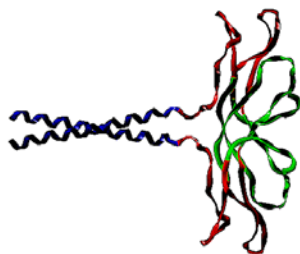
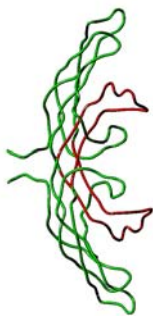


**Glycoprotein hormone homodimers: epitope directed immunogens.** Paul H. Ehrlich<sup>1,2</sup>, Win Lin<sup>1</sup>, Rebecca V. Myers<sup>1</sup>, Michael P. Bernard<sup>1</sup>, William R. Moyle<sup>1</sup>  
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**Introduction:** Monoclonal antibodies have been used extensively to study the structure and function of hCG, a hormone required to maintain early pregnancy and produced by many tumors. Antibodies to several conformational epitopes on the  $\beta$ -subunit have been identified that inhibit the biological activity of hCG. Antibodies to other sites have lower or almost no ability to block LHR mediated hCG. Many antibodies to both sites crossreact with hLH. An “ideal” vaccine would be capable of eliciting a potent immune response to only those hCG-specific epitopes that prevent it from binding to LHR. We made glycoprotein homodimers by replacing  $\beta$ -subunit loop 2 with  $\alpha$ -subunit loop 2 and vice versa. These contain two conformational epitopes on  $\beta$ -subunit loops 1/3 and  $\beta$ -subunit loop 2, respectively. Studies outlined here were designed to test the antigenicity of the former. **Methods:** Constructs termed BABS contained the coding regions for  $\beta$ -subunit loop 1,  $\alpha$ -subunit loop 2,  $\beta$ -subunit loop 3, and seatbelt. Constructs termed GCN4-ABAS<sub>t</sub> encoded a GCN4 dimerization domain,  $\alpha$ -subunit loop 1,  $\beta$ -subunit loop 2,  $\alpha$ -subunit loop 3, and part of the seatbelt. Both were made by standard methods and expressed in COS-7 and CHO cells. BABS was purified over a resin containing a monoclonal antibody to  $\beta$ -subunit loops 1/3, and used to immunize mice. **Results & Discussion:** BABS and GCN4-ABAS<sub>t</sub> were produced efficiently and contained two copies of the expected antibody binding sites. Mice immunized with BABS elicited an immune response to  $\beta$ -subunit loops 1/3, but not to  $\beta$ -subunit loop 2. BABS homodimers had low abilities to bind LHR and would not be expected to elicit ovarian function.



Models of BABS (left) and GCN4-ABAS<sub>t</sub> (right) based on the crystal structure of hCG.  
Color coding: residues derived from the  $\alpha$ -subunit, red; residues derived from the  $\beta$ -subunit, green; residues derived from GCN4, blue.

**Conclusions:** Homodimers can be produced that have two conformational epitopes from any part of the  $\beta$ -subunit. Immunogens that lack residues that elicit hLH crossreacting antibodies should evoke a high titer hCG-specific response. When expressed in a viral vector format, these may form the basis of a potent and inexpensive contraceptive vaccine. Targeting to particular antigen presenting cells may facilitate an hCG-specific cytotoxic T-cell response. **Support: NICHD**