

## Molecular & Cellular Oncology

ISSN: (Print) 2372-3556 (Online) Journal homepage: <http://www.tandfonline.com/loi/kmco20>

# The endoplasmic reticulum may be an Achilles' heel of cancer cells that have undergone an epithelial-to-mesenchymal transition

Yu-xiong Feng, Ethan S Sokol & Piyush B Gupta

To cite this article: Yu-xiong Feng, Ethan S Sokol & Piyush B Gupta (2014) The endoplasmic reticulum may be an Achilles' heel of cancer cells that have undergone an epithelial-to-mesenchymal transition, *Molecular & Cellular Oncology*, 1:2, e961822, DOI: 10.4161/23723548.2014.961822

To link to this article: <https://doi.org/10.4161/23723548.2014.961822>



© 2014 The Author(s). Published with license by Taylor & Francis Group, LLC © Yu-xiong Feng, Ethan S Sokol, and Piyush B Gupta



Accepted author version posted online: 29 Oct 2014.  
Published online: 29 Oct 2014.



Submit your article to this journal [↗](#)



Article views: 335



View related articles [↗](#)



View Crossmark data [↗](#)



Citing articles: 2 View citing articles [↗](#)

# The endoplasmic reticulum may be an Achilles' heel of cancer cells that have undergone an epithelial-to-mesenchymal transition

Yu-xiong Feng<sup>1</sup>, Ethan S Sokol<sup>1,2</sup>, and Piyush B Gupta<sup>1,2,3,4,5,\*</sup>

<sup>1</sup>Whitehead Institute for Biomedical Research, 9 Cambridge Center; Cambridge, MA USA; <sup>2</sup>Department of Biology, Massachusetts Institute of Technology, Cambridge, MA USA; <sup>3</sup>Koch Institute for Integrative Cancer Research, Cambridge, MA USA; <sup>4</sup>Harvard Stem Cell Institute, Cambridge, MA USA; <sup>5</sup>Broad Institute, Cambridge, MA USA

**Keywords:** EMT, ER stress, UPR

**Abbreviations:** ECM, extracellular matrix; EMT, Epithelial-to-mesenchymal transition; ER, endoplasmic reticulum; UPR, unfolded protein response.

In a recent report published in *Cancer Discovery* we identified a novel vulnerability of cancer cells that have undergone an epithelial–mesenchymal transition (EMT) and established that the PERK branch of the unfolded protein response is constitutively activated upon EMT. In this commentary, we summarize and provide context for our findings.

Scientists have known for decades that cancer cells can become invasive and metastatic by undergoing an epithelial-to-mesenchymal transition (EMT).<sup>1</sup> More recently, researchers have discovered that the EMT also confers resistance to radiation and a wide spectrum of chemotherapy drugs, including DNA-damaging agents and targeted inhibitors of specific kinases.<sup>2,3</sup> Moreover, cancer cells that undergo an EMT are, in many cases, functionally indistinguishable from cancer stem-like cells.<sup>4,5</sup> These observations have revealed that, by merely changing their differentiation state, cancer cells can gain the key malignant traits responsible for most cancer-related deaths.

In light of this surprising fact, there is significant interest in finding ways to treat tumors by targeting the EMT. One major focus has been to delineate the ligands, receptors, and downstream signaling proteins that, when activated, induce cells to undergo an EMT; inhibiting these factors could suppress tumor progression by either preventing cancer cells from undergoing an EMT, or by reversing EMT in cells that have already undergone the

transition.<sup>6</sup> A second area of focus has been to delineate the molecular mechanisms by which the EMT causes cells to acquire either invasiveness or drug resistance;<sup>7</sup> while it would not prevent or reverse EMT, inhibiting these mechanisms could provide a therapeutic benefit by suppressing the malignancy of cells that have undergone an EMT.

