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Letter

The Promise and Peril of Natural Killer Cell Therapies in Pulmonary Infection

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The emergence of novel respiratory viruses such as SARS-CoV-2 results in rapid dissemination through virus-naive populations and an accumulation of critically ill patients in hospitals across the globe. In the absence of an effective vaccine or proven treatment, this accelerating threat to human health must be met with swift deployment of experimental therapeutics. Dozens of prospective drugs and other therapies are being tested in clinical trials or applied via off-label or compassionate use protocols for patients with life-threatening complications of infection with SARS-CoV-2. To date, the potential promise for viral control and disease amelioration with each new approach has been balanced by problematic data concerning possible elevated risk for serious adverse events. This is particularly true for natural killer (NK) cells, whose celebrated antiviral capacity and strong safety profile in the oncology clinic may encourage hasty application of these cells against SARS-CoV-2. Critically, a wealth of data in a variety of respiratory infections reveals a frightening proclivity for NK cells to exacerbate lung damage during viral injury. Here, we weigh evidence supporting and discouraging clinical use of NK cells in the present pandemic, as well as in future outbreaks of respiratory pathogens. This assessment aims to provide insights for clinicians considering application of NK cells in the care of SARS-CoV-2 infected patients.

Numerous cellular and biologic therapies harnessing the antitumor capabilities of NK cells have proven both beneficial and safe in treatment of cancer. These approaches include infusion of allogeneic NK-cell products derived from umbilical cord blood, unrelated blood donors, induced pluripotent stem cells, and established cell lines (e.g., NK-92). Adaptive subsets of NK cells, including cytokine-induced memory-like NK cells, exhibit enhanced responsiveness and function in patients (Romee et al., 2016). The antitumor capacity of these therapeutic NK cells can be further enhanced via incorporation of chimeric antigen receptors (CARs) or other engineered components. In the context of numerous hematologic cancers and a handful of solid tumors, NK-cell-based regimens combined with preconditioning (i.e., radiation or chemotherapy) appear highly efficacious with an improved safety profile over parallel approaches using T cells (Liu et al., 2020). The latter feature putatively relates to a reduced capacity of NK cells to trigger cytokine release syndrome (Romee et al., 2016; Liu et al., 2020), a feared complication of CAR T cell approaches. Elevated interleukin-6 (IL-6) production is an important component of both cytokine release syndrome and the harmful cytokine storm elicited during pathogenic SARS-CoV-2 infection (Chen et al., 2020), so the reduced capacity of NK-cell infusions to trigger or amplify these responses is viewed as an advantage over T cell-based therapies to combat severe disease in the present pandemic.

Furthermore, extensive studies provide evidence that early innate functions of NK cells are essential and beneficial in immune defense against respiratory viral infections. These activities include antiviral cytokine production (e.g., interferon [IFN]-γ) and cytolysis of virus-infected cells. At low to intermediate inoculum doses of respiratory syncytial virus (RSV), Sendai virus (parainfluenza virus), and influenza A virus (IAV) infections in mice and hamsters, the activities of NK cells can reduce viral burden and protect from fatal disease (Cong and Wei, 2019).

The relative contributions of conventional NK cells recruited into the lung from the circulation during infection versus the phenotypically unique resident NK cells in human lungs remain undefined. Yet, the hypofunctional status of human-lung-resident NK cells during homeostasis (Marquardt et al., 2017) suggests that persistence of highly active NK cells in the lung may be more harmful than beneficial, potentially worsening lung injury.

