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THE E2DISP ANTIGEN DISPLAY SYSTEM: A NOVEL HIV VACCINE APPROACH

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BACKGROUND: It is widely accepted that an ideal HIV vaccine should elicit broad and robust cytotoxic T lymphocyte (CTL) and neutralizing antibody (NAb) responses. The E2 protein from *Geobacillus stearothermophilus* self-assembles into a virus-like 60-mer particle (1.5 MDa). Preliminary evidence from our group has shown that a novel antigen display system, E2DISP, can be engineered to display the HIV Gag-p17 protein or a T helper epitope from reverse transcriptase (pep23). Both self-assembling particles elicited both antibodies and CTL in HLA-A2 transgenic mice.

METHODS: E2DISP was engineered to display HIV peptides or proteins as N-terminal fusions to E2, with up to 60 copies of one (“pure”) or multiple (“hybrid”) antigens on a single particle. We have expressed as stable E2DISP fusion proteins large domains of HIV Env, Gag, Pol, Nef and Rev ranging in size from 5 to 20 kDa. Purification by ion-exchange chromatography and gel filtration yields up to 3 mg of E2DISP-HIV fusion protein per liter of bacterial culture.

RESULTS: E2Gag-p17 particles elicit robust anti-HIV antibody responses in C57BL6XBalb/C mice following immunization with either pure E2Gag-p17 or hybrid E2Gag-p17-pep23 particles adjuvanted with IFA. Responses were effectively boosted after two doses. Cellular responses were undetectable using intracellular cytokine staining following stimulation of splenic cells with pooled Gag peptides.

CONCLUSIONS: Both pure and hybrid E2DISP-HIV particles are immunogenic in non-transgenic mice. The successful boosting of HIV-specific antibodies despite the presence of antibodies to the E2DISP backbone suggests that higher titer responses may be elicited with additional immunizations. Future studies will evaluate the

immunogenicity of E2Env, E2Nef and E2Rev constructs, and alternative adjuvants will be used to enhance CTL responses. Generation of NAbS will be evaluated following immunizations with E2Env constructs, such as E2Env-gp41EC, which contains a target of broad NAbS, the Membrane Proximal External Region.

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