be clarified[65]. Furthermore, apart from zyxin, there are multiple force sensitive modules present at the FAs that are activated at distinct locations and regulate specific aspects of junction dynamics[110]. A large number of studies have been conducted in this aspects, but the mechanism they corporate with each other in regulating cytoskeletal tension and mediating nuclear activities has not been revealed thoroughly[65,106].

It is worth noticing that though zyxin is mechanoresponsive and a potential candidate of mechanosensor, there is no direct evidence so far that zyxin directly receives and senses force in cells[111]. Although localization and phosphorylation of zyxin are sharply altered in response to mechanical stimuli to cells[10], it is possible that some other molecule may sense force and transduce it into a signal that modulates zyxin behaviors. Detailed data mining and bioinformatic analysis have revealed that FAs are composed of 180 different kinds of molecules, connected to each other within a network containing at least 742 interactions[112]. Zyxin's recruitment to cytoskeletal structures under tension might be sensed and driven by newly revealed conformational changes, actin barbed ends, or post-translational modifications in actin or zyxin-binding partners[36,113].

As mentioned above, accumulating evidence suggests that zyxin has nuclear functions that affect transcription, in addition to their functions at focal adhesion plaques. Zyxin interacts with a variety of nuclear proteins, acting as coactivators of transcription to regulate gene expression[85]. Zyxin may constitute from FAs to the nucleus through some signaling pathways such as the Wnt/β-catenin pathway[114] and the guanylate kinase CASK[115], which have already been clarified their role in mechanotransduction[116-118]. We think further studies ought to reveal which pathways are involved in this process. Moreover, LIM domains have structures related to certain zinc fingers, which are known to mediate DNA binding in several transcription factors. However, zyxin is probably not direct transcription factor, as only the LIM domains of Hic-5 have been shown to have DNA-binding activity, and this has only been demonstrated *in vitro*[119]. Whether zyxin is an actual transcription factor, still needs further exploration.

In certain circumstance, the nuclear misregulation function of zyxin is relevant to pathogenic effects and diseases[9]. Zyxin, via its LIM domain region, interacts with the E6 oncoprotein of HPV type 6, which is commonly associated with genital warts. The excessive cyclic stretch of vascular smooth muscle cells leads to the shift in their phenotype like hypertension. Zyxin modulates the mechanotransduction of vascular smooth muscle cells by influencing the cytoskeletal structure and signaling pathways[55,94]. Bronchial hyperresponsiveness of airway smooth muscle (ASM) is a characteristic feature of asthma[120]. Zyxin is also found to assist the ASM cells to respond to stretch caused by deep inspiration in people with asthma. Based on its ability to repair SFs fragmentation, zyxin maintains the ASM structure, promotes the recovery of contractile force and finally slows airway dilation[121]. Moreover, several studies have revealed the relevance between zyxin and different kinds of cancer. Prostate cancer is a malignant tumor which used to appear mostly in the male urogenital system[122]. Zyxin siRNA treatment inhibited the migration and invasion of DU145 cells. Zyxin expression in tumor tissues is higher than in normal tissues, suggesting that zyxin may participate in the growth and invasiveness process of human prostate cancer[123]. Lung cancer also has high metastatic potential, which is the leading cause of the significant mortality[124]. Zyxin has been determined as a potential early diagnostic marker for non-small-cell lung cancer[125]. Therapies ought to be formulated based on the further understanding of the role of zyxin aiming at these diseases. Future studies must reveal the mechanism of how zyxin serves as a mechanotransductor in force sensing and regulation of the gene expression in several diseases, and that is meaningful for constituting therapeutic strategies to combat the diseases.

Conflict of Interest
The Authors declare that they have no conflict of interests.

Acknowledgements
The work was supported by grants from the National Natural Science Foundation of China (No. 81573409), the Natural Science Foundation of Jiangsu Province (No. BK20161574) and the Brand Specialty Building Program of Jiangsu Higher Education Institutions (Traditional Chinese Medicine).

