Cancer evolution constrained by the immune microenvironment

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Summary

Tumor development is a Darwinian evolutionary process, involving the interplay between cancer subclones and the local immune microenvironment. These complex interactions are highlighted in this issue of *Cell* by the results from a deep analysis of one patient with advanced serous carcinoma of the ovary (Jiménez-Sánchez et al., 2017).

Main Text

Recent years have seen a rejuvenation in the field of cancer evolution, with an increasing number of clinical studies revealing the impact of genetic heterogeneity, the substrate for cancer evolution, on the acquisition of drug resistance, biomarker qualification and clinical outcome (Greaves, 2015).

Coincident with a renewed interest in the field of cancer evolution, and appreciation of the limitations of targeting single genomic alterations, cancer immunotherapy has risen like a phoenix from the flames as the new hope in cancer medicine. An impressive array of antibodies, effectively releasing the breaks from the immune system, have entered clinical practice with a range of FDA approvals across distinct tumor types over the last 18 months, in many cases offering meaningful overall survival benefits (Topalian et al., 2012).

The (re-)emergence of the fields of both cancer evolution and immunotherapy has led to an appreciation that tumor development is not determined solely by genetic alterations in tumor cells, but also by the fitness advantage such mutations confer within a given environment and geographic location (Junttila and de Sauvage, 2013). As such, just as knowledge of ecological niches and geographical barriers are required to understand species evolution, an understanding of immune surveillance and the constraints the local immune environment imposes upon an evolving tumor entity are central to deepen knowledge about a cancer's origin and its fate (Figure 1).

With this in mind, in this issue of Cell, Snyder and Miller and colleagues (Jiménez-Sánchez et al., 2017) perform a deep analysis of one patient with advanced serous carcinoma of the ovary to understand the interactions of an evolving tumor at multiple distinct sites of disease within the constraints of the local tumor microenvironment. The authors focus on an often-overlooked phenomenon in cancer management, namely the occurrence of heterogeneous responses across geographically distinct tumor lesions, with simultaneous regression and progression of tumor metastases at distinct anatomical locations.

The patient underwent surgical debulking, followed by platinum combination therapy, and experienced disease recurrence within 7 months, followed by multiple lines of cytotoxic therapies over the course of the next 3 years. Following exhaustion of all standard lines of therapy, the patient was offered best supportive care and on follow-up off chemotherapy her CA125 tumor marker began to fall with differential growth of her metastatic lesions with a new growth in the vaginal cuff. Owing to her long treatment free interval (a marker of prognosis in this disease which guides management), she underwent a further debulking procedure which revealed widespread metastatic disease involving the liver capsule, splenic hilum, right upper quadrant of the abdomen and recto-vaginal space.

The authors subjected the tissue from the resected specimens to a range of immune-genomic analyses including exome sequencing, RNA expression data, neo-epitope prediction, immunohistochemistry, and in-situ T cell receptor sequencing for tumor infiltrating immune cells.

The sites of metastases (vaginal cuff and liver), which had developed later in the disease-course, after extensive treatment, had the highest tumor mutation and neo-epitope loads, however, no predicted neo-epitopes were found to be shared between regressing and progressing tumors. The differential growth rates at the different metastatic sites was reflected in the propensity for mutations to yield neo-epitopes at different sites, with evidence of immune editing in the regressing right upper quadrant lesion compared to the progressing vaginal cuff and splenic lesions. Immune activation scores (immunophenoscores) and T-cell infiltrate was also higher in regressing lesions than stable or progressing ones. The implications are that while the immune system can successfully prune some tumor branches, others may advance unrestrained from immune micro-environmental predation, consistent with the possibility that genetic heterogeneity might permit immune escape.

Lower immunogenicity scores were found for clonal/truncal somatic mutations than in branched, subclonal, mutations and the primary tumor demonstrated no quantifiable T cell infiltration, as well as negativity for PD-L1 and CD-68. These data mirror recent evidence that clonal neo-epitopes present in every tumor cell are the most potent mediators of immune-predation, and therefore subject to the highest selection pressures to be eliminated (McGranahan et al., 2016). If this is a general phenomenon, this may act as a barrier to the success of immunotherapies in ovarian cancers.

The case study provides evidence of divergent tumor genetics, tumor microenvironments, and immune activation within a single patient and supports a growing body of work suggesting that a single biopsy is insufficient to fully characterize a tumor, its microenvironment and understand its development. The findings, assuming they translate more broadly across patients with this disease, raise several important questions and challenges.

