MINI-REVIEW

Significance of Caveolin-1 Regulators in Pancreatic Cancer

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Abstract

Caveolin-1 is a scaffold protein on the cell membrane. As the main component of caveolae, caveolin-1 is involved in many biological processes that include substance uptake and transmembrane signaling. Many of these processes and thus caveolin-1 contribute to cell transformation, tumorigenesis, and metastasis. Of particular interest are the dual rolesof tumor suppressor and oncogene that caveolin-1 appear to play in different malignancies, including pancreatic cancer. Therefore, analyzing caveolin-1 regulators and understanding their mechanisms of actionis key to identifying novel diagnostic and therapeutic tools for pancreatic cancer. This review details the mechanisms of action of caveolin-1 regulators and the potential significance for pancreatic cancer treatment.

Keywords: Caveolae - caveolin-1 - pancreatic cancer - regulatory factor - significance

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Caveolin-1 and Pancreatic Cancer

Caveolin-1

Caveolae were originally identified as omega-shaped invaginations of the plasma membrane inepithelial cells (Smart et al., 1999), which were discovered by Palade in the 1950s (Casley-Smith et al., 1975). Currently, caveolae are considered integral transmembrane microdomains and critical components for the interactions between integrin receptors and cytoskeleton-associated signaling molecules (Cordes et al., 2007). Further, they are associated with various membranous structures, including the endoplasmic reticulum, Golgi, and plasma membranes (Parat et al., 2004). Caveolae are specialized structures mainly composed of cholesterols and sphingolipids. They are abundant in endothelia, muscle cells, adipocytes, and lung epithelial cells (Okamoto et al., 1998), and are implicated in several endocytic and trafficking mechanisms. The coat proteins required for caveolae formation are the three caveolins: caveolin-1, -2, and -3 (Fujimoto et al., 2000). Caveolin-1 and -2 are ubiquitously expressed in the human body, whereas caveolin-3 is found only in muscle tissue (Anderson et al., 1998). Caveolin-1 is the major structural protein in caveolae (Smart et al., 1994) and acts as a scaffold to organize multiple molecular complexes that regulate a variety of cellular events (Kato et al., 2004) such as cellular transformation, tumorigenesis, cell metastasis, and angiogenesis.

However, the fact that it appears to act as both a tumor suppressor and oncogene, depending on the context, is especially intriguing. In ovarian (Prinetti et al., 2010), colon (Nimri et al., 2012), and breast cancer cells (Rao et al., 2012; Simpkins et al., 2012), caveolin-1 is down-regulated and negatively correlated with the malignant potential of tumor cells. It is up-regulated and promotes cell proliferation and invasion in bladder (Thomas et al., 2011), esophageal (Kato et al., 2002), and prostate cancer cells (Li et al., 2001). High expression of caveolin-1 has favorable prognoses in bile duct cancer and breast cancer (Murakami et al., 2003; Rao et al., 2012), but is correlated with poor prognoses in prostate, esophageal, renal, and non-small cell lung carcinoma (Li et al., 2001; Kato et al., 2002; Steffens et al., 2011). In pancreatic cancer, caveolin-1 is reduced compared to normal pancreaticor precancerous tumor tissue.

Pancreatic cancer

Pancreatic cancer is one of the deadliest cancers (Shi et al., 2012) and has been called the king of cancer because of its poor cure rate and prognosis (Siegel et al., 2012). Compared with other cancers, it has higher resistance to conventional treatments including surgery, radiation, and/or chemotherapy (Diamantidis et al., 2008). Despite the fact that diagnostic techniques are rapidly developing, the early diagnosis of pancreatic cancer remains poor (Luo et al., 2008). Data indicates that the five-year survival rate ranges between 0.4 and 2 percent in the United States (Krechler et al., 2011). Furthermore, 75 percent of the patients who are diagnosed at an advanced stage die within 1 year. Currently, surgical resection is the only treatment that results in long-term survival for pancreatic cancer patients.

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Table 1. Caveolin-1 Regulators

Regulatory Factor	Method	Outcome	References
Forkhead box (FOXO)	PI3K/AKT/FOXO pathway	Promotion	(Boreddy et al., 2011; Roy et al., 2010)
Lipopolysaccharide (LPS)	Inhibit NF-κB activation by preventing the formation of IKK-γ/IKK complex and TLR4/MyD88 signaling	Suppression	(Tiruppathi et al., 2008; Ikebe et al., 2009)
High-density lipoprotein (HDL)	Activate MAP kinase pathway through ERK1/2	Suppression	(Frank et al., 2001)
Stimulatory protein 1 (Sp1)	Affect promoter activity	Promotion	(Dasari et al., 2006)
Estrogen receptorα (ERα)	Methylate caveolin-1 gene promoter	Suppression	(Zschocke et al., 2003)
p53	Bind caveolin-1 promoter sequence with E2F/DP-1	Promotion	(Lee et al., 2012)
Cholesterol	Sterol regulatory element binding protein (SREBP) and Sp1	Suppression	(Llaverias et al., 2004)
Carbon monoxide (CO)	Activate guanylatecyclase and p38 MAPK	Suppression	(Kim et al., 2005)
Vascular endothelial growth factor (VEGF)	VEGF/MEK signal transduction pathway and protein kinase C/MEK/c-myc gene/androgen receptor pathway	Suppression	(Liu et al., 1999)
Epithelial membrane protein 2 (EMP2)	Promote formation of membrane lipid rafts containingGPI-APS	Promotion	(Wadehra et al., 2004)
Endothelial NO synthase (eNOS)	Inhibit the catalytic activity of eNOS	Promotion	(Venema et al., 1997)
Reactive oxygen species (ROS)	Catalase and N-acetylcysteine; prevent formation of caveolin-1-ubiquitin complex	Suppression	(Rungtabnapa et al., 2011)
Src kinase	Phosphorylate caveolin-1	Suppression	(Shields et al., 2011)
Na+/K+-ATPase	Separate regulation of Na+/K+-ATPase in the transport process	Promotion	(Cai et al., 2008)
Breast cancer susceptibility gene 1 (BRCA1)	Anti-caveolin-1 gene in caveolin-1 gene promoter	Promotion	(Wang et al., 2008)
Flotillin-1	Prevent lysosomal degradation	Promotion	(Vassilieva et al., 2009)

