

A functional high-throughput assay for selection and isolation of tumor-specific recombinant antibodies

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ABSTRACT

Viventia isolates specific human antibodies by using the natural human immune response raised against tumor-associated antigen present on cancer cells. Antibodies are identified using a soluble recombinant Fab immune library-based discovery platform called Fusogenics™. The recombinant Fab fragment is fused with a truncated form of *Pseudomonas* exotoxin A and expressed in *E. coli* supernatant. The Fab-PE format allows functional screening using a semi-high throughput system and is also suitable for biological characterization (flow cytometry, TMA staining), preclinical studies and antigen identification by the Unlock™ platform. Immune libraries are generated from enriched B cell population isolated from lymph nodes of cancer patients. After RT-PCR, the heavy and *kappa* light chain variable domains of the plasma B cells are randomly inserted in an expression vector as a dicistronic unit under the control of the arabinose-inducible *araBAD* promoter leading to the secretion of the soluble Fab-PE protein in *E. coli* supernatant. Using an annexinV assay, functional Fab-PE candidates that internalize are selected based upon differential cytotoxicity measured between tumor and normal cells. The screening algorithm, isolation and characterization of tumor-specific recombinant antibodies from a colon immune library will be presented.

Take home message: Fusogenics™ represents a novel screening and discovery platform using a Fab-PE format as a functional assay to identify potentially unique specific antibody fragments.