Comment

Host-directed therapy: tuberculosis vaccine development

The age-old BCG is the only licensed vaccine for tuberculosis despite its extensive and universal use over eight decades. Tuberculosis vaccine development is a major challenge, with 15 different candidate vaccines in phase 1 and 2 trials.[1]

In *The Lancet Respiratory Medicine*, Birahim Pierre Ndiaye and colleagues[2] assess the safety, immunogenicity, and efficacy of a candidate tuberculosis vaccine, modified vaccinia virus Ankara expressing antigen 85A (MVA85A), in adults infected with HIV-1. The authors reported only a low incidence of side effects (one case of tuberculous meningitis possibly related to vaccination). Although interferon γ producing CD4-positive T cells specific for Ag85A peaked after 7 days of vaccination, the authors did not observe a significant decrease in *Mycobacterium tuberculosis* infection among vaccinees.

Vaccine efficacy is multifactorial, and includes the immunological history of previous exposure to *M tuberculosis*; the nutritional status, due to its effect on T-cell metabolism and functionality;[3] age;[3] multiple co-infections, in particular HIV that affects cellular and humoral immune responses;[4] comorbidity with non-communicable diseases; and the type of vector used for vaccination. The nature of the immunising antigen itself plays an important role, because Ag85A is expressed by mycobacteria other than tuberculosis, to which exposure is generally common.

Whether a single antigen is able to generate an immune response effective enough to curb *M tuberculosis* is questionable. Is Ag85A a clinically relevant target antigen to contain or kill off *M tuberculosis*? *M tuberculosis* bacteria contain about 4500 possible target proteins. Ag85A and Ag85B belong to the (secreted) mycolyltransferase family of enzymes. Ag85B has been implicated as a decoy antigen that deviates anti-*M tuberculosis* directed immune responses. Several studies showed that targeting single tumour-associated antigens can be clinically relevant because they can refocus the cellular immune response and facilitate immune responses directed against (unknown) target antigens: the initial driving T-cell response (specific for the immunising antigen) leads to an immunological attack, subsequently resulting in the release of immunogenic material that enables the generation of novel T-cell epitopes. This mechanism, termed determinant spreading, provides the framework of a clinically relevant complex immune recognition, initiated by a single T-cell epitope in patients responding to cancer vaccines.[5]

Perhaps we can learn more from the MVA-initiated target responses from other clinical research areas. A phase 1 trial involving MVA encoding the Epstein-Barr virus antigens EBNA-1 and LMP2 showed that pre-existing anti-MVA immune reactivity did not affect the efficacy of MVA–Epstein-Barr virus-induced immune responses.[6] Similarly, expansion of (pre-existing) CD8-positive T cells against MUC-1, a protein expressed on tumour cells, led to better outcomes in patients with non-small-cell lung cancer.[7] Antitumour cellular and humoral immune responses concomitant with patient survival have also been noted for TroVax, an MVA-vectored vaccine that targets the oncofetal antigen 5T48.[8]

There are commonalities and differences in these approaches. First, immune responses against tumour-associated antigens are low since the vaccination efforts target non-mutated, subdominant antigens. This low response is not an issue for Ag85A, which shows a strong pre-existing immune reactivity.

Second, CD8-positive T-cell responses seem to be elicited after MVA-guided vaccination in patients with cancer.[6,7] However, in Ndiaye and colleagues' study[2] only CD4-positive T-cell-derived interferon γ responses could be detected, at least as defined by intracellular cytokine staining and enzyme-linked immunospot analysis. The frequency of CD4 T cells producing interleukin 17, interleukin 2, and tumour necrosis factor α was low; the dominant T-cell responses were in effect monofunctional. There might be at least one T-cell parameter missing, namely cytotoxic T cells producing the cytolytic molecules granzyme and perforin. This omission could be particularly relevant in individuals with HIV, since studies showed that cytotoxic T-lymphocyte responses in patients with HIV co-infection can be mutually exclusive (ie, they either produce interferon γ or cytotoxic molecules).[9,10] Although the clinical relevance of cytotoxic T lymphocytes in tuberculosis is debated, their clinical significance is underlined by the observation that depletion of CD8-positive T-cell responses affects the onset of active tuberculosis.[9,11]

Third, a similarly debated topic is the induction of humoral immune responses by vaccination. Although a cellular immune response is desirable, anti-tuberculosis antibody responses could help to augment

cellular immune responses while enhancing antigen cross-presentation.[10] This effect has been shown for (intracellular) cancer antigens, where intravenous administration of a monoclonal antibody targeting the nuclear antigen NY-ESO-1 displayed clinical benefit.[12] Ndiaye and colleagues[2] did not identify substantial humoral immune responses after MVA85A vaccination, with only three of 320 vaccinees (about 1%) showing raised concentrations of anti-Ag85A serum antibodies. At this point, it is unclear whether this weak humoral response is related to the nature of the vector, the antigen, or a more general impairment of B-cell responses in HIV-positive individuals.[4]

