BioArchitecture 2:3, 75–87; May/June 2012; © 2012 Landes Bioscience

The actin cytoskeleton as a sensor and mediator of apoptosis

Melissa Desouza, Peter W. Gunning and Justine R. Stehn*

Oncology Research Unit; School of Medical Sciences; The University of New South Wales; Sydney, Australia

Keywords: actin, apoptosis, actin binding proteins, mitochondria, Bcl-2, cancer, multi-drug resistance

Abbreviations: ABP/s, actin-binding protein/s; ADF, actin depolymerizing factor; Bcl-2, B cell lymphoma 2; CIN, chronophin; DISC, death-inducing signaling complex; F-actin, filamentous actin; FADD, fas-associated death domain; G-actin, globular actin; Fractin, fragment actin; HMW, High molecular weight; LIMK, LIM kinase; LMW, Low molecular weight; MLCK, myosin light chain kinase; MDR, multi-drug resistance; N-Gelsolin, NH_2-terminal gelsolin; OMM, outer mitochondrial membrane; mt$\Delta\Psi$, mitochondrial membrane potential; SSH, slingshot homolog; TESK, testicular kinase; TNFα, tumor necrosis factor-alpha; tActin, truncated actin; VDAC, voltage-dependent anion channel

Apoptosis is an important biological process required for the removal of unwanted or damaged cells. Mounting evidence implicates the actin cytoskeleton as both a sensor and mediator of apoptosis. Studies also suggest that actin binding proteins (ABPs) significantly contribute to apoptosis and that actin dynamics play a key role in regulating apoptosis signaling. Changes in the organization of the actin cytoskeleton has been attributed to the process of malignant transformation and it is hypothesized that remodeling of the actin cytoskeleton may enable tumor cells to evade normal apoptotic signaling. This review aims to illuminate the role of the actin cytoskeleton in apoptosis by systematically analyzing how actin and ABPs regulate different apoptosis pathways and to also highlight the potential for developing novel compounds that target tumor-specific actin filaments.

Hallmarks of Apoptosis

Apoptosis or programmed cell death is an essential biological function required during embryogenesis, tissue homeostasis, organ development and immune system regulation.[1-4] The importance of apoptosis in organism development is now recognized by the myriad of pathologies associated with the de-regulation of apoptotic signaling pathways leading to cancer, autoimmune diseases and neurodegenerative diseases.[5-7] Apoptosis can be triggered by the reception of a death signal or by the removal of an anti-apoptotic signal resulting in a cascade of distinct morphological changes. The "apoptotic" cell is isolated from surrounding tissue (cell rounding), followed by chromatin condensation, organelle compaction, membrane blebbing and the formation of intact apoptotic bodies.[8-10] Finally, the externalization of phosphatidyl serine to the outer membrane surface signals to the immune system that the cell is destined for death by phagocytosis.[11,12]

*Correspondence to: Justine R. Stehn; Email: j.stehn@unsw.edu.au
Submitted: 05/14/12; Accepted: 06/01/12
http://dx.doi.org/10.4161/bioa.20975

Successful apoptosis is coordinated by the family of cysteine proteases termed caspases. Caspases cleave numerous cellular substrates by specifically targeting aspartate residues.[13] Caspases are synthesized as inactive zymogens or pro-caspases that are activated in response to specific apoptotic stimuli.[14,15] Activation of the initiator caspases-8 and -9 occurs via the extrinsic and intrinsic apoptosis pathways respectively.[16] The extrinsic pathway is triggered by the ligation of extracellular "death" ligands from the tumor necrosis factor (TNF) family such as CD95/FasL and TNFα with their cognate membrane receptor.[17] Ligand binding to the cell surface triggers the intra-cellular association of Fas associated death domain (FADD) with pro-caspase-8 forming the death-inducing signaling complex (DISC).[18,19] The accumulation of pro-caspase-8 molecules results in their dimerization and auto-processing to produce active caspase-8.[16,20] Caspase-8 can then activate the executioner caspases-3, -6 and -7 which are responsible for widespread proteolytic activity leading to the removal of the apoptotic cell by the immune system.[21] (**Fig. 1**)

The intrinsic apoptosis pathway is chiefly mediated by the Bcl-2 family of pro- and anti-apoptotic proteins (**Fig. 1**). The Bcl-2 super family act as sentinels of cell well-being that detect stress signals such as DNA damage, cytokine/growth factor withdrawal and anoikis (detachment induced cell death).[22] They also ensure the completion of apoptosis by irreversible mitochondrial membrane damage.[23] All members of the mammalian Bcl-2 super family contain conserved BH domains and their classification into three functionally distinct groups is governed by the number of BH domains present.[24] The Bcl-2-like anti-apoptotic proteins (Bcl-2, Bcl-xL, Bcl-w, Mcl-1 and A1) contain BH 3 and 4 domains and protect cells from apoptosis by guarding the outer mitochondrial membrane (OMM).[25,26] The BH3-only proteins (Bim, Bad, Bid/tBid, Bmf, Bik, Hrk and Noxa Puma) which contain a single BH domain, detect cell stress and when activated engage with specific pro-survival Bcl-2 partners neutralizing their pro-survival activity.[27,28] **Table 1** outlines the specific Bcl-2:BH3 interacting partnerships currently established in the apoptosis field. Lastly, the BH1–3 group (Bax, Bak and Bok) regulate mitochondrial membrane permeability and have a specialized

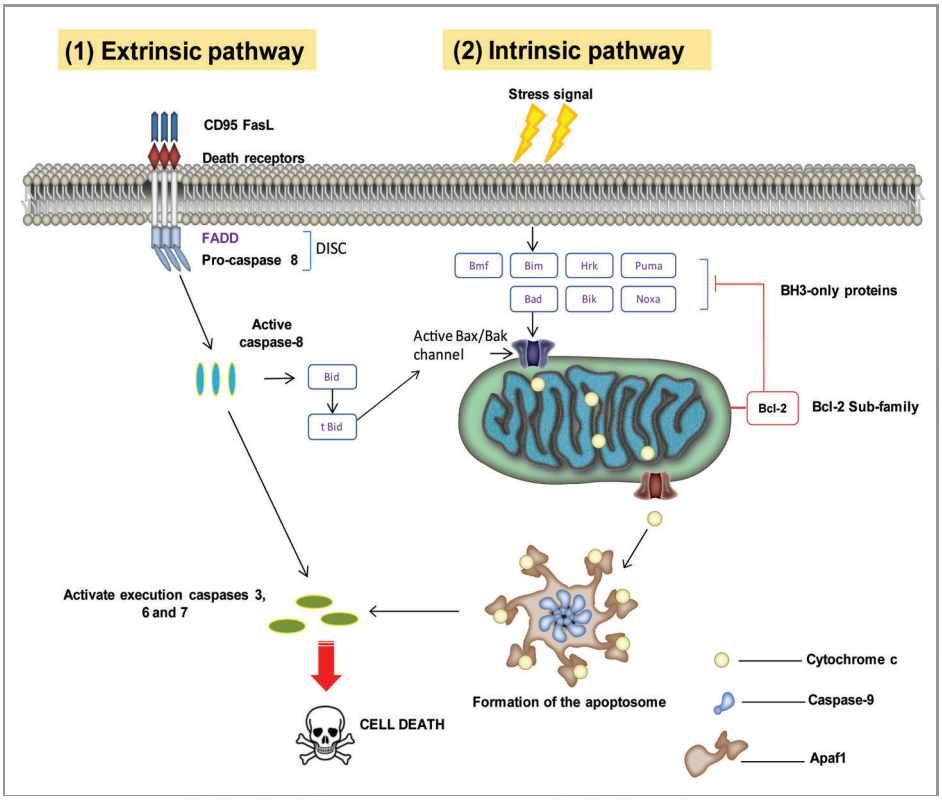

Figure 1. Schematic of the extrinsic and intrinsic apoptosis pathways. (1) The extrinsic pathway is mediated by the ligation of TNF/CD95/Fas ligands to the membrane. This triggers the formation of the death-inducing signaling complex (DISC) composed of FADD and pro-caspase 8. Caspase 8 activation occurs due to the induced proximity of pro-enzyme molecules. Caspase-8 also activates the pro-apoptotic protein Bid which feeds into the intrinsic pathway. (2) The intrinsic pathway is mediated primarily by the Bcl-2 super family. BH3 pro-apoptotic proteins inactivate Bcl-2 pro-survival partners releasing Bax and Bak. Homo-oligomerization of Bax and Bak at the OMM results in the release of cytochrome c and the downstream activation of caspase-9 via a conformational change. Both pathways converge to activate the executioner caspases leading to cell death.

capacity to homo-oligomerize upon activation.[29,30] In healthy cells Bak is located at the surface of the OMM in complex with Mcl-1 and Bax resides within the cytosol.[31] Inactivation of Bcl-2 activity at the OMM via BH3 ligation releases Bak and activates Bax translocation to the mitochondria.[32] Bax and Bak oligomerization at the OMM induces the loss of the mitochondrial membrane potential (mt$\Delta\Psi$) leading to the formation of the mitochondrial permeability transition pore (mPTP).[29,33] Formation of the mPTP induces the release of apoptogenic factors such as cytochrome c, smac/Diablo and apoptosis inducing factor, AIF-1.[34,35] Cytochrome c release is particularly important as it activates the conformational change in Apaf-1 (apoptotic protease activating factor) required for the activation of the second initiator caspase-9.[36,37] Caspase- 9 can also activate caspases-3, -6 and -7 to further provoke the apoptosis response. The actin cytoskeleton has been implicated in regulating apoptosis at multiple stages both upstream and downstream of caspase activation. Knowledge of the nature of actin filament dynamics reveals how actin can both initiate and mediate an apoptotic signal.

Actin Filament Dynamics

The actin cytoskeleton is a structural network of proteins that are essential for multiple biological functions including cell contraction, cell motility, vesicle trafficking, intracellular organization, cytokinesis, endocytosis and apoptosis.[38-41] Actin, the major component of the cytoskeleton, is a 42 kDa globular protein (G-actin) that reversibly polymerizes to form filaments (F-actin). In muscle cells actin is a core component of the sarcomere and interacts with myosin filaments to enable force generation required during muscle contraction.[42] In non-muscle cells, actin isoforms (β and γ) perform a diverse range of functions that enable cell survival and adaptation to a changing environment.[43-46] For a comprehensive review of actin structure and function see reference 47

To cope with the dynamic cellular environment F-actin assembly is in a constant state of flux with G-actin association occurring at the barbed end (+) and dissociation at the pointed end (-).[48,49] Actin filament dynamics are regulated by the action of a large group of proteins termed the actin binding proteins (ABPs). ABPs undertake a range of functions including actin severing, depolymerizing, capping, stabilizing and de novo actin polymerization which enables the actin cytoskeleton to constantly adapt to a changing environment.[39] The Rho GTPases are an important signaling protein family that regulate ABP function to achieve the formation of higher order structures such as stress fibers (actin/myosin bundles), lammelipodia (membrane ruffles at the leading edge) and filopodia (membrane protrusions).[50]

Table 1. Detailed description of the functions of Bcl-2 pro-survival and pro-apoptotic proteins

Bcl-2 pro-survival group		Function		References
Bcl-2, Bcl-w		Inhibits the activity of BH3-only proteins		27
Bcl-xL, Mcl-1, A1		Inhibits the activity of Bak and BH3-only proteins		30
Pro-apoptotic proteins	**Activated by...**	**Sub-cellular changes upon activation**	**Function**	**References**
Bim	Cytokine withdrawal	Release from the microtubule-associated dynein motor complex and localization to the mitochondria	Known as a potent killer since it binds to all Bcl-2 pro-survival proteins allowing Bax and Bak activation	27,85
Bad	Growth factor withdrawal	De-phosphorylated and released from the Scaffold protein 14–3-3	Displaces Bcl-xL from Bax and Bak allowing homo-oligomerization	28,30
Bmf	Anoikis	Release from myosin V actin motor complex and localization to the mitochondria	Displaces Bcl-2 allowing Bak homo-oligomerization	86
Bid	Caspase-8	Cleavage to form 15kDa tBid	Binds directly to Bax and Bak stimulating pore formation and the release of cytochrome c	90
Noxa	DNA damage	p53 expression induces its translocation to the mitochondria	Displaces Mcl-1 from Bak allowing the activation of homo-oligomerization	28,30
Puma	DNA damage	p53 expression induces its translocation to the mitochondria	Displaces Bcl-xL from Bax allowing the activation of homo-oligomerization	28,32
Hrk/DP5	Constitutively active	Upregulation of expression in response to death stimuli	Binds to Bcl-xL and Bcl-2 pro-survival proteins allowing the activation of Bax	177
Bax	Multiple BH3 only proteins (see above)	Translocates to the outer mitochondrial membrane and undergoes homo-oligomerization to from a pore due to conformation change	Pore formation enables the release of cytochrome c from the inter-membrane space of the mitochondria	28
Bak	Multiple BH3 only proteins (see above)	Conformational change in Bak enables homo-oligomerization of Bak molecules forming a pore	Pore formation enables the release of cytochrome c from the inter-membrane space of the mitochondria	28

Biochemical reactions such as phosphorylation and calcium ion (Ca^{2+}) binding are also essential to the regulation of ABP function as most ABPs exist in an active and inactive form.[51] Furthermore phosphoinositides such as PtdIns(4,5)P_2 play a pivotal role in regulating actin functions at the plasma membrane by accumulating within lipid rafts and facilitating F-actin polymerization.[52] It is clear that actin filament dynamics are tightly regulated at numerous stages which is warranted considering the vast array of functions mediated by the actin cytoskeleton. Studying the role of actin and ABPs in apoptosis demonstrates the importance of regulated actin filament organization.