Structure and Expression of Caveolin-1

Caveolin-1, a 21-24kDa integral membrane protein, is a principal component of caveolae membranes in vivo (Liu et al., 2013). Caveolae are involved in constitutive endocytic trafficking. Liquid-ordered domains are formed within the Golgi apparatus and thus the biogenesis of both caveolae and caveolae-related liquid-ordered domains initiate in the Golgi and are transported to the cell surface by vesicular organelles. Caveolin-1 is formed during endocytosis and recycled back to the cell membrane (Smart et al., 1999). Immunofluorescent staining of cells transfected with caveolin-1 indicated that, like the NH2 terminus, the COOH-terminal region is located on the cytoplasmic side of the plasma membrane. Using the anti-peptide antibodies and epitope tags targeting the N- and C-terminal, Glenney et al. found that the N-terminal and C-terminal are both located on the cytoplasmic side of the plasma membrane. The NH2 terminus has a tyrosine that is phosphorylated by V-Src (Glenney et al., 1989) and the C-terminus has a cysteine palmitoylationsite (Dietzen et al., 1995). Studies have revealed that COOH-terminal palmitoylationis crucial for caveolin-1 to attach to the plasma membrane (Sowa et al., 2003). Both phosphorylation and palmitoylation occurintracellularly (Sargiacomo et al., 1993). Caveolin-1 interacts with a variety of signaling molecules, including endothelial nitric oxide synthase (eNOS), heterotrimeric G proteins, adhesion molecules, nonreceptor tyrosine kinases, Src-family tyrosine kinases, and p42/44 mitogen-activated protein kinase (MAPK). Residues 82-101 in the N-terminal region are called the caveolin-1 scaffolding domain (CSD) and serve tobind other molecules to the cell membrane (Arbuzova et al., 2000). Couet et al. found that the CSD was the area where caveolin-1 interacted with signaling molecules indicating that the CSD is the most important functional area ofcaveolin-1 (Couet et al., 1997). Some factors have been identifiedthatinteract with the CSD and regulate caveolin-1 activity.

Caveolin-1 Regulation in the Human Body

In the pre-transcriptional and transcriptional stages, caveolin-1 is regulated mainly through cell signaling pathways. During the post-transcriptional stage, expression is mainly regulated through ubiquitination and lysosomal degradation. Caveolin-1 is degraded in the late endosome and lysosome. Generally speaking, the velocity of degradation is very slow. However, if caveolin-1 assembly is altered, the rate of decomposition is accelerated. The most likely explanation for this phenomenon is cholesterol consumption, which would inhibit the assembly of complete cytoskeletal proteins and cause caveolin-1 to be more easily decomposed (Hayer et al., 2010).

During both transcription and translation, caveolin-1 expression is influenced by a variety of factors (mainly multiple signal transduction pathways) which results in changes to cellular physiological processes. The following sections detail how caveolin-1 expression is regulated.

Significance of pre-transcriptional caveolin-1 regulation in pancreatic cancer

Pre-transcriptional regulation of caveolin-1 is mainly controlled by transcription factors and transcription-related factors.

Forkhead box (FOXO): FOXO transcription factors are of vital importance in cellular proliferation, metabolism, and apoptosis downstream of PTEN, phosphoinositide 3-kinase (PI3K), and AKT (Eijkelenboom et al., 2013). In thestationary phase of cell growth, FOXO induces stable expression of insulin receptors and regulates caveolin-1 through thePI3K/AKT/FOXO pathway (Boreddy et al., 2011). Vanden et al. found that active FOXO binds directly to the caveolin-1promoter regionand activates transcription (Van et al., 2005). In pancreatic cancer, Roy SK et al. found that inhibition of the PI3K/AKT and MAPK/extracellular regulated protein kinase (ERK) pathways activates FOXO transcription and caveolin-1 expression, leading to cell cycle arrest and apoptosis (Roy et al., 2010).

Lipopolysaccharide (LPS): LPS actswith NEMO [an essential modifier-binding domain of nuclear factor-kappa B (NF-κB)] to inhibit the formation of the IKK-γ and IKK complexes, thuspreventing the activation ofNF-κB (a family of transcription factors) and caveolin-1 expressionin vivo (Tiruppathi et al., 2008), NF-κB is a key

factor connecting inflammation with cancer progression. The idea that a tumor itself can act as a stimulator of chronic inflammation is becoming more widely accepted by oncologists (Zhu et al., 2008). Maier et al. found that NF-κB promotes epithelial-mesenchymal transition, migration, and invasion in pancreatic carcinoma cells (Maier et al., 2010). Ikebe et al. found that LPS promotes NF-κB activation and increases invasive ability through the TLR4/MyD88 signaling pathway (Ikebe et al., 2009).

High density lipoprotein (HDL): When NIH/3T3 cells areexposedto HDL, caveolin-1 promoter activity isinhibited. This phenomenon suggests that HDL hasa direct negative impact on caveolin-1 transcription. Further research revealed that HDL can downregulate caveolin-1 expression without affecting caveolin-2 expression by activating the MAP kinase pathway through ERK1/2 activation (Frank et al., 2001). We also know that a high-fat diet is a risk factor for pancreatic cancer and a high-fat diet increases caveolin-1 (Yang et al., 2007).

