Cell Cycle 13:5, 697–698; March 1, 2014; © 2014 Landes Bioscience

GSK-3β: A key regulator of breast cancer drug resistance

Comment on: Sokolosky M, et al. Cell Cycle 2014; 13:820–33; PMID: 24407515; http://dx.doi.org/10.4161/cc.27728

Alberto M Martelli[1,*], Francesca Buontempo[1], and Camilla Evangelisti[2,3]; [1]Department of Biomedical and Neuromotor Sciences; University of Bologna; Bologna, Italy; [2]Institute of Molecular Genetics; National Research Council; Bologna, Italy; [3]Muscoloskeletal Cell Biology Laboratory; IOR; Bologna, Italy; *Email: alberto.martelli@unibo.it; http://dx.doi.org/10.4161/cc.28091

Despite continuous advances in the knowledge of breast cancer pathophysiology, this type of neoplasia remains a leading cause of cancer-related death in women, with more than 450000 deaths every year worldwide.[1] Surgery, coupled with radiation and/or chemotherapy, remains the main approach for breast cancer treatment. However, another common therapeutic option is based on the use of the estrogen receptor (ER) antagonist, tamoxifen.[2] Indeed, approximately 60–70% of early-stage breast cancers overexpress the ER, making their growth dependent on estrogens. Tamoxifen blocks estrogen signaling by competitively binding the ER, thus antagonizing its proliferative and pro-survival effects.

Breast cancers can be inherently drug-resistant or develop resistance after exposure to chemotherapeutic drugs, such as the anthracyline, doxorubicin.[1] Resistance can also develop in patients receiving tamoxifen.[2] Therefore, it is very important to understand how breast cancers become drug- and hormone-resistant, and whether or not their drug-resistance can be reversed. There also is a need for novel targeted therapies for breast cancer patients who develop resistance to traditional therapies.

GSK-3β is a serine/threonine kinase involved in several signal transduction cascades, including the PI3K/Akt/mTOR, Wnt/β-catenin, and MEK/ERK pathways.[3] In particular, Akt and other kinases phosphorylate GSK-3β at Ser9. This phosphorylative event inactivates GSK-3β3. GSK-3β regulates cell cycle progression, differentiation, survival, embryogenesis, migration, and metabolism. However, aberrant GSK-3β activity has been linked with an increasing number of pathologies, including cancer, heart disease, immune system disorders, diabetes, atherosclerosis, and different neurological diseases.[3]

Although its role in cancer remains controversial, GSK-3β has also been implicated in breast cancer development and drug resistance.[4,5] Using ER-positive MCF-7 breast cancer cells, Sokolosky and coworkers have performed a detailed analysis of the roles played by GSK-3β in causing resistance to doxorubicin or tamoxifen.[6] They found that MCF-7 cells overexpressing a kinase-dead (KD) form of GSK-3β were more resistant to doxorubicin and tamoxifen compared with MCF-7 cells overexpressing either wild-type (WT) or constitutively active (CA) GSK-3β. Ectopic expression of GSK-3β(KD) also resulted in increased clonogenic activity of MCF-7 cells in comparison with either GSK-3β(WT) or GSK-3β(CA) overexpression.

Moreover, treatment of parental MCF-7 and MCF-7/GSK-3β(WT) cells with doxorubicin abrogated the phosphorylation of GSK-3β at Ser9. In contrast, Ser9 p-GSK-3β was still detectable in MCF-7/GSK-3β(KD) and MCF-7/GSK-3β(CA) cells. This indicated that one of the effects of doxorubicin on MCF-7 cells was suppression of Ser9 p-GSK-3β, which could then result in increased GSK-3β activity. Downregulation of Ser9 p-GSK-3β levels was detected despite increased Akt activity induced in these cell types by doxorubicin. This finding documented that the control of GSK-3β activity is complex and multi-factorial in MCF-7 cells, as there may be several alternate routes of GSK-3β inactivation that are independent of Akt,[3] including downregulation of protein phosphatase activity or upregulation of other kinases (**Fig. 1**).