References

1) Kshitiz, Park J, Kim P, Helen W, Engler AJ, Levchenko A, Kim DH. Control of stem cell fate and function by engineering physical microenvironments. Integr Biol (Camb) 2012; 4: 1008-1018.
2) Suchyna T, Sachs F. Mechanical and electrical properties of membranes from dystrophic and normal mouse muscle. J Physiol Lond 2007; 581(Pt 1): 369-387.
3) Matheson LA, Maksym GN, Santerre JP, Labow RS. Cyclic biaxial strain affects U937 macrophage-like morphology and enzymatic activities. J Biomed Mater Res A 2006; 76: 52-62.
4) Huang C. Hypertension, mechanical force, and renal disease. Clin Exp Hypertens 2014; 2: 1009.

5) Delmas P. Polycystins: from mechanosensation to gene regulation. Cell 2004; 118: 145-148.
6) DuFort CC, Paszek MJ, Weaver VM. Balancing forces: architectural control of mechanotransduction. Nat Rev Mol Cell Biol 2011; 12: 308-319.
7) Wang N. Review of cellular mechanotransduction. J Phys D Appl Phys 2017; 50: 233002.
8) Hirata H, Tatsumi H, Sokabe M. Mechanical forces facilitate actin polymerization at focal adhesions in a zyxin-dependent manner. J Cell Sci 2008; 121: 2795-2804.
9) Wang Y, Gilmore TD. Zyxin and paxillin proteins: focal adhesion plaque LIM domain proteins go nuclear. Biochim Biophys Acta 2003; 1593: 115-120.
10) Yoshigi M, Hoffman LM, Jensen CC, Yost HJ, Beckerle MC. Mechanical force mobilizes zyxin from focal adhesions to actin filaments and regulates cytoskeletal reinforcement. J Cell Biol 2005; 171: 209-215.
11) Nix DA, Beckerle MC. Nuclear-cytoplasmic shuttling of the focal contact protein, zyxin: a potential mechanism for communication between sites of cell adhesion and the nucleus. J Cell Biol 1997; 138: 1139-1147.
12) Uemura A, Nguyen TN, Steele AN, Yamada S. The LIM domain of Zyxin is sufficient for force-induced accumulation of Zyxin during cell migration. Biophys J 2011; 101: 1069-1075.
13) Burridge K, Fath K, Kelly T, Nuckolls G, Turner C. Focal adhesions – transmembrane junctions between the extracellular-matrix and the cytoskeleton. Annu Rev Cell Biol 1988; 4: 487-525.
14) Guo J, Wang Y, Sachs F, Meng F. Actin stress in cell reprogramming. Proc Natl Acad Sci U S A 2014; 111: E5252-5261.
15) Fogh BS, Multhaupt AH, Couchman JR. Protein kinase C, focal adhesions and the regulation of cell migration. J Histochem Cytochem 2014; 62: 172-184.
16) Shiokawa S, Yoshimura Y, Nagamatsu S, Sawa H, Hanashi H, Sakai K, Noguchi K, Nakamura Y. Functional role of focal adhesion kinase in the process of inplantation. Mol Hum Reprod 1998; 4: 907-914.
17) Oakes PW, Gardel ML. Stressing the limits of focal adhesion mechanosensitivity. Curr Opin Cell Biol 2014; 30: 68-73.
18) Bo LJ, Wei B, Li ZH, Wang ZF, Gao Z, Miao Z. Bioinformatics analysis of miRNA expression profile between primary and recurrent glioblastoma. Eur Rev Med Pharmacol Sci 2015; 19: 3579-3586.