Stimulatory protein 1 (Sp1): Sp1 is one of the two transcription factors that bind thecaveolin-1 gene andaffect promoter activity (Chen et al., 2011). Sp1 is a central transcription factor that regulates a number of pathways critical to tumorigenesis, including tumor cell-cycle progression, apoptosis, angiogenesis, metastasis, and evasion of the immune system (Huang et al., 2012). Dasari et al. showed that oxidative stress enhances Sp1-stimulated caveolin-1 expression. In addition, other studies have shown that p38 MAPK is an oxidative stress-induced upstream regulatory factor of Sp1. Inhibition of p38 MAPK prevents oxidative stress from inducing Sp1-mediated caveolin-1 gene expression and premature cell aging (Dasari et al., 2006). Sp1 activation is also essential for the differential overexpression of vascular endothelial growth factor (VEGF), which is involved in pancreatic cancer angiogenesis and progression (Shi et al., 2001).

Estrogen receptorα (ERα): Estrogens are major promoters of cell proliferation in both normal and neoplastic epithelium. Two major ERs are ERα and Erβ (Kimbro et al., 2008). ERα acts as an estrogen transcription factor that stimulates estrogen target genes and regulates cell progression and growth, especially in breast epithelium (Singh et al., 2005). In neuroepithelioma cells, ectopic ERα inhibits caveolin-1 transcription and the caveolin-1 promoter is methylated (Zschocke et al., 2003). However, because caveolin-1 mutations occur in the early stages of mammary transformation, this observation suggested that caveolin-1 might be an upstream activator of Erα (Sotgia et al., 2006). There may be negative feedback regulation of caveolin-1 as the proliferation of pancreatic cancer cells is highly sensitive to estrogen in vitro (Konduri et al., 2007).

Significance of caveolin-1 transcription regulators in pancreatic cancer

p53: p53 is the strongest tumor suppressor gene and it regulates apoptosis, cell cycle arrest, and senescence (Lee et al., 2011). p53 binds directly to the caveolin-1 promoter with the E2F/DP-1 and Sp1 transcription factors, thusincreasing caveolin-1 expression (Lee et al., 2012). In human pancreatic ductal adenocarcinoma, low p53 transcript levelsareassociated with poor prognosis (Grochola et al., 2011). Much evidence indicates that p53 provokes a classic proapoptotic response by delaying G1-to-S progression (Gupta et al., 2010).

Cholesterol: Cholesterol is also a powerful regulator of gene expression. It carries out this activity by jointly binding the sterol regulatory element binding protein (SREBP) with Sp1. KLF11 [a Krüppel-like factor; also referred to as transforming growth factor-beta early inducible gene 2 (TIEG2)] inhibits Sp1/SREBP cholesterol-dependent gene expression (Llaverias et al., 2004). High cholesterol intake increases in the incidence of pancreatic cancer (Takeyama et al., 2005).

Carbon monoxide (CO): CO (a product of hemeoxygenase activity) is an endogenous gaseous transmitter that exerts anti-proliferative effects (Schwer et al., 2013). CO affects caveolin-1 gene expression by activating guanylatecyclase and p38 MAPK. p38 MAPK down-regulates ERKs that inhibit caveolin-1 gene transcription (Kim et al., 2005). In pancreatic cancer, CO protects cells from apoptosis. Protection is mediated through the generation of cyclic GMP (cGMP) and the activation of cGMP-dependent protein kinases and guanylatecyclase (Gunther et al., 2002).

Vascular endothelial growth factor (VEGF): VEGF is a key mediator of angiogenesis and promotes proliferation, survival, migration of endothelial cells, and blood vessel formation and neovascularization (Ferrara et al., 2002). On the one hand, Liu J et al. found that activation of the VEGF/MEK signal transduction pathway decreased caveolin-1 while leaving caveolin-2 unchanged inhuman umbilical vein endothelial cells (Liu et al., 1999). In prostate cancer cells, the protein kinase C/MEK/c-myc gene/androgen receptor pathway increases caveolin-1 (Wu et al., 2002). On the other hand, caveolin-1 also stimulates expression of VEGFvia AKT activation (Li et al., 2009). VEGF is a well-characterized mediator of tumor angiogenesis andfunctions primarily bybinding and activating the VEGF receptor 2. Angiogenesis is a characteristic of many malignant tumors, including pancreatic cancer (Dineen et al., 2008).

Epithelial membrane protein 2 (EMP2): EMP2, a tetraspan protein, facilitates plasma membrane delivery of certain integrins. EMP2 also contributes to the formation and trafficking of lipid rafts bearing glycosyl-phosphatidyl inositol anchored proteins (GPI-Aps), thus reducing caveolin-1 expression (Wadehra et al., 2004). Down-regulation of caveolin-1 by EMP2 does not affect caveolin-1 translational efficiency, phosphorylation, or degradation. Protein half-life analysis showed that caveolin-1 decomposition was more rapid when mediated by EMP2 (Forbes et al., 2007).

eNOS:The eNOS protein binds caveolin-1 through its CSD (Razani et al., 2002). Caveolin-1 also functions as an eNOS inhibitor with a calcium/calmodulin cofactor (Ju et al., 2002). Enhanced renal caveolin-1 expression is linked to poor eNOS expression (Valles et al., 2007). Increased caveolin-1 is associated with inhibition of the catalytic activity of eNOS (Venema et al., 1997). There may be a feedback regulator to caveolin-1. In the model of pancreatic cancer liver metastasis, eNOS overexpression

attenuates both the number and size of tumors. In vitro, NO promotes tumor cell anoikis and limits invasive capacity (Decker et al., 2008).