Actin Filament Dynamics and Apoptosis

The actin cytoskeleton has been demonstrated as essential during multiple hallmarks of apoptosis with dramatic changes in actin filament organization accompanying different stages of apoptosis.[10,53] Cell rounding, which involves the loss of focal contacts with the extra-cellular environment, requires the formation of a contractile cortex of myosin II decorated actin filaments.[54] Retraction of the actin-myosin II cortex significantly alters membrane dynamics resulting in the formation of membrane blebs.[54,55] Actin-dependent membrane blebbing is reliant upon Rho GTPase signaling[56] with Rho inhibition preventing bleb formation in PC12 cells.[57] However, in Jurkat cells, caspases cleave and activate the Rho effector ROCK1, which can regulate actin-mediated membrane blebbing in a Rho-independent manner.[58] At the final stages of apoptosis the actin cytoskeleton is degraded and phagocytosis of the apoptotic bodies ensues.[55] In vitro microfilament disruption assays utilizing U-937 and HL-60 cells highlighted the importance of actin filament dynamics at the final stages of apoptosis with actin targeting drugs inhibiting apoptotic body formation.[59] The important role of actin in the morphological hallmarks of apoptosis is coupled with mounting evidence demonstrating actin as a mediator and initiator of apoptosis signaling.

Actin as a Mediator of Apoptosis

Actin has been demonstrated as a substrate for cleavage by caspases in mammalian cells, resulting in the formation of actin fragments that are 31 kDa (Fractin) and 14 kDa (tActin).[60,61] Transient transfection of 293 T cells with the expression vector of tActin, but not Fractin, resulted in the appearance of morphological hallmarks of apoptosis such as cell rounding and chromatin condensation.[61] Furthermore, ectopic expression of tActin induced these morphological changes without activating caspases, indicating that actin fragment-mediated cellular shrinkage is an event downstream of the caspase signaling cascade.[61]

Manipulating the actin cytoskeleton via drug intervention has further revealed that changes in actin dynamics can also mediate apoptosis. Jasplakinolide is a potent F-actin stabilizing drug, that is derived from the marine sponge *Jaspis johnstoni*.[62] Treatment of Jurkat cells with jasplakinolide resulted in the appearance of

distinct morphological and biochemical hallmarks of apoptosis including DNA fragmentation, chromatin condensation and caspase activation, suggesting that actin stabilization elicits an apoptotic response.[63] Treatment of leukemic HL-60 cells with jasplakinolide similarly induced distinct nuclear and membrane changes that resembled apoptosis.[64] Jasplakinolide also increased the activity of DNase I, which is responsible for the degradation of nuclear DNA strands during apoptosis.[65] DNase I binds with high affinity to G-actin monomers simultaneously promoting actin depolymerization and DNase I inhibition.[66] The actin polymerizing activity of jasplakinolide may be triggering the release of DNase 1 from G-actin once it has been added to the barbed end resulting in activation of DNase I activity. This hypothesis remains to be investigated. Actin depolymerization has been shown to induce an apoptotic response in numerous cell types. Cytochalasin D belongs to a family of fungal metabolites that binds to the barbed end of F-actin preventing further polymerization.[67] Cytochalasin D was shown to induce caspase-3 mediated apoptosis in T lymphocytes[68] and enhanced the commitment of Jurkat T cells to apoptosis after cytokine withdrawal.[69] Given that cytochalasin D binds to the barbed end of the actin filament, the effects of cytochlasin D are through its disruption of actin filament dynamics and not through a shift in G:F-actin levels. The importance of actin dynamics thus explains why both actin polymerizing and depolymerizing can affect cell survival. Given the profound response of the actin cytoskeleton to changes in dynamics, actin may also play a role in initiating an apoptotic signaling cascade.

Actin as an Initiator of Apoptosis Pathways

A number of studies have demonstrated a role for the actin cytoskeleton in triggering apoptosis upstream of caspases both in the extrinsic and intrinsic pathways. CD95 or FasL is a major ligand of the extrinsic apoptosis pathway. In $CD4^+$ T lymphocytes activation of CD95-mediated apoptosis resulted in the polarization of CD95 at the cell surface. CD95-mediated apoptosis was found to be dependent upon the interaction of actin with CD95 via the actin-associated protein ezrin.[70] Ezrin interacts with membrane associated proteins via the FERM domain located within the N-terminus and is tethered to the actin cytoskeleton at the C-terminus.[71,72] Thus ezrin, which is anchored to CD95 at the cell membrane of T lymphocytes is thought to transduce an extracellular signal to the actin cytoskeleton initiating an apoptosis cascade (**Fig. 2**).[73,74] A reduction in ezrin expression has been correlated with the stimulation of CD95-mediated apoptosis in H9 stem cells and normal T lymphocytes which contradicts with aforementioned studies.[75] The role of ezrin in CD95 mediated apoptosis thus remains inconclusive. What is known is that ezrin activity is phospho-regulated and its phophorylated status may govern its' function in apoptosis.[75]

Actin has also been implicated in the initiation of intrinsic mitochondrial-dependent apoptosis spanning yeast, mammals and plants.[40,76,77] Studying the role of actin in mitochondrial-dependent apoptosis in yeast has been very advantageous due to the presence of a single actin isoform, *ACT1*.[78] Yeast strains bearing point mutations in *ACT1* clearly demonstrated altered susceptibility to mitochondrial damage. Expression of a mutant allele with decreased actin dynamics (*act1-159*) resulted in F-actin aggregation and increased susceptibility to apoptosis due to the accumulation of reactive oxygen species (ROS) and mitochondrial membrane depolarization. Expression of a yeast mutant with increased actin filament dynamics (*act1-157*) did not accumulate ROS and survived in long-term culture.[76,79] This suggests that a dynamic actin cytoskeleton is essential to the maintenance of the $mt\Delta\Psi$ in yeast and that modulation of actin dynamics contributes to oxidative stress. The capacity for actin to regulate the opening of voltage dependent anion channels (VDACs) has been postulated as a mechanism by which actin regulates mitochondrial integrity. In *Neurospora crassa* actin stabilization via phalloidin treatment led to the prolonged opening of VDAC pores resulting in the accumulation of ROS and a loss of $mt\Delta\Psi$.[80]

In mammalian HeLa cells, cytochalasin D treatment induced caspase-mediated cytochrome c release suggesting that actin has a role in regulating mitochondrial membrane permeability in both yeast[79] and mammalian cells.[81] Overexpression of the pro-survival protein Bcl-x_L inhibited apoptosis in jasplakinolide treated CTLL-20 cells[82] and partially attenuated apoptosis in Jurkat cells treated with cytochalasin D[69] implicating the Bcl-2 family in the apoptotic action of cytochalasin D and jasplakinolide. The latrunculins are a group of actin depolymerizing agents that are derived from the Red Sea sponge *Latrunculia magnificans* and sequester G-actin monomers and prevent polymerization via a mechanism that differs from cytochalasin D.[83] Latrunculin A was shown to induce caspase-mediated apoptosis in MCF10A epithelial cells[84] whereas latrunculin B induced apoptosis in re-perfused rat kidney tissue.[85] Latrunculin A treatment of MCF-7 cells was also shown to induce the translocation of Bax to the mitochondrial membrane where it was postulated to undergo oligomerization and mitochondrial membrane pore formation.[84] Therefore the mechanism of action of distinct actin targeting drugs may involve the Bcl-2 family of apoptosis proteins. These reports however do not define the specific signaling pathways linking actin filament changes to Bcl-2 activation. Puthalakath and colleagues have demonstrated a direct link between the Bcl-2 family and actin-mediated apoptosis.[86] Pro-apoptotic Bmf was found to be sequestered with actin-associated myosin V motors and upon cell detachment or cytochalasin D treatment is released from the cytoskeleton resulting in a mitochondrial-dependent apoptotic cascade (**Fig. 2**).[86] The translocation of G-actin to the nucleus has been demonstrated in Latrunculin B treated mast cells[87] and hepatocytes treated with protein synthesis inhibitors.[88] Given the apoptotic effects of Latrunculin B in rat kidney tissue[85] nuclear actin translocation could induce an apoptotic response. However a significant increase in apoptotic cells could not be detected in both studies suggesting that nuclear actin translocation was unable to induce apoptosis. As noted by Utsumi et al.,[89] a role for tActin upstream of caspases has also been identified whereby tActin was subjected to post-translational N-myristoylation, targeting it to the mitochondria during apoptosis (**Fig. 2**).[89] A possible explanation for the presence of actin in both the initial

