Building Better Biotherapeutics and Vaccines by Design: EpiVax, Inc., an Immunology Company

LEONARD MOISE, PhD; ANTHONY MARCELLO; RYAN TASSONE; LESLIE COUSENS, PhD; WILLIAM MARTIN; ANNE S. DE GROOT, MD

ABSTRACT

EpiVax, Inc., is an early-stage informatics and immunology biotechnology company in Providence, Rhode Island. It applies computational tools to harness immunity in three major areas: immunomodulation, biotherapeutic immunogenicity risk assessment and de-risking, and vaccine development. Immunotherapy, bio-better and vaccine candidates under development at EpiVax promise to improve the health outcomes of millions of people affected by devastating immune-related diseases.

KEYWORDS: vaccines, immunoinformatics, immunotherapy, immunomodulation, autoimmune diseases

A BRIEF HISTORY OF EPIVAX

A talented post-baccalaureate, a statistics major, and a professor who aspired to develop an HIV vaccine are at the root of EpiVax. Gabriel Meister, Bill Jesdale and Anne S. De Groot, MD, were members of the TB/HIV Research Lab team in Brown University's BioMed Center that created two novel computer-driven tools, EpiMer and EpiMatrix, between 1992 and 1996. These 'epitope discovery' algorithms generated the foundation for a whole suite of advanced in-silico tools that now form the core of EpiVax, Inc., a privately held immunoinformatics company in Providence. The late Michael Lysaght, an approachable and optimistic Brown professor of biotechnology, was another instrumental person in the company's establishment; he recognized the promise of EpiVax and connected the founders to the Slater Center for Biotechnology, a source of funding that brought the technology out of the academe into the entrepreneurial world in 1998. With the addition of a programming expert (Bill Martin) and a formidable lawyer (Fred Stolle), a company was born.

Fifteen years later, EpiVax has evolved into a powerhouse of ideas that is changing the way that we think about vaccines and biotherapeutics. EpiVax has also been the source of an unusual spin-out, the Institute for Immunology and Informatics (established in 2008) at the University of Rhode Island, which has exclusive access to the EpiVax technology to research and develop vaccines for neglected tropical diseases and other targets. Team members at EpiVax are now working on a second spin-out devoted to another promising technology that may change the treatment of autoimmune disease.

URI

Dr. Anne S. De Groot, at URI's Institute for Immunology and Informatics at the University of Rhode Island (URI), is the CEO of EpiVax, Inc.

IN-SILICO DESIGN FOR VACCINES AND PROTEINS

Vaccines are among the most important inventions of modern medicine, but the technology for making vaccines was based on empirical rather than hypothesis-driven science until 1996, when molecular biology made bacterial and viral proteins interpretable by computers. EpiVax has harnessed the availability of whole genomes to develop bioinformatics algorithms and apply them to a four-point vaccine design strategy. Immunoinformatics tools are first used to sort through thousands of potential vaccine candidates in a pathogen's genome, comparing those sequences to similar pathogens and identifying sequences that would trigger a human immune response. Protein sequences are then mapped for short, linear, putative T cell epitopes. These epitopes are synthesized as peptides and evaluated in vitro and in vivo for human leukocyte antigen (HLA) binding and antigenicity in survivors of infection or vaccinees. Finally, the optimal composition of immunogenic sequences to drive an effective human immune response is computationally derived (iVAX software suite), and prototype epitope-based vaccines are

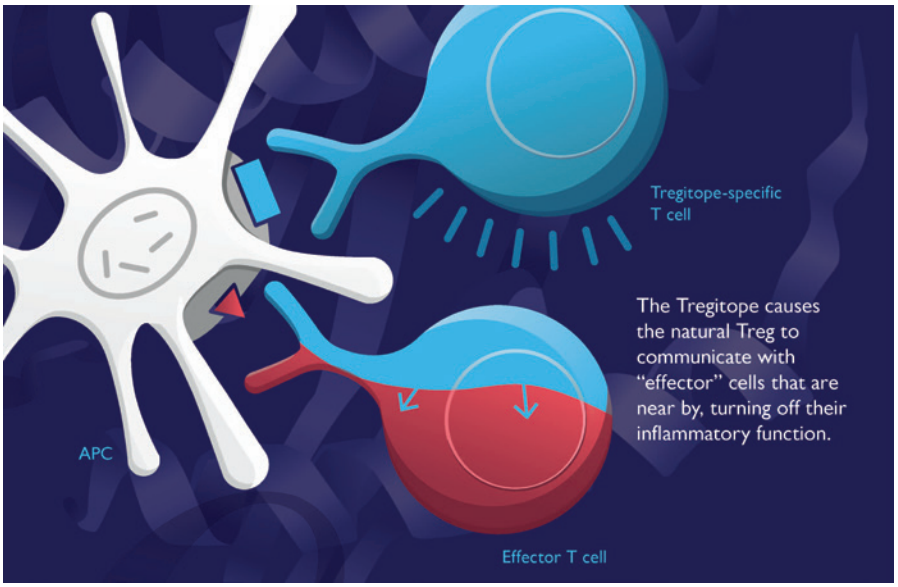
evaluated for immunogenicity and efficacy in humanized transgenic mice. Using this approach, we have demonstrated pre-clinical proof-of-concept for smallpox and tularemia prophylactic vaccination and therapeutic immunization for *H. pylori* infection.[1-4]

The genomes-to-vaccine strategy has two important advantages. First, it strips a pathogen down to the minimal essential antigens, eliciting robust and sustained protective immunity while eliminating non-essential information that could lead to diminished protective immunity and/or immunopathology sometimes associated with whole organism vaccines. This approach may appear to handicap vaccine design because vital elements (i.e., adjuvant, carrier structure) that are normally part of a pathogen are removed, but it creates a valuable opportunity that forms the second advantage. One can combine this novel approach with best-in-class adjuvant and delivery technologies for optimal vaccine construction.

