Polymerase-1 and transcript release factor (CAVIN1) and cancer: where we are and what is next

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Abstract

Polymerase I and transcript release factor (PTRF, also known as CAVIN1) is a unique molecule that has been ascribed with two very different functions; RNA transcription termination and caveolae formation. The role of CAVIN1 in cancer has recently gained researchers' attention and to-date, studies have shown that CAVIN1 may play a role in various malignancies, including prostate, breast and lung, where the expression of CAVIN1 is often detected to be dysregulated. Recent studies have begun to elucidate the underlying molecular and epigenetic signaling that may be associated with the dysregulation of CAVIN1. Here, we summarize the role of CAVIN1 in cancer and discuss possible future research directions in this area. The findings suggest that targeting CAVIN1 may be a potential therapeutic target for several cancers.

Keywords: PTRF, CAVIN1, cancer, epigenetic, signaling

Introduction

Polymerase I and transcript release factor (PTRF, also known as CAVIN1) was first cloned and characterized in 1998 by Jansa and colleagues (1). Recently, it has been proposed by the Human Genome Organization Nomenclature Committee (HGNC) that the official gene name of *PTRF* should be renamed to caveolae associated protein 1 (*CAVIN1*). Therefore, *PTRF* will be referred to as *CAVIN1* in this review.

Successful cloning of *CAVIN1* revealed a 392 amino acid structure that predicts a 44kDa molecular weight protein (1). However, CAVIN1 was detected at 50–60kDa, suggesting post-translational modifications such as phosphorylation, SUMOylation and ubiquitination may be involved in CAVIN1 modification (2). This was supported by the findings that confirm CAVIN1 has several phosphorylation sites (3).

Interestingly, CAVIN1 has been ascribed with very different roles. Initially, CAVIN1 was reported to be involved in transcription termination where it interacts with Transcription Termination Factor-I and also RNA Polymerase I to halt the transcription machinery (1, 4). Thereafter, CAVIN1 was reported to play a critical role in caveola formation at the cell membrane and this was demonstrated across an array of *in vitro* models (5, 6). Expression of CAVIN1 is detected in many tissues and is highly expressed in adipocytes, smooth, cardiac and skeletal muscles and osteoblasts (7). However, CAVIN1 is undetectable in neuronal tissues (7).

In addition to CAVIN1, there are 3 other caveolae associated proteins that to-date, have been characterized and described. They are, caveolae associated protein 2 (CAVIN2, also known as serum deprivation protein response, SDPR), caveolae associated protein 3 (CAVIN3, also known as sdr related gene product that binds to c-kinase, SRBC) and caveolae associated protein 4 (CAVIN4, also known as muscle restricted coiled-coil protein MURC) (8). CAVIN2 has been reported to be involved in inducing the change in caveola morphology that results in the formation of elongated shape caveolae (9). CAVIN3 has been shown to be coupled with caveolin during caveolae budding to form cavicles (vesicles of caveolae) that move along the microtubules to other areas in the cell (10). The expression of CAVIN4 is confined to muscle tissues and is associated with cardiac dysfunction through the Rho/ROCK signaling pathway (10, 11). However, among the four members, only CAVIN1 has polybasic signals, which are reported to be nuclear localization signals (1). Therefore, it appears that CAVIN1 is the only family member that binds transcription factor (12). Additionally, CAVIN1 is the only homolog that is able to trigger caveola formation (13, 14).

The role of CAVIN1 in health and diseases were recently reviewed (15). Since the discovery and characterization of CAVIN1, growing evidence has indicated the important role of this protein in tumorigenesis. Here, we will summarize the evidence that CAVIN1 could be a potential target in cancer.

Breast cancer

CAVIN1 is expressed in normal breast cells but expression is lost in cancer while caveolin 1 (CAV1) is increased (16, 17). CAVIN1 has been described to play a role in inhibiting membrane tubule formation in breast cancer cells (18). Ectopic expression of CAVIN1 in a breast cancer cell line blocked the formation of membrane

tubules and reduced the ability of CAV1, a molecule that is also essential for caveola formation, to form tubules (18). Therefore, the loss of CAVIN1 may explain the increased tendency of re-expressed CAV1 to form tubules in breast cancer cells (18).

Epigenetic mechanisms may play a role in down-regulation of *CAVIN1* in breast cancer (16). In breast cancer cell lines that do not demonstrate CAVIN1 expression, treatment with 5'aza-deoxycytidine resulted in re-expression of CAVIN1 (16). This suggests the association of promoter hypermethylation in down-regulating CAVIN1 in breast cancer (16). CAVIN1 may also be involved in multi-drug resistance in breast cancer (19). In the breast cancer cell line, MCF7, which is resistant to adriamycin (MCF7/ADR), there is an up-regulation of *CAVIN1* and knocking down *CAVIN1*, renders MCF7/ADR cells more susceptible to adriamycin, suggesting a loss of *CAVIN1* confers chemosensitivity in breast cancer cells (19).