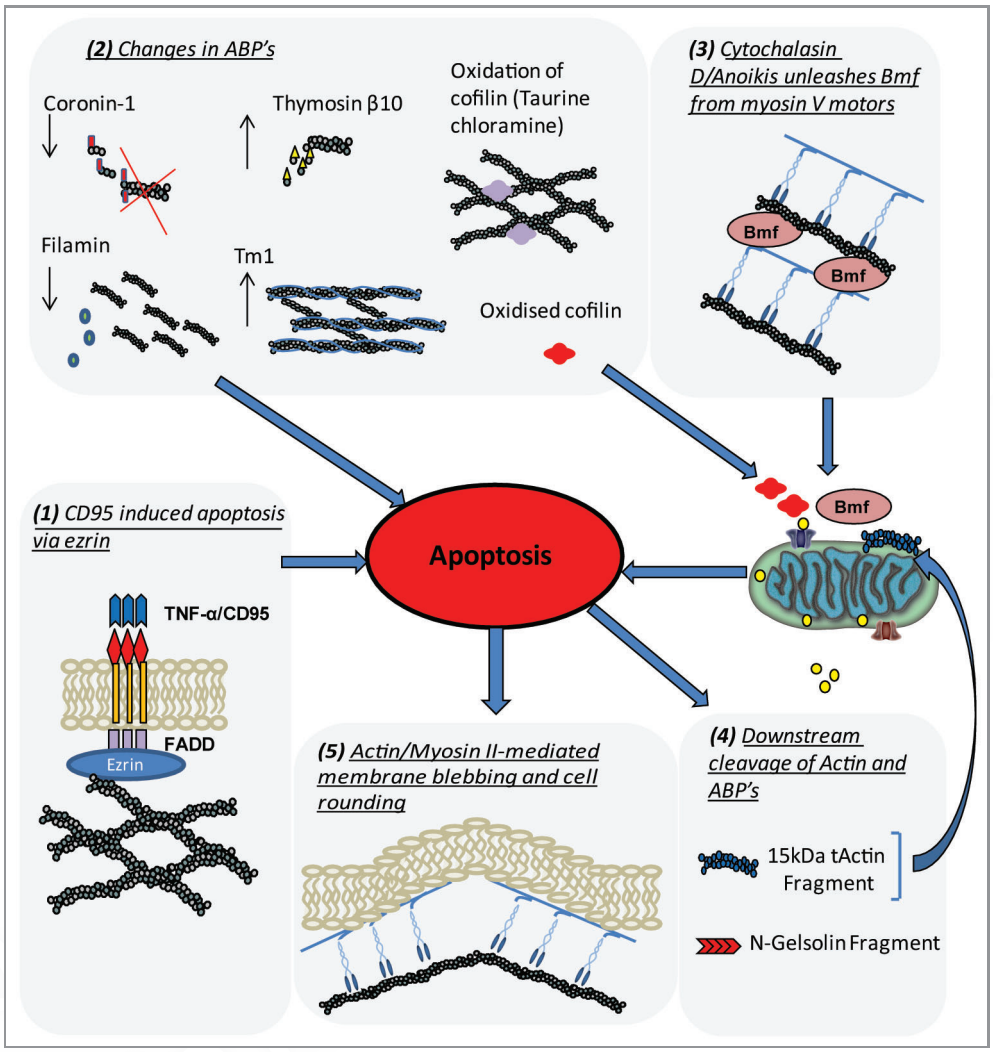

Figure 2. Reception of an apoptotic stimulus induces significant changes in the actin cytoskeleton resulting in the following biochemical and morphological events. (1) The actin-membrane linker protein ezrin has been implicated as a mediator of CD95-mediated apoptosis, however other studies suggest ezrin is a mediator of tumor invasion and metastasis. Thus ezrin may have multiple roles in cellular functioning including apoptosis. (2) Changes in the expression of ABPs (upregulation of thmyosin β10 and Tm1 plus downregulation of coronin 1 and filamin) induce an apoptotic response. Translocation of oxidized cofilin to the mitochondria induces the release of cytochrome c due to mitochondrial membrane permeabilization. (3) Disruption of the microfilament system by cytochalasin D or cell detachment induces the translocation of pro-apoptotic Bmf from the actin cytoskeleton to the mitochondria during apoptosis. (4) Caspase cleavage of actin (tActin) and gelsolin (N-Gelsolin) triggers their N-myristoylation with tActin being translocated to the mitochondria. (5) Actin-myosin II contraction mediates morphological hallmarks of apoptosis including membrane blebbing and cell rounding.

and final stages of apoptosis may involve a positive feedback loop. Initial disruption of the actin cytoskeleton may lead to downstream caspase activation which causes permanent actin filament fragmentation (t-Actin). N-myristolyated tActin may then be responsible for the amplification of apoptosis by inducing irreversible mitochondrial membrane damage. Slee and colleagues have demonstrated that feedback amplification can occur after apoptosis induction via the downstream cleavage of the pro-apoptotic protein Bid.[90] Thus downstream events may play an important role in sustaining an apoptotic cascade and tActin may be a crucial player in this process. Actin filament dynamics are greatly dependent upon ABP regulation suggesting that ABPs may also play a role in actin-mediated apoptosis.

The Role of Actin Binding Proteins in Apoptosis

Several ABPs have been postulated as biomarkers of apoptosis due to alterations in their expression leading to cell death signaling pathways. The ABPs that have been studied in relation to apoptosis are ADF/Cofilin (actin dynamizing), thymosin β (actin sequestering), coronin-1 (actin branching), filamin (actin branching), gelsolin (actin severing and capping), tropomyosin (actin stabilizing) and myosin II (actin filament contraction or bundling).

The ADF/cofilin family regulate actin filament turnover by severing and depolymerizing existing actin filaments thus may increase the G:F-actin ratio.[91] LIM and testicular kinases (LIMK

and TESK I and II) phosphorylate ADF/cofilin at the Ser 3 residue inhibiting G- and F-actin binding[92] whereas slingshot homolog (SSH) and chronophin (CIN) de-phosporylate ADF/cofilin activating cofilin.[93] In relation to apoptosis, cofilin has been demonstrated to be translocated to the mitochondrial membrane in response to the kinase inhibitor staurosporin resulting in the release of cytochrome c and morphological hallmarks of apoptosis. Expression of a phophorylated (inactive) cofilin mutant abolished this mitochondrial targeting of cofilin emphasizing the requirement for active de-phosphorylated cofilin in apoptosis.[94] Oxidation of cofilin by taurine chloramine similarly induced mitochondrial translocation of cofilin resulting in the opening of the mPTP and cytochrome c release.[95] Mutations that removed any of the cysteine residues within cofilin inhibited mitochondrial targeting of cofilin and oxidant-induced apoptosis.[95] Since oxidation of cysteine residues in cofilin resulted in the formation of intermolecular disulphide bonds[96] intermolecular cysteine oxidation may be essential for the mitochondrial targeting of cofilin. Recent studies have identified novel cofilin residues that drive F-actin stabilization induced by nutritional depletion resulting in the accumulation of ROS, mitochondrial fragmentation and Ras hyperactivation.[97] This supports the hypothesis that the actin cytoskeleton is an important biosensor of environmental stresses such as oxidative stress. Second, conserved positively charged residues on cofilin that are not actin binding were shown to be essential for respiratory function further highlighting the potential role of cofilin in sensing oxidative stress.[97] The formation of actin-cofilin rods is a second apoptosis-related role for cofilin whereby ATP depletion resulted in the formation of short actin/cofilin rods.[98,99] Actin-cofilin rods were able to prevent apoptosis by slowing mt$\Delta\Psi$ depletion in hippocampal neurons over a short period of time.[100] However over an extended period of time, this protective mechanism was abolished resulting in rapid loss of mt$\Delta\Psi$ and subsequent apoptosis. Thus persistent actin-cofilin rods contribute to the loss of synapse activity in the neurons of patients suffering from neurodegenerative conditions. The short-term pro-survival role for cofilin in neurons specifically may be a biological conditioning mechanism to reduce the mitochondrial damage experienced by neurons affected by oxidation, micro-ischemia or glutamate excitotoxicity.[98] Whether this short-term pro-survival role for cofilin exists in other cell types remains elusive. Cofilin has also been demonstrated to mediate the apoptosis of hippocampal neurons due to its activation by the scaffold protein RanBP9. Elevated levels of RanBP9 have been implicated in the production of amyloid β peptide which is known to cause neurodegeneration with cofilin expression being essential to RanBP9-mediated apoptosis.[101]

Thymosin β prevents polymerization by attaching to and sequestering G-actin.[102] Elevated expression of thymosin β10 in ovarian tumor cells has been correlated with an increase in sensitivity to apoptosis. The presence of a second ABP, E-tropomodulin, inhibited apoptosis by competing with thymosin β10 for actin binding highlighting the inter-related dependency of ABPs in regulating actin-mediated apoptosis.[103] Thymosin β10 was also shown to accelerate the apoptosis of fibroblasts by disrupting stress fiber formation which further supports the pro-apoptotic role of thymosin β10.[104]

Filamin promotes orthogonal actin branching which strengthens the cell membrane during cellular movement.[105] Filamin cleavage by the T lymphocyte enzyme granzyme B induced an apoptotic response in Jurkat cells that was caspase-independent.[106] In a separate study, filamin-mediated apoptosis of platelet cells was shown to be dependent upon caspase-3 activation in vivo.[107] This contradictory result in regards to caspase dependency may reflect alternative effects based upon the type of apoptotic stress induced. The former study specifically looked at the physiological process of granzyme B activity whereas in the latter study exogenously expressed caspase-3 was utilized (non-physiological process). The utilization of physiologically relevant conditions is therefore important when studying apoptosis pathways.

Coronin-1 regulates the function of the actin nucleating and branching ABP Arp2/3 and is involved with lammelipodial formation required for cell motility.[108] Knockout mouse studies demonstrated that coronin-$1^{-/-}$ cells show an impairment of T lymphocyte migration to the thymus due to an elevated level of apoptosis detected by annexin V staining.[109] Elevated cytochrome c levels were also detected in coronin-$1^{-/-}$ T cells, suggesting that coronin-1 may regulate the survival of migrating cells such as T lymphocytes. A proteomics approach employed by Moriceau and colleagues further identified the presence of a cleavage product of coronin-1 after apoptosis induction suggesting that coronin-1 cleavage may be a downstream response to apoptosis signaling similar to actin.[110] Expression of full length coronin-1 inhibited mitochondrial-mediated apoptosis of mature neutrophils further supporting the pro-survival role of coronin-1 in hematopoietic cells.[110]

Myosin II is an ATP-dependent non-muscle motor that interacts with actin filaments producing a contractile force that is essential during cell rounding and migration.[111] Maintenance of myosin II tension is also crucial to the formation of the contractile ring during cytokinesis.[112] Myosin II activity is regulated by the phosphorylation proteins such as myosin light chain kinase (MLCK) and Rho kinase.[111] Studies have demonstrated a non-redundant role for myosin II phosphorylation in regulating apoptosis in endothelial and epithelial cells. TNFα, a regulator of extrinsic apoptosis, is also responsible for vascular endothelial barrier dysfunction. TNFα triggered the apoptosis of endothelial cells accompanied by the phosphorylation of myosin II leading to an increase in stress fiber formation and the appearance of para-cellular gaps indicative of endothelial barrier dysfunction.[113] Inhibition of myosin II phosphorylation reduced TNFα-induced stress fiber formation and attenuated caspase-8 levels in vitro.[113] As noted by Jin et al.[114] myosin II may regulate TNFα mediated endothelial apoptosis by translocating TNF-receptor to the membrane surface. Further analysis of 3D microvessels revealed that vascular endothelial permeability occurred independently of Rho kinase activity implicating other regulatory elements (e.g., phosphoinositides and Ca^{2+}) in actin/myosin II-dependent vascular permeability in vivo.[115] Myosin II phosphorylation is also essential for the extrusion of apoptotic epithelial

cells from the epithelial barrier during embryonic tissue development. UV irradiation of monolayer MDCK epithelial cells induced the formation of an actin-myosin ring around the edge of apoptotic cells indicative of cell rounding. As this ring of actin and myosin contracted, neighboring live cells moved into the space surrounding the dying cell thus closing the epithelial gap and extruding the apoptotic cell simultaneously. Rho kinase inhibition prevented the extrusion of the apoptotic cell highlighting the importance of myosin II phosphorylation to epithelial cell apoptosis and implicating cross-talk signaling between the actin cytoskeleton of the dying cell and the live neighboring cells.[116]

Gelsolin is a potent actin severing protein that caps the barbed-end of F-actin in the presence of Ca^{2+} preventing further barbed-end polymerization.[117] Gelsolin has been implicated in apoptosis with caspase-3 activation producing an N-terminal gelsolin fragment (N-Gelsolin) with un-regulated actin filament severing capacity.[118] As noted by Chhabra et al., N-Gelsolin specifically induced apoptosis by severing the G-actin:DNase I complex resulting in the nuclear localization and activation of DNase I.[119] The mechanism by which N-Gelsolin releases G-actin bound DNase I remains unknown. N-Gelsolin has also been demonstrated as a pro-survival protein upstream of the mitochondria with its N-myristoylation preventing etoposide induced apoptosis.[120] Elevated expression of gelsolin protected Jurkat cells from apoptosis induced by a variety of mitochondrial targeting agents[121,122] and also prevented apoptosis in neuronal cells with enhanced actin stabilization abolishing this pro-survival effect.[123] Silencing of gelsolin expression in Ras-mutated HCT116 colon cancer cells induced butyric-mediated apoptosis, via caspase activation further supporting the pro-survival role of gelsolin.[124] Resistance to apoptosis was found to be driven by the capacity of gelsolin to inhibit the opening of VDACs, thus preventing mt$\Delta\Psi$ loss and downstream cytochrome c release.[125] It is therefore hypothesized that gelsolin may protect against apoptosis in certain cell types (i.e., neurons, cancer cells), however this hypothesis has not been further explored. What remains certain is that caspase-3 activation releases a pro-apoptotic fragment of gelsolin which completely abolishes its pro-survival role at the mitochondrion and results in the release of DNase-1 from G-actin, but not in the presence of cofilin.[119] Given that gelsolin regulation may involve other ABPs such as tropomyosin, the role of gelsolin in apoptosis may also depend on other proteins within the actin cytoskeleton.[126]

Tropomyosin is a dimerized helical polymer that winds around actin filaments providing structural stability and diverse functioning of actin filaments.[127,128] Tropomyosin isoforms can be classified as high molecular weight (HMW) or low molecular weight (LMW) depending on the gene promoter utilized.[129] Muscle tropomyosins specifically regulate myofibril contraction whereas non-muscle or cytoskeletal tropomyosins are known to regulate numerous cytoskeletal functions due to their spatial and temporal regulation.[127,130] Cytoskeletal tropomyosins have been demonstrated to modulate the activity of other ABPs that are previously mentioned to be involved in apoptosis. Tm5NM1 expression in neuroepithelial cells was found to induce the recruitment of myosin IIA to stress fibers[131] and simultaneously displacing ADF interaction with the actin filament.[132] Conversely elevated levels of the HMW isoforms TmBr3 and Tm3 in neuroepithelial cells promoted ADF interaction with actin filaments resulting in the formation of filopodia which promote cell migration.[131,132] This suggests that certain tropomyosin containing filaments are marked by specific ABP interactions which may be important in apoptosis. Anoikis is a specialized form of apoptosis that is activated when cells dependent on anchorage for survival (e.g., epithelial and endothelial cells) are placed in an anchorage-independent environment.[133] Anoikis represents an important homeostatic function that prevents the migration of detached cells to a foreign location. Studies in mammary epithelial carcinomas have demonstrated that a significant downregulation in the HMW isoform Tm1 correlated with an increased resistance to anoikis perpetuating the survival of mammary carcinoma tissue in vitro.[134] Restoring Tm1 expression in cultured mammary carcinoma cell lines (MCF-7 and MBA-MB231) led to the generation of distinct actin stress fibers and re-sensitized cells to anoikis.[135] The reversion of Tm1 expression was Rho-kinase dependent and resulted in the appearance of more distinct cadheren/catenin containing cell-cell junctions thus enabling the cell to communicate with the extracellular environment.[135,136] Tm1 can therefore act as an important sensor of the extra-cellular environment with unfavorable conditions leading to Tm1-mediated anoikis.