A more direct approach to targeting the EMT would be to search for agents that are selectively lethal to cells that have undergone an EMT. In a high-throughput screen of over 300,000 compounds, we succeeded in identifying a few small molecules with strong EMT-selective toxicity.<sup>8</sup> The discovery of these EMT-selective compounds was not a foregone conclusion because EMT cells were resistant to all test compounds they had been exposed to before the actual screen, and suggested that such agents were exploiting unique vulnerabilities acquired by cells upon EMT. At the time, however, we did not know what any of these vulnerabilities actually were.

In our recent publication, we show that 2 of the EMT-selective compounds identified in the above chemical screen

selectively activate endoplasmic reticulum (ER) stress pathways – collectively termed the unfolded protein response (UPR). By contrast, closely related structural variants of these compounds that are not toxic to EMT cells do not activate ER stress signaling. We further show that EMT sensitizes cells to 4 molecules that are established perturbagens of ER function and to reductions in expression of the ER chaperone BiP. Taken together, our findings identify the first known vulnerability of EMT cells: sensitivity to agents that perturb the function of the ER (Fig. 1).<sup>9</sup>

This Achilles' heel of cells that have undergone EMT appears to be a consequence of physiological changes that occur in cells when they migrate and invade. Invading cells remodel the extracellular matrix (ECM). Upon EMT, cells significantly upregulate the synthesis and secretion of pro-migratory ECM components; this, in turn, significantly increases the protein load within their ER. In our recent publication we show that the increased sensitivity of EMT cells to ER stress is a consequence of this increased ER load, since inhibiting ECM synthesis reduces

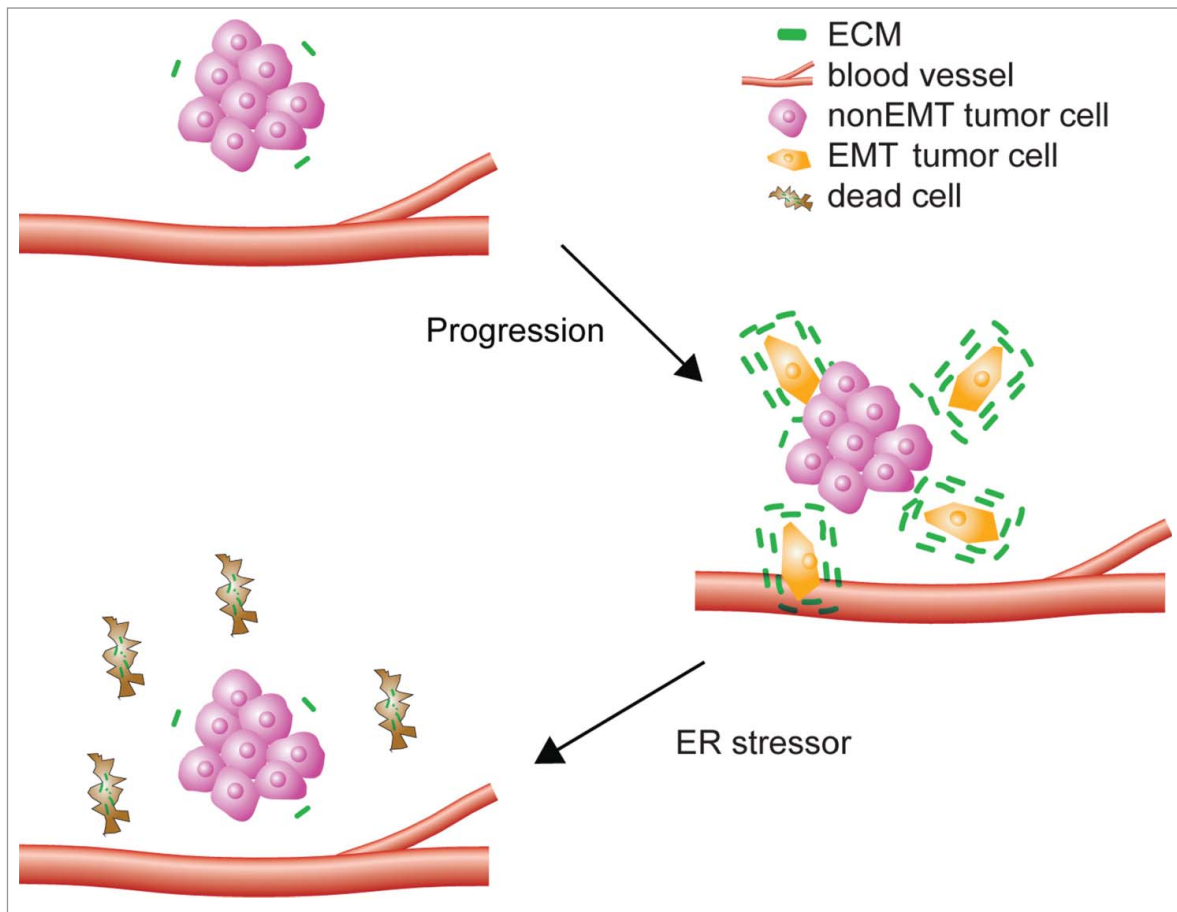
© Yu-xiong Feng, Ethan S Sokol, and Piyush B Gupta

