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Clinical significance of Cofilin-1 in Early Prediction of Type 1 Cardiorenal Syndrome

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Abstract:

Type 1 cardiorenal syndrome (CRS) is a severe complication for acute decompensated heart failure patients. This review aimed at evaluating the feasibility of using urine cofilin-1 as a biomarker for predicting CRS.

Keywords: Cofilin-1, Cardiorenal Syndrome, cardiac.

Introduction:

The organization of cell populations within animal tissues is essential for the morphogenesis of organs during development. Cells recognize three-dimensional positions with respect to the whole organism, and regulate cell shape, motility, migration, polarization, growth, differentiation, gene expression, and cell death according to extracellular signals. These signals include various soluble factors and molecules that direct cell–cell and cell–substrate adhesion. Remodeling of the actin cytoskeleton is essential for these cellular responses and morphological changes, and must be regulated spatially and temporally by a number of complex signaling pathways **(1)**.

The roles of regulatorsof the actin cytoskeleton and its signaling pathwaysduring tissue development and organmorphogenesis have been actively studied in recentyears. The cofilin/actin-depolymerizing factor (ADF)family proteins (cofilin) are conservedfrom yeast to human. Cofilin, one of the essentialactin regulating proteins, binds to both monomericglobular (G)-actins and filamentous (F)-actins, and seversthe F-actin, which causes the depolymerization ofthe F-actin **(2)**.

Since cofilin has a higher affinity forADP-bound actin filaments than ATP-bound filaments,the “aged” actin filaments are selectively disassembled. These activities are essential for generating actin cytoskeleton dynamics in cells. It has been found that the activity of cofilin is spatiotemporally regulated by phosphorylation, lipid binding, and the binding proteins downstream of various signaling pathways. LIM-kinases and Slingshot phosphatases phosphorylate and dephosphorylate cofilin, respectively. The level of phosphorylation of cofilin in cells is altered dynamically during various cellular responses, and the change in the phosphorylation level is critical for the regulation of actin cytoskeleton dynamics **(3)**.

Structure and regulation of cofilin:

Cofilin was purified and identified as a 20-kDa protein, which induces depolymerization of actin filaments in an extract from avian or porcine brain. The cofilin family is composed of non-muscle type cofilin-1 (also named n-cofilin), muscle type cofilin-2 (also named m-cofilin) and actin depolymerizing factor (ADF, also named destrin) in mammals **(4)**.

At least one cofilin protein exists in all eukaryotic cells. Cofilins are globular proteins with a core consisting of four or five b-sheets, which is surrounded by four or five helices. Cofilin amino acid sequence and structure are highly conserved from human to yeast **(5)**.

The ADF-homology (ADF-H) domain, which contains a homologous sequence to cofilin, is conserved in five actin-related protein families, including twinfilin, coactosin actin-binding protein 1 (ABP1), drebrin, and glia maturation factor-like protein (GMF). Although these proteins exhibit actin or actin-related protein 2/3 (Arp2/3) complex binding activities, they do not have actin depolymerizing and severing activities, except for yeast twinfilin. The functions of the ADF-H domain diversified during evolution **(4)**.

Cofilin preferentially binds to ADP-bound F-actin and destabilizes and severs the actin filaments, resulting in depolymerization of the short actin filaments (figure 1). One of the roles of the actin severing activity of cofilin is to generate free actin monomers for polymerization **(6)**.

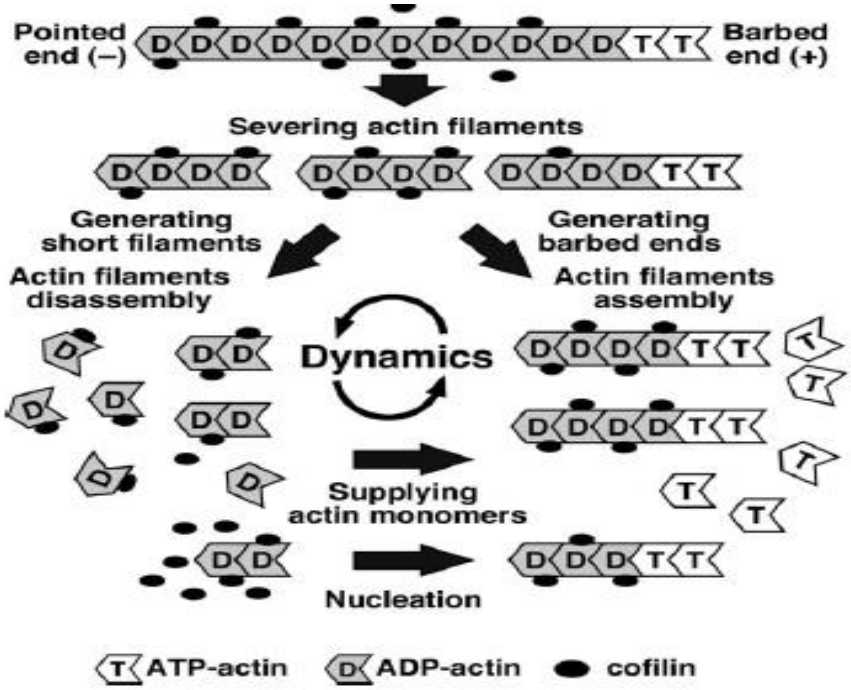

Figure (1): Model of the actin assembly and disassembly by cofilin.Cofilin preferentially binds to ADP-bound actin in an actin filamentand severs the actin filament into short filaments. In one pathway,the short filaments are depolymerized and regenerated toATP-bound actin monomers. The regenerated actin monomerscontribute to assembly of the actin cytoskeleton by increasingthe concentration of actin monomers. In an alternate pathway,the short filaments contribute to increasing the barbed ends andnuclei for actin polymerization. In other pathways, the high concentrationof cofilin promotes the nucleation of actin, leading toactin assembly. Cofilin promotes the dynamics of the actin cytoskeletonby disassembling actin filaments and by supplying actinmonomers, nuclei, and barbed ends.

Cofilin promotes both actin filament disassembly, by severing, and actin polymerization, by supplying actin monomers, resulting in enhanced dynamics of the actin cytoskeleton. An alternative model was proposed in which the severing activity of cofilin generates short filaments and new barbed ends for actin assembly. Furthermore, it was proposed that the high concentration of cofilin promotes the nucleation of actin, leading to actin assembly **(7)**.

When the cellular morphology changes, cofilin promotes the generation of actin monomers, nuclei, and newly barbed ends for the assembly of actin structures such as lamellipodia, filopodia, and stress fibers, and also promotes the turnover of these structures. In contrast, the inactivation of cofilin in response to extracellular signals decelerates the actin turnover and contributes to the assembly of actin structures **(8)**.

