Caveolin-1: A multifunctional regulator of cancer cell proliferation and survival

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Background and rationale

Caveolin-1 is an essential protein constituent of plasma membrane caveolae - non-clathrin-coated, flask-shaped invaginations of the plasma membrane. Caveolin-1 is a principal component of the caveolar coat and a regulator of caveolae-dependent signaling and endocytosis. In addition, caveolin-1 exhibits an unusual ability to interact with and modulate multiple signaling pathways, suggesting that its expression is likely to profoundly affect cell function and cell fate. The expression of caveolin-1 is tightly controlled: it is up regulated in terminally differentiated epithelial cells and, conversely, down regulated upon oncogenic transformation. In addition, heterologous expression of caveolin-1 inhibits mitogenic signaling and abrogates anchorage-independent growth of cancer cells, while antisense suppression of caveolin-1 expression leads to fibroblast transformation. Finally, caveolin-1-null mice exhibit tissue-specific hyperplasia and increased sensitivity to oncogene- and carcinogen-induced tumorigenesis, in the mammary gland and skin, respectively. These results led to the suggestion that caveolin-1 is a growth-inhibitory protein that may act as a tumor-suppressor. However, this idea is inconsistent with the fact that caveolin-1 is highly expressed in many cancer cell lines. This was initially demonstrated by us in human multidrug resistant cancer cells (Lavie et al., 1998) and by others in mouse metastatic prostate cancer cells. A large body of data that has accumulated in recent years reveals that in many forms of cancer caveolin-1 expression is up-regulated. Furthermore, the expression of caveolin-1 is positively correlated with the tumor cell grade and its progression stage and, in some cases, the expression of caveolin-1 is an independent predictor of poor disease prognosis (Liscovitch et al., 2005; Shatz and Liscovitch, 2008).

These data highlight an important question: Why a putative tumor suppressor protein like caveolin-1 is highly expressed in so many cancer cells? One possibility is that in such cancer cells caveolin-1 promotes cell survival. Indeed, the ability of caveolin-1 to effect both growth-inhibitory and survival-promoting actions may explain its divergent expression in early vs. advanced stage human cancers (Fig. 1; Liscovitch et al., 2005). Therefore, the main focus of our current research is to elucidate the function(s) of caveolin-1 in human cancer cell lines and to examine the hypothesis that its expression in advanced stage, multidrug resistant and/or metastatic cancer is related to its pro-survival actions.

Summary of current research

To study the role of caveolin-1 in human cancer cells we have taken two approaches: (i) Overexpression of caveolin-1 in caveolin-negative cells and, conversely, (ii) gene-specific suppression of caveolin-1 expression or function in caveolin-positive cells. We have shown recently that stable expression of caveolin-1 in MCF-7 cells results in attenuation of cell proliferation, inhibition of matrix invasiveness and abrogation of anchorage-independent growth (Fiucci et al., 2002). Surprisingly, further studies revealed that expression of caveolin-1 results in inhibition of anoikis (detachment-induced apoptosis), indicating that caveolin-1 promotes matrix-independent survival. Caveolin-1 expression also prevents detachment-induced activation of p53. Our data indicate that caveolin-1 enhances matrix-independent cell survival by a mechanism that could be mediated by up-regulation of IGF-I receptor expression and signaling (Ravid et al. 2005). Furthermore, we have identified filamin A is a novel, caveolin-1-dependent target of IGF-I. Filamin A is an actin filament cross-linking protein that has been implicated in dynamic remodeling of the actin cytoskeletal network during cell migration and survival. These results raise the attractive possibility that caveolin-1 mediates a novel signaling pathway, leading from cell-surface receptors to filamin A-mediated regulation of cytoskeletal-dependent changes in cancer cell migration and survival (D. Ravid, submitted).

We have also tested the effect(s) of RNAi-mediated suppression and over-expression of dominant negative caveolin-1 mutants (Cav1-P132L and Cav1-Y14F) in human H1299 non-small-cell lung cancer cells that express high levels of endogenous caveolin-1. Our results indicate that RNAi-mediated suppression of caveolin-1 inhibits clonogenic growth of H1299 cells. As clonal growth of cancer cells is largely dependent on autocrine growth/survival factors, the results suggest that caveolin-1 may positively regulate cell response to an autocrine factor that remains to be identified. In contrast, stable expression of Cav1-P132L and Cav1-Y14F inhibit cell migration and invasion in H1299 cells (M. Shatz, submitted).

Caveolin-1 is up regulated in numerous human MDR cancer cells (Lavie et al., 1998). The pro-survival actions of caveolin-1 may enhance the ability of MDR cells to withstand genotoxic and/or oxidative stress. In this context, we have recently shown that specific activators of peroxisome proliferator-activated receptor (PPAR)-γ up-regulate caveolin-1 mRNA and protein expression in human cancer cells (Burgermeister et al., 2003). More recently we have shown that PPARγ activation increases cellular resistance to oxidative stress and to doxorubicin. The up-regulation of caveolin-1 by PPARγ ligands involves both PPARγ-dependent and PPARγ-response element (PPRE)-independent mechanisms and it occurs via a complex pathway that includes trans-activation of the EGF receptor and EGFR-dependent signaling pathways (Tencer et al., 2008).

Finally, we designed a novel, general and simple 'reverse chemical genetic' approach for generation of protein alleles that can be regulated by a small-molecule drug. The new procedure involves insertion into a given protein of a chemical-genetic 'switch' which consists of a genetically-encoded peptide that binds a small-molecule ligand with high affinity. The insertion position is selected empirically to confer ligand-dependent modulation of the mutant protein's activity (Fig. 2). We demonstrated the feasibility of the new method by its application to the TEM-1 β-lactamase antibiotics resistance gene. We generated multiple ligand-sensitive mutants of TEM-1, two of which are inhibited by the ligand, whereas a third is stimulated by the ligand. Our results suggest that the method may be applied to any protein given an appropriate activity assay. Our method is currently being implemented on a novel cancer-related MAP kinase (ERK8) whose cellular function is unknown. In general, 'regulatable' mutant alleles could be utilized to determine the effect(s) of drug-dependent activation or inhibition of the protein on cancer-related cellular phenotypes and help establish its role in cell function. This method will therefore be used to determine the role played by selected proteins, including caveolin-1, in mediating the phenotypic changes that are associated with cancer cell progression to a full-fledged malignant state (Erster et al., 2007).

Fig. 1 *Dynamic changes in caveolin-1 expression during cell differentiation, tumor initiation and tumor progression*

Fig. 2 *Ligand Interaction Scan: Insertion of a chemical-genetic 'switch' into a given protein to generate 'regulatable' protein alleles.*

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