This methodology also forms the core of immunogenicity screening, the process by which protein therapeutics are evaluated for their potential to elicit harmful responses that would impede their effectiveness. Non-vaccine protein therapeutics risk causing harmful immunoreactions, which can render a biologic ineffective and severely compromise patient health. For example, the induction of antibodies known as "inhibitors" against factor VIII in the treatment of hemophilia is a sign of therapeutic protein immunogenicity.[5,6] In 2001, antibodies to a commonly used therapeutic protein drug Erythropoietin, were linked to transfusion dependent anemia.[7] Consequently, unwanted immunoreactions to biologics are a major concern for physicians and drug developers.

EpiVax has thus developed an entire suite of immunoinformatics tools for prospectively identifying and reducing protein therapeutic immunogenicity "in silico," a process that dramatically reduces the time and effort involved, allowing drug developers to accelerate the pre-clinical development of their protein products. The tools are organized in an interactive website called the ISPRI (Interactive Screening and Protein Reengineering Interface) system. Using the ISPRI system, researchers have the ability to screen the protein sequences of product candidates for the presence and immunogenic potential of putative T cell epitopes (EpiMatrix) and epitope clusters (ClustiMer). Protein sequences can be ranked for immunogenic potential in comparison to known proteins on a normalized scale, and an interactive protein reengineering tool (OptiMatrix) allows researchers to modify, or deimmunize, T cell epitope clusters in real time by optimizing the amino acid sequence so that it is no longer able to interact with T cells.

The EpiMatrix toolkit has been extensively validated internally and externally, with several key publications demonstrating the technology and rigorous testing procedures using known protein therapeutic targets.[8,9] In addition, EpiVax incorporates exclusive knowledge of the impact of Tregitopes (T cell regulatory epitopes) on the immunogenicity of protein therapeutics in clinical use, leading to higher accuracy in immunogenicity predictions.



EPIVAX, INC.

TREGITOPES: AN EPIVAX DISCOVERY AND IMMUNOMODULATION POWERHOUSE

The discovery of Tregitopes, or "T Regulatory Epitopes" in one of the most common proteins found in blood (immunoglobulin G, or IgG) can be attributed to keen observation on the part of the scientific team at EpiVax. Tregitopes turned up regularly in the immunogenicity screens that were performed by the scientists at EpiVax as soon as the ISPRI tools were being applied to monoclonal antibody therapeutics, but were only recognized for their regulatory potential by De Groot and Martin in 2008.[10] Tregitopes act as a natural 'off switch' for the immune system. They are naturally part of the arms (Fab) and stem (Fc) of human IgG and are thought to balance the inflammatory triggers that are present in the re-arranged, or hypervariable segments (variable loops) of the antibody arms. Tregitopes are also found in Intravenous IgG (IVIG), a blood-derived product that is used clinically to control autoimmune conditions.[11] Indeed, some of the anti-inflammatory activity of IVIG may be due to the presence of Tregitopes.[12]

The Tregitope discovery has been validated in a range of standard preclinical models and by collaborating laboratories, where Tregitopes have been shown to suppress and treat autoimmune disease and allergies,[13] and to effectively suppress the immunogenicity of co-administered proteins.[14,15] In addition, Tregitopes have been shown to modify immune

responses to biotherapeutics, such as FVIII. In vitro, co-incubation of proteins with Tregitopes leads to suppression of effector cytokine and chemokine secretion, reduced proliferation of effector T cells, and expansion of antigen-specific adaptive Tregs. In vivo, co-administration of Tregitopes with a wide range of proteins (i.e., FVIII, ovalbumin, and autoantigens) leads to antigen-specific suppression of T cell and antibody responses.

Funding for research on Tregitopes has been flowing. For example, EpiVax recently received a Small Business Innovation Research (SBIR) Phase I grant for $600,000 to explore the use of Tregitope in facilitating tolerance to the lifesaving enzyme replacement therapy for Pompe's disease.[16] In 2012 alone, EpiVax scientists were able to obtain $3.4 million in National Institutes of Health (NIH) funding for development of Tregitope therapies; the group has been awarded more than $6 million in grants to develop Tregitopes over the past few years. Once the right formulation of Tregitopes is identified, and they pass the usual regulatory hurdles, their use is expected to have a radical impact on the clinical management of autoimmunity, transplant rejection, and protein replacement therapies.

CONCLUSION

EpiVax will continue to apply the experience gained from these basic research efforts to practical problems in immunotherapy and vaccine design. In the field of protein therapeutics, we are broadly recognized as thought leaders, and we expect to maintain this position through our discovery work on Tregitopes and tolerance. In addition, our work on epitope-driven vaccines – such as the smallpox, Tularemia, and *H. pylori* vaccines in our pipeline – has begun to demonstrate the power of T cell epitopes to generate protective immune responses. We will combine these breakthroughs with advancements in delivery and formulation to bring novel immunomodulatory therapies and vaccines to market.

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Authors

Leonard Moise, PhD, is senior director vaccine research at EpiVax.

Annie De Groot, MD, is CEO/CSO of EpiVax.

Anthony Marcello is an associate, business development at EpiVax.

Ryan Tassone is research associate at EpiVax.

William Martin is chief information officer and chief operating officer at EpiVax.

Leslie Cousens, PhD, is scientific director protein therapeutics at EpiVax.

Correspondence

Anne S. De Groot, MD

EpiVax, Inc.

146 Clifford Street, Providence RI 02903

401-272-2123

Fax 401-272-7562

AnnieD@EpiVax.com