19) Smith MA, Blankman E, Deakin NO, Hoffman LM, Jensen CC, Turner CE, Beckerle MC. LIM domains target actin regulators paxillin and zyxin to sites of stress fiber strain. PLoS One 2013; 8: e69378.
20) Wu C. Focal adhesion: a focal point in current cell biology and molecular medicine. Cell Adh Migr 2007; 1: 13-18.
21) Ngu H, Feng Y, Lu L, Oswald SJ, Longmore GD, Yin FC. Effect of focal adhesion proteins on endothelial cell adhesion, motility and orientation response to cyclic strain. Ann Biomed Eng 2010; 38: 208-222.
22) Burridge K, Wittchen ES. The tension mounts: stress fibers as force-generating mechanotransducers. J Cell Biol 2013; 200: 9-19.
23) Mori M, Nakagami H, Koibuchi N, Miura K, Takami Y, Koriyama H, Hayashi H, Sabe H, Mochizuki N, Morishita R, Kaneda Y. Zyxin mediates actin fiber reorganization in epithelial-mesenchymal transition and contributes to endocardial morphogenesis. Mol Biol Cell 2009; 20: 3115-3124.
24) Huttenlocher A, Horwitz AR. Integrins in cell migration. Cold Spring Harb Perspect Biol 2011; 3: a005074.
25) Zamir E, Geiger B. Components of cell-matrix adhesions. J Cell Sci 2001; 114: 3577-3579.
26) Drees BE, Andrews KM, Beckerle MC. Molecular dissection of zyxin function reveals its involvement in cell motility. J Cell Biol 1999; 147: 1549-1560.
27) Li B, Trueb B. Analysis of the alpha-actinin/zyxin interaction. J. Biol. Chem 2001; 276: 33328-33335.
28) Reinhard M, Zumbrunn J, Jaquemar D, Kuhn M, Walter U, Trueb B. An alpha-actinin binding site of zyxin is essential for subcellular zyxin localization and alpha-actinin recruitment. J Biol Chem 1999; 274: 13410-13418.
29) Steele AN, Sumida GM, Yamada S. Tandem zyxin LIM sequences do not enhance force sensitive accumulation. Biochem Biophys Res Commun 2012; 422: 653-657.
30) Li B, Zhuang L, Trueb B. Zyxin interacts with the SH3 domains of the cytoskeletal proteins LIM-nebulette and Lasp-1. J Biol Chem 2004; 279: 20401-20410.
31) Drees B, Friederich E, Fradelizi J, Louvard D, Beckerle MC, Golsteyn RM. Characterization of the interaction between zyxin and members of the Ena/vasodilator-stimulated phosphoprotein family of proteins. J Biol Chem 2000; 275: 22503-22511.
32) Nix DA, Fradelizi J, Bockholt S, Menichi B, Louvard D, Friederich E, Beckerle MC. Targeting of zyxin to sites of actin membrane interaction and to the nucleus. J Biol Chem 2001; 276: 34759-34767.
33) Kadrmas JL, Beckerle MC. The LIM domain: from the cytoskeleton to the nucleus. Nat Rev Mol Cell Biol 2004; 5: 920-931.
34) Beckerle MC. Zyxin: zinc fingers at sites of cell adhesion. Bioessays 1997; 19: 949-957.
35) Gregory Call S, Brereton D, Bullard JT, Chung JY, Meacham KL, Morrell DJ, Reeder DJ, Schuler JT, Slade AD, Hansen MD. A zyxin-nectin interaction facilitates zyxin localization to cell-cell adhesions. Biochem Biophys Res Commun 2011; 415: 485-489.
36) Smith MA, Hoffman LM, Beckerle MC. LIM proteins in actin cytoskeleton mechanoresponse. Trends Cell Biol 2014; 24: 575-583.