Cofilin activity is regulated by several molecular mechanisms (figure 2). Cofilin is inactivated by phosphorylation at an N-terminal Serine-3 (Ser-3) residue, and by the binding of phosphatidylinositol 4,5-bisphosphate (PI (4,5)P2) and cortactin. Likewise, cofilin is activated by actin-interacting protein-1 (Aip1) and cyclase-associated protein-1 (CAP1) **(9)**.

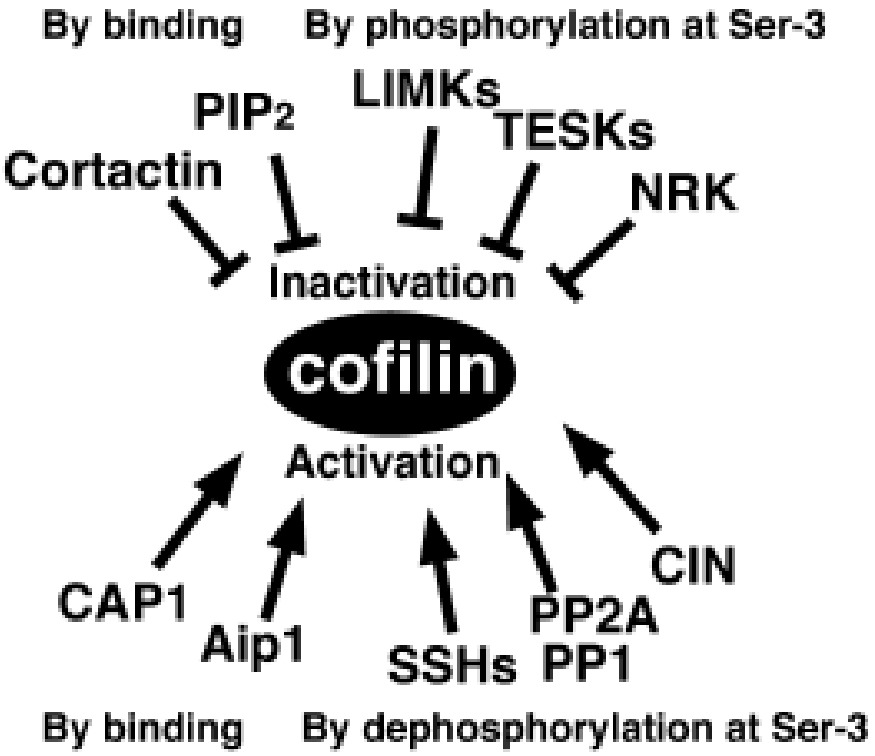

Figure (2): The regulatory molecules of cofilin activity. Cofilin isphosphorylated and inactivated by LIM-kinases (LIMKs), testicularprotein kinases (TESKs), and Nck-interacting kinase (NIK)-relatedkinase (NRK), and by binding of PIP2 and cortactin. In contrast,cofilin is dephosphorylated and reactivated by slingshot proteinphosphatases (SSHs), PP1, PP2A, and chronophin (CIN). Cofilinis also activated by the binding of Aip1 and CAP1.

The phosphorylation level of cofilin is regulated by a number of extracellular signals. Cofilin is phosphorylated at Ser-3 by several kinases. The major kinases of cofilin in mammals are LIM-kinase family proteins, which consist

of LIM domain containing protein kinase 1 (LIMK1), LIMK2, Testicular protein Kinase 1 (TESK1), and TESK2. Nck-interacting kinase (NIK)-related kinase (NRK) also phosphorylates cofilin **(10)**.

On the other hand, cofilin is dephosphorylated by members of the Slingshot (SSH) protein phosphatase family, which includes SSH1, SSH2, and SSH3 in mammals. It was also reported that the haloacid dehalogenase, chronophin (CIN), and the general protein phosphatases, protein phosphatase1 (PP1) and protein phosphatase 2A (PP2A), also dephosphorylate cofilin **(3)**.

Although, cofilin is found in all eukaryotes, from yeast to human, including plants, orthologous LIMK and SSH genes are not found in yeast, Caenorhabditis elegans (C. elegans), Dictyostelium, or in plants. The phospho-regulation of cofilin may have evolved as a mechanism for actin cytoskeleton reorganization during complex multicellular processes in some higher-level organisms **(3)**.

Signal transduction-mediated phosphoregulation of cofilin:

A number of studies have investigated the functions of LIMKs and SSHs, and their respective signaling pathways, during the response to extracellular signals. LIMK1 was discovered while cloning the c-Sea receptor tyrosine kinase from a human cDNA library. The other members, LIMK2, TESK1, and TESK2 were cloned by low-stringency DNA hybridization, using the LIMK1 cDNA as a probe **(11)**.

The LIMK proteins contain tandem, N-terminal LIM domains, which contain Zn-finger motifs, an internal PDZ-like domain and a C-terminal protein kinase domain (figure 3). The TESK proteins are related to the LIMKs and contain the highly homologues kinase domain of LIMKs at the N-terminus and a unique C-terminal proline-rich domain **(11)**.

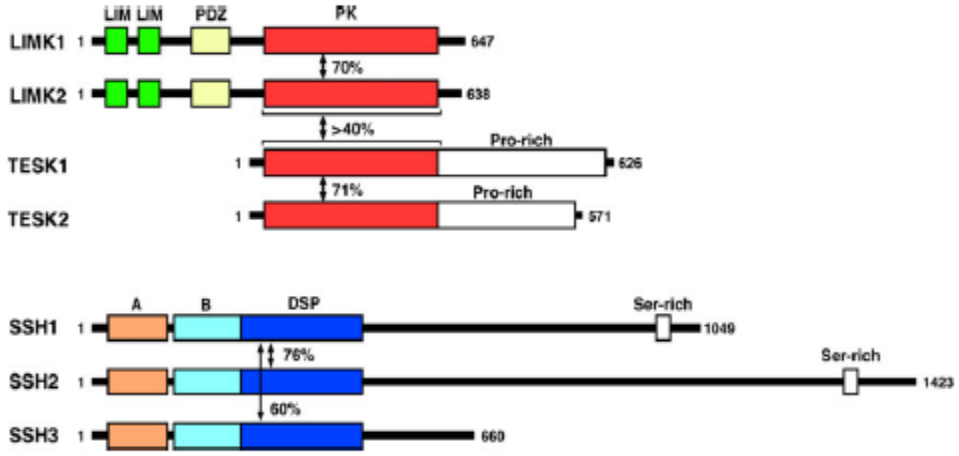

Figure (3): Structures of human LIM-kinases (LIMKs), testicular protein kinases (TESKs), and slingshot protein phosphatases (SSHs). Thenumbers on both sides of the schemata indicate the length of the amino acid sequences of the LIMKs and SSHs. The percentages indicatethe identities of amino acid sequences in the kinase domains. LIM, LIM domain; PDZ, PDZ domain; PK, protein kinase domain;Pro-rich, proline-rich region; A and B, N-terminal conserved domains in SSHs; DSP, dual specificity protein phosphatase domain; Serrich,Serine-rich region.