Reactive oxygen species (ROS): ROS areproduced by cellular aerobic metabolism (Gough et al., 2011). Cells treated with oxidation have increased tyrosine kinase activity and decreased phosphatase activity (Vepa et al., 1997). Rungtabnapa found that catalase and N-acetylcysteine promote the ubiquitination and degradation of caveolin-1. In addition, exogenous hydrogen peroxide prevents the formation of the caveolin-1-ubiquitin complex and inhibits caveolin-1 reduction (Rungtabnapa et al., 2011). Endogenous hydrogen peroxide also prevents the transport of newly synthesized caveolin-1 to the cell membrane. Palmitoylation of caveolin-1 is significantly inhibited in endothelial cells exposed to hydrogen peroxide (Parat et al., 2002). Park JH et al. found that hydrogen peroxide and methyl-beta-cyclodextrin down-regulate caveolin-1. In pancreatic cancer, NADPH oxidase 4-mediated generation of ROS is proposed to have anti-apoptotic activity and thus confer a growth advantage to cancer cells. ROS transmit cell survival signals through the AKT/ASK1 pathway and their depletion leads to apoptosis (Mochizuki et al., 2006).

Src kinase: Src family kinases regulate cell proliferation, adhesion, and motility. They are frequently activated in human cancers and contribute to malignancy and metastasis (Di et al., 2011). Src kinase increases as a function of tumor progression and plays a role in the transition to malignancy. Further, itis associated with phosphorylation of the caveolin-1 gene Y14. More than 60% of pancreatic cancer patients show increased c-Src activity, which is associated with poor prognosis (Shields et al., 2011). Src/Stat3 signaling plays a crucial role in tumor cell survival, proliferation, angiogenesis, and immune suppression (Nam et al., 2012).

Others: There are additional cellular signaling pathways, like transforming growth factor/PI3K, histone deacetylase, and cAMP that are associated with caveolin-down-regulation that are not mentioned here (Zschocke et al., 2005). In contrast, oxidized LDL increases caveolin-1 expression (Wu et al., 2009).

Post-transcriptional regulation

Post-transcriptional regulation of caveolin-1 mainly occurs through the decomposition process that is carried out through thelysosomal and ubiquitination degradation pathways.

Na+/K+-ATPase: High metabolism is a characteristic of malignancies and Na+/K+-ATPase provides energy for cellular metabolism. Na+/K+-ATPase is an important enzyme in the protein transport process. Cai T et al. found that caveolin-1 was significantly reduced on the cell surface when the Na+/K+-ATPase gene was knocked out. This is due to unilateral regulation of the transport process by Na+/K+-ATPase rather than an interaction between Na+/K+-ATPase and caveolin-1 (Cai et al., 2009).

Breast cancer susceptibility gene 1 (BRCA1): BRCA1 is involved in multiple processes, such as cell growth, apoptosis, DNA damage repair, and transcriptional activation. In immunofluorescence studies, Wang Y et al. showed that BRCA1 might inhibit the invasive and metastatic abilities of cancer cells by inducing the redistribution of caveolin-1. In addition, the BRCA1 gene inhibits redistribution of caveolin-1 in the cell membrane and cytoplasm (Wang et al., 2008). BRCA1 mutations have been shown to drastically decrease survival rate in breast and ovarian cancer patients who carry them. A number of studies have shown that the third most common cancer associated with these mutations is pancreatic cancer (Lynch et al., 2005).

Flotillin-1: Flotillins are localized to lipid rafts independent of caveolin-1 and are the principal proteins associate with lipid rafts. These microdomains function in roles such as membrane trafficking, cell morphogenesis,and cell signaling (Evans et al., 2003). Flotillin-1 regulates caveolin-1 levels by preventing its degradation in lysosomes (Vassilieva et al., 2009).

Relationship between Caveolin-1 and Pancreatic Cancer

Pancreatic cancer progression is attributed to genetic and epigenetic alterations and a chaotic tumor microenvironment (Huang et al., 2012). Recent studies suggest that caveolin-1 plays important roles in promoting cancer cell development, migration, invasion, and metastasis (Thomas et al., 2011). Further research also suggests that caveolin-1 can impact cancer biology both positively and negatively. In tumor tissue, both tumor cells and blood vessels express caveolin-1. However, in peritumoral tissue caveolin-1 is mainly expressed in blood vessels and only occasionally expressed in ductal or parenchymal cells. Overexpression of caveolin-1 is associated with tumor size, grade, stage, and increased serum levels of CA19-9 (Tanase et al., 2009).

Caveolin-1 has recently been identified as a tumor metastasis modifier gene that affects cancer cell motility (Koleske et al., 1995; Yang et al., 1999). In contrast, loss of caveolin-1 leads to RhoC-mediated migration and invasion in metastatic pancreatic cancer cells (Thomas et al., 2011). Data also indicates that caveolin-1, with its dual function in cancers, is associated with tumor progression and inhibits proliferation and invasion (Mathew et al., 2011). Tumor cells have significantly higher caveolin-1 levels, especially in the tumor stroma. Caveolin-1 knockdown significantly induces cell apoptosis and enhances the radio sensitivity of cancer cells (Hehlgans et al., 2009). The caveolin-1 gene inhibits invasion of pancreatic carcinoma cellslikely through the Erk/MMP signal pathway, however, the mechanism remains unclear. This suggests that endogenous expression or re-expression of caveolin-1 could act to reduce the potential invasivenessof cancer cells (Han et al., 2010). Together, these findings strongly imply that caveolin-1 plays a critical role in pancreatic cancer development and progression and is a valuable biomarker for the disease. The majority of normal and adjacent normal pancreatic tissue cells are negative for caveolin-1, whereas pancreatic cancer tissue cells and stromal cells are strongly positive for caveolin-1. Caveolin-1 expression is positively correlated with tumor differentiation, disease stage, and tumor metastasis.

