Targeting CXCR4-induced desmoplasia to improve checkpoint inhibition in breast cancer

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T lymphocyte checkpoint inhibition-based therapy represents the great therapeutic advance for cancer in the current decade. Beginning in 2012 with the initial presentation of a phase 3 trial in metastatic melanoma demonstrating the value of CTLA-4–directed therapy, every year has seen the expansion of this therapeutic modality in terms of targets, drugs, and tumors. This has led to improved outcomes for patients with melanoma, lung cancer, bladder cancer, Merkel cell cancer, renal cell carcinoma, hepatoma, and Hodgkin disease. Initially a therapy for metastatic cancers, checkpoint inhibitor therapy is now moving to the early-disease setting in select high-risk populations.

Metastatic breast cancer has been a latecomer to the immuno-oncology party. Partly this represented a bias among drug developers that breast cancer was a less promising target than many other human cancers. Breast cancer is a disease with many available targeted therapeutics [for estrogen receptor (ER)-positive and human epidermal growth factor receptor 2 (HER2)-positive disease] as well as a broad array of chemotherapeutic agents, all with established therapeutic benefit. In addition, breast cancer is a less heavily mutated cancer than other cancers for which checkpoint inhibition has proved successful.

This changed recently with the results of the IMpassion130 trial (NCT02425891), a randomized controlled trial in first-line metastatic triple-negative breast cancer (1). In this trial, patients received nanoparticle albumin-bound (nab)-paclitaxel (a microtubule-targeting chemotherapeutic agent) alone or in combination with atezolizumab, a monoclonal antibody targeting programmed cell death ligand 1 (PD-L1). While ineffective in PD-L1–negative cancers, the addition of atezolizumab improved progression-free survival from 5.0 to 7.5 mo and overall survival from 15.5 to 25 mo in patients with PD-L1–positive tumors. Checkpoint inhibition has arrived in metastatic breast cancer.

Despite these positive results, much remains to be done to render this therapeutic approach successful for the majority of patients with metastatic breast cancer. Most patients receiving the combination of chemotherapy and PD-L1 targeting are not long-term survivors, and we do not yet know whether the plateau in progression-free survival with cancers such as melanoma will be seen in breast cancer.

It is in this context that Chen et al. (2), in PNAS, ask simple but profoundly important questions: Might therapeutic failure relate to the inability of effector T cells to physically engage with their tumor target? In particular, might the presence of a dense fibrotic stroma (or desmoplasia) represent an immunosuppressive barrier for T cells? And, following on this, might we be able to reverse this immunosuppressive state by reducing desmoplasia, allowing improved access by activated T cells to metastatic cancers?

The answer to all these questions, the authors argue, is yes. Beginning with an analysis of the The Cancer Genome Atlas database of human breast cancers, Chen et al. (2) identify genes associated with stromal T lymphocyte exclusion. Among these was the CXCL12 receptor CXCR4. Previous studies have shown an important role for both fibrosis and the CXCL12/CXCR4 axis in the metastatic process and in immune suppression within the tumor microenvironment (3). Chen et al. (2) examine desmoplasia and CXCR4 both in the clinic and in preclinical models of breast cancer. In the clinic, comparison of primary and metastatic tumors demonstrates increased desmoplasia in metastases, an association between CXCR4 and PD-L1 expression, and the relative absence of cytotoxic T lymphocytes in metastases.

In the preclinical MCa-M3C murine breast cancer model, the authors demonstrate that inhibition of CXCR4 with plerixafor (AMD3100) results in decreased fibroblast recruitment and desmoplasia by tumors and in reduced profibrotic and immunosuppressive gene expression (in two mouse models). Lastly, they show that CXCR4 blockade decreases immunosuppression, decreases metastasis, and improves T cell infiltration and response to checkpoint inhibition, with subsequent improvement in mouse survival. The authors make a good case for both the role of the CXCL12/CXCR4 axis

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in immunosuppression, and the potential targeting of the axis for therapeutic benefit in combination with checkpoint inhibitor therapy.

Questions Raised by the Study

This very interesting paper raises as many questions as it answers. These issues relate to the study as presented and to larger issues for the field.

Beginning with Chen et al.'s (2) study itself, the number of patients with paired primary and metastatic disease that form the clinical basis of this study is small ($n = 17$), and as such, there are severe limitations to the conclusions one might draw from any biomarker analysis. For instance, all of the paired samples came from patients with lung and liver metastases (i.e., no bone metastases were studied). Although we know that at the genomic level, breast cancer represents a family of diseases rather than a single disease, the small numbers studied here do not allow any meaningful analysis of intrinsic subtypes. The disease-free survival curves, with their rapid, cliff-like fall-off in the CXCR4-high arm suggest possible selection bias in the tissue samples employed.