LIMKs and TESKs phosphorylate cofilin at Ser-3 and inactivate its actin binding activity. LIMK1 is activated and contributes to lamellipodia or stress fiber formation downstream of Rho signaling. LIMKs are phosphorylated at a conserved threonine residue (Thr-508 in human LIMK1 or Thr-505 in human LIMK2) in the activation loop of the kinase domain and activated by Rho-associated kinase (ROCK), p21-activated kinase-1, -2,-4 (PAK1, PAK2, and PAK4), and myotonic dystrophykinase-related Cdc42-binding kinase-a (MRCKa) **(12)**, which are downstreameffectors of RhoA, Rac1 and Cdc42, respectively (figure 4). LIMK1 is also activated downstream of Ca2+signaling. LIMK1 is phosphorylated at the same Thr-508 residue by Ca2+/calmodulin-dependent protein kinase (CaMK) II and IV **(13)**.

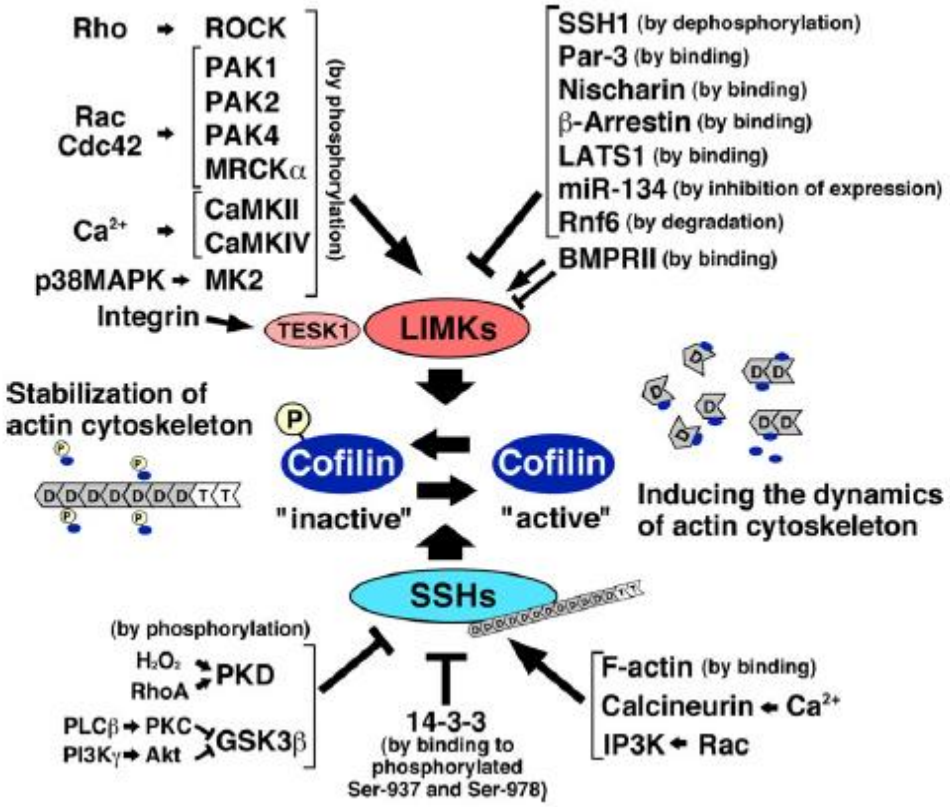

Figure (4): The regulatory mechanisms ofLIM-kinases (LIMKs), testicular proteinkinases (TESKs), and slingshot proteinphosphatases (SSHs). LIMKs are phosphorylatedand activated by the indicatedupstream kinases (*upper left*) and areinactivated by molecules with their indicatedactions (*upper right*). TESK1 is activateddownstream of an integrin signal.SSHs is activated and inactivated by theindicated molecules (*bottom*). Activation ofLIMKs and inactivation of SSHs stabilizethe actin cytoskeleton, and, conversely,inactivation of LIMKs and activation ofSSHs accelerate the dynamics of the actincytoskeleton via a mechanism thatcorresponds to the dephosphorylation ofcofilin.

Another phosphorylation site for theactivation of LIMK1 has been identified. LIMK1 isphosphorylated at the Ser-323 residue by p38 mitogen-activated protein kinase (MAPK)-activated proteinkinase-2 (MAPKAPK-2/MK2) downstream of p38-MAPkinase in response to vascular endothelial growth factor (VEGF) in vascular endothelial cells. BMP signal also regulates LIMK1activity. Bone morphogenic protein (BMP)-4 and -7stimulation activates LIMK1 **(14)**.

However, two groups reportedopposite mechanisms. **Foletta et al. (15)** reported that bindingof LIMK1 to the cytoplasmic C-terminal domain ofBMP receptor II (BMPRII) inhibits LIMK1 and thatLIMK1 is activated when released from BMPRII by the binding of BMP4 to BMPRII. **Lee-Hoeflich et al. (16)** reported that LIMK1 binds to the cytoplasmicdomain of BMPRII and is activated downstreamof Cdc42 when BMP7 activates Cdc42 through BMP receptor I.

In both models, LIMK1 is activated by theBMP stimulation. In contrast, LIMK1 is inactivated byseveral mechanisms. LIMK1 is dephosphorylatedand inactivated by Slingshot-1 phosphatase. LIMKs are also inactivated bythe binding of polarity protein Par-3, Nischarin, b-arrestin, and the tumor suppressor protein, LATS1. The degradation ofLIMK1 by the ubiquitin-proteasome system is inducedby the RING finger E3 ubiquitin ligase, Rnf6. The translation of LIMK1 protein is posttranscriptionallysuppressed by the microRNA, miR-134 **(17)**.

TESK1 is activated by integrin-mediated cell adhesionto the extracellular matrix. Although TESKs are predominantly expressedin the testes, the functions and the regulatory mechanismsof TESKs in mammals are not well known. ADrosophila orthologue of TESK, Center divider (Cdi)was analyzed for a possible role in development **(11)**.

To precisely regulate actin cytoskeleton reorganization, dephosphorylation of cofilin has to be spatiotemporallyregulated by various signaling mechanisms. Slingshot cofilin phosphatase was originally identified by genetic studies of a Drosophila mutant with abnormal morphologies of bristles, wing hairs, and eyes. Slingshot was named for the characteristicbifurcated bristles and hairs. The other family members, SSH2 and SSH3, were isolated from mammals **(18)**.

The SSH proteins contain aconserved N-terminal domain, a protein phosphatase domain, which is distantly related to the MAPK phosphatases (MKPs). SSH1 and SSH2, but not SSH3,have a long C-terminal domain containing a Ser-richregion **(3)**.

