Redefining Protein Therapeutics with an Expanded Amino Acid Universe

Therapeutic proteins are used to treat a wide variety of diseases, from cancer and heart disease to cystic fibrosis and diabetes. These proteins currently are produced using microbial fermentation in cell cultures and by transgenic animals or plants. They represent a nearly $200 billion market, yet product stability and unintended immune responses remain significant obstacles to treatment. For example, many therapeutics suffer from short half-lives that require frequent dosing, which can lead to increased side effects and noncompliance to treatment. Many other therapies produce immune reactions that impair therapeutic activity or endanger the patient. Often, these challenges arise from a limitation inherent to all proteins: they are made from the same 20 amino acid building blocks.

Thanks to funding from the National Science Foundation (NSF), Boston-based biotechnology start-up GRO Biosciences (GRObio) is working to overcome this intrinsic limitation of protein-based therapies by expanding the amino acid alphabet to include non-standard amino acids (NSAAs).

At the core of its technology is the Genomically Recoded Organism (GRO) platform. The platform consists of the first production organisms (*e.g., E. coli*) with modified genomes and engineered protein translational machinery for producing NSAA proteins at commercial scale. GROs are designed so that all instances of one or more triplets of DNA nucleotides, called codons, are swapped to synonymous codons. The company likens this process to a genomic "find-replace" procedure that leaves the organism healthy and productive but liberates target codons to direct the incorporation of NSAAs. The company also removes translational machinery within its GROs that would impair productivity by competing for the target codons. The resulting GROs can produce NSAA protein therapeutics with unprecedented efficiency and scalability.

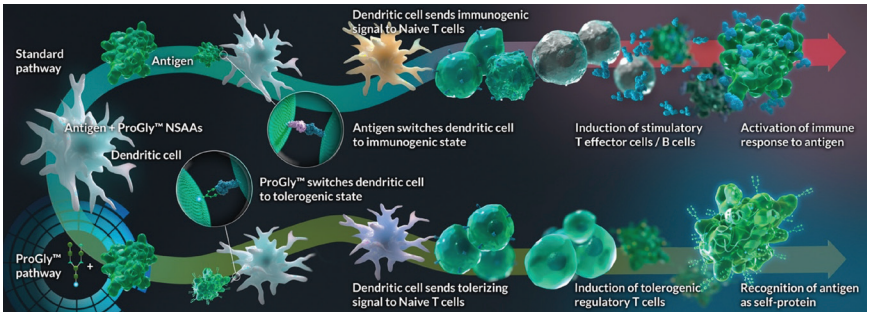
To develop the NSAA protein translational machinery and to streamline development of NSAA protein therapeutics, the company has constructed a biofoundry that combines computational protein design tools with automated robotics workflows. The biofoundry allows GRObio to build scalable NSAA protein "factories" from trillions of candidates.

The company is currently developing therapeutic pipelines for two NSAA families. The DuraLogic NSAA product family enhances and maintains the three-dimensional structure of proteins needed for therapeutic activity. DuraLogic NSAAs impart proteins with unprecedented duration of activity, providing therapeutic benefits such as flatter pharmacodynamic profiles and relaxed dosing schedules.

The ProGly platform chemistry comprises glycan-containing NSAAs that can induce or inhibit an immune reaction. The GRO platform enables precise placement of ProGly NSAAs on the protein surface necessary to elicit a defined immune response. The company's first ProGly NSAAs can control whether a protein is recognized by the immune system as "self" or "non-self" — a novel modality for eliminating autoimmunity and anti-drug antibodies.

"GRObio's technology isn't confined to a single disease state or patient population — it allows the company to focus on medical conditions where they can make a unique impact on patient outcomes. With ProGly, the company has the potential to create entirely new therapies to reverse autoimmune disease, and also to provide protein therapeutics that are free from immunogenic reactions," says Tracey Lodie, Chief Scientific Officer at Quell Therapeutics and Clinical Translation Advisor to GRObio.

GRObio anticipates initiating their first clinical trials in 2024. The company is developing new production GROs that will produce proteins with multiple different NSAAs simultaneously, allowing researchers to, for example, combine DuraLogic and ProGly chemistries, or to integrate entirely new therapeutic capabilities. CEP



▲ GRObio's ProGly platform chemistry can re-educate the immune system to treat autoimmune disease or to eliminate anti-drug antibodies against otherwise promising protein therapeutics. In the standard pathway (in the upper panel), the immune system recognizes an antigen as a foreign protein and mounts an immune response. In the ProGly pathway, ProGly non-standard amino acids (NSAAs) carry sugar signatures called glycans that signal to the immune system that the antigen is a self-protein, reversing the immune response.

This technology was funded through the NSF Small Business Innovation Research Program.

This article was prepared by the National Science Foundation in partnership with *CEP*.