IN BRIEF

⤷ **COVID-19**

Crossreactivity not so helpful after all?

Crossreactive CD4+ T cells to SARS-CoV-2, thought to originate from immune responses to common cold coronaviruses (CCCs), have been reported in up to 80% of SARS-CoV-2-naive individuals. This led to the hypothesis that encounters with CCCs may provide a degree of cross-protective immune memory. Now, a study in *Immunity* shows that SARS-CoV-2-crossreactive CD4+ T cells can be detected in almost all individuals tested and that these cells generally have a low functional avidity. At the same time, the authors identified highly expanded populations of low-avidity CD4+ T cells as a hallmark of severe COVID-19. This challenges the idea of a protective function of crossreactive CD4+ T cells and even raises the possibility that these cells may contribute to the risk of developing severe COVID-19. However, given the correlative nature of the study, causal links remain to be verified.

ORIGINAL ARTICLE Bacher, P. et al. Low-avidity CD4+ T cell responses to SARS-CoV-2 in unexposed individuals and humans with severe COVID-19. *Immunity* **53**, 1258–1271 (2020)

⤷ **COVID-19**

Genetic clues for predisposition to severe disease

The course of disease in individuals infected with SARS-CoV-2 is hugely variable, ranging from asymptomatic infections to severe COVID-19 and death. The GenOMICC genome-wide association study, published in *Nature*, now identifies significant associations of severe disease with several genes involved in antiviral defence mechanisms or in host-driven inflammatory lung injury. These include a cluster of genes that encode antiviral restriction enzyme activators (*OAS1*, *OAS2* and *OAS3)*, *TYK2*, encoding a tyrosine kinase, the dipeptidyl peptidase gene *DPP9* and the interferon receptor gene *IFNAR2*. Mendelian randomization revealed a causal link between severe disease, low expression of *IFNAR2* and high expression of *TYK2*. Moreover, a transcriptome-wide association study showed the monocyte/macrophage chemotactic receptor CCR2 is associated with severe COVID-19. These findings indicate opportunities for the potential repurposing of existing drugs.

ORIGINAL ARTICLE Pairo-Castineira, E. et al. Genetic mechanisms of critical illness in Covid-19. *Nature* https://doi.org/10.1038/s41586-020-03065-y (2020)

⤷ **COVID-19**

Deciphering the protective features of the antibody response

The serological features that determine clinical outcomes in patients with COVID-19 are currently ill defined and there has been considerable controversy regarding the duration of antibody responses to SARS-CoV-2. A study in *Science Immunology* now reports a longitudinal investigation of plasma samples from 79 hospitalized patients with COVID-19, as well as 175 outpatients and asymptomatic SARS-CoV-2-positive individuals. Overall, outpatients and asymptomatic individuals had higher ratios of spike protein receptor-binding domain-specific IgG versus nucleoprotein-targeted IgG antibodies than hospitalized patients. In hospitalized patients, increases in antibody titres correlated with decreases in viral titres, but antibody responses during acute illness were insufficient to predict outcomes. In all patients, antibody titres started to wane around 1 month after disease onset.

ORIGINAL ARTICLE Röltgen, K. et al. Defining the features and duration of antibody responses to SARS-CoV-2 infection associated with disease severity and outcome. *Sci. Immunol.* **5**, eabe0240 (2020)

⤷ **AUTOIMMUNITY**

Targeting pathogenic T cells by metabolic checkpoint inhibition

CD4+ T cells that react inappropriately to microbiota antigens are thought to drive pathology of inflammatory bowel disease. A study in *Science Immunology* describes a new approach to selectively eliminate these microbiota-specific pathogenic T cells by activating them in the presence of metabolic inhibition.

Naive and memory CD4+ T cells undergo a profound metabolic transition to aerobic glycolysis when stimulated through the T cell receptor (TCR) that supports their activation and expansion. This metabolic checkpoint is primarily controlled by the mammalian target of rapamycin (mTOR) complex. Zhao et al. hypothesized that interfering with this metabolic checkpoint during T cell activation — using the mTOR inhibitor rapamycin or metformin to activate the negative regulator of mTOR, 5′-AMP-activated protein kinase — would lead to death or anergy of pathogenic naive and memory CD4+ T cells.