Caveolin-1 is also an oncogene that could promote invasion. In summary, a variety of data indicates that caveolin-1 might be a good candidate for a prognostic tumor marker and a potential target for therapeutic intervention (Bailey et al., 2008).

Five year view

Caveolin-1 is involved in a variety of cellular signal pathways and transmembrane transport. It is generally accepted that signaling proteins are proposed to use conserved caveolin-binding motifs (CBMs) to associate with caveolae via CSD. However, Collins BM et al. found CBM/CSD-dependent interactions are unlikely to mediate caveolar signaling (Collins et al., 2012). Its precise role as a tumor suppressor or oncogene in different human malignancies remains elusive. Caveolin-1 regulates a variety of cellular events that include cellular transformation, tumorigenesis, cell metastasis, and angiogenesis. It is interesting to note that caveolin-1 is reduced in pancreatic cancer compared to normal tissue in precancerous tumors, such as pancreatic intraductal papillary-mucinous neoplasms (Terris et al., 2002). Caveolin-1 contributes to cellular resistance against genotoxic agents and thus its knockdown sensitizes human pancreatic tumor cells to ionizing radiation (Cordes et al., 2007). Based on this observation, caveolin-1 appears to be a tumor suppressor in pancreatic cancer. However, it is highly expressed in invasive tumors compared to noninvasive tumors (Terris et al., 2012). Huang C et al. found the FoxM1-caveolin signaling promotes pancreatic cancer invasion and metastasis (Huang et al., 2012). This review detailed the mechanisms that regulate caveolin-1 expression *in vivo* and their significance in pancreatic cancer. Other mechanisms are being actively explored. At present, a variety of factors, such as HDL, SREBP1, and epidermal growth factor receptor, that regulate the expression of caveolin-1 have been identified, but the precise regulatory mechanisms remain unclear. In-depth research in this field will improve our understanding of pancreatic cancer and potentially highlight novel diagnostic methods and anti-cancer strategies.

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References

Anderson RG (1998). The caveolae membrane system. *Annu Rev Biochem*, **67**, 199-225.

Arbuzova A, Wang L, Wang J, et al (2000). Membrane binding of peptides containing both basic and aromatic residues. Experimental studies with peptides corresponding to the scaffolding region of caveolin and the effector region of MARCKS. *Biochemistry-Us*, **39**, 10330-9.

Bailey KM, Liu J (2008). Caveolin-1 up-regulation during epithelial to mesenchymal transition is mediated by focal adhesion kinase. *J Biol Chem*, **283**, 13714-24.

Boreddy SR, Pramanik KC, Srivastava SK (2011). Pancreatic tumor suppression by benzyl isothiocyanate is associated with inhibition of PI3K/AKT/FOXO pathway. *Clin Cancer Res*, **17**, 1784-95.

Cai T, Wang H, Chen Y, et al (2008). Regulation of caveolin-1 membrane trafficking by the Na/K-ATPase. *J Cell Biol*, **182**, 1153-69.

Casley-Smith JR, Casley-Smith JR (1975). The fine structure of the blood capillaries of some endocrine glands of the hagfish, Eptatretus stouti: implications for the evolution of blood and lymph vessels. *Rev Suisse Zool*, **82**, 35-40.

Chen W, Chen Y, Qin L, et al (2011). Transcription factor Sp1 is essential for the regulation of the porcine caveolin-1 gene. *Dna Cell Biol*, **30**, 491-7.

Collins BM, Davis MJ, Hancock JF, et al (2012). Structure-based reassessment of the caveolin signaling model: do caveolae regulate signaling through caveolin-protein interactions? *Dev Cell*, **23**, 11-20.

Cordes N, Frick S, Brunner TB, et al (2007). Human pancreatic tumor cells are sensitized to ionizing radiation by knockdown of caveolin-1. *Oncogene*, **26**, 6851-62.

Couet J, Li S, Okamoto T, et al (1997). Identification of peptide and protein ligands for the caveolin-scaffolding domain. Implications for the interaction of caveolin with caveolae-associated proteins. *J Biol Chem*, **272**, 6525-33.

Dasari A, Bartholomew JN, Volonte D, et al (2006). Oxidative stress induces premature senescence by stimulating caveolin-1 gene transcription through p38 mitogen-activated protein kinase/Sp1-mediated activation of two GC-rich promoter elements. *Cancer Res*, **66**, 10805-14.

Decker NK, Abdelmoneim SS, Yaqoob U, et al (2008). Nitric oxide regulates tumor cell cross-talk with stromal cells in the tumor microenvironment of the liver. *Am J Pathol*, **173**, 1002-12.

Di Florio A, Adesso L, Pedrotti S, et al (2011). Src kinase activity coordinates cell adhesion and spreading with activation of mammalian target of rapamycin in pancreatic endocrine tumour cells. *Endocr Relat Cancer*, **18**, 541-54.

Diamantidis M, Tsapournas G, Kountouras J, et al (2008). New aspects of regulatory signaling pathways and novel therapies in pancreatic cancer. *Curr Mol Med*, **8**, 12-37.

Dietzen DJ, Hastings WR, Lublin DM (1995). Caveolin is palmitoylated on multiple cysteine residues. Palmitoylation is not necessary for localization of caveolin to caveolae. *J Biol Chem*, **270**, 6838-42.

Dineen SP, Sullivan LA, Beck AW, et al (2008). The Adnectin CT-322 is a novel VEGF receptor 2 inhibitor that decreases tumor burden in an orthotopic mouse model of pancreatic cancer. *BMC Cancer*, **8**, 352.