In summary ABPs are essential in regulating numerous key apoptotic processes such as cell rounding, membrane blebbing, caspase activation and mitochondrial membrane permeabilization. ABPs are also important in regulating specialized death pathways such as anoikis and epithelial cell extrusion. This further highlights the importance of actin filament dynamics in regulating apoptosis signaling via modulation of ABP function. As tumor cells have developed mechanisms to evade apoptosis, the transformed phenotype has been used extensively to further characterize the role of actin and ABPs in apoptosis signaling pathways.

Changes in the Actin Cytoskeleton upon Transformation

The actin cytoskeleton is dramatically re-modeled upon cellular transformation permitting metastatic properties such as anchorage-independent growth and enhanced cell migration.[137] A stabilized actin cytoskeleton has been demonstrated as an activator of Ras signaling resulting in apoptosis driven by the production of ROS and loss of the mt$\Delta\Psi$.[138] This discovery highlights the actin cytoskeleton as a trigger of Ras signaling and given the importance of Ras in tumorogenesis, actin may mediate tumor-associated processes such as cell migration via Ras signaling.[138] ABPs such as tropomyosin, gelsolin and cofilin show varied expression profiles in both malignant cell types and in virally transformed cells implicating these proteins as potential biomarkers of malignancy. The exact phenotypic changes in ABPs associated with transformation remains complex. The expression of gelsolin in breast, urothelial and oral carcinomas has been described as biphasic with an early downregulation in gelsolin

followed by a substantial elevation in gelsolin with prolonged metastasis.[126,139,140] Cofilin overexpression has been confirmed in numerous tumor cell types with elevated cofilin levels potentially correlating with tumor cell migration.[141,142] However a down-regulation in cofilin was found in the highly metastatic hepatocellular carcinoma[143] contradicting previous studies. A more defined role for cofilin in metastasis may be achieved by examining the entire output from the cofilin pathway which includes inhibitors such as LIMK I and II and stimulators such SSH and CIN phophatase.[144,145] Several studies have demonstrated that increased expression of LIMK I contributed to the invasive capacity of prostate and breast cancer cells highlighting LIMK I as a potential malignant biomarker.[146,147] Varying the local concentration of cofilin can significantly alters its function with high levels of active cofilin enhancing F-actin stability by stimulating actin nucleation rather than enhancing turnover and lower levels of cofilin favoring actin severing.[148] This novel paradigm may also influence the role of the cofilin system in tumorogenesis. Downregulation of multiple HMW tropomyosins (Tm1, 2 and 3) has been detected in virally transformed chicken and rat fibroblasts,[149,150] cultured human tumor cells[136,151] and in tumor derived patient samples.[134] Restoration of Tm2 and Tm3 increased the appearance of distinct microfilament bundles and adhesion proteins with Tm2 expression also restoring anchorage-dependent growth.[152] This correlated with reports highlighting a role for Tm1 in anoikis.[134,136] Further studies have demonstrated an increased reliance on LMW Tm5NM1 in transformed cells[153] with reduced TM5NM1 expression leading to a reduction in melanoma cell motility.[154] Together these studies suggest that certain ABPs (such as gelsolin, cofilin and Tm5NM1) have an important role in permitting tumorogenic hallmarks such as enhanced motility and anchorage independent growth. It is now well established that the evasion of apoptosis contributes to the prolonged survival and metastasis of tumor cells.[155] Given that the actin cytoskeleton changes dramatically upon cellular transformation investigating apoptosis pathways in tumor cells may further elucidate a direct link between the actin cytoskeleton, apoptosis signaling and tumor cell survival.

Targeting the Actin Cytoskeleton for Chemotherapy

De-regulation of apoptosis by oncogenic transformation is partly responsible for the ability of tumor cells to evade normal apoptotic signaling pathways.[155] Overexpression of Bcl-2 and other pro-survival proteins has been detected in multiple tumor cell types (androgen independent prostate cancer cells, B pancreatic cancer cells, B cell lymphoma) and correlated with the prolonged survival of B lymphoid tumors.[156] Furthermore, synergistic activation of the proto-oncogene cMyc and Bcl-2 accelerated lymphoma tumorogenesis.[157] In contrast the loss of pro-apoptotic factors such as Bim and Puma has been correlated with the survival of tumor cells, with re-expression of Bim suppressing the activity of cMyc in leukemic cells[158] and Puma re-expression increasing the sensitivity of melanoma cells to apoptosis.[159] The generation of specific BH3-only mimetic compounds thus represents a potential anti-tumor therapy that could restore sensitivity of tumor cells to apoptosis. This type of therapy has been proven successful in vivo with the high affinity BH3 mimicking compound ABT-737 triggering Bax/Bak dependent apoptosis in a mouse lymphoma model.[160] Indeed many cytotoxic therapies including DNA damaging agents exert their anti-tumor effects by inducing either extrinsic CD95 or Bcl-2 mediated apoptosis.[161] However, in a panel of apoptosis resistant tumor cells, the activation of CD95 apoptosis permitted tumorogenic hallmarks such as cell invasion and migration through cellular barriers.[162] This highlighted a novel anti-apoptotic role for CD95 in metastasis.[163] Furthermore, Rac1 stimulated CD95 activity in developing neurons suggesting that Rho GTPases may regulate the invasive potential of CD95.[164] Rac activation was found to be dependent upon association with ezrin suggesting that cellular transformation may convey a tumorogenic role for ezrin in actin-mediated tumor cell invasion.[165] Thus ezrin may have a more global role in transducing extracellular signals to the actin cytoskeleton and that transformation alters this signaling pathway to promote tumor cell survival. Given the mounting evidence implicating the actin cytoskeleton in both apoptosis and tumorogenesis, targeting actin filaments represents an attractive anti-cancer therapy. Studies have demonstrated the anti-tumor effects of numerous actin targeting drugs both in vitro and in vivo. The cytochalasins were shown to inhibit cytokine stimulated melanoma cell motility in vitro[166] and conferred an anti-proliferative effect in an in vivo mouse melanoma model.[167] Jasplakinolide (actin stabilizing drug) has also been shown to possess a potent anti-proliferative capacity in a panel of prostate cancer cells accompanied by distinct apoptotic changes such as multi-nucleated cells and actin filament disruption.[168] This study utilized phalloidin as its marker of actin filaments, however phalloidin F-actin binding is out-competed by the binding of Jasplaknolide.[62] Therefore the observed actin filament disruption may be incorrect if filaments are already saturated with Jasplakinloide leading to the inhibition of phalloidin binding. More recently, latrunculin A has been demonstrated as an effective anti-tumor agent in both in vitro and in vivo models of gastric cancer.[169] Due to the high sequence similarity between all actin isoforms indiscriminate targeting of the global actin filament population has hampered the success of these compounds in pre-clinical trials.[170] To circumvent this problem, an ideal approach would be to target a sub-population of actin filaments involved in distinct functions such as cytokinesis or proliferation.[171] Given the dramatic changes in ABP expression discussed previously, ABPs could be utilized as novel targets for chemotherapeutic drug design.[172] More specifically tumor cells downregulate their HMW tropomyosins and show an increased reliance upon the LMW isoforms such as Tm5NM1/2. A novel strategy would be to target actin filaments containing Tm5NM1 to improve the specificity toward tumor cells.[171] Inhibitors of LIMK I have also been postulated as a second target for actin-based chemotherapy because elevated expression of LIMK I was associated with the malignant phenotype.[173] Given the high regulatory nature of the ADF/cofilin family and the conflicting expression patterns of cofilin in tumor cells, upstream targeting of LIMK may be a more effective strategy.

In the realm of chemotherapy, multi-drug resistance (MDR) is an emerging issue and changes in the actin cytoskeleton have been identified in MDR-specific cell lines. An altered actin cytoskeleton has been detected in a specific sub-population of MDR osteosarcoma cells with increased resistance correlating to the appearance of cells with distinct actin filament bundles.[174] Furthermore, drug resistance in leukemic cells treated with the anti-microtubule drug vincristine produced fragments of actin that were identified via proteomic analysis.[175] Reduced expression of the γ-actin isoform was specifically detected in leukemic cells that were resistant to another anti-microtubule drug vinblastine. Consequently a lower level of γ-actin in vinblastine-resistant cells correlated with a worse prognosis in relapse patients diagnosed with acute lymphoblastic leukemia.[176] In relation to ABPs, mutational analysis of cofilin revealed the existence of positively charged surfaces that can regulate the activity of the drug transporter PDR1 without compromising mitochondrial function.[97] This implicates cofilin as a potential player in the acquisition of multi-drug resistance with human cofilin CFL-2 being identified as a prognostic marker for non-small cell lung cancer drug resistance. Collectively, these reports suggest that changes in the organization of the actin cytoskeleton facilitate the survival of drug resistant cells resulting in a worse prognosis for cancer patients. Thus actin filament changes in MDR-specific tumor cells may represent a point of vulnerability in the actin cytoskeleton that could be treated by drug intervention.

Summary

Given the important role of the actin cytoskeleton in cellular homeostasis, it is not surprising that actin also has an important role in apoptosis. In the yeast system, the role of actin and cofilin in sensing oxidative stress has been well established. However in mammalian cells apoptotic mechanisms are more complex and defining a global role for actin in mammalian apoptosis remains challenging. It is clear that actin initiates and mediates mammalian apoptosis via the intrinsic and extrinsic pathways and final degradation of actin filaments amplifies the apoptosis signaling cascade. Actin dynamics is the crucial determinant of whether a cell succumbs to insult or resists with ABPs such as gelsolin, cofilin and tropomyosin conveying an important regulatory function in apoptosis. Given that the actin cytoskeleton significantly changes upon cellular transformation and the correlative changes in actin filament architecture in drug resistant cells, drugs that target MDR tumor-specific actin filaments could be utilized in combination with routine therapies to enhance their effectiveness in patients.

Disclosure of Potential Conflicts of Interest

The authors of this review have no affiliation or financial involvement with any organization or entity with a financial interest in the subject material discussed in this review. No writing assistance was utilized in the production of this review.

Acknowledgments

This work was supported by grants from the NH&MRC awarded to Peter W. Gunning and Justine R. Stehn. We acknowledge our major funding body 'The Kids Cancer Project' for supporting this work. Justine Stehn is supported by a Kids Cancer Project C4 Fellowship. Melissa Desouza has been awarded an Australian Postgraduate Award. We also thank Shane Whittaker for his assistance in the preparation of this review.

