Another Breast Cancer Entity Confirmed: Genomics of Invasive Lobular Breast Cancer

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Invasive lobular carcinoma (ILC) accounts for up to 15% of all invasive breast cancers.[1] We have long recognized the unique clinical characteristics of ILC, but an increased understanding of its biology in contrast with the more common breast cancer histologic type, infiltrating ductal carcinoma (IDC), has emerged. The article that accompanies this editorial by Desmedt et al[2] reinforces and extends this understanding.

ILC is characterized by loss of normal cell-cell adhesion, which results in discohesive cells that fail to form glandular structures. The defining genetic feature that distinguishes ILC from IDC is *CDH1* mutation or loss of either mRNA or protein expression of the cell adhesion molecule E-cadherin; 95% of ILCs demonstrate some form of E-cadherin alteration.[3,4] Most ILCs are of the classical histologic type, which is low grade and possesses a low to intermediate mitotic index,[1] whereas less common ILC variants, such as the solid and mixed nonclassical types, often are high grade and associated with a poorer outcome than classical ILC.[5] ILC of all types usually expresses estrogen receptors (ERs) and progesterone receptors and are rarely clinically human epidermal growth factor receptor 2 (HER2) positive. Perhaps related to *CDH1* loss, which in other tumor types confers increased risk of invasion and metastasis,[6,7] some studies have suggested that patients with ILC have similar or worse long-term outcomes compared with patients with stage-matched IDC.[8,9] Metastatic ILC also has unusual patterns of spread to sites such as the ovaries, lining of the GI tract, and peritoneum.[1,8] All these features are consistent with a unique biology.

Despite many studies that investigated the genomic and mutational landscape of breast cancer, lobular breast tumors have been largely underrepresented.[4,10] Several studies have examined transcriptional profile differences that distinguish ILC from IDC; however, with the exception of the loss of *CDH1* expression, few ILC-distinguishing features have been identified.[11-16] Desmedt et al[2] genomically profiled the largest cohort of primary invasive lobular breast tumors analyzed to date, which included 430 tumors analyzed for mutations and 170 for copy number aberrations, to identify and characterize the recurrent genomic alterations present in these tumors. The results of this large study in conjunction with remarkably consistent findings from 127 ILCs and 490 IDCs in The Cancer Genome Atlas (TCGA) project[3] and 144 ILCs from a European cohort[17] have clear biologic and clinical implications with regard to genetic alterations that drive lobular breast cancer.

The most frequent significantly altered gene identified in this study was *CDH1* in which loss-of-function mutations occurred in 65% of cases and loss of heterozygosity or deletion was identified in 94% of patients. These findings are consistent with other studies and underscore the role of E-cadherin loss in contributing to the characteristic discohesive morphology associated with ILC.[3]

Given that the majority of ILCs are hormone receptor positive, endocrine therapy is key to treatment. ILC appears to derive particular benefit from treatment with aromatase inhibitors compared with tamoxifen, which is possibly related to relative tamoxifen resistance.[18,19] The current study and TCGA analysis[3] identified alterations that affect ER signaling, including mutations in ER modulators that could lead to altered or divergent ER-mediated signaling. Of note, the authors found that focal copy number gains at 6q25.1, including *ESR1*, occur in 25% of lobular tumors and that copy number changes corresponded to increased expression of an *ESR1* mRNA gene expression signature. Although TCGA analysis did not indicate copy number gain as significantly enriched in ILC, it revealed increased ER activity in the form of significantly higher levels of total and phosphorylated ER protein. Beyond *ESR1* and ER expression, ILC-specific mutations that affect ER cofactors were identified. In both TCGA and the current study, ILC was characterized by an increased incidence of *FOXA1* mutations, whereas IDC was enriched for *GATA3* mutations. Similarly, lower *GATA3* mRNA and protein expression in ILC and reduced methylation at ER promoters were observed in the TCGA cohort.[3] Because both *GATA3* and *FOXA1* modulate ER transcriptional activity, the results suggest preferential binding of ER to *FOXA1*-bound promoter sites in ILC and to *GATA3*-bound sites in IDC, which could lead to differential *ESR1*-regulated gene expression.[3,20] Both altered ER protein expression and unique mutation patterns in ER transcription complex family members can affect response to endocrine therapy.

Beyond *CDH1* and ER signaling, 50% of ILCs harbor mutations in one of three key genes that regulate the phosphatidylinositol 3-kinase (PI3K) pathway. Specifically, Desmedt et al[2] identified an increased frequency of lobular-specific mutations in *PIK3CA*, the PI3K-negative regulator *PTEN*, and the downstream kinase *AKT1*. An increased incidence of *PTEN* loss-of-function mutations or loss of heterozygosity as well as a high frequency of

PIK3CA mutations in ILC was observed in the TCGA and European studies.[3,17] In TCGA, loss or mutation of *PTEN* corresponded with decreased PTEN protein expression and a concomitant increase in activated or phosphorylated Akt protein expression in lobular tumors. In fact, the levels of phosphorylated Akt protein expression identified in lobular tumors was comparable to the expression of these proteins in basal-like or HER2-enriched ductal tumors, which have been shown to have the highest levels of PI3K/Akt signaling among IDC tumors.[4] Beyond protein expression, integrative analysis of TCGA genomic data identified multiple alterations that contribute to aberrant Akt/mammalian target of rapamycin signaling in 45% of ILC cases. Previous studies have linked loss of E-cadherin with Akt activation and epidermal growth factor receptor (EGFR) expression. Results from TCGA that demonstrated overexpression of phosphorylated EGFR in conjunction with data that describe the increased incidence of PI3K/Akt pathway mutations from both the current study and TCGA suggest that the ILC tumor environment provides a favorable cellular context for altered Akt signaling[3,4,21,22] and that the targeting of the PI3K/Akt pathway may be particularly relevant in ILC.

The study by Desmedt et al[2] is the first to characterize *HER2* and *HER3* mutations in ILC. Overall, 14% of ILC tumors were either *HER2* amplified or carried a mutation in *HER2* or *HER3*. These data suggest that ILC has the highest incidence of *HER2*-mutated, nonamplified tumors among all breast cancer subsets. In the TCGA study, 10% of ILCs had amplified or mutated *HER2,* and reverse phase protein assays revealed increased EGFR activity in ILC. Together, these studies suggest that *ERBB* family signaling may significantly contribute to ILC biology. Additional analyses are necessary to fully illuminate the context and extent of alterations that affect this family of genes as well as to understand the clinical relevance of these alterations.

In the TCGA and European studies,[3,17] transcriptome analyses found at least two molecularly distinct subsets of ILC, including an immune-related subset that appeared to have both high expression of lymphocyte signaling pathway components and negative immune response regulators. This finding may have consequences for the use of immune checkpoint inhibitors or other immune-based approaches in ILC.

Desmedt et al,[2] in concert with similar initiatives published within the past few months,[3] have extensively characterized the genomic landscape of ILC and have produced remarkably consistent results that identify at least three significantly altered and therapeutically relevant pathways. These are an important first step to understanding ILC as well as the potential treatment implications of this unique biologic subset of breast cancer. It is now clear that ILC and IDC are distinct molecular diseases, a fact that should be considered in future experimental and therapeutic initiatives.

AUTHORS' DISCLOSURES OF POTENTIAL CONFLICTS OF INTEREST

Disclosures provided by the authors are available with this article at www.jco.org.

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