Sincethe phenotypes of animals with mutations in actin-related proteins often include abnormal bristles, slingshotwas identified as a cofilin phosphatase. Three mammalian SSH proteins dephosphorylate cofilin at Ser-3. Among the mammalian SSHs, the mechanisms of activation and the functionsof SSH1 are the best characterized **(3)**.

SSH1 localizes to F-actin-rich structures, such asstress fibers, lamellipodia, and cortical actin networksin cultured cells. SSH1 has an F-actin binding activityand the ability of SSH1 to dephosphorylate cofilin isstrongly activated by binding to F-actin **(19)**.

SSH1 is activated bythe stimulation of the chemoattractant, SDF-1, through Rac-phosphoinositide 3-kinase (PI3K) signaling in Jurkat T cells. Increasing the Ca2+permeability of cells using a calcium ionophore activatescalcineurin, which induces SSH1 activation andcofilin dephosphorylation **(20)**.

SSH1 is also negatively regulated by phosphorylationand by binding 14-3-3 proteins. 14-3-3 proteinbinds to phosphorylated Ser-937 and Ser-978 residuesin the C-terminal, serine-rich region of humanSSH1 and inhibits the binding of SSH1 to actin filaments.The dissociation of 14-3-3 protein from SSH1facilitates neuregulin-induced lamellipodia formation **(21)**.

Protein kinase D (PKD) isactivated by hydrogen peroxide (H2O2) and phosphorylatesSSH1 at Ser-937 and Ser-978 downstream ofRhoA, resulting in the induction of actin polymerization. PKD alsophosphorylates the conserved Ser-402 of SSH1 withinthe phosphatase domain, thereby inactivating SSH1. In addition, PKD phosphorylatesand activates PAK4, thereby activating LIMK1 **(22)**.

Tang et al. (23) indicated that PKD is a negativeregulator of SSH1 and elevates the level of phospho-cofilin. Glycogen synthase kinase 3b (GSK3b) phosphorylates the N-terminal region of SSH2 and inhibits SSH2 activity. When neutrophilis stimulated by chemoattractants, GSK3b isphosphorylated and inactivated by the PLCb-PKC

andPI3Kb-Akt signaling pathways, resulting in the activationof SSH2 **(23)**. The activity and the localization of LIMKs/TESKs and SSHs are coordinately regulated downstreamof various signaling pathways.

Expression patterns of cofilin; LIMKs, TESKs, and SSHs:

Cofilin-1 and ADF are expressed in almost all tissues and cells. The ratio of the amount of cofilin-1 and ADF differs in each cell type and within each developing period. The differences in the ratios correlate with the different phenotypes of cofilin-deficient and ADF-deficient mice. Muscle type cofilin-2 is specifically expressed in skeletal and cardiac muscles **(24)**.

limk1 mRNA is predominantly expressed in neural tissues, and both limk1 and limk2 mRNAs are widely expressed in various developing and adult tissues **(25)**. tesk1 and tesk2 mRNAs are predominantly expressed in testes and are detected at lower levels in other tissues **(11)**. The ssh1,ssh2, and ssh3 mRNAs are widely expressed in varioustissues with distinctly different patterns **(26)**.

Role of cofilin-1 in prediction of type 1 cardiorenal syndrome:

Cofilin-1 is an essential structural protein of renal tubular cells and plays a crucial role in mediating cell actin-associated motility and shape. For ςthis reason, cofilin-1 is thought to be one of the most important mediators for epithelial-mesenchymal transition or de-differentiated renal tubular cells, which are critical for AKI and long-term renal function loss **(27)**.

The higher intracellular cofilin-1 level indicates renal cells changed in both phenotype and function in nephropathy due to the inflammatory process. Additionally, adding angiotensin in cell culture induces cofilin-1-related cell cycle arrest, which has been the most important biomarker to AKI in recent years **(28)**.

Therefore, the close relationships between cofilin-1 and epithelial-mesenchymal transition and cell cycle arrest assume the central role of urine cofilin-1 level in detecting CRS under the proper measurement method. Moreover, cofilin-1 also correlates with the severity of HF as atrial natriuretic peptide, and the dual role of cofilin-1 in heart and renal failure may also be the reason why cofilin-1 could be used as a biomarker among CRS patients **(29)**.

Chen et al. (30) detected urine cofilin-1 as a biomarker for predicting CRS among patients in the coronary care unit (CCU) and concluded that the gold nanoparticle-based immunoassay localized surface plasmon-coupled fluorescence biosensor (LSPCFB) could exploit the potential of urine cofilin-1 as a single biomarker to predict CRS among CCU patients.

Clinical significance of cofilin:

Cancer:

For the past two decades, scientists have extensively studied the role of cofilin in tumor invasion ability, but this research has continued and has carried out new research directions in cancer metastasis. The lamellar pseudopodia play a major role in driving the migration of cancer cells, and cortical proteins regulate the formation of dendritic pseudopodia through phosphorylation and dephosphorylation, a number of studies have shown that metastasis of cancers is associated with cofilin-related mechanisms that regulate the movement of these cells **(9)**.

Studies have been conducted to screen cofilin-1 as differentially expressed proteins in melanoma **(31)** and ovarian cancer tissues **(32)**. The expression of activated cofilin-1 was significantly increased in cell lines such as breast

cancer (MTLn3) **(33)**, lung cancer (A549) **(34)** and prostate cancer (PC3) **(35)**, while the expression of phosphorylated inactivated cofilin-1 was significantly decreased.

Activated cofilin-1 could induce the formation of invasive pseudopodia, determine the migration direction of cancer cells, promote the proliferation and invasion of cancer cells, and decrease the activation free cofilin-1 after knockout of cell lines, cell invasive pseudopodia production decreased or maturation disorder, cancer cell proliferation, invasion ability greatly weakened. Cofilin-1 high expression is related to the formation of drug resistance mechanism after chemotherapy in lung adenocarcinoma and ovarian cancer, and provides a target for drug therapy **(36)**.

The ROCK/Lin11, ISL-1 and LIMK/cofilin signaling cascades are thought to be crucial for the regulation of this link in the cofilin molecular regulatory mechanisms of cancer metastasis. More broadly, the factors that activate the signaling cascade mainly include receptor tyrosine kinases, G protein-coupled receptors, integrins and their ligands, growth factors, hormones, fibronectin, collagen, and laminin. This also provides a theoretical basis for ROCK/LIMK/cofilin signaling proteins as good candidates for cancer prevention strategies or treatment **(37)**.

Sengelaub et al. (38) have shown that these molecules are associated with known signaling pathways that affect cofilin, like PLCY1, PLCβ1 which are considered another pathway for cofilin activation in breast cancer. LIMK controls cancer cell migration by connecting signals from Rho families, altering cofilin activity and regulating actin dynamics. Whereas in lung cancer A549 cells and human osteoblastoma cells, there is a cofilin signaling pathway that responds to folate levels and stromal cell-derived factor-1 to regulate cell motility, respectively.