Fortunately, there are other datasets looking at CXCR4 in paired breast cancer samples. For example, Szekely et al. (4) examined a somewhat larger number of paired primary and metastatic samples and, consistent with Chen et al. (2), tumor infiltrating lymphocyte counts were significantly lower in metastases than in primary breast tumors, suggesting that immune exclusion is a real phenomenon. In addition, CXCR4 expression, while not increased in the metastatic site, is maintained compared with the primary tumor and may continue to have biologic relevance.

Larger paired genomic datasets have recently become available and should provide interesting new information regarding the immune microenvironment in metastatic disease. The tumor microenvironment can differ by organ site (lung vs. liver vs. bone) and by tumor type. For example, in primary breast cancer, ER-positive breast cancers, particularly luminal A cancers, are associated with greater tumor fibrosis than are ER-negative tumors (5), and one would be unsurprised to see similar patterns in metastatic sites. Increased fibrosis in primary tumors has also been associated with higher likelihood of bone metastasis (6), so it would be unsurprising to see differential expression of CXCR4 by metastatic site.

At the preclinical level, there are similar issues with the Chen et al. (2) study. The principal murine model used here is hardly representative of the breast cancer seen in the clinic. It is a classic laboratory model, with rapid metastasis and death, as opposed to the far more gradual course of human cancers. It examines metastasis to a single site (the lung) and does not take into account the targeted therapy approaches (e.g., for ER and HER2) that make up the majority of breast cancer treatments and that might affect the makeup of the metastatic microenvironment (antiestrogen therapy, for instance, reduces fibrosis). And, as with all limited model systems, one is always concerned with broader applicability. By way of contrast, Brooks et al. (7) examined fibrosis across 11 triple-negative breast cancer metastasis model systems and concluded that overall metastasis-induced fibrosis was limited and therefore unlikely to represent an important therapeutic target.

The Way Forward with CXCR4 Inhibition in the Clinic

Even allowing for these concerns, the results of this study are interesting and offer a clinically testable hypothesis. With the advent of a positive checkpoint inhibitor trial, it is reasonable to expect that some combination of chemotherapy and checkpoint inhibitor therapy (such as the combination of nab-paclitaxel and atezolizumab used in the IMpassion130 trial) will become a standard-of-care therapy. This opens the door for clinical trials of CXCR4 inhibition as a means of improving clinical benefit.

The availability of relatively nontoxic CXCR4 antagonists suggests a simple testable clinical hypothesis well worth examining in breast cancer.

That this is a reasonable prospect is demonstrated by the increasing clinical interest in agents targeting CXCR4. Numerous such agents (e.g., BL-8040, LY2510924, and USL311) are currently in clinical trials across several disease types. In metastatic breast cancer, there is already intriguing published data employing the CXCR4 antagonist balixafortide. Pernas et al. (8) performed a phase 1, single-arm, dose-escalation study, combining the microtubule-targeting agent eribulin with balixafortide in patients with HER2-negative metastatic breast cancer. Objective responses were seen in 30% of patients in the overall study and in 38% of patients at the highest combined dose level. These impressive results certainly warrant further testing. Based on these data, the Food and Drug Administration granted Fast Track designation for balixafortide. Because the CXCR4 antagonist adds little in the way of drug toxicity, it is not a great stretch to incorporate this agent in trials with a chemotherapy/checkpoint inhibitor combination.

Though the way forward seems clear, questions remain. The basic premise of this approach is that desmoplasia represents a major immunosuppressive barrier for checkpoint inhibitor-based therapy. Both clinical and laboratory researchers are appropriately skeptical of monoform explanations of drug resistance, and such skepticism is certainly warranted here. One of the reasons that breast cancer was not first on anyone's list of potential targets for checkpoint inhibition was that its overall tumor mutational burden (TMB) is low compared with many of the currently successful targets, with the implication that high TMB is associated with an increased number of T cell-targeting surface epitopes. Resistance, therefore, might simply represent the fact that many breast cancers are inherently immune deserts.

Even within breast cancer, TMB may differ widely, with some breast cancers therefore being unlikely targets. ER-positive breast cancers have, overall, lower TMB than triple-negative breast cancers. As mentioned, ER-positive cancers are more commonly associated with tumor fibrosis than ER-negative tumors. If a well-differentiated, slow-growing, low-TMB ER-positive tumor fails to respond to checkpoint inhibitor therapy, then is the culprit fibrosis or rather the lack of valid immune targets in a cancer that is doing its best to mimic a normal milk duct?

Nevertheless, Chen et al. (2) offer us a fascinating way forward in the immuno-oncology space. It is already clear that while checkpoint inhibition will play a role in the treatment of metastatic breast cancer, currently available data do not suggest that this therapy is a panacea. New approaches are still needed if we are to optimize checkpoint inhibitor therapy. The availability of relatively nontoxic CXCR4 antagonists suggests a simple testable clinical hypothesis well worth examining in breast cancer.

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