Fourth, the immune cell repertoire available for T-cell responses is crucial. Vaccines can only expand specific T-cell populations present at the time of vaccination. This effect could be limited by the reduced T-cell receptor repertoire in people infected with HIV,[13] which might be paralleled in patients with cancer-induced and chemotherapy-induced lymphopenia. The availability of fewer T cells translates to less competition for anti-apoptotic cytokines (ie, interleukin 7 and interleukin 15), enabling antigen-specific T cells to expand and acquire immune effector functions.[14]

Ndiaye and colleagues[2] show that MVA85A vaccination in patients with HIV is safe, and induces a predominantly monofunctional CD4-positive T-cell response that peaks about 7 days after vaccination. However, they state that the study was underpowered to assess efficacy, and the flavour (ie, the the functional profile) of MVA85A vaccination-induced cellular immune response is one of the lessons learned. A pivotal exercise in the art of developing vaccines against complex pathogens for individuals with a complex immunological background (ie, multiple *M tuberculosis* exposures, co-infections, non-communicable disease comorbidity) is to develop robust and clinically relevant platforms that capture these multifaceted factors. Ndiaye and colleagues[2] have achieved this, and this work will undeniably aid the design of future tuberculosis vaccine trials. Furthermore, manipulation of the host immune response at the time of vaccination (eg, by removal of regulatory T cells) is worthwhile in view of its success in vaccine trials against self antigens.[15] This approach is just one facet of how simple modifications can gear vaccine-induced immune responses. Therefore, the study by Ndiaye and colleagues[2] provides a valuable starting point to strategically tailor the immune response, with the caveat that we are still left to our own devices until we can formulate the ideal profile of a clinically relevant and protective anti-tuberculosis specific immune response.

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1 Kaufmann SH, Lange C, Rao M, et al. Progress in tuberculosis vaccine development and host-directed therapies—a state of the art review. *Lancet Respir Med* 2014; **2:** 301–20.

2 Ndiaye BP, Thienemann F, Ota M, et al. Safety, immunogenicity, and efficacy of the candidate tuberculosis vaccine MVA85A in healthy adults infected with HIV-1: a randomised, placebo-controlled, phase 2 trial. *Lancet Respir Med* 2015; published online Feb 26. http://dx.doi.org/10.1016/S2213-2600(15)00037-5.

3 Pearce EL, Poffenberger MC, Chang CH, Jones RG. Fueling immunity: insights into metabolism and lymphocyte function. *Science* 2013; **342:** 1242454.

4 Ashenafi S, Aderaye G, Zewdie M, et al. BCG-specific IgG-secreting peripheral plasmablasts as a potential biomarker of active tuberculosis in HIV negative and HIV positive patients. *Thorax* 2013; **68:** 269–76.

5 Butterfield LH, Ribas A, Dissette VB, et al. Determinant spreading associated with clinical response in dendritic cell-based immunotherapy for malignant melanoma. *Clin Cancer Res* 2003; **9:** 998–1008.

6 Hui EP, Taylor GS, Jia H, et al. Phase I trial of recombinant modified vaccinia Ankara encoding Epstein-Barr viral tumor antigens in nasopharyngeal carcinoma patients. *Cancer Res* 2013; **73:** 1676–88.

7 Ramlau R, Quoix E, Rolski J, et al. A phase II study of Tg4010 (Mva-Muc1-Il2) in association with chemotherapy in patients with stage III/IV non-small cell lung cancer. *J Thoracic Oncol* 2008; **3:** 735–44.

8 Amato RJ, Stepankiw M. Clinical efficacy of TroVax in the treatment of progressive castration-resistant prostate cancer. *Clin Med Insights Oncol* 2012; **6:** 67–73.

9 Brighenti S, Andersson J. Induction and regulation of CD8+ cytolytic T cells in human tuberculosis and HIV infection. *Biochem Biophys Res Commun* 2010; **396:** 50–57.

10 Chan J, Mehta S, Bharrhan S, et al. The role of B cells and humoral immunity in *Mycobacterium tuberculosis* infection. *Semin Immunol* 2014; **26:** 588–600.

11 Appay V, Nixon DF, Donahoe SM, et al. HIV-specific CD8(+) T cells produce antiviral cytokines but are impaired in cytolytic function. *J Exp Med* 2000; **192:** 63–75.

12 Sabbatini P, Tsuji T, Ferran L, et al. Phase I trial of overlapping long peptides from a tumor self-antigen and poly-ICLC shows rapid induction of integrated immune response in ovarian cancer patients. *Clin Cancer Res* 2012; **18:** 6497–508.

13 Baum PD, Young JJ, Schmidt D, et al. Blood T-cell receptor diversity decreases during the course of HIV infection, but the potential for a diverse repertoire persists. *Blood* 2012; **119:** 3469–77.

14 Dudley ME, Wunderlich JR, Robbins PF, et al. Cancer regression and autoimmunity in patients after clonal repopulation with antitumor lymphocytes. *Science* 2002; **298:** 850–54.

15 Walter S, Weinschenk T, Stenzl A, et al. Multipeptide immune response to cancer vaccine IMA901 after single-dose cyclophosphamide associates with longer patient survival. *Nat Med* 2012; **18:** 1254–61.