\*Correspondence to: Piyush B Gupta; Email: pgupta@wi.mit.edu

Submitted: 07/23/2014; Revised: 07/28/2014; Accepted: 07/29/2014

<http://dx.doi.org/10.4161/23723548.2014.961822>

This is an Open Access article distributed under the terms of the Creative Commons Attribution-Non-Commercial License (<http://creativecommons.org/licenses/by-nc/3.0/>), which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited. The moral rights of the named author(s) have been asserted.



**Figure 1.** Epithelial-to-mesenchymal transition sensitizes cancer cells to endoplasmic reticulum stress.

the sensitivity of EMT cells to ER stress. Inhibiting ECM synthesis, however, also prevents EMT cells from migrating. Because ECM synthesis is essential for invasion, our findings suggest that increased sensitivity to ER stress may be a general feature of metastatic cancer cells.

Because some highly secretory cell types appear to selectively utilize UPR pathways,<sup>10</sup> we examined whether there was any evidence of this occurring upon EMT. We found that the highly secretory EMT cells specifically activate the PERK branch of the UPR at low levels, even in the absence of any treatment with exogenous ER stressors. In contrast, there was no detectable activation of the IRE1 or ATF6 branches of the UPR upon EMT. Treatment with ER stressors greatly increased the level of PERK signaling in EMT cells, while also strongly activating both IRE1 and ATF6 signaling.

These findings led naturally to the question of the role of PERK signaling in

EMT cell biology. The PERK branch of the UPR pathway is critical for the function of many secretory cells including osteoblasts and  $\beta$  cells. PERK loss of function causes reduced secretion in osteoblasts and cell death in  $\beta$  cells, manifesting in animal models as decreased bone density and diabetes, respectively. Although PERK inhibition did not affect the survival or growth of EMT cells, we found that inhibiting PERK activity increased the sensitivity of EMT cells to ER stress. Moreover, we found that EMT cells required PERK signaling to form tumorspheres and to migrate.

The observation that the EMT program leads to selective activation of the PERK branch of the UPR was supported by analysis of gene expression data from uncultured patient tumors. Analysis of expression data from 800 tumors – spanning a range of tumor types including breast, colon, gastric, and lung cancer – revealed a strong positive correlation

between the expression of EMT genes and PERK pathway genes. In contrast, no significant correlation was observed between EMT genes and IRE1 pathway genes in the same set of tumor expression data.

Our findings have two implications for the treatment of invasive tumors. First, they suggest that agents that promote ER malfunction should be explored for their potential to selectively eradicate cancer cells that have undergone an EMT. Second, they suggest that PERK pathway inhibitors may prove useful for suppressing several of the malignant traits associated with EMT.

#### Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

#### Acknowledgments

We thank Tom DiCesare for assistance with graphical design.

