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Editorial

Breast cancer: the menacing face of Janus kinase

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Janus, the two-faced Roman god of gates and doors, beginnings and endings, symbolizes important transitions. How appropriate therefore that Janus kinase 2 has been discovered to control the transition from normal mammary gland development to breast cancer in a recent manuscript from the Schreiber laboratory published in *Cell Death and Differentiation*.[1] The lifetime risk of breast cancer for women in the western world is 1 in 8 and the incidence rate is rising toward epidemic levels with 1.4 million women diagnosed annually with this disease worldwide http://www.cancerresearchuk.org/cancer-info/cancerstats/world/breast-cancer-world/. Breast cancer is a heterogeneous disease with three main histological subtypes: estrogen receptor alpha/progesterone receptor (ERα/PR) positive, HER2 overexpressing, and triple negative tumors, which do not express ERα, PR or HER2. Six subtypes have been identified on the basis of gene expression signatures, each with a different prognosis.[2] Patients with luminal subtype tumors, which are ERα positive and account for over 70% of breast cancers, have the best prognosis and are treated clinically with anti-estrogen therapies such as tamoxifen or aromatase inhibitors. However, despite an overall good prognosis for ERα-positive breast cancer, ~25% of women develop resistance to anti-estrogen therapy, have recurrent tumors and succumb to metastatic disease.[3] Furthermore, ERα+ breast cancer is itself heterogeneous. There is thus a pressing need to understand the origins of ERα+ tumors and the molecular mechanisms that give rise to their diversity and differential response to anti-estrogen therapy.

Murine breast cancer models have been invaluable in delineation of both the genes and signaling pathways that regulate tumorigenesis. However, although ERα+ tumors are the most common type of breast cancer in women, there is a paucity of experimental mouse models of ERα+ tumors. Recently, the Schreiber laboratory described a new mouse model of ERα+ mammary carcinoma that arises in mice deficient for Stat1.[4] Stat1 is a member of the Stat family of latent transcription factors, which bind to cytokine and growth factor receptors on their engagement by ligand, resulting in activation of receptor-associated JAK kinases that in turn tyrosine phosphorylate Stats, which dimerize, translocate to the nucleus and bind to promoters of target genes. Genetic ablation of Stat1 resulted in the spontaneous development of mammary tumors, with a long latency, that recapitulate many of the features and gene expression profiles of ERα+ luminal breast cancer. Thus, Stat1 is an unexpected tumor suppressor in mammary gland and this correlates with reduced expression levels of STAT1 in 45% of ERα+ human breast tumors, suggesting that Stat1$^{-/-}$ mice may be a useful model of ERα+ breast cancer. Conversely, other members of the Stat family of transcription factors, in particular Stat3 and Stat5, are potent oncogenes in mammary gland and appear to have reciprocal effects on target gene expression.[5–7] During normal mammary gland development, Stat5 is essential for alveologenesis, the process by which differentiated milk-producing cells arise during pregnancy,[8] whereas Stat3 is a critical mediator of cell death during post-lactational regression of the gland.[9,10] Stats 3 and 5 are generally thought of as having opposing functions and, despite recognizing a similar DNA-binding motif, bind distinct promoters.[10]

This Stat1$^{-/-}$ ERα+ tumor model has now been characterized in more detail and the authors show that control of Jak2 activity by the Stat1-SOCS1 axis is essential to maintain mammary gland homeostasis and that deletion of Stat1 and the concomitant loss of SOCS1, a negative regulator of Jak2, results in hyperactivation of Jak2 and unopposed signaling through the prolactin receptor (PrlR). This corroborates a previous study showing that loss of a single allele of SOCS1 can rescue lactation failure that occurs in PrlR$^{+/-}$ mice.[11,12] Interestingly, persistent PrlR signaling is a feature also of human ERα+ and ERα− breast cancer cells[13] and elevated serum levels of prolactin (Prl) have been associated with increased risk of invasive ERα+ tumors and poor long-term survival.[14] Furthermore, over 95% of human breast cancers overexpress PrlR and human breast cancer cells have been shown to upregulate local synthesis of Prl.[15]

The primary transcription factor downstream of PrlR in mammary gland is Stat5a.[16] However, in Stat1$^{-/-}$ tumors, pJak2, pStat3 and pStat5a/b were all detected with pStat3 and pStat5 being observed in a proportion of the tumor cells raising the possibility that they are either co-activated in a subset of cells or that they are activated in two discrete populations. This distinction may be critically important to understanding the perturbation of signaling pathways in these tumors. Previous studies demonstrated that breast tumors exhibiting both activated STAT3 and STAT5 were more differentiated than tumors with just pSTAT3.[5] As only Stat5