Li and Chen (39) implicated that Pak1 is associated with the regulation of the Limk1/cofilin pathway, which is responsible for governing cell motility and morphologic changes, thereby facilitating cancer metastasis. They also suggested the correlation between cancer metastasis and ROCK/LIMK/cofilin signaling proteins, so some scholars have proposed therapeutic strategies to use ROCK study of tumor metastasis prevention.

Neurological diseases:

ADF/cofilin plays an important role in the improvement of the structure and function of the nervous system, such as growth cone extension, axonal transport, cell tail retraction and so on. Cofilin effects on nervous system diseases are mainly carried out in three aspects:

(1) Involvement in neuronal differentiation: Talens-Visconti have found that the Rho protein family is widely distributed in the nervous system, and the RhoE molecules can promote the remodeling of actin to induce neuronal differentiation by inhibiting the signal pathway **(37)**.

(2) Participation in neuronal movements: lee and other studies have found that the directionalmovement of neurons is closely related to the dynamic changes of actin. The leading edge of neurons pushes the neurons forward through F-actin remodeling and pulls the neurons forward at the base through the depolymerization of the neurons. Marsick studies have found that nerve growth factors and netrin-1 are injected into temporal retinal nerve cells to activate cofilin signaling pathways in nerve cells to accelerate actin polymerization. Growth cone cells (vegetative cone) that are suitable for growth in the environment eventually turn to growth **(40)**.

(3) The plasticity involved in synapses: actin cytoskeleton plays an important role in the structure and function of postsynaptic and synaptic plasticity **(41)**.

Cofilin can accelerate the depolymerization of actin monomers by severing actin filaments, thus improving the turnover and deformation ability of actin. This characteristic makes cofilin have the ability to regulate the dynamic changes of dendrites and spinous processes of neurons. Zhang studies have found that when the Ser3 site of cofilin protein is in a non-phosphorylated state, the protein is activated, which causes the morphological structure of dendritic spine to deform **(41)**.

Due to the role of these cofilin in the development of nervous system diseases, cofilin is mainly involved in the pathogenesis of epilepsy, Parkinson's disease, Alzheimer's disease, spinal cord injury, ischemic brain injury and neuropathic pain. In addition, it was also found that cofilin can play an important role in regulating the progression of spinal cord lateral bundle-related diseases **(42)**.

Xiao et al. (43) found that the expression level of cyclin (cell division cycle 42, cdc42) in patients with epilepsy was significantly increased, which could cdc42 activate LIM1 and promote the phosphorylation of cofilin protein. The formation of neural cones and processes leads to the formation of abnormal neural circuits.

An LRRK2-PKA-cofilin signaling pathway is associated with Parkinson's disease. Parisiadou found out: after LRRK2 gene knockout, the activity of SPNs protein kinase A and postsynaptic density protein-95 in striatal projection neurons decreased, and the expression of the latter decreased, which led to the hyperphosphorylation of cofilin and hindered the maturation of dendrites **(44)**.

Cofilin is associated with chronic neuroinflammation during Alzheimer's disease, a process that exacerbates the oxidative cascade of neurodegeneration by cofilin- actin complexes to accelerate mitochondrial decline and ATP depletion and destroy essential actin dynamics. The Mst3b/LIMK1/cofilin system and involved in the pathogenesis and repair of spinal cord injury. When the Mst3b/LIMK1/cofilin system in the injured spinal cord cells is overactivated, the injured spinal cord cells have more growth cone branches, while in the contrast test, the pathway is inhibited by silencing Mst3b molecules.It was found that there were very few neural cones in this kind of spinal cord cells. The pathogenesis of ischemic brain injury is similar to that of this disease, both of which are due to the imbalance of cofilin and p-cofilin. This physiological process is related to hypoxic-ischemic brain damage, and may also be related to the occurrence of learning and memory dysfunction **(45)**.

Qiu et al. (46) have found that when various factors stimulate the sciatic nerve to cause its injury, the Rho/LIMK/cofilin system will be activated, causing neurogenic pain in the body, while in contrast tests, the use of Simvastatin to treat injured neurons can reduce the activity of the Rho/LIMK/cofilin system. Symptoms of neurogenic pain are also alleviated.

Shaw and Bamburg (47) have found it possible to treat neurological diseases by regulating cofilin activity and controlling actin. During treatment, cofilin activity-related neurological diseases can be treated by developing targeted and cell-specific peptide reagents. Current research needs to focus on developing NLG1-CTD peptides with higher affinity to bind and lead to SPAR degradation, but this peptide may provide a neuron-specific cofilin phosphorylation enhancer that works upstream of LIMK activation, and possibly long term stability through the use of D-amino acids, might be the future direction for research in this area.

Kidney disease:

The phenotypic characteristics of the inherent cells in renal tissue can be changed with different physiological and pathological states. Except for glomerular endothelial cells, all the other inherent cells have reported cytoskeletal proteins as phenotypic markers. As an important member of the cytoskeletal protein, silk fibroin also plays an

important role in renal disease by regulating the actin cytoskeleton of various inherent cells of kidney. Cofilin-related renal diseases are associated with glomerular epithelial cells and renal tubular epithelial cells.

First, actin dynamics determines and maintains the normal morphology of renal podocytes during development, as well as its recovery after damage, and plays an important role in the branches of ureteral buds during renal development. The loss of cofilin-1 and destrin in the epithelium of the ureter bud in mice caused normal epithelial tissue to be disturbed, cell migration to be inhibited, and branch morphogenesis of the kidney was inhibited in the early stage, resulting in renal failure **(48)**.

Mutant mice with specific knockdown of podocyte cofilin were unable to form podocyte secondary podocytes, resulting in typical podocyte fusion and a large amount of proteinuria and renal impairment were observed at three months **(49)**.

Lee et al. (50) confirmed that TGF-β stimulation can cause phosphorylation of cofilin in mice and human podocytes. **Garg et al. (51)** have confirmed in vitro that cofilin and nephrin are located on the cell membrane together, which can be dephosphorylated and activated by PI3K/SSH1L pathway to maintain the normal morphology and function of podocytes. The specific knockout of podocyte cofilin gene can lead to podocyte injury and proteinuria. In mouse and human podocytes, cofilin and synaptic polar proteins are distributed in the same position. The therapeutic effect of cyclosporine A on massive proteinuria in nephrotic syndrome is related to the immune regulation of T cells and the stabilization of podocyte actin cytoskeleton by acting on synaptic polar proteins. Cyclosporine A can inhibit purine mycin-induced fusion of rat podocytes and decrease the expression of synaptic polar protein, reduce proteinuria, and increase the expression of cofilin. The effect of cyclosporine A on reducing urinary protein may be to stabilize the actin cytoskeleton of podocytes by up regulating the expression of cofilin.