37) Velyvis A, Qin J. LIM domain and its binding to target proteins. Madame Curie Bioscience Database Internet. Austin (TX): Landes Bioscience; 2000-2013.

38) Hotulainen P, Lappalainen P. Stress fibers are generated by two distinct actin assembly mechanisms in motile cells. J Cell Biol 2006; 173: 383-394.

39) Beckerle MC. Spatial control of actin filament assembly: lessons from Listeria. Cell 1998; 95: 741-748.

40) Blystone SD. Integrating an integrin: a direct route to actin. Biochim Biophys Acta 2004; 1692: 47-54.

41) Calderwood DA, Shattil SJ, Ginsberg MH. Integrins and actin filaments: reciprocal regulation of cell adhesion and signaling. J Biol Chem 2000; 275: 22607-22610.

42) Shang Y, Guo XX, Li WW, Rao W, Chen ML, Mu LN, Li SJ. Cucurbitacin-B inhibits neuroblastoma cell proliferation through up-regulation of PTEN. Eur Rev Med Pharmacol Sci 2014; 18: 3297-3303.

43) Kuo JC, Han X, Hsiao CT, Yates JR 3rd, Waterman CM. Analysis of the myosin-II-responsive focal adhesion proteome reveals a role for beta-Pix in negative regulation of focal adhesion maturation. Nat Cell Biol 2011; 13: 383-393.

44) Schiller HB, Friedel CC, Boulegue C, Fässler R. Quantitative proteomics of the integrin adhesome show a myosin II-dependent recruitment of LIM domain proteins. EMBO Rep 2011; 12: 259-266.

45) Guo WH, Wang YL. Retrograde fluxes of focal adhesion proteins in response to cell migration and mechanical signals. Mol Biol Cell 2007; 18: 4519-4527.

46) Kim-Kaneyama JR, Suzuki W, Ichikawa K, Ohki T, Kohno Y, Sata M, Nose K, Shibanuma M. Uni-axial stretching regulates intracellular localization of Hic-5 expressed in smooth-muscle cells in vivo. J Cell Sci 2005; 118: 937-949.

47) Hoffman LM, Jensen CC, Chaturvedi A, Yoshigi M, Beckerle MC. Stretch-induced actin remodeling requires targeting of zyxin to stress fibers and recruitment of actin regulators. Mol Biol Cell 2012; 23: 1846-1859.

48) Mise N, Savai R, Yu H, Schwarz J, Kaminski N, Eickelberg O. Zyxin is a transforming growth factor-β (TGF-β)/Smad3 target gene that regulates lung cancer cell motility via integrin α5β1. J Biol Chem 2012; 287: 31393-31405.

49) Colombelli J, Besser A, Kress H, Reynaud EG, Girard P, Caussinus E, Haselmann U, Small JV, Schwarz US, Stelzer EH. Mechanosensing in actin stress fibers revealed by a close correlation between force and protein localization. J Cell Sci 2009; 122: 1665-1679.

50) Burridge K, Guilluy C. Focal adhesions, stress fibers and mechanical tension. Exp Cell Res 2016; 343: 14-20.

51) Hansen MD, Beckerle MC. Opposing roles of zyxin/LPP ACTA repeats and the LIM domain region in cell-cell adhesion. J Biol Chem 2006; 281: 16178-16188.

52) Nguyen TN, Uemura A, Shih W, Yamada S. Zyxin-mediated actin assembly is required for efficient wound closure. J Biol Chem 2010; 285: 35439-35445.

53) Schmeichel KL, Beckerle MC. The LIM domain is a modular protein-binding interface. Cell 1994; 79: 211-219.

54) Sun N, Huiatt TW, Paulin D, Li Z, Robson RM. Synemin interacts with the LIM domain protein zyxin and is essential for cell adhesion and migration. Exp Cell Res 2010; 316: 491-505.

55) Sun Z, Huang S, Li Z, Meininger GA. Zyxin is involved in regulation of mechanotransduction in arteriole smooth muscle cells. Front Physiol 2012; 3: 472.

56) Yi J, Kloeker S, Jensen CC, Bockholt S, Honda H, Hirai H, Beckerle MC. Members of the zyxin family of LIM proteins interact with members of the p130 Cas family of signal transducers. J Biol Chem 2002; 277: 9580-9589.

57) Krause M, Dent EW, Bear JE, Loureiro JJ, Gertler FB. Ena/VASP proteins: regulators of the actin cytoskeleton and cell migration. Annu Rev Cell Dev Biol 2003; 19: 541-564.

58) Reinhard M, Halbrügge M, Scheer U, Wiegand C, Jockusch BM, Walter U. The 46/50 kDa phosphoprotein VASP purified from human platelets is a novel protein associated with actin filaments and focal contacts. EMBO J 1992; 11: 2063-2070.