References

1. Hardy K. Apoptosis in the human embryo. Rev Reprod 1999; 4:125-34; PMID:10521149; http://dx.doi.org/10.1530/ror.0.0040125
2. Henson PM, Hume DA. Apoptotic cell removal in development and tissue homeostasis. Trends Immunol 2006; 27:244-50; PMID:16584921; http://dx.doi.org/10.1016/j.it.2006.03.005
3. Vaux DL, Korsmeyer SJ. Cell death in development. Cell 1999; 96:245-54; PMID:9988219; http://dx.doi.org/10.1016/S0092-8674(00)80564-4
4. Krammer PH, Behrmann I, Daniel P, Dhein J, Debatin KM. Regulation of apoptosis in the immune system. Curr Opin Immunol 1994; 6:279-89; PMID:8011211; http://dx.doi.org/10.1016/0952-7915(94)90102-3
5. Lowe SW, Lin AW. Apoptosis in cancer. Carcinogenesis 2000; 21:485-95; PMID:10688869; http://dx.doi.org/10.1093/carcin/21.3.485
6. Rathmell JC, Thompson CB. Pathways of apoptosis in lymphocyte development, homeostasis, and disease. Cell 2002; 109(Suppl):S97-107; PMID:11983156; http://dx.doi.org/10.1016/S0092-8674(02)00704-3
7. Mattson MP. Apoptosis in neurodegenerative disorders. Nat Rev Mol Cell Biol 2000; 1:120-9; PMID:11253364; http://dx.doi.org/10.1038/35040009
8. Kerr JF, Wyllie AH, Currie AR. Apoptosis: a basic biological phenomenon with wide-ranging implications in tissue kinetics. Br J Cancer 1972; 26:239-57; PMID:4561027; http://dx.doi.org/10.1038/bjc.1972.33
9. Wyllie AH. Apoptosis: an overview. Br Med Bull 1997; 53:451-65; PMID:9374030; http://dx.doi.org/10.1093/oxfordjournals.bmb.a011623
10. Häcker G. The morphology of apoptosis. Cell Tissue Res 2000; 301:5-17; PMID:10928277; http://dx.doi.org/10.1007/s004410000193
11. Martin SJ, Reutelingsperger CP, McGahon AJ, Rader JA, van Schie RC, LaFace DM, et al. Early redistribution of plasma membrane phosphatidylserine is a general feature of apoptosis regardless of the initiating stimulus: inhibition by overexpression of Bcl-2 and Abl. J Exp Med 1995; 182:1545-56; PMID:7595224; http://dx.doi.org/10.1084/jem.182.5.1545
12. Fadok VA, Daleke DL, Henson PM, Bratton DL, Bratton DL, de Cathelineau A. Loss of phospholipid asymmetry and surface exposure of phosphatidylserine is required for phagocytosis of apoptotic cells by macrophages and fibroblasts. J Biol Chem 2001; 276:1071-7; PMID:10986279; http://dx.doi.org/10.1074/jbc.M003649200
13. Fuentes-Prior P, Salvesen GS. The protein structures that shape caspase activity, specificity, activation and inhibition. Biochem J 2004; 384:201-32; PMID:15450003; http://dx.doi.org/10.1042/BJ20041142
14. Earnshaw WC, Martins LM, Kaufmann SH. Mammalian caspases: structure, activation, substrates, and functions during apoptosis. Annu Rev Biochem 1999; 68:383-424; PMID:10872455; http://dx.doi.org/10.1146/annurev.biochem.68.1.383
15. Nicholson DW. Caspase structure, proteolytic substrates, and function during apoptotic cell death. Cell Death Differ 1999; 6:1028-42; PMID:10578171; http://dx.doi.org/10.1038/sj.cdd.4400598
16. Taylor RC, Cullen SP, Martin SJ. Apoptosis: controlled demolition at the cellular level. Nat Rev Mol Cell Biol 2008; 9:231-41; PMID:18073771; http://dx.doi.org/10.1038/nrm2312
17. Nagata S. Fas ligand-induced apoptosis. Annu Rev Genet 1999; 33:29-55; PMID:10690403; http://dx.doi.org/10.1146/annurev.genet.33.1.29
18. Muzio M, Chinnaiyan AM, Kischkel FC, O'Rourke K, Shevchenko A, Ni J, et al. FLICE, a novel FADD-homologous ICE/CED-3-like protease, is recruited to the CD95 (Fas/APO-1) death–inducing signaling complex. Cell 1996; 85:817-27; PMID:8681377; http://dx.doi.org/10.1016/S0092-8674(00)81266-0
19. Boldin MP, Goncharov TM, Goltsev YV, Wallach D. Involvement of MACH, a novel MORT1/FADD-interacting protease, in Fas/APO-1- and TNF receptor-induced cell death. Cell 1996; 85:803-15; PMID:8681376; http://dx.doi.org/10.1016/S0092-8674(00)81265-9
20. Muzio M, Stockwell BR, Stennicke HR, Salvesen GS, Dixit VM. An induced proximity model for caspase-8 activation. J Biol Chem 1998; 273:2926-30; PMID:9446604; http://dx.doi.org/10.1074/jbc.273.5.2926

21. Hengartner MO. The biochemistry of apoptosis. Nature 2000; 407:770-6; PMID:11048727; http://dx.doi.org/10.1038/35037710
22. Borner C. The Bcl-2 protein family: sensors and checkpoints for life-or-death decisions. Mol Immunol 2003; 39:615-47; PMID:12493639; http://dx.doi.org/10.1016/S0161-5890(02)00252-3
23. Newmeyer DD, Ferguson-Miller S. Mitochondria: releasing power for life and unleashing the machineries of death. Cell 2003; 112:481-90; PMID:12600312; http://dx.doi.org/10.1016/S0092-8674(03)00116-8
24. Huang DC, Strasser A. BH3-Only proteins-essential initiators of apoptotic cell death. Cell 2000; 103:839-42; PMID:11136969; http://dx.doi.org/10.1016/S0092-8674(00)00187-2
25. Reed JC. Bcl-2 and the regulation of programmed cell death. J Cell Biol 1994; 124:1-6; PMID:8294493; http://dx.doi.org/10.1083/jcb.124.1.1
26. Adams JM. Ways of dying: multiple pathways to apoptosis. Genes Dev 2003; 17:2481-95; PMID:14561771; http://dx.doi.org/10.1101/gad.1126903
27. Bouillet P, Strasser A. BH3-only proteins—evolutionarily conserved proapoptotic Bcl-2 family members essential for initiating programmed cell death. J Cell Sci 2002; 115:1567-74; PMID:11950875
28. Chen L, Willis SN, Wei A, Smith BJ, Fletcher JI, Hinds MG, et al. Differential targeting of prosurvival Bcl-2 proteins by their BH3-only ligands allows complementary apoptotic function. Mol Cell 2005; 17:393-403; PMID:15694340; http://dx.doi.org/10.1016/j.molcel.2004.12.030
29. Dewson G, Kluck RM. Mechanisms by which Bak and Bax permeabilise mitochondria during apoptosis. J Cell Sci 2009; 122:2801-8; PMID:19795525; http://dx.doi.org/10.1242/jcs.038166
30. Willis SN, Adams JM. Life in the balance: how BH3-only proteins induce apoptosis. Curr Opin Cell Biol 2005; 17:617-25; PMID:16243507; http://dx.doi.org/10.1016/j.ceb.2005.10.001
31. Willis SN, Chen L, Dewson G, Wei A, Naik E, Fletcher JI, et al. Proapoptotic Bak is sequestered by Mcl-1 and Bcl-xL, but not Bcl-2, until displaced by BH3-only proteins. Genes Dev 2005; 19:1294-305; PMID:15901672; http://dx.doi.org/10.1101/gad.1304105
32. Melino G, Bernassola F, Ranalli M, Yee K, Zong WX, Corazzari M, et al. p73 Induces apoptosis via PUMA transactivation and Bax mitochondrial translocation. J Biol Chem 2004; 279:8076-83; PMID:14634023; http://dx.doi.org/10.1074/jbc.M307469200
33. Zamzami N, Kroemer G. The mitochondrion in apoptosis: how Pandora's box opens. Nat Rev Mol Cell Biol 2001; 2:67-71; PMID:11413468; http://dx.doi.org/10.1038/35048073
34. Kakkar P, Singh BK. Mitochondria: a hub of redox activities and cellular distress control. Mol Cell Biochem 2007; 305:235-53; PMID:17562131; http://dx.doi.org/10.1007/s11010-007-9520-8
35. Green DR, Evan GI. A matter of life and death. Cancer Cell 2002; 1:19-30; PMID:12086884; http://dx.doi.org/10.1016/S1535-6108(02)00024-7
36. Li P, Nijhawan D, Budihardjo I, Srinivasula SM, Ahmad M, Alnemri ES, et al. Cytochrome c and dATP-dependent formation of Apaf-1/caspase-9 complex initiates an apoptotic protease cascade. Cell 1997; 91:479-89; PMID:9390557; http://dx.doi.org/10.1016/S0092-8674(00)80434-1
37. Srinivasula SM, Ahmad M, Fernandes-Alnemri T, Alnemri ES. Autoactivation of procaspase-9 by Apaf-1-mediated oligomerization. Mol Cell 1998; 1:949-57; PMID:9651578; http://dx.doi.org/10.1016/S1097-2765(00)80095-7
38. Khaitlina SY. Functional specificity of actin isoforms. Int Rev Cytol 2001; 202:35-98; PMID:11061563; http://dx.doi.org/10.1016/S0074-7696(01)02003-4
39. Pollard TD, Cooper JA. Actin and actin-binding proteins. A critical evaluation of mechanisms and functions. Annu Rev Biochem 1986; 55:987-1035; PMID:3527055; http://dx.doi.org/10.1146/annurev.bi.55.070186.005011
40. Franklin-Tong VE, Gourlay CW. A role for actin in regulating apoptosis/programmed cell death: evidence spanning yeast, plants and animals. Biochem J 2008; 413:389-404; PMID:18613816; http://dx.doi.org/10.1042/BJ20080320
41. Yarar D, Waterman-Storer CM, Schmid SL. A dynamic actin cytoskeleton functions at multiple stages of clathrin-mediated endocytosis. Mol Biol Cell 2005; 16:964-75; PMID:15601897; http://dx.doi.org/10.1091/mbc.E04-09-0774
42. Huxley HE. The mechanism of muscular contraction. Science 1969; 164:1356-65; PMID:4181952; http://dx.doi.org/10.1126/science.164.3886.1356
43. Schevzov G, Lloyd C, Gunning P. High level expression of transfected beta- and gamma-actin genes differentially impacts on myoblast cytoarchitecture. J Cell Biol 1992; 117:775-85; PMID:1577857; http://dx.doi.org/10.1083/jcb.117.4.775
44. Cooper JA. The role of actin polymerization in cell motility. Annu Rev Physiol 1991; 53:585-605; PMID:2042972; http://dx.doi.org/10.1146/annurev.ph.53.030191.003101
45. Hoock TC, Newcomb PM, Herman IM. Beta actin and its mRNA are localized at the plasma membrane and the regions of moving cytoplasm during the cellular response to injury. J Cell Biol 1991; 112:653-64; PMID:1993736; http://dx.doi.org/10.1083/jcb.112.4.653
46. Hill MA, Gunning P. Beta and gamma actin mRNAs are differentially located within myoblasts. J Cell Biol 1993; 122:825-32; PMID:8349732; http://dx.doi.org/10.1083/jcb.122.4.825
47. Dominguez R, Holmes KC. Actin structure and function. Annu Rev Biophys 2011; 40:169-86; PMID:21314430; http://dx.doi.org/10.1146/annurev-biophys-042910-155359
48. Pollard TD, Borisy GG. Cellular motility driven by assembly and disassembly of actin filaments. Cell 2003; 112:453-65; PMID:12600310; http://dx.doi.org/10.1016/S0092-8674(03)00120-X
49. Wang YL. Exchange of actin subunits at the leading edge of living fibroblasts: possible role of treadmilling. J Cell Biol 1985; 101:597-602; PMID:4040521; http://dx.doi.org/10.1083/jcb.101.2.597
50. Hall A, Nobes CD. Rho GTPases: molecular switches that control the organization and dynamics of the actin cytoskeleton. Philos Trans R Soc Lond B Biol Sci sciences 2000; 355:965-70.
51. Janmey PA. Phosphoinositides and calcium as regulators of cellular actin assembly and disassembly. Annu Rev Physiol 1994; 56:169-91; PMID:8010739; http://dx.doi.org/10.1146/annurev.ph.56.030194.001125
52. Rozelle AL, Machesky LM, Yamamoto M, Driessens MH, Insall RH, Roth MG, et al. Phosphatidylinositol 4,5-bisphosphate induces actin-based movement of raft-enriched vesicles through WASP-Arp2/3. Curr Biol 2000; 10:311-20; PMID:10744973; http://dx.doi.org/10.1016/S0960-9822(00)00384-5
53. Laster SM, Mackenzie JM, Jr. Bleb formation and F-actin distribution during mitosis and tumor necrosis factor-induced apoptosis. Microsc Res Tech 1996; 34:272-80; PMID:8743415; http://dx.doi.org/10.1002/(SICI)1097-0029(19960615)34:3<272::AID-JEMT10>3.0.CO;2-J
54. Charras GT, Hu CK, Coughlin M, Mitchison TJ. Reassembly of contractile actin cortex in cell blebs. J Cell Biol 2006; 175:477-90; PMID:17088428; http://dx.doi.org/10.1083/jcb.200602085
55. Coleman ML, Olson MF. Rho GTPase signalling pathways in the morphological changes associated with apoptosis. Cell Death Differ 2002; 9:493-504; PMID:11973608; http://dx.doi.org/10.1038/sj.cdd.4400987
56. Coleman ML, Sahai EA, Yeo M, Bosch M, Dewar A, Olson MF. Membrane blebbing during apoptosis results from caspase-mediated activation of ROCK I. Nat Cell Biol 2001; 3:339-45; PMID:11283606; http://dx.doi.org/10.1038/35070009
57. Mills JC, Stone NL, Erhardt J, Pittman RN. Apoptotic membrane blebbing is regulated by myosin light chain phosphorylation. J Cell Biol 1998; 140:627-36; PMID:9456322; http://dx.doi.org/10.1083/jcb.140.3.627
58. Sebbagh M, Renvoizé C, Hamelin J, Riché N, Bertoglio J, Bréard J. Caspase-3-mediated cleavage of ROCK I induces MLC phosphorylation and apoptotic membrane blebbing. Nat Cell Biol 2001; 3:346-52; PMID:11283607; http://dx.doi.org/10.1038/35070019
59. Cotter TG, Lennon SV, Glynn JM, Green DR. Microfilament-disrupting agents prevent the formation of apoptotic bodies in tumor cells undergoing apoptosis. Cancer Res 1992; 52:997-1005; PMID:1737363
60. Mashima T, Naito M, Noguchi K, Miller DK, Nicholson DW, Tsuruo T. Actin cleavage by CPP-32/apopain during the development of apoptosis. Oncogene 1997; 14:1007-12; PMID:9070648; http://dx.doi.org/10.1038/sj.onc.1200919
61. Mashima T, Naito M, Tsuruo T. Caspase-mediated cleavage of cytoskeletal actin plays a positive role in the process of morphological apoptosis. Oncogene 1999; 18:2423-30; PMID:10229193; http://dx.doi.org/10.1038/sj.onc.1202558
62. Bubb MR, Senderowicz AM, Sausville EA, Duncan KL, Korn ED. Jasplakinolide, a cytotoxic natural product, induces actin polymerization and competitively inhibits the binding of phalloidin to F-actin. J Biol Chem 1994; 269:14869-71; PMID:8195116
63. Odaka C, Sanders ML, Crews P. Jasplakinolide induces apoptosis in various transformed cell lines by a caspase-3-like protease-dependent pathway. Clin Diagn Lab Immunol 2000; 7:947-52; PMID:11063504
64. Rao JY, Jin YS, Zheng Q, Cheng J, Tai J, Hemstreet GP, 3rd. Alterations of the actin polymerization status as an apoptotic morphological effector in HL-60 cells. J Cell Biochem 1999; 75:686-97; PMID:10572251; http://dx.doi.org/10.1002/(SICI)1097-4644(19991215)75:4<686::AID-JCB14>3.0.CO;2-F
65. Peitsch MC, Polzar B, Stephan H, Crompton T, MacDonald HR, Mannherz HG, et al. Characterization of the endogenous deoxyribonuclease involved in nuclear DNA degradation during apoptosis (programmed cell death). EMBO J 1993; 12:371-7; PMID:8428592
66. Weber A, Pennise CR, Pring M. DNase I increases the rate constant of depolymerization at the pointed (-) end of actin filaments. Biochemistry 1994; 33:4780-6; PMID:8161537; http://dx.doi.org/10.1021/bi00182a005
67. Cooper JA. Effects of cytochalasin and phalloidin on actin. J Cell Biol 1987; 105:1473-8; PMID:3312229; http://dx.doi.org/10.1083/jcb.105.4.1473
68. Suria H, Chau LA, Negrou E, Kelvin DJ, Madrenas J. Cytoskeletal disruption induces T cell apoptosis by a caspase-3 mediated mechanism. Life Sci 1999; 65:2697-707; PMID:10622279; http://dx.doi.org/10.1016/S0024-3205(99)00538-X
69. Celeste Morley S, Sun GP, Bierer BE. Inhibition of actin polymerization enhances commitment to and execution of apoptosis induced by withdrawal of trophic support. J Cell Biochem 2003; 88:1066-76; PMID:12616543; http://dx.doi.org/10.1002/jcb.10449