Eijkelenboom A, Burgering BM (2013). FOXOs: signalling integrators for homeostasis maintenance. *Nat Rev Mol Cell Biol*, **14**, 83-97.

Evans WT, Coyer RL, Sandusky MF, et al (2003). Characterization of membrane rafts isolated from rat sertoli cell cultures: caveolin and flotillin-1 content. *J Androl*, **24**, 812-21.

Ferrara N (2002). VEGF and the quest for tumour angiogenesis factors. *Nat Rev Cancer*, **2**, 795-803.

Forbes A, Wadehra M, Mareninov S, et al (2007). The tetraspan protein EMP2 regulates expression of caveolin-1. *J Biol Chem*, **282**, 26542-51.

Frank PG, Galbiati F, Volonte D, et al (2001). Influence of caveolin-1 on cellular cholesterol efflux mediated by high-density lipoproteins. *Am J Physiol Cell Physiol*, **280**, C1204-14.

Fujimoto T, Kogo H, Nomura R, et al (2000). Isoforms of

caveolin-1 and caveolar structure. *J Cell Sci*, **113**, 3509-17.
Glenney JJ (1989). Tyrosine phosphorylation of a 22-kDa protein is correlated with transformation by Rous sarcoma virus. *J Biol Chem*, **264**, 20163-6.
Gough DR, Cotter TG (2011). Hydrogen peroxide: a Jekyll and Hyde signalling molecule. *Cell Death Dis*, **2**, e213.
Grochola LF, Taubert H, Greither T, et al (2011). Elevated transcript levels from the MDM2 P1 promoter and low p53 transcript levels are associated with poor prognosis in human pancreatic ductal adenocarcinoma. *Pancreas*, **40**, 265-70.
Gunther L, Berberat PO, Haga M, et al (2002). Carbon monoxide protects pancreatic beta-cells from apoptosis and improves islet function/survival after transplantation. *Diabetes*, **51**, 994-9.
Gupta S, Sathishkumar S, Ahmed MM (2010). Influence of cell cycle checkpoints and p53 function on the toxicity of temozolomide in human pancreatic cancer cells. *Pancreatology*, **10**, 565-79.
Han F, Zhu HG (2010). Caveolin-1 regulating the invasion and expression of matrix metalloproteinase (MMPs) in pancreatic carcinoma cells. *J Surg Res*, **159**, 443-50.
Hayer A, Stoeber M, Ritz D, et al (2010). Caveolin-1 is ubiquitinated and targeted to intralumenal vesicles in endolysosomes for degradation. *J Cell Biol*, **191**, 615-29.
Hehlgans S, Eke I, Storch K, et al (2009). Caveolin-1 mediated radioresistance of 3D grown pancreatic cancer cells. *Radiother Oncol*, **92**, 362-70.
Huang C, Qiu Z, Wang L, et al (2012). A novel FoxM1-caveolin signaling pathway promotes pancreatic cancer invasion and metastasis. *Cancer Res*, **72**, 655-65.
Huang C, Xie K (2012). Crosstalk of Sp1 and Stat3 signaling in pancreatic cancer pathogenesis. *Cytokine Growth Factor Rev*, **23**, 25-35.
Ikebe M, Kitaura Y, Nakamura M, et al (2009). Lipopolysaccharide (LPS) increases the invasive ability of pancreatic cancer cells through the TLR4/MyD88 signaling pathway. *J Surg Oncol*, **100**, 725-31.
Ju H, Zou R, Venema VJ, et al (1997). Direct interaction of endothelial nitric-oxide synthase and caveolin-1 inhibits synthase activity. *J Biol Chem*, **272**, 18522-5.
Kato K, Hida Y, Miyamoto M, et al (2002). Overexpression of caveolin-1 in esophageal squamous cell carcinoma correlates with lymph node metastasis and pathologic stage. *Cancer*, **94**, 929-33.
Kato T, Miyamoto M, Kato K, et al (2004). Difference of caveolin-1 expression pattern in human lung neoplastic tissue. Atypical adenomatous hyperplasia, adenocarcinoma and squamous cell carcinoma. *Cancer Lett*, **214**, 121-8.
Kim HP, Wang X, Nakao A, et al (2005). Caveolin-1 expression by means of p38beta mitogen-activated protein kinase mediates the antiproliferative effect of carbon monoxide. *Proc Natl Acad Sci U S A*, **102**, 11319-24.
Kimbro KS, Duschene K, Willard M, et al (2008). A novel gene STYK1/NOK is upregulated in estrogen receptor-alpha negative estrogen receptor-beta positive breast cancer cells following estrogen treatment. *Mol Biol Rep*, **35**, 23-7.
Koleske AJ, Baltimore D, Lisanti MP (1995). Reduction of caveolin and caveolae in oncogenically transformed cells. *Proc Natl Acad Sci U S A*, **92**, 1381-5.
Konduri S, Schwarz RE (2007). Estrogen receptor beta/alpha ratio predicts response of pancreatic cancer cells to estrogens and phytoestrogens. *J Surg Res*, **140**, 55-66.
Krechler T, Horejs J, Ulrych J, et al (2011). Current status of pancreatic cancer diagnosis. *Cas Lek Cesk*, **150**, 587-93.
Lee KB, Byun HJ, Park SH, et al (2012). CYR61 controls p53 and NF-kappaB expression through PI3K/Akt/mTOR pathways in carboplatin-induced ovarian cancer cells. *Cancer Lett*, **315**, 86-95.
Lee SH, Park BJ (2011). p53 activation by blocking Snail: a novel pharmacological strategy for cancer. *Curr Pharm Des*, **17**, 610-17.
Li L, Ren C, Yang G, et al (2009). Caveolin-1 promotes autoregulatory, Akt-mediated induction of cancer-promoting growth factors in prostate cancer cells. *Mol Cancer Res*, **7**, 1781-91.
Li L, Yang G, Ebara S, et al (2001). Caveolin-1 mediates testosterone-stimulated survival/clonal growth and promotes metastatic activities in prostate cancer cells. *Cancer Res*, **61**, 4386-92.
Liu J, Razani B, Tang S, et al (1999). Angiogenesis activators and inhibitors differentially regulate caveolin-1 expression and caveolae formation in vascular endothelial cells. Angiogenesis inhibitors block vascular endothelial growth factor-induced down-regulation of caveolin-1. *J Biol Chem*, **274**, 15781-5.
Liu L, Xu HX, Wang WQ, et al (2013). Cavin-1is essential for the tumor-promoting effect of caveolin-1 and enhances itsprognostic potency in pancreatic cancer. *Oncogene*. doi: 10.1038/onc.2013.223.
Llaverias G, Vazquez-Carrera M, Sanchez RM, et al (2004). Rosiglitazone upregulates caveolin-1 expression in THP-1 cells through a PPAR-dependent mechanism. *J Lipid Res*, **45**, 2015-24.
Luo G, Long J, Zhang B, et al (2008). Stroma and pancreatic ductal adenocarcinoma: An interaction loop. *Biochim Biophys Acta*, **1826**, 170-8.
Lynch HT, Deters CA, Snyder CL, et al (2005). BRCA1 and pancreatic cancer: pedigree findings and their causal relationships. *Cancer Genet Cytogenet*, **158**, 119-25.
Maier HJ, Schmidt-Strassburger U, Huber MA, et al (2010). NF-kappaB promotes epithelial-mesenchymal transition, migration and invasion of pancreatic carcinoma cells. *Cancer Lett*, **295**, 214-28.
Mathew R (2011). Cell-specific dual role of caveolin-1 in pulmonary hypertension. *Pulm Med*, **2011**, 573432.
Mochizuki T, Furuta S, Mitsushita J, et al (2006). Inhibition of NADPH oxidase 4 activates apoptosis via the AKT/apoptosis signal-regulating kinase 1 pathway in pancreatic cancer PANC-1 cells. *Oncogene*, **25**, 3699-707.
Murakami S, Miyamoto M, Hida Y, et al (2003). Caveolin-I overexpression is a favourable prognostic factor for patients with extrahepatic bile duct carcinoma. *Br J Cancer*, **88**, 1234-8.
Nam S, Wen W, Schroeder A, et al (2012). Dual inhibition of Janus and Src family kinases by novel indirubin derivative blocks constitutively-activated Stat3 signaling associated with apoptosis of human pancreatic cancer cells. *Mol Oncol*, 7, 369-78.
Nimri L, Barak H, Graeve L, et al (2012). Restoration of caveolin-1 expression suppresses growth, membrane-type-4 metalloproteinase expression and metastasis-associated activities in colon cancer cells. *Mol Carcinog*. doi: 10.1002/mc.21927.
Okamoto T, Schlegel A, Scherer PE, et al (1998). Caveolins, a family of scaffolding proteins for organizing "preassembled signaling complexes" at the plasma membrane. *J Biol Chem*, **273**, 5419-22.
Parat MO, Fox PL (2004). Oxidative stress, caveolae and caveolin-1. *Subcell Biochem*, **37**, 425-41.
Parat MO, Stachowicz RZ, Fox PL (2002). Oxidative stress inhibits caveolin-1 palmitoylation and trafficking in endothelial cells. *Biochem J*, **361**, 681-8.
Prinetti A, Aureli M, Illuzzi G, et al (2010). GM3 synthase overexpression results in reduced cell motility and in

