All that glitters is not gold: the need to consider desirable and undesirable immune aspects of oncolytic virus therapy

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Oncolytic viruses (OVs), a novel class of anticancer therapeutic agents, can overturn cancer-mediated immunosuppression and initiate antitumor immunity. Contrary to this paradigm, our recent study illustrates that oncolytic reovirus transiently augments cancer-associated immunosuppression immediately following its therapeutic administration. To achieve the optimum efficacy for OV-based anticancer therapies, the pathophysiological as well as clinical implications of this phenomenon need to be considered.

OVs preferentially target and kill cancer cells in a process called oncolysis, and many of them are currently undergoing clinical trials as anticancer therapeutics. In addition to this tumor-killing activity, OVs further invoke a sequence of immunological events that overturn various tumor-induced immunosuppressive mechanisms and ultimately develop tumor-specific immune response. Such OV-induced mechanisms and ultimately develop tumor-specific immune response. OVs further invoke a sequence of immunological events that overturn various tumor-induced immunosuppressive mechanisms and ultimately develop tumor-specific immune response. Such OV-induced antitumor immunity promotes additional cancer cell death and can protect the host against possible tumor relapse. Thus, OVs induce an attack on cancer cells through two distinct mechanisms: direct oncolysis and antitumor immune response.

To evade immune-mediated elimination of cancer cells, tumors have developed a wide range of immunosuppressive strategies to create a tumor microenvironment (TME) that actively resists the development of anticancer immunity. These strategies include modulation of cytokine production and/or response, dysregulation of immune checkpoints, development of tissue barriers, generation of immunosuppressive cells (e.g. regulatory T cells [Tregs], myeloid-derived suppressor cells [MDSCs]), and alterations in tumor antigen processing and presentation. In the context of OV-based oncotherapy, cancer-associated immunosuppression plays contrasting roles. On one hand, it provides an infection-vulnerable niche that promotes replication of the virus. On the other hand, it hampers the establishment of OV-driven antitumor immune responses and represents a major hurdle in the development of a clinically meaningful anticancer immunity. Thus, the strategic management of cancer-associated immunosuppression during all stages of OV-based oncotherapy is of utmost importance.

MDSCs represent key orchestrators of cancer-associated immunosuppression due to their ability to directly suppress innate and adaptive immunity. MDSCs comprise a heterogeneous population of immunosuppressive cells of myeloid origin. In healthy hosts, MDSCs originate from the bone marrow and subsequently differentiate into mature granulocytes, macrophages, or dendritic cells in the periphery. However, under pathological conditions such as cancer, infection, trauma, and sepsis, MDSCs display aberrant differentiation and maturation processes, and accumulate at the site of affliction. With respect to cancer progression, these cells have the remarkable ability to inhibit tumor surveillance and killing by T cells via direct suppression of the latter and indirectly through recruitment of Tregs and depletion of essential nutrients for T cell activity. Furthermore, MDSCs have been reported to regulate innate immune responses either through the regulation of NK cells or cytokine production of macrophages. Currently, two major subpopulations of MDSCs have been widely recognized: monocytic MDSCs (M-MDSCs) and granulocytic/polymorphonuclear MDSCs (G-MDSCs). In mice, these subpopulations are depicted by the variation in the surface expression of CD11b, Gr-1, Ly6G, and Ly6C, where the G-MDSC population displays the CD11b⁺, Gr-1⁺, Ly6G⁺, Ly6Clow phenotype, and the M-MDSC population displays the CD11b⁺, Gr-1⁺, Ly6G⁺, Ly6Chigh phenotype. Importantly, MDSCs with such distinct phenotypes also bear differential functional signatures. For instance, G-MDSCs predominantly suppress antigen-specific T cells via reactive oxygen species (ROS) production. In contrast, M-MDSCs generally suppress effector T cells through high expression of enzymes arginase (ARG1), inducible nitric oxide synthase (iNOS), and reactive nitrogen species. Thus, the elucidation of phenotypic heterogeneity of MDSCs provides further clues on MDSC-mediated immunosuppressive capacities, and guides the
designing of appropriate MDSC-modulating therapeutic interventions.

Recently, we observed that the therapeutic administration of oncolytic reovirus transiently augments cancer-associated immunosuppression in mice with ovarian peritoneal carcinomatosis. We showed that reovirus drives an immediate recruitment and accumulation of highly suppressive CD11b<sup>+</sup>, Gr-1<sup>+</sup>, Ly6Chigh myeloid cells, which differ from the tumor-associated CD11b<sup>+</sup>, Gr-1<sup>+</sup>, Ly6Clow resident myeloid cell population. Additionally, reovirus drives the selective chemotaxis (ex vivo) of the CD11b<sup>+</sup>, Gr-1<sup>+</sup>, Ly6C<sup>high</sup> population in the TME, and promotes the expression of numerous pro-MDSC factors known to be involved in the survival, proliferation, and chemotaxis of MDSCs. Most importantly, CD11b<sup>+</sup>, Gr-1<sup>+</sup>, Ly6C<sup>high</sup> myeloid cells specifically potentiate the suppression of T cell proliferation and cytotoxicity, and are associated with the absence of IFNγ response in the TME early during oncotherapy. Considering the fact that the development of an antitumor T cell response is largely dictated by the accompanying TME as well as the preceding innate immune response, our findings are of critical importance while devising interventions that promote a robust antitumor immunity.

In recent times, anticancer immunotherapeutic potential of OVs (as well as other pathogens including bacteria<sup>6</sup>) has been well appreciated. The interaction of these pathogens with the immune system of the cancer-bearing host is found to induce beneficial antitumor activities that are otherwise absent. However, it should be noted that the outcome of this interaction (pathogen-immune system-cancer) is bound to be different from the outcome of the interaction between pathogen and the immune system of the naïve, non-cancer-bearing hosts (pathogen-immune system). This hypothesis is further supported by the fact that the immune system of the cancer-bearing host has a multitude of phenotypical and functional impairments as compared to the immune system of the naïve host. Of note, similar recruitment of MDSCs during acute viral infections in naïve hosts has been previously described<sup>6,9</sup> and is hypothesized to play a role in either controlling the unnecessary immune-mediated collateral damage to the host or allowing the immune evasion advantage to the virus.

In the context of OV-based cancer therapies, however, virus-mediated suppression of the immune system could be both beneficial (by promoting virus-mediated oncolysis) and detrimental (by mitigating virus-induced antitumor immunity) at the same time. At present, various immune-modulating approaches are being devised with the ultimate objective of enhancing the OV-induced antitumor benefits. Such interventions must also take into consideration the contrasting roles of OV-induced MDSCs during their therapeutic regimens. In this regard, we recently showed that combining reovirus oncotherapy with chemotherapeutic gemcitabine, known to inhibit MDSCs, delays the development of disease pathology and prolongs the survival of ovarian...
cancer bearing hosts. It therefore seems clear that only through the strategic management of such immunosuppressive entities can we promote the optimum efficacy for OV-based oncotherapies (summarized in Fig. 1).

Disclosure of Potential Conflicts of Interest
No potential conflicts of interest were disclosed.

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