Prostate cancer

Several studies have demonstrated the importance of CAVIN1 in prostate cancer. Firstly, Hill *et al* (2008) reported that the expression of *CAVIN1* was down-regulated in prostate cancer PC3 cells and that *CAVIN1* is essential for caveola formation in this organ (6). Then, Gould *et al* (2010) described the changes in CAVIN1 expression in a panel of cell lines and clinical tissues and showed that the expression of CAVIN1 is lost in prostate cancer (20). When CAVIN1 is over-expressed in PC3 cells, this impairs the migratory capacity of these cells. The impaired migration has been demonstrated to be an effect of down-regulation of matrix metalloproteinase-9 (MMP9) related to the over-expressing of CAVIN1 (14).

Furthermore, it has been shown that CAVIN1 expression selectively impairs the recruitment of actin cytoskeletal proteins to the detergent-resistant membrane, resulting in altered cholesterol distribution within the prostate cancer cells (21). This suggests that CAVIN1 may modulate the membrane microenvironment to accentuate secretion pathways, possibly at the step of endoplasmic reticulum sorting/exit (21). Additionally, CAVIN1 has been shown to neutralize the effect of CAV1 that resides outside of caveolae. This was shown by the reduction of anchorage independent growth which occurs when CAVIN1 and CAV1 are co-expressed in prostate cancer cells (22). On the other hand, CAVIN1 reduces prostate cancer cell migration through reduction of focal adhesion kinase (FAK) stabilization within focal adhesions. However, this effect is reversed when exogenous Galectin-3 is introduced into CAVIN1 expressing prostate cancer cells, where stabilization of FAK occurs, as observed in focal adhesions (23). Importantly, CAVIN1 is reported to decrease angiogenesis and lymph-angiogenesis in prostate cancer in both *in vitro* and *ex vivo* models (24).

Collectively, these discoveries highlight the importance of CAVIN1 in inhibiting tumor progression and metastasis by reducing the aggressive phenotypes of prostate cancer cells. Dysregulation of CAVIN1 occurs in prostate cancer and loss of expression may drive prostate tumorigenesis. Re-expression of, or preventing loss of CAVIN1 could be a potential therapeutic target in prostate cancer.

Lung cancer

CAVIN1 has been shown to be differentially expressed between normal human bronchial epithelial lung cells and tumorigenic human bronchial epithelial lung

cells (25). Using fresh frozen tissues, CAVIN1 was shown to be expressed in normal lung tissue, but no expression of CAVIN1 in non-small cell lung tumor specimens was identified (26). Pathway analysis revealed CAVIN1 is associated with EGFR signaling and co-immunoprecipitation confirmed the interaction between CAVIN1 and EGFR in lung cancer suggesting the importance of CAVIN1 in the ERBB/ HER signaling pathway (27). Recently, microRNA-187 was shown to directly target CAVIN1 in lung cancer and contribute to epithelial-mesenchymal transition, cell growth and invasion (28, 29).

Other cancers

Apart from the 3 main cancers discussed above. CAVIN1 is also implicated in an array of cancers suggesting the rising importance of CAVIN1 in malignancies other than breast, prostate and lung (16, 22, 27). In pancreatic cancer, the tumor promoting role of CAVIN1 is dependent on the expression of CAV1 where the combination of CAVIN1 and CAV1 predicts for poor survival outcome (30). In colon cancer, CAVIN1 is reported to negatively regulate the tumorigenic activities of colorectal cell lines (31). CAVIN1 suppresses the AKT/mTOR pathway, through decreasing phosphorylation of AKT, mTOR, and expression of their downstream target MMP9 (31).The expression of CAVIN1 and CAV1 are up-regulated in the recurring glioblastoma and knock down of CAVIN1 in glioblastoma cell lines increases chemosensitivity of cells to chemo-therapeutics (32). Interestingly, co-expression of CAV1 with CAVIN1 is essential for aggressive phenotypes in rhabdomyosarcoma (33).

Studies are beginning to investigate the regulation of CAVIN1 expression in cancer. From an epigenetic perspective, differential hypermethylation of CpG islands (cytosine and guanine rich region in the gene promoter) in CAVIN1 in Ewing sarcoma cells correlates with transcriptional silencing. Re-expression of CAVIN1 reduces the aggressive phenotype of Ewing sarcoma cells (34). Additionally, CAVIN1 is a direct target gene of miRNA-217 in cutaneous squamous cell carcinoma. This was demonstrated by the down-regulated expression of CAVIN1 in clinical tissues and this decrease is associated with a significant inverse correlation between miRNA-217 and CAVIN1 expression. Furthermore, over-expression of CAVIN1 inhibits the oncogeneic effect of miRNA-217's in cutaneous squamous cell carcinoma cell line. Taken together, these findings suggest that CAVIN1 may be a target that is frequently dysregulated in tumorigenesis.