What is even more interesting, MCF-7/GSK-3β(KD) cells displayed an elevated sensitivity to the mTORC1 inhibitor, rapamycin, compared with MCF-7/GSK-3β(WT) or MCF-7/GSK-3β(CA) cells, while concurrent MEK/ERK inhibition alleviated doxorubicin and tamoxifen resistance in MCF-7/GSK-3β(KD) cells (**Fig. 1**). Overall, these results demonstrated that GSK-3β is a key regulatory molecule in sensitivity of breast cancer cells to chemo-,

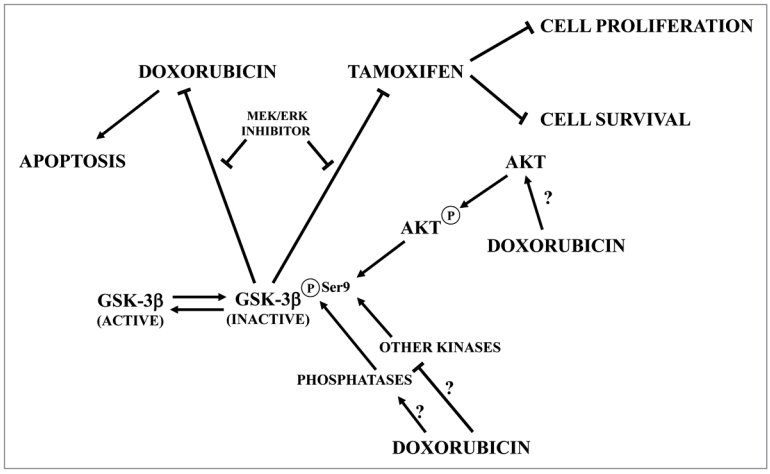

Figure 1. Schematic of the effects of GSK-3β inactivation on MCF-7 cell sensitivity to doxorubicin or tamoxifen. Arrows indicate activating events, whereas perpendicular lines indicate inhibitory events. At present, it is not clear how doxorubicin could dephosphorylate GSK-3β and activate Akt at the same time. Doxorubicin could either inhibit other protein kinases, targeting Ser9 p-GSK-3β, or stimulate protein phosphatases.

hormonal, and targeted therapy (**Fig. 1**). Moreover, they fit well with the conclusions of an immunohistochemical study performed on paraffin-embedded samples from 72 consecutive invasive mammary carcinomas, which documented how higher Ser9 p-GSK-3β levels correlated with a worse clinical outcome in ER-positive cases.[7] GSK-3β-controlled drug resistance may be mediated by the BH3-only member of the Bcl-2 family, Mcl-1. Indeed, the expression levels of this anti-apoptotic protein inversely correlated with GSK-3β activity (assessed by immunostochemical staining for p-GSK-3β) in breast cancer specimens, and active GSK-3β was found to be required for proteasome-mediated Mcl-1 degradation.[8]

In conclusion, the findings by Sokolosky et al.[6] unequivocally demonstrated that loss of GSK-3β kinase activity could dramatically increase the drug and hormonal resistance of breast cancer cells. However, this may confer an Achilles' heel by sensitizing cancer cells to targeted therapy with small-molecule kinase inhibitors. Although further studies are needed to determine the clinical relevance of the complex interactions between GSK-3β, mTORC1, and MEK/ERK, it could be envisaged that mTORC1 and MEK/ERK inhibitors should be able to potentiate the effects of chemo- and hormonal therapy, thereby presenting an attractive treatment route for overcoming GSK-3β-mediated drug-resistance in breast cancer.

References

1. McCubrey JA, et al. Adv Enzyme Regul 2010; 50:285-307; PMID:19895837; http://dx.doi.org/10.1016/j.advenzreg.2009.10.016
2. Steelman LS, et al. Cell Cycle 2011; 10:3003-15; PMID:21869603; http://dx.doi.org/10.4161/cc.10.17.17119
3. Phukan S, et al. Br J Pharmacol 2010; 160:1-19; PMID:20331603; http://dx.doi.org/10.1111/j.1476-5381.2010.00661.x
4. Farago M, et al. Cancer Res 2005; 65:5792-801; PMID:15994955; http://dx.doi.org/10.1158/0008-5472.CAN-05-1021
5. Dong J, et al. Cancer Res 2005; 65:1961-72; PMID:15753396; http://dx.doi.org/10.1158/0008-5472.CAN-04-2501
6. Sokolosky M, et al. Cell Cycle; 13:820-33; PMID: 24407515; http://dx.doi.org/10.4161/cc.27728
7. Armanious H, et al. Hum Pathol 2010; 41:1657-63; PMID:20709358; http://dx.doi.org/10.1016/j.humpath.2010.04.015
8. Ding Q, et al. Cancer Res 2007; 67:4564-71; PMID:17495324; http://dx.doi.org/10.1158/0008-5472.CAN-06-1788