59) Bachmann C, Fischer L, Walter U, Reinhard M. The EVH2 domain of the vasodilator-stimulated phosphoprotein mediates tetramerization, F-actin binding, and actin bundle formation. J Biol Chem 1999; 274: 23549-23557.

60) Hüttelmaier S, Mayboroda O, Harbeck B, Jarchau T, Jockusch BM, Rüdiger M. The interaction of the cell-contact proteins VASP and vinculin is regulated by phosphatidylinositol-4, 5-bisphosphate. Curr Biol 1998; 8: 479-488.

61) Hansen SD, Mullins RD. VASP is a processive actin polymerase that requires monomeric actin for barbed end association. J Cell Biol 2010; 191: 571-584.

62) Machner MP, Urbanke C, Barzik M, Otten S, Sechi AS, Wehland J, Heinz DW. ActA from Listeria monocytogenes can interact with up to four Ena/VASP homology 1 domains simultaneously. J Biol Chem 2001; 276: 40096-40103.

63) Hoffman LM, Jensen CC, Kloeker S, Wang CL, Yoshigi M, Beckerle MC. Genetic ablation of zyxin causes Mena/VASP mislocalization, increased motility, and deficits in actin remodeling. J Cell Biol 2006; 172: 771-782.

64) Smith MA, Blankman E, Gardel ML, Luettjohann L, Waterman CM, Beckerle MC. A zyxin-mediated mechanism for actin stress fiber maintenance and repair. Dev Cell 2010; 19: 365-376.

65) Grange J, Moody JD, Ascione MP, Hansen MD. Zyxin-VASP interactions alter actin regulatory activity in zyxin-VASP complexes. Cell Mol Biol Lett 2013; 18: 1-10.

