

De-Immunized Bouganin: An innovative, antibody-directed, cytotoxic payload for a safer and more efficacious treatment of cancer

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ABSTRACT

Tumor-targeting antibodies, carrying a cytotoxic payload offer a very promising alternative mechanism of action to conventional anti-cancer agents. A major obstacle to effective systemic drug delivery is the induction of a human anti-toxin antibody response (HATA). Viventia has developed a portfolio of fully-human cancer-specific antibodies that can be genetically tailored for optimal tumor penetration. Thus, the development of a de-immunized payload is critical to the development of immuno-cytotoxic drugs that can be delivered repeatedly via systemic route without the appearance of neutralizing antibodies. Bouganin is a plant-derived 29 kDa single-chain type I ribosome-inactivating protein (RIP) that is able to arrest protein synthesis by the deadenylation of ribosomal RNA resulting in apoptosis. Unlike other well-known members of this toxin group, native bouganin is one of the least toxic and therefore, is eminently suitable for engineering in a variety of molecular formats. Using Biovation's unique "peptide threading" technology, peptides covering the entire sequence of bouganin were synthesized and used in a T cell proliferation assay. The T cell assay was performed using PBMCs from twenty donors. The PBMC donors were selected to give an optimal coverage of MHC class II allotypes; for these donors the allotypic coverage was in excess of 85%. Analysis of the stimulation indices identified 3 potential human T-cell epitopes. In addition, a structural model of bouganin, based upon the crystal structure of Pokeweed antiviral protein, was made to assist in the identification of any steric effects that could arise from epitope mutagenesis. Site-directed mutagenesis was done at each of the three identified sites and the mutated peptides were tested for immunogenicity in the T-cell proliferation assay. Results showed the peptide mutants to be non-immunogenic, but of similar potency when compared to the wild-type molecule. Bouganin in its de-immunized form (de-bouganin) should permit repeat systemic administration when linked to Viventia's fully-human tumor-specific antibodies (Armed Antibodies™).