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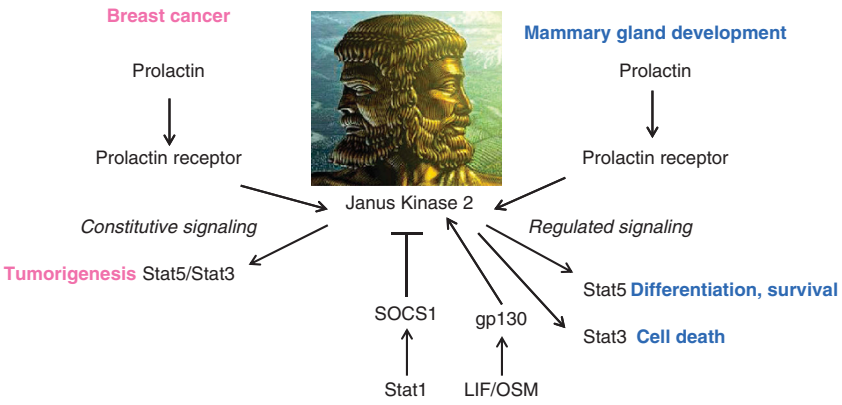

Figure 1 Summary of outcomes downstream of the prolactin receptor and JAK2 in breast cancer and in normal mammary gland. During pregnancy, engagement of the prolactin receptor by its ligand prolactin results in phosphorylation of Stat5, which regulates alveologenesis and differentiation of luminal epithelial cells and provides also a survival signal to the epithelium. On the initiation of post-lactational regression (involution), prolactin levels drop precipitously and Stat3 becomes phosphorylated in response to elevated levels of LIF, whereupon it induces cell death. Notably, signaling is regulated by negative feedback through multiple mechanisms including the SOCS family of proteins. In contrast, in the absence of SOCS1, JAK2 becomes constitutively active resulting in coincident phosphorylation of Stat3 and Stat5, promoting the development of ERα+ tumors

engages with the PrlR in normal mammary gland during pregnancy, when Stat3 is present at high levels but is not phosphorylated, this suggests that active Jak2 is bound also to receptors that engage Stat3 such as gp130/LIFR in Stat1-deficient mammary cells. It is well established that both Stat1 and Stat3 are activated downstream of the common gp130 receptor chain but in many contexts, gp130 is preferentially bound by Stat3, so in this instance it is interesting that deficiency of Stat1 has a profound effect on levels of pStat3.[17]

Using knockdown or inhibition of Jak2, the authors demonstrated that Jak2 is required for phosphorylation of Stat3 and Stat5 and that persistent activation of the PrlR-Jak2-Stat3/5a/5b axis provides a potent survival signal to tumor-derived cells that have presumably become addicted to Stat3 and/or Stat5a/5b, as observed in other human tumors and cell lines.[18] As these studies utilized cell lines, the requirement for PrlR signaling *in vivo* was investigated by treating Stat1$^{-/-}$ tumor-bearing animals with a Jak2 inhibitor. This blocked, as anticipated, pStat3 and pStat5 and resulted in an increase in cleaved caspase 3-positive cells and a remarkable and prolonged inhibition of tumor growth for 7 months following inhibitor withdrawal. Furthermore, Jak2 drives tumor growth in both hormone-dependent and hormone-independent Stat1$^{-/-}$ tumors.

This interesting and important work provides a useful model for further studies on the initiation and progression of ERα+ breast tumors (Figure 1). JAK2 inhibitors are currently undergoing clinical trials and these could be a valuable addition to the clinician's armory for tamoxifen-resistant breast cancers. Also, given the demonstration in this manuscript of the prophylactic efficacy of a JAK2 inhibitor, women at risk for breast cancer could receive anti-JAK2 therapy. In this context, it is notable that Jak2 has been shown to be required for initiation but not the maintenance of mammary tumors driven by Prl overexpression,[19] as genetic ablation of Jak2 before, but not after, neoplastic transformation abolished tumorigenesis. The discrepancy between these two studies suggests that signaling through Jak2, downstream of receptors other than PrlR in the Stat1$^{-/-}$ mice, results in tumors that are dependent on Jak2.

Further work is required to determine whether inhibition or ablation of either Stat5 or Stat3 alone is sufficient to inhibit tumor growth or whether it is the combination of both of these Stats that is required to generate ERα+ tumors. Given the competition between Stat5 and Stat3 for binding to specific promoters, and their overall distinct sets of transcriptional targets, it will be informative to investigate whether relative levels of these Stats dictate the clinical outcome. This ERα+ breast tumor model described by the Schreiber laboratory further endorses the view that there is an intricate interplay between Janus kinases and their corresponding Stats during mammary tumorigenesis. The irrefutable conclusion that can be drawn from this work is that perturbing the delicate balance between the activities of JAK2, STAT1, STAT3 and STAT5 leads to breast cancer development and targeting this pathway provides opportunities for therapeutic intervention.

Conflict of Interest

The authors declare no conflict of interest.

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