caveolin-1 upregulation in human ovarian carcinoma cells. *Glycobiology*, **20**, 62-77.

Rao X, Evans J, Chae H, et al (2012). CpG island shore methylation regulates caveolin-1 expression in breast cancer. *Oncogene*. doi: 10.1038/onc.2012.474.

Razani B, Woodman SE, Lisanti MP (2002). Caveolae: from cell biology to animal physiology. *Pharmacol Rev,* **54**, 431-67.

Roy SK, Srivastava RK, Shankar S (2010). Inhibition of PI3K/AKT and MAPK/ERK pathways causes activation of FOXO transcription factor, leading to cell cycle arrest and apoptosis in pancreatic cancer. *J Mol Signal*, **5**, 10.

Rungtabnapa P, Nimmannit U, Halim H, et al (2011). Hydrogen peroxide inhibits non-small cell lung cancer cell anoikis through the inhibition of caveolin-1 degradation. *Am J Physiol Cell Physiol*, **300**, C235-45.

Sargiacomo M, Sudol M, Tang Z, et al (1993). Signal transducing molecules and glycosyl-phosphatidylinositol-linked proteins form a caveolin-rich insoluble complex in MDCK cells. *J Cell Biol,* **122**, 789-807.

Schwer CI, Stoll P, Rospert S, et al (2013). Carbon monoxide releasing molecule-2 CORM-2 represses global protein synthesis by inhibition of eukaryotic elongation factor eEF2. *Int J Biochem Cell Biol*, **45**, 201-12.

Shi Q, Le X, Abbruzzese JL, et al (2001). Constitutive Sp1 activity is essential for differential constitutive expression of vascular endothelial growth factor in human pancreatic adenocarcinoma. *Cancer Res*, **61**, 4143-54.

Shi S, Yao W, Xu J, et al (2012). Combinational therapy: new hope for pancreatic cancer? *Cancer Lett*, **317**, 127-35.

Shields DJ, Murphy EA, Desgrosellier JS, et al (2011). Oncogenic Ras/Src cooperativity in pancreatic neoplasia. *Oncogene*, **30**, 2123-34.