Kolavennu et al. (52) showed that high glucose and high osmotic pressure can activate the Rho-ROCK signaling pathway and inactivate the phosphorylation of cofilin in diabetes mellitus, which affects actin dynamics and is one of the pathogenic factors of diabetic nephropathy.

Secondly, **Thirone et al. (53)** have found that cofilin promotes actin depolymerization and plays a key role in maintaining the polarity and function of proximal tubular epithelial cells. Cofilin proteins play an important role in the acidification mechanism of renal tubular epithelial cells and the assembly and transportation macromolecular substances by depolymerizing filamentous actin.

When porcine proximal tubular epithelial cells were cultured in high glucose environment for 6 h, the of intracellular p-cofilin and LIMK1 increased in a time-dependent manner, which indicated that high glucose glucose could increase the phosphorylation of cofilin by activating RhoA/ROCK/LIMK1 signaling pathway **(54)**.

Actin kinetics changes damage the polarity and function of proximal tubular epithelial cells. Recent studies also indicate that cofilin may regulate the inflammatory response of renal tissue in hypertensive nephropathy by affecting the nuclear factor kappa-B activity of renal tubular epithelial cells and nuclear translocation. grape seed proanthocyanidins can regulate this inflammatory response through antioxidant mechanisms **(55)**.

Heart disease:

Cofilin and actin alterations, as well as their interactions, have been shown to be important for the study of the pathogenesis of human myocardial disease. For example, the study noted that drug-stimulated cofilin-2 phosphorylation and gene overexpressed phosphorylation protein promote accumulated “stress-like” fibers and severely damaged cardiac cell contraction. Cofilin-1 also correlates with the severity of heart failure as an atrial

natriuretic peptide, and the dual role of cofilin-1 in heart and renal failure may also be the reason why cofilin-1 could be used as a biomarker among cardiorenal syndrome patients **(30)**.

Aggregates with presenilin-1 and cofilin-2 have been identified in up to one-third of idiopathic dilated cardiomyopathy cases studied, indicating the potential predominance of misfolded proteins in heart failure **(56)**.

But, the role of cofilin in heart disease is still lacking, and many intermediate links are unclear. Some studies have pointed out that cofilin-2 is a specific target for miR-21, and hydrogen sulfide can protect ischemia and inflammatory injury after myocardial ischemia by inducing microRNA-21. And 28 days after myocardial infarction, the expression of cofilin-2 promoting apoptosis has changed significantly **(57)**. **Xiao et al. (43)** have shown that cofilin-2 and Bax form a protein complex in cardiomyocytes in response to oxidative stress; therefore, it suggests that cofilin-2 may function as a Bax transporter of mitochondria. But overall, cofilin studies on the direction of heart disease are lacking.

Osteoarthritis:

Cytoskeleton is a protein fiber grid system in eukaryotic cells, which plays an important role in the biomechanical or biochemical pathway of cell-specific stress conduction. Actin filaments are the main components of the cytoskeleton, providing the main viscoelastic solid features of chondrocytes. Destruction of the chondrocyte cytoskeleton is considered to be an important factor leading to a degeneration of articular cartilage. Cytoskeleton is involved in stress conduction in chondrocytes and may convert mechanical signals into biochemical signals, in which ADF/cofilin phosphorylation/dephosphorylation signaling pathways play an important role. When chondrocytes are subjected to certain physical or chemical stimulation, the free G-actin polymerizes to form F-actin, and microfilaments are formed through their own helix to complete the rearrangement of the cytoskeleton. Cofilin genes and proteins are highly expressed in chondrocyte model cells induced by mechanical force in the early stage of osteoarthritis, suggesting that cofilin gene expression is related to the biological characteristics of chondrocytes in the early stage of osteoarthritis **(58)**.

Meanwhile, the cytoskeleton of osteoarthritis chondrocytes was rearranged under low concentration of leptin, which up regulated the protein content of cofilin and LIMK and inhibited the activity of cofilin protein. Moreover, cofilin-1 is associated with many inflammatory conditions, such as osteoarthritis and inflammatory pain, and some investigations indicated that overexpression of cofilin-1 can decrease glucocorticoid receptor expression and nuclear factor kappa-B activity **(59)**.

Additionally, stabilizing actin filaments by inhibiting gene expression of the two main actin depolymerizing factors: cofilin 1 and destrin in hMSCs, enhanced cell viability and differentiation into osteoblastic cells in vitro, as well as heterotopic bone formation in vivo **(60)**.

Concluding remarks:

Through the discussion of the latest mechanism of cofilin action, we believe that the advantages of cofilin in clinical practice are gradually reflected, and the most used at present is its research as a biomarker of tumor, nervous system disease and heart/kidney disease. But we also cannot ignore the significance of cofilin as actin-binding protein that severs actin filaments to cell movement. As a result, the presence of cofilin is very important in the rearrangement of chondrocytes and neurons, which also provides a basis for targeted clinical treatment of osteoarthropathy and nervous system diseases. Current diseases resulting from changes in activity due to phosphorylation of actin-binding proteins, are often possible to control actin-binding activity by designing targeted drugs, and its potential cannot be underestimated **(61)**.

For example, phosphorylation of many actin-binding proteins is regulated by RhoA/ROCK and the cAMP/PKA pathways, these signaling pathways also play an important role. Because cofilin protein is widely existed in human body, when using its phosphorylation inhibitor as a treatment method, it often has systemic side effects **(62)**.

Peng et al. (58) reported that warm acupuncture in rat knee osteoarthritis by down-regulating the expression of chondrocyte cytoskeleton protein ROCK, p-cofilin and phosphorylated LIMK1 to reduce arthritis injury in rats with knee osteoarthritis. This kind of treatment is worthy of further discussion.

