Systemic Immune Responses Induced After Immunization with HSV-2 Antigens Serve as Surrogates for Responses in the Murine Genital Tract

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ABSTRACT

GEN-003 is a subunit vaccine currently in phase 2 clinical trials for immunotherapy of HSV-2-induced genital herpes. Here we describe several methods to assess both humoral and T cell responses in murine genital tracts that can further characterize GEN-003 immunogenicity and support future vaccine development.

Extending our assessment of humoral responses by quantifying serum IgG responses, we developed a miniaturized ELISA method that measures both IgG and secretory IgA antibodies in vaginal lavages of immunized mice. In addition, we quantified with GEN-003 and adjuvant GB1A1, mucosal IgG antibodies specific for both gD and ICAP antigens were induced at comparable levels, mirroring systemic IgG responses in these animals. Secretory IgA responses tended to be greater for gD than ICAP, although this difference was non-significant.

Similarly, antigen-specific IFNγ responses in cell preparations from spleens or genital tracts of mice immunized with GEN-003 or a novel antigen GB209 together with adjuvant GB2A followed the same patterns when quantified by ELISpot. Flow cytometry revealed these genital tract responses were derived from CD4+ T cells, as relative frequencies of CD3+CD4+IFNγ+ cells after restimulation with overlapping peptides spanning the sequences of gD, ICAP and GB209 matched ELISpot counts. In addition, most of these antigen-specific T cells were polyfunctional, expressing at least three out of the four markers measured (IFNγ, IL-2, TNFα, CD107). As for antibodies, local polyfunctional CD4+ T cell responses were accordant with systemic responses.

While intracellular cytokine staining of genital tract lymphocytes is feasible, cell quantities limit its regular use during vaccine candidate screening. Nevertheless, it is a powerful tool to assess immunogenic profiles of selected candidates.

Our data show comparable results of antigen-specific humoral and cellular immune responses locally in genital tracts and systemically in sera and spleens. Thus, systemic immunity can be used as surrogate for mucosal responses during early vaccine development in mice.

INTRODUCTION

Genocea’s subunit vaccine GEN-003 is currently in phase 2 clinical trials for the treatment of recurrent genital herpes caused by HSV-2. It is comprised of two antigens, ICAP2-3 and gD2TMR, and the saponin-based adjuvant Matrix®.

In a dose optimization trial, GEN-003 reduced genital lesion rates by up to 65% after 12 months as compared to pre dosing baseline levels2. While the systemic immune responses to GEN-003 have been well characterized in animal models and humans, the understanding of local immune response in the genital mucosa is less complete. GEN-003 is most promising for not only boost vaccine antigen-specific antibody titers, but also to stimulate T cell responses2, whose role in disease control is increasingly appreciated5.

In this context, the analysis of immune responses in the genital tract will provide deeper insight into the mechanisms of action of GEN-003. In addition, the characterization of mucosal immune responses may also facilitate the rational design of novel vaccine candidates with improved efficacy. These next generation vaccines may contain different or additional antigens as well as an adjuvant with an improved activity profile.

Here, we describe both humoral and T cell responses in the genital tract of mice immunized with several vaccine candidates and compare them to systemic responses to vaccination.

REFERENCES

1. Sabido et al., J. Virol. 2013
2. Fife et al., ASM Microbe 2016
3. Linda et al., Cytokine Immunology 2013