Siegel R, DeSantis C, Virgo K, et al (2012). Cancer treatment and survivorship statistics. *CA Cancer J Clin*, **62**, 220-41.

Simpkins SA, Hanby AM, Holliday DL, et al (2012). Clinical and functional significance of loss of caveolin-1 expression in breast cancer-associated fibroblasts. *J Pathol*, **227**, 490-8.

Singh RR, Kumar R (2005). Steroid hormone receptor signaling in tumorigenesis. *J Cell Biochem*, **96**, 490-505.

Smart EJ, Graf GA, McNiven MA, et al (1999). Caveolins, liquid-ordered domains, and signal transduction. *Mol Cell Biol*, **19**, 7289-304.

Smart EJ, Ying YS, Conrad PA, et al (1994). Caveolin moves from caveolae to the Golgi apparatus in response to cholesterol oxidation. *J Cell Biol*, **127**, 1185-97.

Sotgia F, Rui H, Bonuccelli G, et al (2006). Caveolin-1, mammary stem cells, and estrogen-dependent breast cancers. *Cancer Res*, **66**, 10647-51.

Sowa G, Pypaert M, Fulton D, et al (2003). The phosphorylation of caveolin-2 on serines 23 and 36 modulates caveolin-1-dependent caveolae formation. *Proc Natl Acad Sci U S A*, **100**, 6511-6.

Steffens S, Schrader AJ, Blasig H, et al (2011). Caveolin 1 protein expression in renal cell carcinoma predicts survival. *BMC Urol*, **11**, 25.

Takeyama Y (2005). Dietary intake as a risk factor for pancreatic cancer in Japan: high cholesterol and low vitamin C diet. *J Gastroenterol*, **40**, 324-5.

Tanase CP, Dima S, Mihai M, et al (2009). Caveolin-1 overexpression correlates with tumour progression markers in pancreatic ductal adenocarcinoma. *J Mol Histol*, **40**, 23-9.

Terris B, Blaveri E, Crnogorac-Jurcevic T, et al (2002). Characterization of gene expression profiles in intraductal papillary-mucinous tumors of the pancreas. *Am J Pathol*, **160**, 1745-54.

Thomas S, Overdevest JB, Nitz MD, et al (2011). Src and caveolin-1 reciprocally regulate metastasis via a common downstream signaling pathway in bladder cancer. *Cancer Res*, **71**, 832-41.

Tiruppathi C, Shimizu J, Miyawaki-Shimizu K, et al (2008). Role of NF-kappaB-dependent caveolin-1 expression in the mechanism of increased endothelial permeability induced by lipopolysaccharide. *J Biol Chem*, **283**, 4210-8.

Valles PG, Manucha W, Carrizo L, et al (2007). Renal caveolin-1 expression in children with unilateral ureteropelvic junction obstruction. *Pediatr Nephrol*, **22**, 237-48.

van den Heuvel AP, Schulze A, Burgering BM (2005). Direct control of caveolin-1 expression by FOXO transcription factors. *Biochem J*, **385**, 795-802.

Vassilieva EV, Ivanov AI, Nusrat A (2009). Flotillin-1 stabilizes caveolin-1 in intestinal epithelial cells. *Biochem Biophys Res Commun*, **379**, 460-5.

Venema VJ, Zou R, Ju H, et al (1997). Caveolin-1 detergent solubility and association with endothelial nitric oxide synthase is modulated by tyrosine phosphorylation. *Biochem Biophys Res Commun*, **236**, 155-61.

Vepa S, Scribner WM, Natarajan V (1997). Activation of protein phosphorylation by oxidants in vascular endothelial cells: identification of tyrosine phosphorylation of caveolin. *Free Radic Biol Med*, **22**, 25-35.

Wadehra M, Goodglick L, Braun J (2004). The tetraspan protein EMP2 modulates the surface expression of caveolins and glycosylphosphatidyl inositol-linked proteins. *Mol Biol Cell*, **15**, 2073-83.

Wang Y, Yu J, Zhan Q (2008). BRCA1 regulates caveolin-1 expression and inhibits cell invasiveness. *Biochem Biophys Res Commun*, **370**, 201-6.

Wu CC, Wang SH, Kuan II, et al (2009). OxLDL upregulates caveolin-1 expression in macrophages: Role for caveolin-1 in the adhesion of oxLDL-treated macrophages to endothelium. *J Cell Biochem*, **107**, 460-72.

Wu D, Terrian DM (2002). Regulation of caveolin-1 expression and secretion by a protein kinase cepsilon signaling pathway in human prostate cancer cells. *J Biol Chem*, **277**, 40449-55.

Yang G, Truong LD, Wheeler TM, et al (1999). Caveolin-1 expression in clinically confined human prostate cancer: a novel prognostic marker. *Cancer Res*, **59**, 5719-23.

Yang N, Ying C, Xu M, et al (2007). High-fat diet up-regulates caveolin-1 expression in aorta of diet-induced obese but not in diet-resistant rats. *Cardiovasc Res,* **76**, 167-74.

Zhu XD, Zhang JB, Zhuang PY, et al (2008). High expression of macrophage colony-stimulating factor in peritumoral liver tissue is associated with poor survival after curative resection of hepatocellular carcinoma. *J Clin Oncol*, **26**, 2707-16.

Zschocke J, Bayatti N, Behl C (2005). Caveolin and GLT-1 gene expression is reciprocally regulated in primary astrocytes: association of GLT-1 with non-caveolar lipid rafts. *Glia*, **49**, 275-87.

Zschocke J, Manthey D, Bayatti N, et al (2003). Functional interaction of estrogen receptor alpha and caveolin isoforms in neuronal SK-N-MC cells. *J Steroid Biochem Mol Biol*, **84**, 167-70.