## Funding

This research was supported by grants from the Richard and Susan Smith Family

Foundation and the Breast Cancer Alliance (to PB Gupta), and the National Science

Foundation Graduate Research Fellowship (Grant No. 1122374; to ES Sokol).

## References

1. Kalluri R, Weinberg RA. The basics of epithelial-mesenchymal transition. *J Clin Invest* 2009; 119: 1420-8; PMID:19487818; <http://dx.doi.org/10.1172/JCI39104>
2. Thomson S, Buck E, Petri F, Griffin G, Brown E, Ramnarine N, Iwata KK, Gibson N, Haley JD. Epithelial to mesenchymal transition is a determinant of sensitivity of non-small-cell lung carcinoma cell lines and xenografts to epidermal growth factor receptor inhibition. *Cancer Res* 2005; 65:9455-62; PMID:16230409; <http://dx.doi.org/10.1158/0008-5472.CAN-05-1058>
3. Yang AD, Fan F, Camp ER, van Buren G, Liu W, Somcio R, Gray MJ, Cheng H, Hoff PM, Ellis LM. Chronic oxaliplatin resistance induces epithelial-to-mesenchymal transition in colorectal cancer cell lines. *Clin Cancer Res* 2006; 12:4147-53; PMID:16857785; <http://dx.doi.org/10.1158/1078-0432.CCR-06-0038>
4. Gupta PB, Onder TT, Jiang G, Tao K, Kuperwasser C, Weinberg RA, Lander ES. Identification of selective inhibitors of cancer stem cells by high-throughput screening. *Cell* 2009; 138:645-59; PMID:19682730; <http://dx.doi.org/10.1016/j.cell.2009.06.034>
5. Mani SA, Guo W, Liao MJ, Eaton EN, Ayyanan A, Zhou AY, Brooks M, Reinhard F, Zhang CC, Shipitsin M, et al. The epithelial-mesenchymal transition generates cells with properties of stem cells. *Cell* 2008; 133:704-15; PMID:18485877; <http://dx.doi.org/10.1016/j.cell.2008.03.027>
6. Scheel C, Eaton EN, Li SH, Chaffer CL, Reinhardt F, Kah KJ, Bell G, Guo W, Rubin J, Richardson AL, et al. Paracrine and autocrine signals induce and maintain mesenchymal and stem cell states in the breast. *Cell* 2011; 145:926-40; PMID:21663795; <http://dx.doi.org/10.1016/j.cell.2011.04.029>
7. Hwang WL, Yang MH, Tsai ML, Lan HY, Su SH, Chang SC, Teng HW, Yang SH, Lan YT, Chiou SH, et al. SNAIL regulates interleukin-8 expression, stem cell-like activity, and tumorigenicity of human colorectal carcinoma cells. *Gastroenterology* 2011; 141:279-91, 91 e1-5.
8. Germain AR, Carmody LC, Morgan B, Fernandez C, Forbeck E, Lewis TA, Nag PP, Ting A, VerPlank L, Feng Y, et al. Identification of a selective small molecule inhibitor of breast cancer stem cells. *Bioorg Med Chem Lett* 2012; 22:3571-4; PMID:22503247; <http://dx.doi.org/10.1016/j.bmcl.2012.01.035>
9. Feng YX, Sokol ES, Del Vecchio CA, Sanduja S, Claessen JH, Proia TA, Jin DX, Reinhardt F, Ploegh HL, Wang Q, et al. Epithelial-to-mesenchymal transition activates PERK-eIF2alpha and sensitizes cells to endoplasmic reticulum stress. *Cancer Discov* 2014; 4: 702-15; PMID:24705811; <http://dx.doi.org/10.1158/2159-8290.CD-13-0945>
10. Walter P, Ron D. The unfolded protein response: from stress pathway to homeostatic regulation. *Science* 2011; 334:1081-6; PMID:22116877; <http://dx.doi.org/10.1126/science.1209038>