66) Niebuhr K, Ebel F, Frank R, Reinhard M, Domann E, Carl UD, Walter U, Gertler FB, Wehland J, Chakraborty T. A novel prolinerich motif present in ActA of Listeria monocytogenes and cytoskeletal proteins is the ligand for the EVH1 domain, a protein module present in the Ena/VASP family. EMBO J 1997; 16: 5433-5444.
67) Reinhard M, Rudiger M, Jockusch BM, Walter U. VASP interaction with vinculin: a recurring theme of interactions with proline-rich motifs. FEBS Lett 1996; 399: 103-107.
68) Moody JD, Grange J, Ascione MP, Boothe D, Bushnell E, Hansen MD. A zyxin head-tail interaction regulates zyxin-VASP complex formation. Biochem Biophys Res Commun 2009; 378: 625-628.
69) Crawford AW, Michelsen JW, Beckerle MC. An interaction between zyxin and alpha-actinin. J Cell Biol 1992; 116: 1381-1393.
70) Shih W, Yamada S. N-cadherin as a key regulator of collective cell migration in a 3D environment. Cell Adh Migr 2012; 6: 513-517.
71) Watanabe-Nakayama T, Saito M, Machida S, Kishimoto K, Afrin R, Ikai A. Requirement of LIM domains for the transient accumulation of paxillin at damaged stress fibres. Biol Open 2013; 2: 667-674.
72) Wang N, Butler JP, Ingber DE. Mechanotransduction across the cell surface and through the cytoskeleton. Science 1993; 260: 1124-1127.
73) Cukierman E, Pankov R, Stevens DR, Yamada KM. Taking cell-matrix adhesions to the third dimension. Science 2001; 294: 1708-1712.
74) Friedland JC, Lee MH, Boettiger D. Mechanically activated integrin switch controls α5β1 function. Science 2009; 323: 642-644.
75) del Rio A, Perez-Jimenez R, Liu R, Roca-Cusachs P, Fernandez JM, Sheetz MP. Stretching single talin rod molecules activates vinculin binding. Science 2009; 323: 638-641.
76) Sawada Y, Tamada M, Dubin-Thaler BJ, Cherniavskaya O, Sakai R, Tanaka S, Sheetz MP. Force sensing by mechanical extension of the Src family kinase substrate p130Cas. Cell 2006; 127: 1015-1026.
77) Hu K, Ji L, Applegate KT, Danuser G, Waterman-Storer CM. Differential transmission of actin motion within focal adhesions. Science 2007; 315: 111-115.
78) Dingyu W, Fanjie M, Zhengzheng D, Baosheng H, Chao Y, Yi P, Huiwen W, Jun G, Gang H. Regulation of intracellular structural tension by talin in the axon growth and regeneration. Mol Neurobiol 2016; 53: 4582-4595.
79) Hirata H, Tatsumi H, Lim CT, Sokabe M. Force-dependent vinculin binding to talin in live cells: a crucial step in anchoring the actin cytoskeleton to focal adhesions. Am J Physiol Cell Physiol 2014; 306: C607-620.
80) Hytonen VP, Vogel V. How force might activate talin's vinculin binding sites: SMD reveals a structural mechanism. PLoS Comput Biol 2008; 4: e24.
81) Nuckolls GH, Turner CE, Burridge K. Functional studies of the domains of talin. J Cell Biol 1990; 110: 1635-1644.
82) Lin ZF, Shen XY, Lu FZ, Ruan Z, Huang HL, Zhen J. Reveals new lung adenocarcinoma cancer genes based on gene expression. Eur Rev Med Pharmacol Sci 2012; 16: 1249-1256.
83) Chan MW, Arora PD, Bozavikov P, McCulloch CA. FAK, PIP5KIgamma and gelsolin cooperatively mediate force-induced expression of alpha-smooth muscle actin. J Cell Sci 2009; 122: 2769-2781.
84) Riveline D, Zamir E, Balaban NQ, Schwarz US, Ishizaki T, Narumiya S, Kam Z, Geiger B, Bershadsky AD. Focal contacts as mechanosensors: externally applied local mechanical force induces growth of focal contacts by an mDia1-dependent and ROCK-independent mechanism. J Cell Biol 2001; 153: 1175-1186.
85) Ezzell RM, Goldmann WH, Wang N, Parashurama N, Ingber DE. Vinculin promotes cell spreading by mechanically coupling integrins to the cytoskeleton. Exp Cell Res 1997; 231: 14-26.
86) Mammoto A, Huang S, Ingber DE. Filamin links cell shape and cytoskeletal structure to Rho regulation by controlling accumulation of p190RhoGAP in lipid rafts. J Cell Sci 2007; 120: 456-467.
87) Zhao XH, Laschinger C, Arora P, Szászi K, Kapus A, McCulloch CA. Force activates smooth muscle alpha-actin promoter activity through the Rho signaling pathway. J Cell Sci 2007; 120: 1801-1809.
88) Lele TP, Pendse J, Kumar S, Salanga M, Karavitis J, Ingber DE. Mechanical forces alter zyxin unbinding kinetics within focal adhesions of living cells. J Cell Physiol 2006; 207: 187-194.
89) Huang TT, Kudo N, Yoshida M, Miyamoto S. A nuclear export signal in the N-terminal regulatory domain of IκBα controls cytoplasmic localization of inactive NF-κB/IκBα complexes. Proc Natl Acad Sci U S A 2000; 97: 1014-1019.
90) Zeile WL, Condit RC, Lewis JI, Purich DL, Southwick FS. Vaccinia locomotion in host cells: evidence for the universal involvement of actin-based motility sequences ABM-1 and ABM-2. Proc Natl Acad Sci U S A 1998; 95:13917-13922.
91) Choi YH, McNally BT, Igarashi P. Zyxin regulates migration of renal epithelial cells through activation of hepatocyte nuclear factor-1β. Am J Physiol Renal Physiol 2013; 305: F100-110.
92) Schwartz MA. Integrins and extracellular matrix in mechanotransduction. Cold Spring Harb Perspect Biol 2010; 2: a005066.
93) Cattaruzza M, Lattrich C, Hecker M. Focal adhesion protein zyxin is a mechanosensitive modulator of gene expression in vascular smooth muscle cells. Hypertension 2004; 43: 726-730.
94) Petit MM, Fradelizi J, Golsteyn RM, Ayoubi TA, Menichi B, Louvard D, Van de Ven WJ, Friederich E. LPP, an actin cytoskeleton protein related to zyxin, harbors a nuclear export signal and transcriptional activation capacity. Mol Biol Cell 2000; 11: 117-129.