To investigate this in the setting of experimental colitis, the authors engineered a multi-epitope peptide (MEP) comprising multiple flagellin peptides from commensal bacteria that could activate flagellin-specific TCR-transgenic CD4+ T cells. Application of this MEP to transgenic CD4+ T cells in vitro together with rapamycin inhibited mTOR signalling, reduced glucose uptake and promoted cell death of more than 90% of proliferating cells. Moreover, concomitant MEP and rapamycin favoured differentiation of the remaining CD4+ T cells into regulatory T (T_{reg}) cells, which were shown to provide antigen-specific and bystander suppressive effects.

To test the effects of rapamycin in vivo, colitis was induced by adoptive transfer of flagellin-specific TCR-transgenic CD4+ T cells

⤷ **COVID-19**

Immune readouts from the Oxford COVID-19 vaccine

The vast majority of COVID-19 candidate vaccines are designed to target the SARS-CoV-2 spike (S) protein, but the precise vaccine-mediated immune correlates of protection remain to be determined. Two recent reports from the Oxford COVID-19 vaccine team detail the immune outcomes observed in a phase I/II trial of their ChAdOx1 nCoV-19 vaccine, in which volunteers received a single standard dose or various two-dose regimens.

The ChAdOx1 nCoV-19 vaccine comprises a non-replicating chimpanzee adenovirus vector (ChAdOx1) that is genetically modified to express the full-length S protein of SARS-CoV-2. Trial participants were healthy adults aged between 18 and 55 years, with the paper by Ewer et al. describing the immune responses seen in 88 individuals who received either a single dose of ChAdOx1 nCoV-19 or a control vaccine. The paper by Barrett et al. details immune responses in 52 volunteers who were vaccinated with a standard dose of ChAdOx1 nCoV-19 and then received a standard dose (*n* = 20) or half-dose (*n* = 32) booster 56 days later. Previously published data on trial participants who received two standard doses 28 days apart were also included for comparison.

A key finding in the single-dose paper is that a sole vaccination induced S-protein-reactive CD4+ T and CD8+ T cells with a T helper 1 (T_H1)-type cytokine bias as well as CD8+ T cells with a cytotoxic phenotype. This is important as T_H1-type immunity is thought to mediate protective antiviral immunity whereas T_H2-type responses have been linked with potentially adverse vaccine effects.

to *Rag1*^{−/−} mice. Mice receiving MEP activation plus rapamycin were fully protected from the development of colitis, and this protection was associated with a significant reduction in the number of effector CD4^{+} T cells in the colon compared with control mice. Rapamycin treatment also prevented the formation of antigen-specific memory T cells when given on initial antigen encounter, as well as the differentiation of T follicular helper (T$_{FH}$) cells, but favoured the production of T$_{reg}$ cells.

By combining rapamycin and metformin, the authors achieved an even greater inhibitory effect, being able to block the expansion of pre-existing memory T cells and T$_{FH}$ cells following secondary antigen encounter and shift the remaining population to a regulatory phenotype. RNA sequencing of cells treated with MEP plus metabolic inhibition confirmed the effects at the level of the transcriptome, revealing a downregulation of pro-proliferation and pro-inflammatory genes and an upregulation of pro-apoptotic genes.

Finally, the observation that flagellin-specific memory CD4^{+} T cells isolated from patients with Crohn's disease were similarly inhibited by rapamycin and metformin during antigen restimulation supports this approach as a potential therapy for inflammatory bowel disease.

Lucy Bird

ORIGINAL ARTICLE Zhao, Q. et al. CD4^{+} T cell activation and concomitant mTOR metabolic inhibition can ablate microbiota-specific memory cells and prevent colitis. *Sci. Immunol.* **5**, eabc6373 (2020)

Robust B cell activation and proliferation were also observed after a single dose and anti-S protein IgG (predominantly the T$_H$1-associated IgG1 and IgG3 isotypes) were detected by day 14 and maintained at day 56. Notably, these antibodies showed neutralizing activity against SARS-CoV-2 and their avidity for the S protein increased between days 28 and 56. A single vaccination also induced S protein-specific IgM and IgA. No sex-specific or age-related differences in vaccine responses were observed.