Firstly, the heterogeneity observed in both response and immune infiltration across different sites raises important clinical challenges for the use of cytotoxic-, targeted-, and immunotherapies. The work emphasizes the pressing need to distinguish stable immune parameters from dynamic ones, both in terms of designing optimal immune-based combinations as well as to drive effective companion biomarker development (Topalian et al., 2016). An appreciation of the heterogeneity of immune parameters between different

metastatic sites, including spatial distribution of T cells within and around a tumor and checkpoint molecule expression such as PDL1, will be important in the development of new biomarkers, predictive of response to immune based therapies. Such studies highlight the critical role of extensive tumor sampling of metastatic disease, linked with imaging analyses, to understand this diverse landscape in relation to tumor response. An essential model to study such complexity will be through autopsy analyses of patients with end-stage cancers, in programs such as PEACE in the United Kingdom.

Secondly, the findings presented by Miller and colleagues emphasize the importance of developing a deeper understanding of the impact of treatment on tumor evolution and immune predation. The divergent growth rates at different tumor sites speaks to the need to understand how therapy itself can contribute to heterogeneous responses. The repeated rounds of platinum chemotherapy may directly sculpt the cancer genome by inducing heterogeneous mutations, which may be immunogenic, and it may also act as a selection barrier, altering the evolutionary trajectory and selecting for growth of specific subclones within specific immune environments.

Finally, the results also highlight the need to further explore molecular mechanisms underpinning immune evasion and its relationship with the genomic landscape and extent of heterogeneity. Can branched tumor evolution and diversity within tumors be traced back to a failure of immune predation or do cancer mutational processes, which foster cell-to-cell variation, actively provide a means of immune evasion?

Aristotle, Darwin and Wallace, among others, recognized the importance of the analysis of geographically isolated island populations to developing insights into nature's rulebook and speciation (Stott, 2012). This case study emphasizes the importance of an integrative approach to understanding the molecular mechanisms governing evolution of geographically distinct cell populations manifested as the interaction between the tumor and its immune microenvironment. A deeper understanding of this interplay will be required to harness an adaptive immune system to combat cancer.

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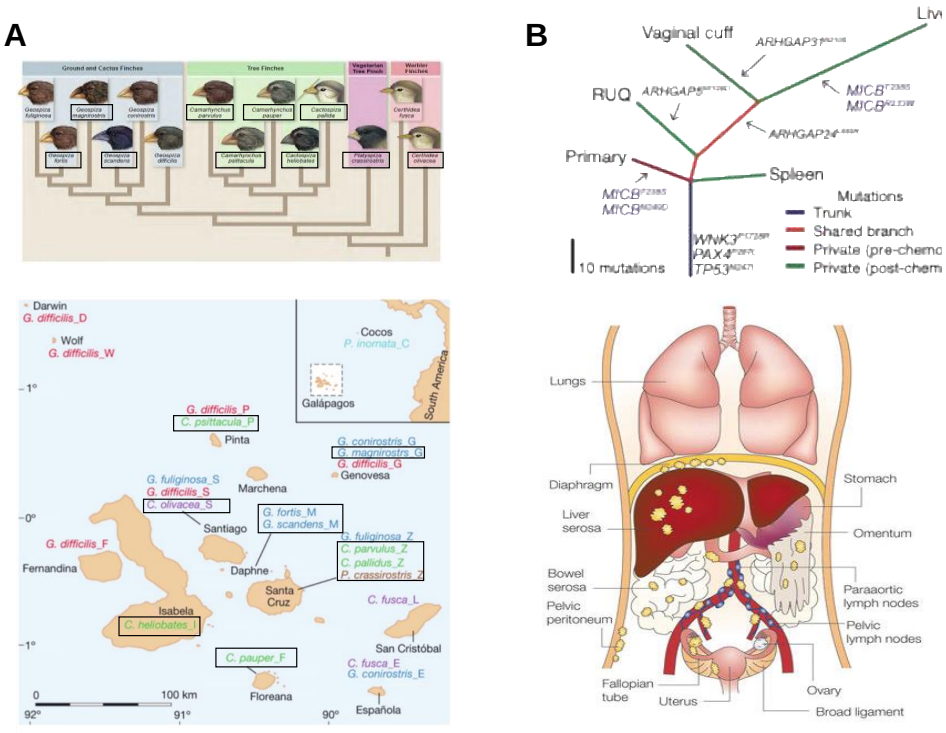

Figure legends

Figure 1 Analysis of geographically isolated populations sheds light on evolution.

A) Darwin's formulation of the theory of evolution by natural selection was influenced by his discovery of several distinct species of finches, adapted to specific ecological niches on different Galapagos islands. The top panel shows the phylogenetic relationship between different finch species, and the bottom panel depicts the different islands where these species are found. For simplicity, only finch species found on only one island are depicted.

B) The analysis of geographically distinct tumor sites and their microenvironmental constraints can aid our understanding of tumor evolution. The bottom panel shows the different tumor sites and the top panel depicts the phylogenetic relationship between cancer cells found at each site. Just as each Galapagos island exhibits specific ecological niches, geographically distinct tumor sites likely exhibit specific immune microenvironments.