Living Plants: One-Stop Shop for Protein Manufacturing

Chemical manufacturing facilities housing green plants perform all of the processes formerly handled by stainless steel equipment. Production is energy efficient, fueled by photosynthesis, as living plants secrete raw materials (*e.g.*, high-purity spider silk proteins) for downstream production and processing. Thanks to Wisconsin-based ag-biotech startup PhylloTech, this surreal vision may not be so far-fetched.

PhylloTech has discovered a protein-manufacturing system within the leaves of green plants that produces and selectively secretes proteins to the leaf surface. These proteins could be collected and used as environmentally friendly agricultural pest control for other crops. The company is also modifying the technology to enable plants to produce and secrete other chemicals from their leaves as well — thereby transforming the way specialty chemicals are produced.

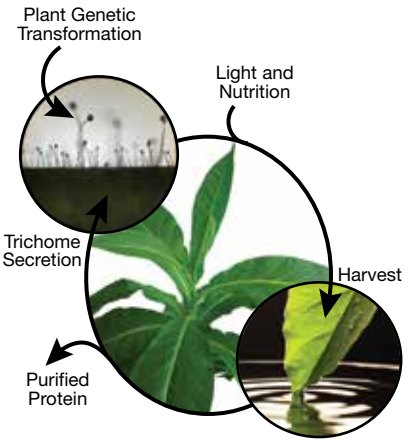
"Current approaches to express and purify targets from bacterial, yeast, and mammalian cell systems are complex, as well as capital intensive, with high downstream processing costs," says Ryan Shepherd, co-founder and CEO of PhylloTech. "We wanted to redefine protein production with sustainable manufacturing processes that avoid high extraction and purification costs. This is now possible with our plant-based trichome bioproduction system."

The new process involves trichomes, or leaf hairs abundant on the aerial surfaces of many plant species. Glandular trichomes, which consist of a biosynthetically active gland cell atop a multicellular stalk, often produce and secrete specific biochemicals to the leaf surface, or the phylloplane. As a doctoral student at the Univ. of Kentucky, Shepherd began characterizing phylloplane proteins, and found that plants such as *Nicotiana tabacum* (tobacco) secrete large amounts of pure proteins to their leaf surfaces. One predominant family of proteins, which he termed phylloplanins, is produced by the gland cells of specialized glandular trichomes and possesses antimicrobial properties.

Shepherd and his team founded PhylloTech to commercialize phylloplanins as environmentally friendly controls for agricultural pathogens. A recent Small Business Innovation Research (SBIR) grant from the National Science Foundation has allowed the company to modify the natural system to biomanufacture other target compounds, including complex enzymes, antimicrobials, and even highly tailored chemicals such as small molecules and metabolites.

PhylloTech has genetically modified an existing pathway in *N. tabacum* that naturally drives phylloplanin expression in the plant's trichome glands to secrete various proteins to the leaf surface. From there, the proteins can be easily recovered by spraying the leaves with water. The harvest process is nondestructive, and manufacturers can harvest proteins throughout the entire lifespan of the plants.

"Our technology is unique because not only do we genetically engineer the plant to produce the protein, but the plant isolates the target protein as well," says Danielle Stacy, PhylloTech's R&D manager. "We let the plants do most of the purification work for us so that we only recover the desired target in an enriched state. Some downstream processing is still required, but in most instances, the target can be used directly."



▲ PhylloTech's trichome bioproduction and purification process involves altering a natural pathway in tobacco plants to selectively produce target proteins in their trichome glands. The proteins are secreted to the leaf surface, where they are harvested nondestructively. Image courtesy of PhylloTech.

Depending on the desired protein, the production costs of PhylloTech's trichome bioproduction system could be as much as 80–90% less than those of standard expression systems, which typically involve large fermentation systems, require substantial infrastructure, and involve expensive cell lysis and chromatographic techniques. PhylloTech's system is holistic and requires little infrastructure. Plants are ideal production platforms because they are easy to maintain, provide substantial production surface area on their large leaves, have simple growth needs, and can produce glycosylated, endotoxin-free targets.

Shepherd and his team consider the technology to be in the advanced development stage, and they already have numerous plant lines generating biomaterials and industrial enzymes. PhylloTech is now constructing a plant-growth facility to scale up its production capacity.

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