70. Parlato S, Giammarioli AM, Logozzi M, Lozupone F, Matarrese P, Luciani F, et al. CD95 (APO-1/Fas) linkage to the actin cytoskeleton through ezrin in human T lymphocytes: a novel regulatory mechanism of the CD95 apoptotic pathway. EMBO J 2000; 19:5123-34; PMID:11013215; http://dx.doi.org/10.1093/emboj/19.19.5123
71. Tsukita S, Yonemura S. Cortical actin organization: lessons from ERM (ezrin/radixin/moesin) proteins. J Biol Chem 1999; 274:34507-10; PMID:10574907; http://dx.doi.org/10.1074/jbc.274.49.34507
72. Turunen O, Wahlström T, Vaheri A. Ezrin has a COOH-terminal actin-binding site that is conserved in the ezrin protein family. J Cell Biol 1994; 126:1445-53; PMID:8089177; http://dx.doi.org/10.1083/jcb.126.6.1445
73. Algeciras-Schimnich A, Peter ME. Actin dependent CD95 internalization is specific for Type I cells. FEBS Lett 2003; 546:185-8; PMID:12832037; http://dx.doi.org/10.1016/S0014-5793(03)00558-1
74. Algrain M, Turunen O, Vaheri A, Louvard D, Arpin M. Ezrin contains cytoskeleton and membrane binding domains accounting for its proposed role as a membrane-cytoskeletal linker. J Cell Biol 1993; 120:129-39; PMID:8416983; http://dx.doi.org/10.1083/jcb.120.1.129
75. Kuo WC, Yang KT, Hsieh SL, Lai MZ. Ezrin is a negative regulator of death receptor-induced apoptosis. Oncogene 2010; 29:1374-83; PMID:19935704; http://dx.doi.org/10.1038/onc.2009.417
76. Gourlay CW, Ayscough KR. A role for actin in aging and apoptosis. Biochem Soc Trans 2005; 33:1260-4; PMID:16246093; http://dx.doi.org/10.1042/BST20051260
77. Thomas SG, Huang S, Li S, Staiger CJ, Franklin-Tong VE. Actin depolymerization is sufficient to induce programmed cell death in self-incompatible pollen. J Cell Biol 2006; 174:221-9; PMID:16831890; http://dx.doi.org/10.1083/jcb.200604011
78. Wertman KF, Drubin DG, Botstein D. Systematic mutational analysis of the yeast ACT1 gene. Genetics 1992; 132:337-50; PMID:1427032
79. Belmont LD, Drubin DG. The yeast V159N actin mutant reveals roles for actin dynamics in vivo. J Cell Biol 1998; 142:1289-99; PMID:9732289; http://dx.doi.org/10.1083/jcb.142.5.1289
80. Xu X, Forbes JG, Colombini M. Actin modulates the gating of Neurospora crassa VDAC. J Membr Biol 2001; 180:73-81; PMID:11284205; http://dx.doi.org/10.1007/s002320010060
81. Paul C, Manero F, Gonin S, Kretz-Remy C, Virot S, Arrigo AP. Hsp27 as a negative regulator of cytochrome C release. Mol Cell Biol 2002; 22:816-34; PMID:11784858; http://dx.doi.org/10.1128/MCB.22.3.816-834.2002
82. Posey SC, Bierer BE. Actin stabilization by jasplakinolide enhances apoptosis induced by cytokine deprivation. J Biol Chem 1999; 274:4259-65; PMID:9933626; http://dx.doi.org/10.1074/jbc.274.7.4259
83. Morton WM, Ayscough KR, McLaughlin PJ. Latrunculin alters the actin-monomer subunit interface to prevent polymerization. Nat Cell Biol 2000; 2:376-8; PMID:10854330; http://dx.doi.org/10.1038/35014075
84. Martin SS, Leder P. Human MCF10A mammary epithelial cells undergo apoptosis following actin depolymerization that is independent of attachment and rescued by Bcl-2. Mol Cell Biol 2001; 21:6529-36; PMID:11533241; http://dx.doi.org/10.1128/MCB.21.19.6529-6536.2001
85. Genescà M, Sola A, Hotter G. Actin cytoskeleton derangement induces apoptosis in renal ischemia/reperfusion. Apoptosis 2006; 11:563-71; PMID:16528472; http://dx.doi.org/10.1007/s10495-006-4937-1
86. Puthalakath H, Villunger A, O'Reilly LA, Beaumont JG, Coultas L, Cheney RE, et al. Bmf: a proapoptotic BH3-only protein regulated by interaction with the myosin V actin motor complex, activated by anoikis. Science 2001; 293:1829-32; PMID:11546872; http://dx.doi.org/10.1126/science.1062257
87. Pendleton A, Pope B, Weeds A, Koffer A. Latrunculin B or ATP depletion induces cofilin-dependent translocation of actin into nuclei of mast cells. J Biol Chem 2003; 278:14394-400; PMID:12566455; http://dx.doi.org/10.1074/jbc.M206393200
88. Meijerman I, Blom WM, de Bont HJ, Mulder GJ, Nagelkerke JF. Induction of apoptosis and changes in nuclear G-actin are mediated by different pathways: the effect of inhibitors of protein and RNA synthesis in isolated rat hepatocytes. Toxicol Appl Pharmacol 1999; 156:46-55; PMID:10101098; http://dx.doi.org/10.1006/taap.1998.8616
89. Utsumi T, Sakurai N, Nakano K, Ishisaka R. C-terminal 15 kDa fragment of cytoskeletal actin is posttranslationally N-myristoylated upon caspase-mediated cleavage and targeted to mitochondria. FEBS Lett 2003; 539:37-44; PMID:12650923; http://dx.doi.org/10.1016/S0014-5793(03)00180-7
90. Slee EA, Keogh SA, Martin SJ. Cleavage of BID during cytotoxic drug and UV radiation-induced apoptosis occurs downstream of the point of Bcl-2 action and is catalysed by caspase-3: a potential feedback loop for amplification of apoptosis-associated mitochondrial cytochrome c release. Cell Death Differ 2000; 7:556-65; PMID:10822279; http://dx.doi.org/10.1038/sj.cdd.4400689
91. Bamburg JR. Proteins of the ADF/cofilin family: essential regulators of actin dynamics. Annu Rev Cell Dev Biol 1999; 15:185-230; PMID:10611961; http://dx.doi.org/10.1146/annurev.cellbio.15.1.185
92. DesMarais V, Ghosh M, Eddy R, Condeelis J. Cofilin takes the lead. J Cell Sci 2005; 118:19-26; PMID:15615780; http://dx.doi.org/10.1242/jcs.01631
93. Huang TY, DerMardirossian C, Bokoch GM. Cofilin phosphatases and regulation of actin dynamics. Curr Opin Cell Biol 2006; 18:26-31; PMID:16337782; http://dx.doi.org/10.1016/j.ceb.2005.11.005
94. Chua BT, Volbracht C, Tan KO, Li R, Yu VC, Li P. Mitochondrial translocation of cofilin is an early step in apoptosis induction. Nat Cell Biol 2003; 5:1083-9; PMID:14634665; http://dx.doi.org/10.1038/ncb1070
95. Klamt F, Zdanov S, Levine RL, Pariser A, Zhang Y, Zhang B, et al. Oxidant-induced apoptosis is mediated by oxidation of the actin-regulatory protein cofilin. Nat Cell Biol 2009; 11:1241-6; PMID:19734890; http://dx.doi.org/10.1038/ncb1968
96. Pfannstiel J, Cyrklaff M, Habermann A, Stoeva S, Griffiths G, Shoeman R, et al. Human cofilin forms oligomers exhibiting actin bundling activity. J Biol Chem 2001; 276:49476-84; PMID:11679578; http://dx.doi.org/10.1074/jbc.M104760200
97. Kotiadis VN, Leadsham JE, Bastow EL, Gheeraert A, Whybrew JM, Bard M, et al. Identification of new surfaces of Cofilin that link mitochondrial function to the control of multi-drug resistance. J Cell Sci 2012. In press. PMID:22344251; http://dx.doi.org/10.1242/jcs.099390
98. Minamide LS, Striegl AM, Boyle JA, Meberg PJ, Bamburg JR. Neurodegenerative stimuli induce persistent ADF/cofilin-actin rods that disrupt distal neurite function. Nat Cell Biol 2000; 2:628-36; PMID:10980704; http://dx.doi.org/10.1038/35023579
99. Ashworth SL, Southgate EL, Sandoval RM, Meberg PJ, Bamburg JR, Molitoris BA. ADF/cofilin mediates actin cytoskeletal alterations in LLC-PK cells during ATP depletion. Am J Physiol Renal Physiol 2003; 284:F852-62; PMID:12620926
100. Bernstein BW, Chen H, Boyle JA, Bamburg JR. Formation of actin-ADF/cofilin rods transiently retards decline of mitochondrial potential and ATP in stressed neurons. Am J Physiol Cell Physiol 2006; 291:C828-39; PMID:16738008; http://dx.doi.org/10.1152/ajpcell.00066.2006
101. Woo JA, Jung AR, Lakshmana MK, Bedrossian A, Lim Y, Bu JH, et al. Pivotal role of the RanBP9-cofilin pathway in Aβ-induced apoptosis and neurodegeneration. Cell Death Differ 2012. In press. PMID:22361682; http://dx.doi.org/10.1038/cdd.2012.14
102. Safer D, Sosnick TR, Elzinga M. Thymosin beta 4 binds actin in an extended conformation and contacts both the barbed and pointed ends. Biochemistry 1997; 36:5806-16; PMID:9153421; http://dx.doi.org/10.1021/bi970185v
103. Rho SB, Chun T, Lee SH, Park K, Lee JH. The interaction between E-tropomodulin and thymosin beta-10 rescues tumor cells from thymosin beta-10 mediated apoptosis by restoring actin architecture. FEBS Lett 2004; 557:57-63; PMID:14741341; http://dx.doi.org/10.1016/S0014-5793(03)01438-8
104. Hall AK. Thymosin beta-10 accelerates apoptosis. Cell Mol Biol Res 1995; 41:167-80; PMID:8589757
105. D'Addario M, Arora PD, Fan J, Ganss B, Ellen RP, McCulloch CA. Cytoprotection against mechanical forces delivered through beta 1 integrins requires induction of filamin A. J Biol Chem 2001; 276:31969-77; PMID:11423540; http://dx.doi.org/10.1074/jbc.M102715200
106. Browne KA, Johnstone RW, Jans DA, Trapani JA. Filamin (280-kDa actin-binding protein) is a caspase substrate and is also cleaved directly by the cytotoxic T lymphocyte protease granzyme B during apoptosis. J Biol Chem 2000; 275:39262-6; PMID:11050075; http://dx.doi.org/10.1074/jbc.C000622200
107. Umeda T, Kouchi Z, Kawahara H, Tomioka S, Sasagawa N, Maeda T, et al. Limited proteolysis of filamin is catalyzed by caspase-3 in U937 and Jurkat cells. J Biochem 2001; 130:535-42; PMID:11574073; http://dx.doi.org/10.1093/oxfordjournals.jbchem.a003016
108. Cai L, Marshall TW, Uetrecht AC, Schafer DA, Bear JE. Coronin 1B coordinates Arp2/3 complex and cofilin activities at the leading edge. Cell 2007; 128:915-29; PMID:17350576; http://dx.doi.org/10.1016/j.cell.2007.01.031
109. Föger N, Rangell L, Danilenko DM, Chan AC. Requirement for coronin 1 in T lymphocyte trafficking and cellular homeostasis. Science 2006; 313:839-42; PMID:16902139; http://dx.doi.org/10.1126/science.1130563
110. Moriceau S, Kantari C, Mocek J, Davezac N, Gabillet J, Guerrera IC, et al. Coronin-1 is associated with neutrophil survival and is cleaved during apoptosis: potential implication in neutrophils from cystic fibrosis patients. J Immunol 2009; 182:7254-63; PMID:19454722; http://dx.doi.org/10.4049/jimmunol.0803312
111. Bresnick AR. Molecular mechanisms of nonmuscle myosin-II regulation. Curr Opin Cell Biol 1999; 11:26-33; PMID:10047526; http://dx.doi.org/10.1016/S0955-0674(99)80004-0
112. Ma X, Kovács M, Conti MA, Wang A, Zhang Y, Sellers JR, et al. Nonmuscle myosin II exerts tension but does not translocate actin in vertebrate cytokinesis. Proc Natl Acad Sci U S A 2012; 109:4509-14; PMID:22393000; http://dx.doi.org/10.1073/pnas.1116268109
113. Petrache I, Verin AD, Crow MT, Birukova A, Liu F, Garcia JG. Differential effect of MLC kinase in TNF-alpha-induced endothelial cell apoptosis and barrier dysfunction. Am J Physiol Lung Cell Mol Physiol 2001; 280:L1168-78; PMID:11350795