The two-dose paper shows that a second vaccination enhances the titres of anti-S antibodies and their neutralizing activity and further promotes T$_H$1-type T cell responses. Moreover, the booster dose enhances the functional capacity of anti-S antibodies to support antibody-dependent phagocytosis, complement deposition and natural killer cell activation, which have been linked with protective immunity in preclinical studies and with better survival of hospitalized patients with COVID-19. Boosting with a half dose was found to be less effective than a standard dose boost, but giving a second standard dose at day 56 had a similar immune-enhancing effect to a second standard dose given at day 28. Importantly, the second dose of the vaccine was shown to be safe and, in fact, better tolerated than the prime dose. This contrasts to what has been observed for booster shots with other COVID-19 vaccines.

The authors conclude that a two-dose regimen for the vaccine is more effective at promoting immunity to SARS-CoV-2 and also likely to be well tolerated. Moreover, these data suggest that the booster dose should still be effective if delivered at 8 weeks after the initial vaccination.

Yvonne Bordon

ORIGINAL ARTICLES Barrett, J. R. et al. Phase 1/2 trial of SARS-CoV-2 vaccine ChAdOx1 nCoV-19 with a booster dose induces multifunctional antibody responses. *Nat. Med.* https://doi.org/10.1038/s41591-020-01179-4 (2020) | Ewer, K. J. et al. T cell and antibody responses induced by a single dose of ChAdOx1 nCoV-19 (AZD1222) vaccine in a phase 1/2 clinical trial. *Nat. Med.* https://doi.org/10.1038/s41591-020-01194-5 (2020)

⮞ **PREPRINT WATCH**

Sensing our Z-RNA

Our cells contain an increasingly appreciated repertoire of receptors that identify and respond to foreign nucleic acids, usually following viral infection. How are these receptors prevented from inadvertently responding to unusual forms of host nucleic acids and triggering unwanted type I interferon responses? One key mechanism depends on adenosine deaminase acting on RNA 1 (ADAR1), which marks endogenous double-stranded RNA (dsRNA) as 'self' by editing adenosine to inosine to avoid cytosolic sensing by the RNA sensor melanoma differentiation-associated protein 5 (MDA5). ADAR1 edits dsRNA in its canonical form, but the Zα domain of ADAR1 can also recognize Z-RNA, a left-handed conformer derived from short interspersed nuclear elements (SINEs), which are non-coding retrotransposons that constitute a large part of the human genome. Two simultaneous preprints (non-peer-reviewed) now describe the importance of ADAR1 in recognizing Z-RNA.

The preprint by de Reuver et al. shows that spontaneous MDA5-dependent activation of the type I interferon pathway occurs when ADAR1 is unable to recognize Z-RNA. The authors were able to dissect the Z-RNA-binding function of ADAR1 by modifying the Zα domain using carefully engineered mutations that mimic the loss of ADAR1 function observed in patients with Aicardi-Goutières syndrome but leave the protein structure unaffected.

Defective binding of Z-RNA to ADAR1 exclusively activated the MDA5–mitochondrial antiviral-signalling protein (MAVS) pathway, leading to spontaneous type I interferon production in both a human cell line and mice expressing ADAR1 mutated in the Zα domain. The authors show that, by losing the ADAR1–Z-RNA interaction, the capacity of ADAR1 to edit Z-RNA is also lost, suggesting cytosolic sensing of unedited SINE-derived dsRNAs.

Another preprint by Tang et al. draws similar conclusions, but also distinguishes between paracrine and autocrine effects in different cell types for the interferon signature found in ADAR1 Zα domain mutant mice, and suggests that neutrophils may initiate the type I interferon response. In both preprints, the spontaneous interferon response when ADAR1 is unable to recognize Z-RNA confers greater protection against viral infections.

Together, these preprints highlight the importance of the intracellular processes that mask self-nucleic acids from cytosolic sensors in the complex, precise and tight regulation of the type I interferon response. In addition, exploring how by-products from the transcription of retroelements are modulated is important for epigenetic therapy, which has been shown to stimulate SINE expression.

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ORIGINAL ARTICLES de Reuver, R. et al. ADAR1 interaction with Z-RNA promotes editing of endogenous double-stranded RNA and prevents MDA5-dependent immune activation. Preprint at *bioRxiv* https://doi.org/10.1101/2020.12.04.411702 (2020) | Tang, Q. et al. Recognition of Z-RNA by ADAR1 limits interferon responses. Preprint at *bioRxiv* https://doi.org/10.1101/2020.12.04.411793 (2020)
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