References:

1. Etienne-Manneville, S. and Hall, A. Rho GTPases in cell biology. Nature 2002; 420: 629–635.
2. Bernstein, B. W. and Bamburg, J. R. ADF/cofilin: a functional node in cell biology. Trends Cell Biol 2010; 20: 187–195.
3. Mizuno, K. Signaling mechanisms and functional roles of cofilin phosphorylation and dephosphorylation. Cell. Signal. 2013; 25: 457–469.
4. Poukkula, M., Kremneva, E., Serlachius, M. and Lappalainen, P. Actin-depolymerizing factor homology domain: a conserved fold performing diverse roles in cytoskeletal dynamics. Cytoskeleton 2011; 68: 471–490.
5. Ono, S. Mechanism of depolymerization and severing of actin filaments and its significance in cytoskeletal dynamics. Int. Rev. Cytol 2007; 258: 1–82.
6. Kiuchi, T., Nagai, T., Ohashi, K. & Mizuno, K. Measurements of spatiotemporal changes in G-actin concentration reveal its effect on stimulus-induced actin assembly and lamellipodium extension. J. Cell Biol. 2011; 193: 365–380.
7. Oser, M. and Condeelis, J. The cofilin activity cycle in lamellipodia and invadopodia. J. Cell. Biochem. 2009; 108: 1252–1262.
8. Ohashi, K., Sampei, K., Nakagawa, M., Uchiumi, N., Amanuma, T., Aiba, S., Oikawa, M. and Mizuno, K. Damnacanthal, an effective inhibitor of LIM-kinase, inhibits cell migration and invasion. Mol. Biol. Cell 2014; 25: 828–840.
9. Oser, M., Yamaguchi, H., Mader, C. C., Bravo-Cordero, J. J., Arias, M., Chen, X., DesMarais, V., Van Rheenen, J., Koleske, A. J. and Condeelis, J. Cortactin regulates cofilin and N-WASp activities to control the stages of invadopodium assembly and maturation. J. Cell Biol. 2009; 186: 571–587.
10. Nakano, K., Kanai-Azuma, M., Kanai, Y., Moriyama, K., Yazaki, K., Hayashi, Y. and Kitamura, N. Cofilin phosphorylation and actin polymerization by NRK/NESK, a member of the germinal center kinase family. Exp. Cell Res. 2003; 287: 219–227.
11. Toshima, J., Toshima, J. Y., Amano, T., Yang, N., Narumiya, S. and Mizuno, K. Cofilin phosphorylation by protein kinase testicular protein kinase 1 and its role in integrin-mediated actin reorganization and focal adhesion formation. Mol. Biol. Cell 2001; 12: 1131–1145.
12. Ahmed, T., Shea, K., Masters, J. R., Jones, G. E. and Wells, C. M. A PAK4-LIMK1 pathway drives prostate cancer cell migration downstream of HGF. Cell. Signal 2008; 20: 1320–1328.

13. Saito, A., Miyajima, K., Akatsuka, J., Kondo, H., Mashiko, T., Kiuchi, T., Ohashi, K. and Mizuno, K. CaMKIIb-mediated LIM-kinase activation plays a crucial role in BDNF-induced neuritogenesis. Genes Cells 2013; 18: 533–543.
14. Kobayashi, M., Nishita, M., Mishima, T., Ohashi, K. and Mizuno, K. MAPKAPK-2-mediated LIM-kinase activation is critical for VEGF-induced actin remodeling and cell migration. EMBO J 2006; 25: 713–726.
15. Foletta, V. C., Lim, M. A., Soosairajah, J., Kelly, A. P., Stanley, E. G., Shannon, M., He, W., Das, S., Massague, J. and Bernard, O. Direct signaling by the BMP type II receptor via the cytoskeletal regulator LIMK1. J. Cell Biol. 2003; 162: 1089–1098.
16. Lee-Hoeflich, S. T., Causing, C. G., Podkowa, M., Zhao, X., Wrana, J. L. and Attisano, L. Activation of LIMK1 by binding to the BMP receptor, BMPRII, regulates BMPdependent dendritogenesis. EMBO J 2004; 23: 4792–4801.
17. Ding, Y., Milosavljevic, T. and Alahari, S. K. Nischarin inhibits LIM kinase to regulate cofilin phosphorylation and cell invasion. Mol. Cell. Biol 2008; 28: 3742–3756.
18. Niwa, R., Nagata-Ohashi, K., Takeichi, M., Mizuno, K. and Uemura, T. Control of actin reorganization by Slingshot, a family of phosphatases that dephosphorylate ADF/cofilin. Cell 2002; 108: 233–246.
19. Kurita, S., Watanabe, Y., Gunji, E., Ohashi, K. and Mizuno, K. Molecular dissection of the mechanisms of substrate recognition and F-actin-mediated activation of cofilinphosphatase Slingshot-1. J. Biol. Chem. 2008; 283: 32542–32552.
20. Wang, Y., Shibasaki, F. & Mizuno, K. Calcium signalinduced cofilin dephosphorylation is mediated by Slingshot via calcineurin. J. Biol. Chem. 2005; 280: 12683–12689.
21. Nagata-Ohashi, K., Ohta, Y., Goto, K., Chiba, S., Mori, R., Nishita, M., Ohashi, K., Kousaka, K., Iwamatsu, A., Niwa, R., Uemura, T. and Mizuno, K. A pathway of neuregulininduced activation of cofilin-phosphatase Slingshot and cofilin in lamellipodia. J. Cell Biol. 2004; 165: 465–471.
22. Spratley, S. J., Bastea, L. I., Doppler, H., Mizuno, K. and Storz, P. Protein kinase D regulates cofilin activity through p21-activated kinase 4. J. Biol. Chem. 2011; 286: 34254–34261.
23. Tang, W., Zhang, Y., Xu, W., Harden, T. K., Sondek, J., Sun, L., Li, L. and Wu, D. A PLCb/PI3Kc-GSK3 signaling pathway regulates cofilin phosphatase slingshot2 and neutrophil polarization and chemotaxis. Dev. Cell 2011; 21: 1038–1050.
24. Gurniak, C. B., Perlas, E. and Witke, W. The actin depolymerizing factor n-cofilin is essential for neural tube morphogenesis and neural crest cell migration. Dev. Biol. 2005; 278: 231–241.
25. Mori, T., Okano, I., Mizuno, K., Tohyama, M. and Wanaka, A. Comparison of tissue distribution of two novel serine/threonine kinase genes containing the LIM motif (LIMK-1 and LIMK-2) in the developing rat. Brain Res. Mol. Brain Res. 1997l; 45: 247–254.
26. Kousaka, K., Kiyonari, H., Oshima, N., Nagafuchi, A., Shima, Y., Chisaka, O. and Uemura, T. Slingshot-3 dephosphorylates ADF/cofilin but is dispensable for mouse development. Genesis 2008; 46: 246–255.