114. Jin Y, Atkinson SJ, Marrs JA, Gallagher PJ. Myosin ii light chain phosphorylation regulates membrane localization and apoptotic signaling of tumor necrosis factor receptor-1. J Biol Chem 2001; 276:30342-9; PMID:11384975; http://dx.doi.org/10.1074/jbc.M102404200
115. Adamson RH, Curry FE, Adamson G, Liu B, Jiang Y, Aktories K, et al. Rho and rho kinase modulation of barrier properties: cultured endothelial cells and intact microvessels of rats and mice. J Physiol 2002; 539: 295-308; PMID:11850521; http://dx.doi.org/10.1113/jphysiol.2001.013117
116. Rosenblatt J, Raff MC, Cramer LP. An epithelial cell destined for apoptosis signals its neighbors to extrude it by an actin- and myosin-dependent mechanism. Curr Biol 2001; 11:1847-57; PMID:11728307; http://dx.doi.org/10.1016/S0960-9822(01)00587-5
117. McGough AM, Staiger CJ, Min JK, Simonetti KD. The gelsolin family of actin regulatory proteins: modular structures, versatile functions. FEBS Lett 2003; 552:75-81; PMID:14527663; http://dx.doi.org/10.1016/S0014-5793(03)00932-3
118. Kothakota S, Azuma T, Reinhard C, Klippel A, Tang J, Chu K, et al. Caspase-3-generated fragment of gelsolin: effector of morphological change in apoptosis. Science 1997; 278:294-8; PMID:9323209; http://dx.doi.org/10.1126/science.278.5336.294
119. Chhabra D, Nosworthy NJ, dos Remedios CG. The N-terminal fragment of gelsolin inhibits the interaction of DNase I with isolated actin, but not with the cofilin-actin complex. Proteomics 2005; 5:3131-6; PMID:16021605; http://dx.doi.org/10.1002/pmic.200401127
120. Sakurai N, Utsumi T. Posttranslational N-myristoylation is required for the anti-apoptotic activity of human tGelsolin, the C-terminal caspase cleavage product of human gelsolin. J Biol Chem 2006; 281:14288-95; PMID:16556605; http://dx.doi.org/10.1074/jbc.M510338200
121. Ohtsu M, Sakai N, Fujita H, Kashiwagi M, Gasa S, Shimizu S, et al. Inhibition of apoptosis by the actin-regulatory protein gelsolin. EMBO J 1997; 16:4650-6; PMID:9303309; http://dx.doi.org/10.1093/emboj/16.15.4650
122. Koya RC, Fujita H, Shimizu S, Ohtsu M, Takimoto M, Tsujimoto Y, et al. Gelsolin inhibits apoptosis by blocking mitochondrial membrane potential loss and cytochrome c release. J Biol Chem 2000; 275:15343-9; PMID:10809769; http://dx.doi.org/10.1074/jbc.275.20.15343
123. Harms C, Bösel J, Lautenschlager M, Harms U, Braun JS, Hörtnagl H, et al. Neuronal gelsolin prevents apoptosis by enhancing actin depolymerization. Mol Cell Neurosci 2004; 25:69-82; PMID:14962741; http://dx.doi.org/10.1016/j.mcn.2003.09.012
124. Klampfer L, Huang J, Sasazuki T, Shirasawa S, Augenlicht L. Oncogenic Ras promotes butyrate-induced apoptosis through inhibition of gelsolin expression. J Biol Chem 2004; 279:36680-8; PMID:15213223; http://dx.doi.org/10.1074/jbc.M405197200
125. Kusano H, Shimizu S, Koya RC, Fujita H, Kamada S, Kuzumaki N, et al. Human gelsolin prevents apoptosis by inhibiting apoptotic mitochondrial changes via closing VDAC. Oncogene 2000; 19:4807-14; PMID:11039896; http://dx.doi.org/10.1038/sj.onc.1203868
126. Rao J, Seligson D, Visapaa H, Horvath S, Eeva M, Michel K, et al. Tissue microarray analysis of cytoskeletal actin-associated biomarkers gelsolin and E-cadherin in urothelial carcinoma. Cancer 2002; 95:1247-57; PMID:12216092; http://dx.doi.org/10.1002/cncr.10823
127. Gunning P, O'Neill G, Hardeman E. Tropomyosin-based regulation of the actin cytoskeleton in time and space. Physiol Rev 2008; 88:1-35; PMID:18195081; http://dx.doi.org/10.1152/physrev.00001.2007
128. Gunning PW, Schevzov G, Kee AJ, Hardeman EC. Tropomyosin isoforms: divining rods for actin cytoskeleton function. Trends Cell Biol 2005; 15: 333-41; PMID:15953552; http://dx.doi.org/10.1016/j.tcb.2005.04.007
129. Ball EH, Kovala T. Mapping of caldesmon: relationship between the high and low molecular weight forms. Biochemistry 1988; 27:6093-8; PMID: 3191109; http://dx.doi.org/10.1021/bi00416a039
130. O'Neill GM, Stehn J, Gunning PW. Tropomyosins as interpreters of the signalling environment to regulate the local cytoskeleton. Semin Cancer Biol 2008; 18: 35-44; PMID:17942320; http://dx.doi.org/10.1016/j.semcancer.2007.08.004
131. Bryce NS, Schevzov G, Ferguson V, Percival JM, Lin JJ, Matsumura F, et al. Specification of actin filament function and molecular composition by tropomyosin isoforms. Mol Biol Cell 2003; 14:1002-16; PMID: 12631719; http://dx.doi.org/10.1091/mbc.E02-04-0244
132. Creed SJ, Desouza M, Bamburg JR, Gunning P, Stehn J. Tropomyosin isoform 3 promotes the formation of filopodia by regulating the recruitment of actin-binding proteins to actin filaments. Exp Cell Res 2011; 317:249-61; PMID:21036167; http://dx.doi.org/10.1016/j.yexcr.2010.10.019
133. Frisch SM, Screaton RA. Anoikis mechanisms. Curr Opin Cell Biol 2001; 13:555-62; PMID:11544023; http://dx.doi.org/10.1016/S0955-0674(00)00251-9
134. Raval GN, Bharadwaj S, Levine EA, Willingham MC, Geary RL, Kute T, et al. Loss of expression of tropomyosin-1, a novel class II tumor suppressor that induces anoikis, in primary breast tumors. Oncogene 2003; 22:6194-203; PMID:13679858; http://dx.doi.org/10.1038/sj.onc.1206719
135. Bharadwaj S, Thanawala R, Bon G, Falcioni R, Prasad GL. Resensitization of breast cancer cells to anoikis by tropomyosin-1: role of Rho kinase-dependent cytoskeleton and adhesion. Oncogene 2005; 24:8291-303; PMID:16170368; http://dx.doi.org/10.1038/sj.onc.1208993
136. Mahadev K, Raval G, Bharadwaj S, Willingham MC, Lange EM, Vonderhaar B, et al. Suppression of the transformed phenotype of breast cancer by tropomyosin-1. Exp Cell Res 2002; 279:40-51; PMID: 12213212; http://dx.doi.org/10.1006/excr.2002.5583
137. Yamaguchi H, Condeelis J. Regulation of the actin cytoskeleton in cancer cell migration and invasion. Biochim Biophys Acta 2007; 1773:642-52.
138. Gourlay CW, Ayscough KR. Actin-induced hyperactivation of the Ras signaling pathway leads to apoptosis in Saccharomyces cerevisiae. Mol Cell Biol 2006; 26:6487-501; PMID:16914733; http://dx.doi.org/10.1128/MCB.00117-06
139. Thor AD, Edgerton SM, Liu S, Moore DH, 2nd, Kwiatkowski DJ. Gelsolin as a negative prognostic factor and effector of motility in erbB-2-positive epidermal growth factor receptor-positive breast cancers. Clin Cancer Res 2001; 7:2415-24; PMID: 11489821
140. Shieh DB, Chen IW, Wei TY, Shao CY, Chang HJ, Chung CH, et al. Tissue expression of gelsolin in oral carcinogenesis progression and its clinicopathological implications. Oral Oncol 2006; 42:599-606; PMID: 16753328; http://dx.doi.org/10.1016/j.oraloncology.2005.10.021
141. Turhani D, Krapfenbauer K, Thurnher D, Langen H, Fountoulakis M. Identification of differentially expressed, tumor-associated proteins in oral squamous cell carcinoma by proteomic analysis. Electrophoresis 2006; 27:1417-23; PMID:16568407; http://dx.doi.org/10.1002/elps.200500510
142. Martoglio AM, Tom BD, Starkey M, Corps AN, Charnock-Jones DS, Smith SK. Changes in tumorigenesis- and angiogenesis-related gene transcript abundance profiles in ovarian cancer detected by tailored high density cDNA arrays. Mol Med 2000; 6:750-65; PMID:11071270
143. Ding SJ, Li Y, Shao XX, Zhou H, Zeng R, Tang ZY, et al. Proteome analysis of hepatocellular carcinoma cell strains, MHCC97-H and MHCC97-L, with different metastasis potentials. Proteomics 2004; 4: 982-94; PMID:15048980; http://dx.doi.org/10.1002/pmic.200300653
144. van Rheenen J, Condeelis J, Glogauer M. A common cofilin activity cycle in invasive tumor cells and inflammatory cells. J Cell Sci 2009; 122:305-11; PMID:19158339; http://dx.doi.org/10.1242/jcs.031146
145. Wang W, Eddy R, Condeelis J. The cofilin pathway in breast cancer invasion and metastasis. Nat Rev Cancer 2007; 7:429-40; PMID:17522712; http://dx.doi.org/10.1038/nrc2148
146. Davila M, Frost AR, Grizzle WE, Chakrabarti R. LIM kinase 1 is essential for the invasive growth of prostate epithelial cells: implications in prostate cancer. J Biol Chem 2003; 278:36868-75; PMID:12821664; http://dx.doi.org/10.1074/jbc.M306196200
147. Yoshioka K, Foletta V, Bernard O, Itoh K. A role for LIM kinase in cancer invasion. Proc Natl Acad Sci U S A 2003; 100:7247-52; PMID:12777619; http://dx.doi.org/10.1073/pnas.1232344100
148. Andrianantoandro E, Pollard TD. Mechanism of actin filament turnover by severing and nucleation at different concentrations of ADF/cofilin. Mol Cell 2006; 24:13-23; PMID:17018289; http://dx.doi.org/10.1016/j.molcel.2006.08.006
149. Matsumura F, Lin JJ, Yamashiro-Matsumura S, Thomas GP, Topp WC. Differential expression of tropomyosin forms in the microfilaments isolated from normal and transformed rat cultured cells. J Biol Chem 1983; 258:13954-64; PMID:6315714
150. Hendricks M, Weintraub H. Tropomyosin is decreased in transformed cells. Proc Natl Acad Sci U S A 1981; 78:5633-7; PMID:6272310; http://dx.doi.org/10.1073/pnas.78.9.5633
151. Bhattacharya B, Prasad GL, Valverius EM, Salomon DS, Cooper HL. Tropomyosins of human mammary epithelial cells: consistent defects of expression in mammary carcinoma cell lines. Cancer Res 1990; 50:2105-12; PMID:2317800
152. Gimona M, Kazzaz JA, Helfman DM. Forced expression of tropomyosin 2 or 3 in v-Ki-ras-transformed fibroblasts results in distinct phenotypic effects. Proc Natl Acad Sci U S A 1996; 93:9618-23; PMID:8790379; http://dx.doi.org/10.1073/pnas.93.18.9618
153. Miyado K, Sato M, Taniguchi S. Transformation-related expression of a low-molecular-mass tropomyosin isoform TM5/TM30nm in transformed rat fibroblastic cell lines. J Cancer Res Clin Oncol 1997; 123:331-6; PMID:9222299
154. Miyado K, Kimura M, Taniguchi S. Decreased expression of a single tropomyosin isoform, TM5/TM30nm, results in reduction in motility of highly metastatic B16-F10 mouse melanoma cells. Biochem Biophys Res Commun 1996; 225:427-35; PMID: 8753779; http://dx.doi.org/10.1006/bbrc.1996.1190
155. Hanahan D, Weinberg RA. Hallmarks of cancer: the next generation. Cell 2011; 144:646-74; PMID: 21376230; http://dx.doi.org/10.1016/j.cell.2011.02.013

