Notching tumor: Signaling through Notch receptors improves antitumor T cell immunity

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ABSTRACT
Notch signaling is crucial for lymphocyte effector and memory differentiation. While tumor suppress Notch signaling in antitumor lymphocytes, recent studies show that the pharmacological Delta-like ligand-1 multivalent cluster or proteasome inhibitor bortezomib can restore Notch–NF-κB signaling in T cells of tumor-bearing hosts with a potential to overcome cancer cell resistance to therapy.

Tumors suppress Notch signaling in T lymphocytes to escape from immune surveillance. Three recent studies show that it is possible to restore the cross-regulatory loops of Notch–NF-κB signaling in lymphoid cells of tumor-bearing hosts by Delta-like Notch ligand-1 (DLL1) and proteasome inhibitor bortezomib. Such pharmacological manipulations of host immunity enhanced antitumor T cell responses that could improve the outcome of adoptive cell therapy and avoid resistance of cancer cells to chemotherapeutic agents such as tyrosine kinase inhibitors (TKIs).

Notch is a highly conserved signaling system important for the development and differentiation of multiple organs during embryogenesis. The Notch pathway includes a complex repertoire of four transmembrane Notch receptors (Notch 1–4), five ligands such as Delta-like ligands (DLL)-1, 3, 4 and Jagged-1 and 2 as well as multiple non-canonical signaling partners. Engagement of Notch with its ligands brings about receptor conformational changes that results in the cleavage of Notch intracellular domain (NICD) by γ-secretase leading to the release of NICD and its translocation into the nucleus where it interacts with various transcription factors such as recombination signal binding protein for immunoglobulin κ J region (RBP-Jκ) unleashing the so-called “canonical” signaling pathway. In a “non-canonical” route, NICD drives transcription via other signaling molecules such as NF-κB, PI3K, mTORC2, Akt, β-catenin, etc.

Upon activation, T cells upregulate Notch molecules, whereas the inhibition of Notch signaling results in decreased proliferation and effector function of CD4+ and CD8+ T lymphocytes. Further, Notch enhances IL-2 synthesis and upregulates CD25, the high affinity IL-2 receptor α chain, CD25. Activation of Notch can regulate Th1, Th2, and Th17 differentiation. It can also promote differentiation of naive CD8+ T cells into cytotoxic and memory T lymphocytes via upregulation of the transcription factor Eomesodermin responsible for guiding expression of lytic molecules such as IFNγ, granzymes, and perforins. Therefore, interplay of Notch pathway in the transcription and translation of important cellular proteins effectively translates into regulating the differentiation of immune cells and development of the antitumor immunity (Fig. 1).

Dual roles of Notch in tumor development have been established with tumor-promoting implications in T cell acute lymphoblastic leukemia (T-ALL) and various solid tumors including breast cancer, medulloblastoma, colorectal cancer, non-small cell lung carcinoma, and melanoma. Tumor-suppressive properties of Notch have also been described in a wide variety of cells such as hematopoietic cells, skin, pancreatic epithelium, and hepatocytes. A novel tumor escape mechanism from T cell immunity includes downregulation of DLL1 and DLL4 in hematopoietic microenvironment whereas restoration of the cognate Notch receptor signaling leads to the suppression of tumor.

The study by Biktasova et al.1 extended the relevance of Notch signaling for cancer immunotherapy by demonstrating that Notch agonists may offer therapeutic benefits by overcoming tumor-induced T cell immunosuppression. Authors used DLL1-Fc fusion proteins composed of the extracellular domain of mouse or human DLL1 and the Fc part of mouse IgG2A or human IgG1, respectively. To formulate DLL1 clusters, DLL1-Fc, biotinylated anti-IgG antibodies, and NeutrAvidin were utilized. Authors found that clustered DLL1 administered systemically in Lewis lung carcinoma (LLC)-bearing mice promoted infiltration of T cells into tumors and increased T cell activation.
and function as measured by CD25 upregulation, Th1 differentiation, antigen-specific INFγ production, and the pool of central memory T cells. Further, mice treated with clustered DLL1 displayed diminished tumor vascularization, thereby restricting tumor growth and metastasis. A strong immune response was elicited following adoptive transfer of tumor antigen-specific T cells into immunodeficient SCID-NOD mice bearing palpable tumors. Notably, lymphocytes isolated from DLL1-treated animals attenuated tumor growth in recipients in contrast to naive T cells in the control group showing no sign of therapeutic intervention. Thus, restoration of Notch signaling in antitumor lymphocytes orients the immune response toward strong tumor-inhibitory effects.

Authors further confirmed potential effectiveness of DLL1-based therapy in an EGFRL858R transgenic mouse model, clinically relevant to the condition associated with L858R resistance to EGFR TKI treatment.9,10 This calls for additional therapeutics with the EGFRL858R genotype demonstrated residual disease after TKI treatment.9,10 This calls for additional therapeutics needed to control TKI resistance. In this regard, combined TKI/clustered DLL1 therapy showed promising results with decreased lung tumor burden and significantly improved progression-free survival (40% animals survived beyond 5 weeks in the group of combined therapy versus 100% mice dead by the end of 2nd week in the group of TKI treatment alone). These results correlated with enhanced infiltration of INFγ-producing T cells and CD11b+CD11chigh dendritic cells in lungs of mice, which received combined TKIs/clustered DLL1 therapy.

Despite detailed analysis of the role of Notch activation in resuscitating immune response compromised by tumor growth, mechanistic pathways involved in this process remain unclear. It has been shown in this study that DLL1-mediated activation of immune cells infiltrating tumor is associated with the expression of downstream Notch targets Hes1, Hey1 and Delta1 along with the upregulation of T-bet, a transcriptional mediator of Stat1 necessary for Th1 differentiation (Fig. 1). In other studies investigating the immune-stimulatory potential of anticancer proteasome inhibitor drug bortezomib, our group demonstrated that bortezomib mediates its therapeutic effects by enhancing cross-regulatory loops of Notch1/2 signaling with NF-κB in tumor-infiltrating CD8+ T lymphocytes as revealed by the use of inhibitors of γ-secretase (blocking canonical Notch pathway) and NF-κB (Fig. 1).2,3 Although the molecular mechanisms underlying these effects need to be dissected, these studies highlight the clinical relevance of augmenting non-canonical downstream Notch signaling pathways by pharmacological agents such as DLL1 clusters or bortezomib in adoptive T cell immunotherapies.

Disclosure of potential conflicts of interest
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