95) Aoto H, Sasaki H, Ishino M, Sasaki T. Nuclear translocation of cell adhesion kinase h/proline-rich tyrosine kinase 2. Cell Struct Funct 2002; 27: 47-61.

96) Suresh Babu S, Wojtowicz A, Freichel M, Birnbaumer L, Hecker M, Cattaruzza M. Mechanism of stretch-induced activation of the mechanotransducer zyxin in vascular cells. Sci Signal 2012; 5: 91.

97) Wójtowicz A, Babu SS, Li L, Gretz N, Hecker M, Cattaruzza M. Zyxin mediation of stretch-induced gene expression in human endothelial cells. Circ Res 2010; 107: 898-902.

98) Ghosh S, Kollar B, Nahar T, Suresh Babu S, Wojtowicz A, Sticht C, Gretz N, Wagner AH, Korff T, Hecker M. Loss of the mechanotransducer zyxin promotes a synthetic phenotype of vascular smooth muscle cells. J Am Heart Assoc 2015; 4: e001712.

99) Yang L, Guerrero J, Hong H, DeFranco DB, Stallcup MR. Interaction of the tau2 transcriptional activation domain of glucocorticoid receptor with a novel steroid receptor coactivator, Hic-5, which localizes to both focal adhesions and the nuclear matrix. Mol Biol Cell 2000; 11: 2007-2018.

100) Wang Y, Gilmore TD. LIM domain protein Trip6 has a conserved nuclear export signal, nuclear targeting sequences, and multiple transactivation domains. Biochim Biophys Acta 2001; 1538: 260-272.

101) Degenhardt YY, Silverstein S. Interaction of zyxin, a focal adhesion protein, with the e6 protein from human papillomavirus type 6 results in its nuclear translocation. J Virol 2001; 75: 11791-11802.

102) Kato T, Muraski J, Chen Y, Tsujita Y, Wall J, Glembotski CC, Schaefer E, Beckerle M, Sussman MA. Atrial natriuretic peptide promotes cardiomyocyte survival by cGMP-dependent nuclear accumulation of zyxin and Akt. J Clin Invest 2005; 115: 2716-2730.

103) Chan CB, Liu X, Tang X, Fu H, Ye K. Akt phosphorylation of zyxin mediates its interaction with acinus-S and prevents acinus-triggered chromatin condensation. Cell Death Differ 2007; 14: 1688-1699.

104) Hiesberger T, Bai Y, Shao X, McNally BT, Sinclair AM, Tian X, Somlo S, Igarashi P. Mutation of hepatocyte nuclear factor-1beta inhibits Pkhd1 gene expression and produces renal cysts in mice. J Clin Invest 2004; 113: 814-825.

105) Youn H, Kim EJ, Um SJ. Zyxin cooperates with PTOV1 to confer retinoic acid resistance by repressing RAR activity. Cancer Lett 2013; 331: 192-199.

106) Hervy M, Hoffman LM, Jensen CC, Smith M, Beckerle MC. The LIM protein zyxin binds CARP-1 and promotes apoptosis. Genes Cancer 2010; 1: 506-515.

107) Fujita Y, Yamaguchi A, Hata K, Endo M, Yamaguchi N, Yamashita T. Zyxin is a novel interacting partner for SIRT1. BMC Cell Biol 2009; 10: 6.

108) Martynova NIu, Ermolina LV, Eroshkin FM, Gioeva FK, Zaraĭskiĭ AG. Transcriptional factor Xanf1 interacts with the focal adhesion protein zyxin in the early development of the Xenopus laevis brain. Bioorg Khim 2008; 34: 573-576.

109) Hervy M, Hoffman L, Beckerle MC. From the membrane to the nucleus and back again: bifunctional focal adhesion proteins. Curr Opin Cell Biol 2006; 18: 524-532.

