Ticking a new box in enzyme chemistry

Going beyond its comfort zone in biocatalysis, Almac immobilizes an enzyme and manufactures a polymer for Alcresta's lipase cartridge device

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With their first start-up, Alnara Pharmaceuticals, biotech entrepreneurs Alexey Margolin and Robert Gallotto developed a pancreatic enzyme replacement therapy, liprotamase, to treat exocrine pancreatic insufficiency—an inability to break down fats experienced by people with cystic fibrosis and other diseases. The product, which includes a microbially derived lipase enzyme, replaced a lipase that for nearly 100 years had been derived from pig pancreases.

After Eli Lilly and Company acquired Alnara in 2010, the entrepreneurs had a new idea: address concerns that ingesting pancreatic enzyme replacements can cause bowel obstruction and inflammation.

Their new venture, Alcresta Therapeutics, launched in 2011 to develop a device that breaks down fats externally by connecting to a feeding tube. The result was Relizorb, a cartridge launched in 2016 that employs a lipase enzyme immobilized on polymer beads. Triglycerides in the feeding formula pass through the device and break down to fatty acids and monoglycerides before ingestion, with none of the enzyme leaving the cartridge.

Convinced from bench experiments that the device would work, the partners prepared to scale up for clinical and commercial production. They secured food-grade lipase from a Japanese supplier but needed to immobilize the enzyme on a polymer bead they'd designed. David Brown, vice president of technical operations at Alcresta, whose task was to find a contract services firm, found that enzyme immobilization is a peculiar niche expertise.

Brown, who worked at Alnara and was involved in developing the fermentation process for liprotamase, was familiar with the contract development and manufacturing organization (CDMO) market by the time he rejoined Margolin and Gallotto at Alcresta. "When Alnara was sold to Lilly, I went to Patheon," he says. There, he immersed himself in the world of finished drug development and manufacturing services.

Alcresta placed him in new territory. "It was really kind of an exciting time for me because I had just come out of this classical CDMO world, and I was following the trends of what big pharma needed from CDMOs," he says. "The whole world of enzymes, and enzyme immobilization in particular, was much smaller and not necessarily of interest to the big firm I represented."

After exploring the market, Brown identified 28 service providers with enzyme-immobilization expertise, but only 4 of them also had experience with suspension polymerization—the process for synthesizing the bead. The company Alcresta eventually signed with, Almac Sciences, wasn't on the short list.

"Companies with experience in both immobilization and suspension polymerization recognized they were in a unique position and were larger companies," Brown says. "Frankly, they wanted to make a bigger deal of it than Alcresta could stomach at the time."

As he widened his search he decided to take another look at Almac. Brown saw that the Northern Irish firm's biocatalysis group had expertise in using enzymes as catalysts and might be ready for a new challenge: "They had never made a product for a product's sake."

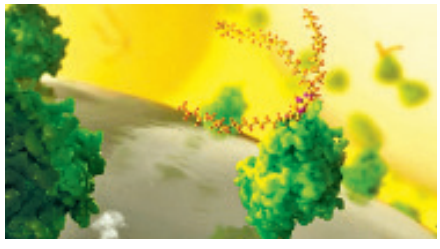
According to Tom Moody, vice president of technology development and commercialization at Almac, the biocatalysis division was primed for the job.

"They were looking for someone with analytical chemistry around enzyme transformation, someone who understood enzyme production and purification, someone who knew about how to take R&D projects that involve chemistry and biochemistry right through to production," he says.

"They wanted someone who can immobilize the enzymes, who understands heterogeneous catalysis, someone who could do design-of-experiment studies for any robust process that can be scaled commercially, and someone who can look at a second-generation product, which we are currently doing. And they wanted someone who can produce at commercial scale and deliver multiton of product," he continues. "We ticked all the boxes."

Relizorb

- **Generic name:** Immobilized lipase cartridge
- **Commercial launch:** 2016
- **Innovation:** Feeding-tube device eliminating ingestion of lipase-replacement therapies
- **Components**: Immobilized lipase enzyme bonded to methacrylate polymer beads



The enzyme (green) breaks down triglycerides while bonded to the surface of a polymer bead.

A technician works on a multipurpose batch reactor at Almac's Arran Chemical facility, where the company works on immobilizing the enzyme on polymer beads for Alcresta Therapeutics.

CREDIT: ALCRESTA (TRIGLYCERIDES); ALMAC (REACTOR)

The commercial production box was ticked, Moody says, thanks to Almac Group's 2015 acquisition of Arran Chemical. Although Almac manufactures to the US Food and Drug Administration's current good manufacturing practice (cGMP) quality standard at its headquarters in Northern Ireland, it operates Arran as a non-cGMP site for the production of drug intermediates and nonpharmaceutical chemicals. The work for Alcresta is done at Arran to the ISO 9001 quality standard.

And it requires a lot of chemistry, Moody points out. "A lot of people hear biocatalysis, and they think it's a black box," he says. "Fundamentally, it's synthetic organic chemistry. Given the way we formulate some of these processes, rather than call them biocatalysts, we call them catalysts."

Almac went to work quickly on the box it could not tick—suspension polymerization. "It was quite a steep learning curve," recalls Scott Wharry, senior research and manufacturing team leader in the company's biocatalysis group, noting that Alcresta wanted the enzyme-immobilization provider to be a key supplier of the beads.

"There was lots of background reading and speaking to various people with experience and just planning a systematic approach to investigate the various components of the reaction," Wharry says.

Engineers at Almac assisted in developing a model that could be scaled up for commercial production. "We ended up designing and installing a reactor specifically for that," Wharry says. "After developing the process and making appropriately sized beads at laboratory scale, we increased in scale until we were manufacturing at approximately 1,000 L scale." Although the beads are now made by another firm that is not involved in enzyme immobilization, Almac remains ready as a supplier.

Ensuring product purity is a key challenge, according to Wharry. "We did an extensive bit of analytical work to help Alcresta with the regulatory bodies to ensure no residual components of the bead formation or residual protein would wash off the beads and into the patient."

The immobilization is a standard covalent bonding reaction, Wharry says, but there are some nuances. "The immobilization is a tricky process because you have to get to a certain level of protein attachment," he says. "You need to make sure there is no leachable protein or nonbonding protein or other impurity." Nor can there be microbial growth on the beads, he says.

"There are certain functional groups on the bead that allow the enzyme we are grafting to retain high activity," Wharry continues. "There is an epoxide on the bead, and you have amine functionality on the protein. We are covalently binding the enzyme to the surface."

"Once it's grafted," Moody adds, "it's grafted."

Brown concurs that the chemistry is tricky. "This is wet chemistry in tanks, yet it's two phases, solid and liquid, with some flammable solvents," he says. "And the beads are somewhat fragile and require delicate handling." At 0.5 mm across, the methacrylate polymer beads are larger than the polymers that are usually produced through suspension polymerization.

"We have an effective surface area of 250 m^2/g, which is about the size of a tennis court," Brown says. "The beads that got us to clinical trials were an incredible achievement."

So, he suggests, was finding the right service firm to connect the enzyme to the bead. Brown says he valued Almac's willingness to develop an immobilized enzyme as a product rather than as a catalyst and its willingness to take on a unique challenge. Beyond its chemistry expertise, Brown adds, the quality that most sold Alcresta on Almac can be summed up in one word: attitude.

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