COVID and Immunotherapy

INCIDENTAL FINDING OF COLORECTAL CANCER IN A COVID-19 PATIENT, FOLLOWED BY DEEP PROFILING OF SARS-COV-2-ASSOCIATED IMMUNE LANDSCAPE AND TUMOUR MICROENVIRONMENT

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Background

Reports suggest that cancer patients may be more vulnerable to COVID-19, with increased disease severity and higher mortality rate.1–3 Although this is likely multifactorial, the exact pathogenesis has not been clearly elucidated. Studies have shown increased ACE2 expression in tumours as compared to normal tissues,4–5 thereby providing increased viral binding. Moreover, other mechanisms of cancer immunotherapy including treatment- and disease-related immunosuppression and functional exhaustion have been reported in patients with concomitant cancer and COVID-19; contributing to greater COVID-19 disease severity.6–8 There is still much to be revealed about the interplay between COVID-19, cancer and the immune system. These insights will give us greater understanding of the immunopathological processes underlying COVID-19 in cancer patients and their clinical relevance.

Methods

A 45-year-old South Asian male diagnosed with COVID-19, with incidental discovery of stage II T3N0 caecal adenocarcinoma was consented for our study. The patient had experienced mild symptoms throughout the course of the disease, and underwent laparoscopic right hemicolectomy 10 days after recovery from COVID-19. His blood, lymph nodes, normal tissue and tumour samples were obtained for further analysis (figure 1). Multiplex immunohistochemistry was performed to understand SARS-CoV-2-associated tumour immune microenvironment. Moreover, to simulate ex vivo SARS-CoV-2 infection, dissociated cells from blood, lymph nodes, and tissue samples were stimulated with SARS-CoV-2 peptides or control for 16 hours. This was followed by 25-colour flow cytometry analysis for immune markers and cytokines. We then compared unstimulated with stimulated cells to study SARS-CoV-2-elicited immune response.

Results

Multiplex immunohistochemistry demonstrated upregulated expression of ACE2 in the tumour as compared to adjacent normal tissue, whilst SARS-CoV-2 was detected only in adjacent normal tissue but not within the tumour (figure 2). We also observed SARS-CoV-2 in other organs such as appendix and lymph nodes; and the presence of tertiary lymphoid structure, abundant T cells and NK cells within the proximity of the tumour (figure 2). Additionally, upon stimulation with SARS-CoV-2 peptides, we successfully elicited SARS-CoV-2-specific CD4+ T cells expressing immune markers such as granzyme B, TNF-α and IFN-γ (figure 3). Deep profiling of
Background CD73, an ecto-5’-nucleotidase involved in ATP metabolism, converts AMP into adenosine. ATP could induce production of interferon-beta, which induces cellular resistance to viral infection and triggers apoptosis of virus-infected cells. In COVID-19, SARS-CoV-2 causes severe respiratory syndrome by effectively inhibiting interferon activity, leading to impaired anti-viral response. Moreover, lung injury seen in severe COVID-19 patients might be caused by excess adenosine. Inhibition of CD73 is believed to increase extracellular ATP, thereby countering COVID-19. Furthermore, independent of its inhibitory effects on CD73, preclinical results from other anti-CD73 mAb suggested CD73 blockade activates lymphocytes, induced antibody production from B cells and enhanced lymphocyte trafficking, thereby, stimulated the production of anti-SARS-CoV-2 antibodies leading to the rapid clearance of the virus. We developed AK119, a humanized monoclonal antibody targeting CD73, as an immunotherapy agent for the treatment of COVID-19.

Methods Evaluation of AK119 activity to bind to the CD73 antigen was performed by using ELISA, Forbebio, and FACS assay. The activity of AK119 to inhibit enzymatic activity of CD73 was evaluated by cell-based enzyme assay; and the activity of AK119 to induce internalization of CD73 and enhance CD69 and CD83 expression on B cell were performed by using FACS assays. We also investigated the potential of AK119 to promote immunoglobulin production from human B cells.

Results AK119 could effectively bind to human CD73 with high affinity, which is comparable or superior to 10.3AA, a leading anti-CD73 antibody, in protein-based assays. AK119 inhibited CD73 enzymatic activity on MDA-MB-231 cells (IC50_AK119 27.60 nM; IC50_10.3AA 15.99 nM) and U87-MG cells (IC50_AK119 0.2448 nM; IC50_10.3AA 0.0691 nM), with a higher maximal inhibition rates of 108.26% in MDA-MB-231 cells and 96.24% in U87-MG cells compared with 10.3AA (77.02% and 75.77%, respectively). AK119 effectively induced CD73 internalization in MDA-MB-231 cells and U87-MG cells, and the internalization rate of CD73 was 60.75% and 82.39%, respectively; for 10.3AA, the internalization rate was 50.53% and 73.65%, respectively. Moreover, AK119 could stimulate approximately 4-5 fold up-regulation of CD69 (figure 1A) and CD83 (figure 1B) that are markers of B cell activation; and, AK119 significantly promoted IgG production from B cells.

Conclusions In summary, in comparison to a leading CD73 antibody currently in clinical trial, AK119 demonstrated more complete CD73 inhibition; and more dramatic B cell activation and antibody production. Thus, AK119 presented desirable preclinical activities. The safety and pharmacokinetics of single ascending doses of AK119 will be evaluated in healthy volunteers in an upcoming Phase 1, First-in-Human study (NCT04516564).

Trial Registration NCT04516564

REFERENCES


