Cell Research (2007) 17: 289-290

www.nature.com/cr

COMMENTARY

EMT or apoptosis: a decision for TGF-β

Jianguo Song[1]

[1] *Laboratory of Molecular Cell Biology, Institute of Biochemistry and Cell Biology, Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences, Shanghai 200031, China*

Cell Research (2007) **17:**289-290. doi:10.1038/cr.2007.25; published online 10 April 2007

Apoptosis and epithelial-to-mesenchymal transition (EMT) are very fundamental physiological processes. They are both independent and interrelated events in normal development and in maintaining body homeostasis. In recent years, EMT has emerged as a focus of studies in cancer research, and increasing data indicate that EMT functions as a central step in the invasion and metastasis of some tumor cells. Besides the cytoskeleton rearrangement and subsequent morphological changes, EMT is often characterized by the dissolution or attenuation of epithelial adhesion and other polarized structures, for example, tight junctions, and by accordingly the acquisition of migratory and invasive properties of the cells undergoing EMT. Along with these changes, EMT can be associated with transcriptional or expressional alterations of some epithelial and mesenchymal genes.

Dysregulation of apoptosis and EMT are linked with various pathological processes, such as fibrosis of liver and kidney, vascular diseases, abnormal development of embryos, tumor formation and progression [1-5]. Transforming growth factor-β (TGF-β) is a potent pleiotropic molecule that is implicated in diverse biological processes. Interestingly, TGF-β is involved in the tight control of both apoptosis and EMT, which are key to TGF-β's role in physiological and pathological events. Thus, exploring the mechanisms of apoptosis and EMT and their regulation by TGF-β can be of great importance in seeking new therapeutic targets for tumor and other diseases.

TGF-β1 treatment induced strong apoptosis of mouse hepatocytes (AML-12) in dose- and time-dependent manners. The apoptotic response of cells was only apparent after 24 h and lasted for several days, until the death of most cells. Then a question was raised: "why TGF-β1 only induces the death of a portion of cells but not of most or all cells after a certain period of treatment"? In our follow-up studies, we observed that TGF-β1 not only induces the apoptosis but also simultaneously induces the EMT of AML-12 cells. This fact provided a partial but clear answer to the above question: some cells underwent EMT in response to the TGF-β1 treatment, so they survived. Then another question arose: how can a factor (TGF-β1) induce these concurrent but distinct events in the same type of cells, or in other words, why did the same type of cells respond differentially to the same factor under the same experimental conditions? We think that the most likely factor that determines the different responses and thus the different fates of cells to the same stimuli could be the differences that lie in the themselves. As it has been well known that cell cycle progression is associated with alterations in cellular components and corresponding signaling events, there might be a relationship between the cell cycle progression and TGF-β1-induced apoptosis and EMT. Although increasing evidence showed that the cell cycle state is an important factor for cellular responses to extracellular stimuli, very little attention has been given to two distinct cellular responses induced by the same stimuli for the same cells, for example, in the case of TGF-β1-induced concomitant apoptosis and EMT.

To test the above possibility, we examined the potential contribution of cell cycle stage. Firstly, we synchronized the cells at G1/S or G2/M phase and then examined TGF-β1 induced EMT and apoptosis. Interestingly, we observed TGF-β1-induced EMT in both unsynchronized cells and those synchronized at G1/S phase, whereas TGF-β1 did not induce EMT in cells synchronized at G2/M phase [6]. The results were confirmed by either morphological alterations or molecular marks of EMT. Consistently, synchronization of cells at G1/S phase markedly reduced apoptotic response

Correspondence:Jianguo Song
Tel: 86-21-54921167
E-mail: jgsong@sibs.ac.cn

of cells, and in sharp contrast, a dramatic increase of apoptosis was induced in cells synchronized at G2/M phase as measured by fluorescence-activated cell sorter (FACS) and DNA fragmentation assays. These results indicate that TGF-β1-induced apoptosis and EMT are closely related with the cell cycle stage; apoptosis was induced mostly in cells at G2/M phase, whereas EMT was only induced in cells at G1/S phase. In addition, TGF-β1-induced caspase activity in cells synchronized at G2/M phase is significantly higher than in cells synchronized at G1/S phase, and inhibition of apoptosis by a caspase inhibitor has no effect on TGF-β1-induced EMT. These data indicate that TGF-β1-induced concurrent apoptosis and EMT are independent of each other.