156. Coultas L, Strasser A. The role of the Bcl-2 protein family in cancer. Semin Cancer Biol 2003; 13:115-23; PMID:12654255; http://dx.doi.org/10.1016/S1044-579X(02)00129-3
157. Strasser A, Harris AW, Bath ML, Cory S. Novel primitive lymphoid tumours induced in transgenic mice by cooperation between myc and bcl-2. Nature 1990; 348:331-3; PMID:2250704; http://dx.doi.org/10.1038/348331a0
158. Egle A, Harris AW, Bouillet P, Cory S. Bim is a suppressor of Myc-induced mouse B cell leukemia. Proc Natl Acad Sci U S A 2004; 101:6164-9; PMID:15079075; http://dx.doi.org/10.1073/pnas.0401471101
159. Karst AM, Dai DL, Martinka M, Li G. PUMA expression is significantly reduced in human cutaneous melanomas. Oncogene 2005; 24:1111-6; PMID:15690057; http://dx.doi.org/10.1038/sj.onc.1208374
160. van Delft MF, Wei AH, Mason KD, Vandenberg CJ, Chen L, Czabotar PE, et al. The BH3 mimetic ABT-737 targets selective Bcl-2 proteins and efficiently induces apoptosis via Bak/Bax if Mcl-1 is neutralized. Cancer Cell 2006; 10:389-99; PMID:17097561; http://dx.doi.org/10.1016/j.ccr.2006.08.027
161. Fulda S, Debatin KM. Extrinsic versus intrinsic apoptosis pathways in anticancer chemotherapy. Oncogene 2006; 25:4798-811; PMID:16892092; http://dx.doi.org/10.1038/sj.onc.1209608
162. Barnhart BC, Legembre P, Pietras E, Bubici C, Franzoso G, Peter ME. CD95 ligand induces motility and invasiveness of apoptosis-resistant tumor cells. EMBO J 2004; 23:3175-85; PMID:15272306; http://dx.doi.org/10.1038/sj.emboj.7600325
163. Steller EJ, Borel Rinkes IH, Kranenburg O. How CD95 stimulates invasion. Cell Cycle 2011; 10:3857-62; PMID:22064519; http://dx.doi.org/10.4161/cc.10.22.18290
164. Ruan W, Lee CT, Desbarats J. A novel juxtamembrane domain in tumor necrosis factor receptor superfamily molecules activates Rac1 and controls neurite growth. Mol Biol Cell 2008; 19:3192-202; PMID:18508927; http://dx.doi.org/10.1091/mbc.E08-02-0161
165. Takahashi K, Sasaki T, Mammoto A, Takaishi K, Kameyama T, Tsukita S, et al. Direct interaction of the Rho GDP dissociation inhibitor with ezrin/radixin/moesin initiates the activation of the Rho small G protein. J Biol Chem 1997; 272:23371-5; PMID:9287351; http://dx.doi.org/10.1074/jbc.272.37.23371
166. Stracke ML, Soroush M, Liotta LA, Schiffmann E. Cytoskeletal agents inhibit motility and adherence of human tumor cells. Kidney Int 1993; 43:151-7; PMID:8094471; http://dx.doi.org/10.1038/ki.1993.25
167. Bousquet PF, Paulsen LA, Fondy C, Lipski KM, Loucy KJ, Fondy TP. Effects of cytochalasin B in culture and in vivo on murine Madison 109 lung carcinoma and on B16 melanoma. Cancer Res 1990; 50:1431-9; PMID:2302708
168. Senderowicz AM, Kaur G, Sainz E, Laing C, Inman WD, Rodríguez J, et al. Jasplakinolide's inhibition of the growth of prostate carcinoma cells in vitro with disruption of the actin cytoskeleton. J Natl Cancer Inst 1995; 87:46-51; PMID:7666463; http://dx.doi.org/10.1093/jnci/87.1.46
169. Konishi H, Kikuchi S, Ochiai T, Ikoma H, Kubota T, Ichikawa D, et al. Latrunculin a has a strong anticancer effect in a peritoneal dissemination model of human gastric cancer in mice. Anticancer Res 2009; 29:2091-7; PMID:19528469
170. Bai R, Verdier-Pinard P, Gangwar S, Stessman CC, McClure KJ, Sausville EA, et al. Dolastatin 11, a marine depsipeptide, arrests cells at cytokinesis and induces hyperpolymerization of purified actin. Mol Pharmacol 2001; 59:462-9; PMID:11179440
171. Stehn JR, Schevzov G, O'Neill GM, Gunning PW. Specialisation of the tropomyosin composition of actin filaments provides new potential targets for chemotherapy. Curr Cancer Drug Targets 2006; 6:245-56; PMID:16712460; http://dx.doi.org/10.2174/156800906776842948
172. Bonello TT, Stehn JR, Gunning PW. New approaches to targeting the actin cytoskeleton for chemotherapy. Future Med Chem 2009; 1:1311-31; PMID:21426105; http://dx.doi.org/10.4155/fmc.09.99
173. Ross-Macdonald P, de Silva H, Guo Q, Xiao H, Hung CY, Penhallow B, et al. Identification of a nonkinase target mediating cytotoxicity of novel kinase inhibitors. Mol Cancer Ther 2008; 7:3490-8; PMID:19001433; http://dx.doi.org/10.1158/1535-7163.MCT-08-0826
174. Takeshita H, Kusuzaki K, Ashihara T, Gebhardt MC, Mankin HJ, Hirasawa Y. Actin organization associated with the expression of multidrug resistant phenotype in osteosarcoma cells and the effect of actin depolymerization on drug resistance. Cancer Lett 1998; 126:75-81; PMID:9563651; http://dx.doi.org/10.1016/S0304-3835(97)00539-9
175. Verrills NM, Walsh BJ, Cobon GS, Hains PG, Kavallaris M. Proteome analysis of vinca alkaloid response and resistance in acute lymphoblastic leukemia reveals novel cytoskeletal alterations. J Biol Chem 2003; 278:45082-93; PMID:12949081; http://dx.doi.org/10.1074/jbc.M303378200
176. Verrills NM, Po'uha ST, Liu ML, Liaw TY, Larsen MR, Ivery MT, et al. Alterations in gamma-actin and tubulin-targeted drug resistance in childhood leukemia. J Natl Cancer Inst 2006; 98:1363-74; PMID:17018783; http://dx.doi.org/10.1093/jnci/djj372
177. Inohara N, Ding L, Chen S, Nunez G. harakiri, a novel regulator of cell death, encodes a protein that activates apoptosis and interacts selectively with survival-promoting proteins Bcl-2 and Bcl-X(L). EMBO J 1997; 16:1686-94; PMID:9130713; http://dx.doi.org/10.1093/emboj/16.7.1686