27. Rozenbaum Z, Benchetrit S, Minha S, Neuman Y, Shlezinger M, Goldenberg I, Mosseri M, Pereg D. The effect of admission renal function on the treatment and outcome of patients with acute coronary syndrome. Cardiorenal Med 2017; 7: 169–178.
28. Ronco C. Cell-cycle arrest biomarkers: the light at the end of the acute kidney injury tunnel. Nephrol Dial Transplant 2016; 31: 3–5.
29. Huntley BK, Heublein DM, Sandberg SM, Burnett JC. Cofilin-1: a possible new biomarker for human heart failure. J Card Fail 2007; 13: S101.
30. Chen HY, Chou C, Chang CH. Urine cofilin-1 detection for predicting type 1 cardiorenal syndrome in the coronary care unit: a gold nanoparticle- and laser-based approach. Cardiorenal Med 2018; 8(4): 302-310.
31. Guan M, Chen X, Ma Y (2015): MDA-9 and GRP78 as potential diagnostic biomarkers for early detection of melanoma metastasis. Tumour Biol 2015; 36(4): 2973-2982.
32. Chen P, Zeng M, Zhao Y. Upregulation of Limk1 caused by microRNA-138 loss aggravates the metastasis of ovarian cancer by activation of Limk1/cofilin signaling. Oncol Rep 2014; 32(5): 2070-2076.
33. Huang X, Pan Q, Sun D. O-GlcNAcylation of cofilin promotes breast cancer cell invasion. J Biol Chem 2013; 288(51): 36418-36425.
34. Oleinik NV, Helke KL, Kistner-Griffin E. Rho GTPases RhoA and Rac1 mediate effects of dietary folate on metastatic potential of A549 cancer cells through the control of cofilin phosphorylation. J Biol Chem 2014; 289(38): 26383-26394.
35. Collazo J, Zhu B, Larkin S. Cofilin drives cell-invasive and metastatic responses to TGF-beta in prostate cancer. Cancer Res 2014; 74(8): 2362-2373.
36. Becker M, De Bastiani M A, Muller C B. High cofilin-1 levels correlate with cisplatin resistance in lung adenocarcinomas. Tumour Biol 2014; 35(2): 1233-1238.
37. Lee M H, Kundu J K, Chae J I. Targeting ROCK/LIMK/cofilin signaling pathway in cancer. Arch Pharm Res 2019; 42(6): 481-491.
38. Sengelaub CA, Navrazhina K, Ross JB. PTPRN2 and PLCbeta1 promote metastatic breast cancer cell migration through PI(4,5)P2-dependent actin remodeling. EMBO J 2016; 35(1): 62-76.
39. Li H, Chen C. Quercetin has antimetastatic effects on gastric cancer cells via the interruption of uPA/uPAR function by modulating NF-kappab, PKC-delta, ERK1/2, and AMPKalpha. Integr Cancer Ther 2018; 17(2): 511-523.
40. Talens-Visconti R, Peris B, Guerri C. RhoE stimulates neurite-like outgrowth in PC12 cells through inhibition of the RhoA/ROCK-I signalling. J Neurochem 2010; 112(4): 1074-1087.
41. Hotulainen P, Hoogenraad CC. Actin in dendritic spines: connecting dynamics to function. J Cell Biol 2010; 189(4): 619-629.
42. Sivadasan R, Hornburg D, Drepper C. C9ORF72 interaction with cofilin modulates actin dynamics in motor neurons. Nat Neurosci 2016; 19(12): 1610-1618.

43. Xiao F, Wang XF, Li JM. Overexpression of N-WASP in the brain of human epilepsy. Brain Res 2008; 1233: 168-175.
44. Parisiadou L, Yu J, Sgobio C. LRRK2 regulates synaptogenesis and dopamine receptor activation through modulation of PKA activity. Nat Neurosci 2014; 17(3): 367-376.
45. Bamburg JR, Bernstein BW. Actin dynamics and cofilin-actin rods in alzheimer disease. Cytoskeleton (Hoboken) 2016; 73(9): 477-497.
46. Qiu Y, Chen WY, Wang ZY. Simvastatin attenuates neuropathic pain by inhibiting the RhoA/LIMK/cofilin pathway. Neurochem Res 2016; 41(9): 2457-2469.
47. Shaw AE, Bamburg JR. Peptide regulation of cofilin activity in the CNS: a novel therapeutic approach for treatment of multiple neurological disorders. Pharmacol Ther 2017; 175: 17-27.
48. Morita R, Kihira M, Nakatsu Y. Coordination of cellular dynamics contributes to tooth epithelium deformations. PLoS One 2016; 11(9): e161336.
49. Ashworth S, Teng B, Kaufeld J. Cofilin-1 inactivation leads to proteinuria: Studies in zebrafish, mice and humans. PLoS One 2010; 5(9): e12626.
50. Lee J, Ko M, Joo CK. Rho plays a key role in TGF-beta1-induced cytoskeletal rearrangement in human retinal pigment epithelium. J Cell Physiol 2008; 216(2): 520-526.
51. Garg P, Verma R, Cook L. Actin-depolymerizing factor cofilin-1 is necessary in maintaining mature podocyte architecture. J Biol Chem 2010; 285(29): 22676-22688.
52. Kolavennu V, Zeng L, Peng H. Targeting of RhoA/ROCK signaling ameliorates progression of diabetic nephropathy independent of glucose control. Diabetes 2008; 57(3): 714-723.
53. Thirone AC, Speight P, Zulys M. Hyperosmotic stress induces Rho/Rho kinase/LIM kinase-mediated cofilin phosphorylation in tubular cells: key role in the osmotically triggered F-actin response. Am J Physiol Cell Physiol 2009; 296(3): C463-C475.
54. Ishibashi F. High glucose increases phosphocofilin via phosphorylation of LIM kinase due to Rho/Rho kinase activation in cultured pig proximal tubular epithelial cells. Diabetes Res Clin Pract 2008; 80(1): 24-33.
55. Wang QZ, Gao HQ, Liang Y. Cofilin1 is involved in hypertension-induced renal damage via the regulation of NF-kappaB in renal tubular epithelial cells. J Transl Med 2015; 13: 323.
56. Parry TL, Melehani JH, Ranek MJ. Functional amyloid signaling via the inflammasome, necrosome, and signalosome: new therapeutic targets in heart failure. Front Cardiovasc Med 2015; 2: 25.
57. Nguyen K, Chau VQ, Mauro AG. Hydrogen sulfide therapy suppresses cofilin-2 and attenuates ischemic heart failure in a mouse model of myocardial infarction. J Cardiovasc Pharmacol Ther 2020; 25(5): 472-483.
58. Peng R, Li J, Li J. Warm acupuncture improves arthritic injury by down-regulating expression of skeleton proteins in rats with knee osteoarthritis. Acupunct Res 2020; 45(2): 105–110.
59. Yang L, Li FF, Han YC, et al. Cannabinoid receptor CB2 is involved in tetrahydrocannabinol-induced anti-inflammation against lipopolysaccharide in MG-63 cells. Mediators Inflamm 2015; 362126.

60. Chen L, Shi K, Frary CE. Inhibiting actin depolymerization enhances osteoblast differentiation and bone formation in human stromal stem cells. Stem Cell Res 2015; 15(2): 281–289.
61. Yin LM, Ulloa L, Yang YQ. Transgelin-2: biochemical and clinical implications in cancer and asthma. Trends Biochem Sci 2019; 44(10): 885–896.
62. Yin LM, Duan TT, Ulloa L. Ezrin orchestrates signal transduction in airway cells. Rev Physiol Biochem Pharmacol 2018; 174: 1–23.