B cell responses in Recurrent Respiratory Papillomatosis patients treated with DNA immunotherapy INO-3107

Authors: Saba N, Sylvester AJ, Reuschel EL, Marcus SA, Wisotsky S, Dolgoter A, Reed KS, Tan GS, Skolnik JM, Gillespie EB.

Background: Recurrent respiratory papillomatosis (RRP) is a chronic, debilitating disease of the airway primarily caused by infection with human papillomavirus (HPV) types 6 and/or 11 and characterized by recurrent, benign tumor growth with potential for malignant transformation. Current standard of care consists of repeated surgical removal of papillomas, which can lead to lasting airway damage and impaired vocal function. Thus, a non-surgical approach to treat RRP is paramount. Previously, we described T cell responses associated with an overall clinical response rate of 81% (26/32) to INO-3107, a DNA immunotherapy designed to generate T cells capable of targeting HPV-infected cells, in adult RRP patients during a Phase 1/2 trial (NCT04398433). Here, we describe B cell responses in these patients.

Methods: INO-3107 was administered during study weeks 0, 3, 6 and 9. Peripheral blood mononuclear cells (PBMCs) were obtained at screening/day 0, weeks 6, 9, 11, 26 and 52. Formalin-fixed, paraffin-embedded papilloma tissue was obtained prior to INO-3107 treatment (Scr) and at the end of the 52-week study (EOS). Both PBMCs and tissue were subjected to RNA and BCR sequencing, which additionally underwent single sample gene set enrichment analysis and CloneTrack analysis, respectively. Clinical response was defined as any reduction in frequency of RRP surgical interventions in the 52 weeks following dose 1 of INO-3107 (Y1) compared to the 52 weeks prior.

Results: Following INO-3107 treatment, 85% (23/27) of patients exhibited B cell expansion in PBMCs that was sustained through Y1 for responders, which in contrast began to contract at week 26 for non-responders. Enrichment of B cell signatures, inclusive of total, naïve, memory, and plasma b cells, in papilloma tissue increased significantly by EOS in responders compared to non-responders. Increases in tissue BCR clone counts from Scr to EOS correlated significantly with clinical response during Y1. BCR sequences detected in tissue taken at EOS displayed a low degree of overlap with those detected at Scr. Newly detected BCR sequences in EOS tissue were present in PBMCs prior to EOS. Some of these B cell clones were detectable in PBMCs and papilloma tissue only after INO-3107 treatment.

Conclusions: B cell immunogenicity may play a role in mediating clinical responses to DNA immunotherapy INO-3107 for the treatment of RRP. INO-3107 treatment induced expansion and emergence of B cells in the blood of RRP patients, which trafficked to and infiltrated papilloma tissue by EOS as evidenced by increased B cell enrichment and clone counts. These immune responses were associated with improved clinical outcomes. These results suggest that INO-3107 engages B cells equipped with the potential to promote activation of T cell responses described

previously (doi: 10.1038/s41467-025-56729-6) and that they may play a role in long-term immunity against RRP.