110) Oldenburg J, van der Krogt G, Twiss F, Bongaarts A, Habani Y, Slotman JA, Houtsmuller A, Huveneers S, de Rooij J. VASP, zyxin and TES are tension-dependent members of Focal Adherens Junctions independent of the α-catenin-vinculin module. Sci Rep 2015; 5: 17225.

111) Zeller Kathrin Stephanie. Integrin signaling in cell adhesion and mechanotransduction. Regulation of P13K, AKT and ROS. Uppsala University Publications, 2012.

112) Zaidel-Bar R, Geiger B. The switchable integrin adhesome. J Cell Sci 2010; 123: 1385-1388.

113) Janoštiak R, Pataki AC, Brábek J, Rösel D. Mechanosensors in integrin signaling: the emerging role of p130Cas. Eur J Cell Biol 2014; 93: 445-454.

114) Moon RT, Bowerman B, Boutros M, Perrimon N. The promise and perils of Wnt signaling through β-catenin. Science 2002; 296: 1644-1646.

115) Hsueh YP, Wang TF, Yang FC, Sheng M. Nuclear translocation and transcription regulation by the membrane-associated guanylate kinase CASK/LIN-2. Nature 2000; 404: 298-302.

116) Yao XL, Li L, He XL, Cui L, Kuang W, Tang M. Activation of β-catenin stimulated by mechanical strain and estrogen requires estrogen receptor in mesenchymal stem cells (MSCs). Eur Rev Med Pharmacol Sci 2014; 18: 3149-3155.

117) Simmons RD, Kumar S, Jo H. The role of endothelial mechanosensitive genes in atherosclerosis and omics approaches. Arch Biochem Biophys 2016; 591: 111-131.

118) Jiang M, Qiu J, Zhang L, Lü D, Long M, Chen L, Luo X. Changes in tension regulates proliferation and migration of fibroblasts by remodeling expression of ECM proteins. Exp Ther Med 2016; 12: 1542-1550.

119) Nishiya N, Sabe H, Nose K, Shibanuma M. The LIM domains of hic-5 protein recognize specific DNA fragments in a zinc-dependent manner in vitro. Nucleic Acids Res 1998; 26: 4267-4273.

120) Terzano C, Laurendi G, Capoccetta G, Rapisarda M, Pacilio R, Petroianni A. The smooth muscle and airway hyperresponsiveness. Eur Rev Med Pharmacol Sci 2003; 7: 9-26.

121) Rosner SR, Pascoe CD, Blankman E, Jensen CC, Krishnan R, James AL, Elliot JG, Green FH, Liu JC, Seow CY, Park JA, Beckerle MC, Paré PD, Fredberg JJ, Smith MA. The actin regulator zyxin reinforces airway smooth muscle and accumulates in airways of fatal asthmatics. PLoS One 2017; 12: e0171728.

122) Song XF, Chang H, Liang Q, Guo ZF, Wu JW. ZEB1 promotes prostate cancer proliferation and invasion through ERK1/2 signaling pathway. Eur Rev Med Pharmacol Sci 2017; 21: 4032-4038.

123) Kei Yamamoto, Yasuki Hori, Jieun Seo, Saho Hashimoto, Yoshihiro Endo, Tadashi Nittami, Yusuke Ito, Hitoshi Ishiguro, Hiroji Uemura, Masatoshi Watanabe. Abstract 522: Involvement of Zyxin in aggressiveness of human prostate cancer. Cancer Res 2015; 75(15 Supplement): 522.

124) Zhuge BZ, Du BR, Meng XL, Zhang YQ. MASTL is a potential poor prognostic indicator in ER+ breast cancer. Eur Rev Med Pharmacol Sci 2017; 21: 2413-2420.

125) Kim YJ, Sertamo K, Pierrard MA, Mesmin C, Kim SY, Schlesser M, Berchem G, Domon B. Verification of the biomarker candidates for non-small-cell lung cancer using a targeted proteomics approach. J Proteome Res 2015; 14: 1412-1419.