Perspective

It is intriguing that a molecule can play two very different roles in a cell. CAVIN1 is such a molecule: it is involved in RNA transcription termination that takes place in the nucleus and caveola formation at the plasma membrane (1, 4-6). The role of CAVIN1 in tumorigenesis has attracted researchers' attention and efforts have been put in to decipher the underlying mechanisms in order to exploit CAVIN1 as a therapeutic target. **Figure 1** summarizes the molecular mechanisms of CAVIN1 that have been deciphered to date in the context of tumorigenesis.

The generation of the *CAVIN1* knockout mouse has provided a useful tool for researchers to understand the role of CAVIN1 in health and disease development. Although *CAVIN1* knockout mouse demonstrate phenotypes that are similar to those seen in humans e.g. congenital lipodystrophy phenotypes, glucose

intolerance and hyperinsulinemia (35), the mice do not spontaneously develop cancer. This greatly limits the understanding of the role of CAVIN1 in this *in vivo* model. Although cancer cells have been successfully grafted into an *in vivo* model and several CAVIN1 prostate cancer xenograft models have been established (14, 22, 24), pushing CAVIN1 towards a more translational model remains a challenge. Perhaps the next step to take would be to establish patient-derived xenograft models to elucidate the effect of existing or novel therapeutics on CAVIN1 positive tumors.

Seeing that CAVIN1 is implicated in a variety of cancers, the use of CAVIN1 as a cancer biomarker seems to be legitimate. However, challenges remain as most of the cancers (especially prostate) experience a loss of CAVIN1 rather than up-regulated CAVIN1 expression. Therefore, the use of CAVIN1 as a biomarker in cancer may be less than ideal. Accumulating evidence suggests that CAVIN1 may be regulated by epigenetic mechanisms (16, 28, 29). Therefore, it is possible that epigenetic reversing drugs could be use as a treatment modality for cancer patients who are negative for CAVIN1. Furthermore, specific micro-RNAs that target CAVIN1 are possible areas to explore as cancer biomarkers.

Conclusion

CAVIN1 is a unique molecule that is ascribed with two different roles: RNA transcription termination and caveola formation at the plasma membrane. Although the molecular mechanism of CAVIN1 has been deciphered, a better understanding of the value of CAVIN1 in the translational setting will allow better exploitation of this molecule in cancer therapeutics.

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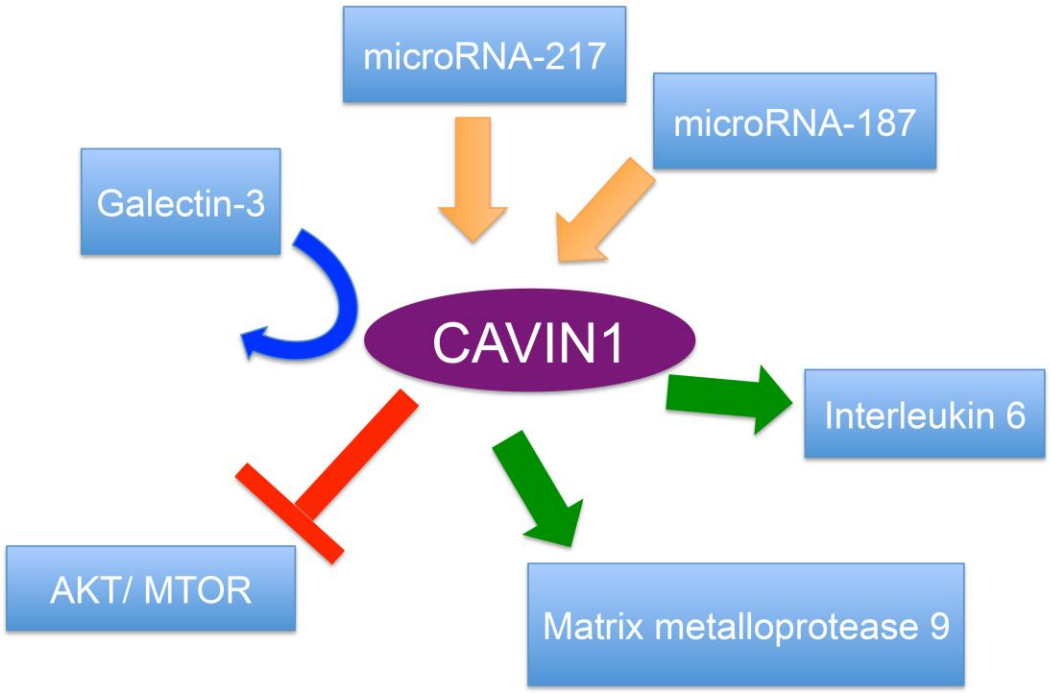

Figure 1: Summary of the molecular and epigenetic mechanisms that are associated with CAVIN1. Orange arrows indicate the molecules that regulate CAVIN1 expression. Green arrows indicate molecules that are down-stream of CAVIN1. The AKT/MTOR pathway is inhibited by CAVIN1. Galectin-3 has been shown to negate the effect of CAVIN1 in promoting prostate cancer cell motility.