Indeed, NK cells can exacerbate lung injury and reduce survival of mice during respiratory infections that are characterized by higher titers of virus and exaggerated inflammatory responses. Exuberant NK-cell activity, including IFN-γ production, contributes to this aggravated lung inflammation during both IAV and RSV infections (Cong and Wei, 2019; Li et al., 2012; Abdul-Careem et al., 2012). Moreover, elevated IL-2 and IL-18 amplify these pathological activities of NK cells during these infections and promote interstitial pneumonia (Okamoto et al., 2002; Harker et al., 2010; McKinstry et al., 2019). Irreversible damage of the lungs by NK cells may be more than just an unfortunate side effect of IFN-γ production, as the robust cytolytic elimination of virus-infected airway epithelial cells by NK cells is a critical antiviral function that may exceed the functional and regenerative capacity of the lung. Of note, the low numbers of NK cells detected in peripheral blood of patients with severe SARS-CoV-2 infections (Wang et al., 2020) may reflect recruitment of pathogenic NK cells to the lungs rather than a true decrease in total NK cell numbers. Indeed, single-cell RNA sequencing of lung bronchoalveolar lavage fluid (BAL) demonstrated higher frequencies of NK cells in the lungs of patients with severe SARS-CoV-2 infections (Liao et al., 2020). In total, the potential for NK-cell based therapies to cause substantial harm to the lungs may outweigh the potential benefits of the possible antiviral activities of these cells.

At the time of this writing, three global trials incorporating NK-cell based cellular therapies have been initiated. These include infusion of allogeneic NK cells (https://clinicaltrials.gov/ct2/show/NCT04344548), placenta-derived (Kang et al., 2013) cord blood NK cells (https://clinicaltrials.gov/ct2/show/NCT04280224), and NK cells bearing an innovative CAR designed to engage the SARS-CoV-2 via its putative cellular receptor (ACE2) (https://clinicaltrials.gov/ct2/show/NCT04324996). The reasonable desire to treat patients with severe SARS-CoV-2 infections using these experimental therapies will conflict with the need to learn as much as possible about their safety. The timelines of these experimental studies are compressed (<2 months) and will be carried out on only a few severely ill patients. This may make it challenging to distinguish harmful treatment outcomes from a general lack of efficacy if trial participants succumb to disease. Thus, while hopes remain high that the potent antiviral functionality of these NK cells will provide clinical benefit, the potential for damage to the lungs, liver, and kidneys should be serious concerns and monitored very carefully in these trials.

Importantly, the timing of therapeutic administration of NK cells will also dictate the balance between their beneficial antiviral and detrimental pathologic effects. During Sendai parainfluenza virus infection of mice, early infusion of NK cells partially controlled low-dose infection, whereas therapy initiation at a later stage of infection increased viral replication and associated morbidity (Mostafa et al., 2018). Similarly, patient-specific factors are likely to impact the efficacy of NK-cell based therapeutics, as severe SARS-CoV-2 infection is associated with hypoxia and elevated IL-6 (Chen et al., 2020), which can significantly impair the function of NK cells (Cifaldi et al., 2015). The potential role of elevated IL-6 in poor outcome of SARS-CoV-2 infections has even led to clinical trials investigating the utility of drugs that inhibit IL-6 (e.g., tocilizumab; https://clinicaltrials.gov/ct2/show/NCT04335071) or JAK signaling (e.g., tofacitinib; https://clinicaltrials.gov/ct2/show/NCT04332042) in the treatment of critical illness in SARS-CoV-2 patients. Of note, tofacitinib and related JAK inhibitors can reduce the number and function of NK cells, highlighting how clinical benefit of such drugs could stem, in part, from depletion of potentially pathogenic NK cells. A final consideration in translation of the clinical success of NK cells in oncology to treatment of severe SARS-CoV-2 infections lies in the paucity of information regarding both efficacy and safety of such NK-cell infusions performed in the absence of the lymphodepleting preconditioning regimens commonly coupled to this approach in cancer therapy.

These data and observations highlight the complexities of using NK-cell-based therapies in the treatment of current and future pandemic respiratory viral infections. Data from each case, including lung-specific viral burden and cytokine measurements before and after therapy, will be critical for the interpretation and future implementation of NK-cell therapeutics. Trial design and candidate selection for NK-cell infusion in SARS-CoV-2 patients should be carefully considered and should weigh potential canonical antiviral benefits against the elevated risk for pathogenic consequences of NK cells in the context of severe respiratory viral infection. In a time of true uncertainty, with vulnerable and desperate patients who are willing to accept almost any experimental therapy, it is critical for the scientific and medical communities to ensure that the scientific support for novel interventions is strong, the risks and benefits of novel therapies are well balanced, and trial protocols are robust and well monitored. Patients are not currently able to live by the ancient saying *caveat emptor* (let the buyer beware), so the scientific community must do so for them.

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