TGF-β1-induced EMT of cells at G1/S phase explained how some cells survived after the TGF-β1 treatment under the same condition. It is understandable that in regular cell cultures without synchronization, cells are highly heterogeneous in terms of cell cycle phases and will therefore respond differentially to TGF-β1, leading to different cell fates. It has been widely known that growth arrest is required for cells to undergo differentiation. As TGF-β1-induced EMT occurred in G1/S phase, TGF-β1-induced G1/S phase growth arrest may provide cells a precondition for undergoing EMT. This possibility is supported by our finding that TGF-β1 also induces EMT in several other epithelial cells (MDCK, A-549, Mv1Lu), because TGF-β1 can strongly inhibit the proliferation of all these cell lines. This fact also raises another possibility: the true nature of TGF-β-induced growth arrest of epithelial cells is to induce EMT. In other words, the fundamental role of TGF-β in these cells may be the induction of EMT, but not growth arrest.

TGF-β1-induced apoptosis and EMT could be mutually exclusive processes in physiological context, because these two events have been shown to contribute differentially to the effects of TGF-β on tumor progression and embryonic development. TGF-β-induced EMT leads to migration and invasion of local epithelial cells. This results in an escape of apoptotic fates of these cells, and is important for organogenesis and tumor metastasis. Our studies indicate that the different effects of TGF-β can take place simultaneously. It is thus highly possible that TGF-β's double effects (induction of growth arrest/apoptosis and EMT) are not necessarily dependent on the stages of cancer development and embryogenesis, as its net effect is contingent on the cellular context and the specific state of cells. The sensitivity of tumor cells to TGF-β may be on one hand determined by the genetic basis, such as gene mutation, TGF-β receptor deletion or other alterations, and on the other hand determined by the cell cycle state. In early embryogenesis, the differentiation, migration or apoptotic responses to TGF-β may also be regulated, at least partly, by the cell cycle stage. Thus, in addition to specific components of the TGF-β signaling pathway, cell cycle phase could also be a factor in the consideration of strategies of clinical therapy, including the cancer treatment. These findings provided a new insight into the mechanism of TGF-β-mediated apoptosis and EMT, and unveiled the mystical nature of cellular responses to TGF-β treatment, which is of great importance in understanding the multifunctional effects of TGF-β and how different cellular fates of the same cell type could be induced.

References

1 GrÜnert S, Jechlinger M, Beug H. Diverse cellular and molecular mechanisms contribute to epithelial plasticity and metastasis. Nat Rev Mol Cell Biol 2003; **4**:657-665.
2 Kalluri R, Neilson EG. Epithelial-mesenchymal transition and its implications for fibrosis. J Clin Invest 2003; **12**:1776-1784.
3 Thiery JP. Epithelial-mesenchymal transitions in development and pathologies. Curr Opin Cell Biol 2003; **15**:740-746.
4 Prindull G, Zipori D. Environmental guidance of normal and tumor cell plasticity: epithelial mesenchymal transitions as a paradigm. Blood 2004; **103**:2892-2899.
5 Huber MA, Kraut N, Beug H. Molecular requirements for epithelial-mesenchymal transition during tumor progression. Curr Opin Cell Biol 2005; **17**:1-11.
6 Yang Y, Pan X, Lei W, Wang J, Song J. Transforming growth factor-beta1 induces epithelial-to-mesenchymal transition and apoptosis via a cell cycle-dependent mechanism. Oncogene 2006; **25**:7235-7244.