Lysin in wait

By Stephen Parmley, Senior Writer

As researchers chase new ways to combat antibiotic resistance, two companies are tackling the problem with anti-infectives based on phage lysins—enzymes that kill bacteria selectively and are not prone to bacterial resistance. **Micreos B.V.** and **ContraFect Corp.** are taking different tacks to getting proof of concept in patients, but for both companies, finding the right formulation might be the key to getting lysins approved for systemic use.

Although lysins have been on the research scene for several years, companies have struggled to create viable therapies from them because of poor solubility and purity.

The enzymes are natural products of bacteriophages—viruses that infect and destroy bacteria. Lysins act by breaking down specific carbohydrates in bacterial cell walls, which results in bacterial lysis. Because the lysins' targets are essential to host viability, the targets are not tolerant to mutation, and lysins have thus far not been found to induce resistance.

According to Mark Woolhouse, a professor of infectious disease epidemiology at **The University of Edinburgh**, "Phage therapy had been seen as a possible way of treating bacterial infections for many decades, but the success of antibiotics restricted phage therapy to something of a sideshow."

He added, "The advent and dramatic spread of antibiotic resistance changed the situation."

According to Vincent Fischetti, a major advantage of lysins over antibiotics is that lysins work almost immediately. He said that when a patient has organisms in the blood—as with sepsis or bacteremia—and is treated with antibiotics, it will take several hours to clear the organisms, whereas treatment with lysins will clear the bacteria within minutes. As a result, he said, the chances of survival with lysin treatment should be much greater than with antibiotics.

Fischetti is a professor and head of the laboratory of bacterial pathogenesis and immunology at **The Rockefeller University**.

Killer formulations

ContraFect's approach is to create lysins against *Staphylococcus aureus* that are soluble and highly expressed when produced in *Escherichia coli* and can be readily purified in active form. Most recombinant lysins that digest *S. aureus* are poorly expressed and exhibit low solubility.

The company's lead compound, CF-301, was developed in collaboration with Fischetti and colleagues at Rockefeller. Earlier this year, the team showed in a mouse model of *S. aureus* bacteremia with a high bacterial inoculum that CF-301 provided similar protection to high doses of Cubicin daptomycin.[1] Treatment with CF-301 alone resulted in 17%–50% survival, whereas treatment with daptomycin alone resulted in 7%–31% survival.

Cubicin is marketed by **Cubist Pharmaceuticals Inc.** and **Merck & Co. Inc.** for bacteremia and skin and skin structure infections (SSSIs).

Fischetti told *SciBX* that the ContraFect lysins also have shown efficacy in a mouse wound infection model. "We put *S. aureus* in wounds, added lysins and saw almost complete destruction of the bacteria, and we put a lot of organisms in."

According to ContraFect CSO Michael Wittekind, the company expects to begin Phase I trials in 2015 evaluating CF-301 safety in healthy subjects. In Phase II trials the company will evaluate the combination of CF-301 and antibiotics for the treatment of *S. aureus* bacteremia, including endocarditis, caused by methicillin-resistant *S. aureus* (MRSA) and methicillin-sensitive *S. aureus* (MSSA).

ContraFect filed an IND for the compound in 2013, but it was placed on clinical hold until additional preclinical studies could be performed to assess the risks of the original trial design. Last week, ContraFect submitted a complete response to the FDA. The company declined to comment on the specifics of the letter.

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> *—Michael Wittekind, ContraFect Corp.*

Micreos, by contrast, is focusing on compound purity in creating a clinically viable lysin formulation acceptable for systemic exposure.

The company already has a marketed lysin—Staphefekt XDR.300—that was approved this year for treatment of *S. aureus* and MRSA skin infections on intact skin and is used for conditions such as eczema. However, the drug cannot be used on open wounds or in surgery because it is not sufficiently pure for systemic exposure.

Micreos is developing a new formulation of Staphefekt for wounds and surgical sites and is increasing the drug's purity.

Bjorn Herpers, a clinical microbiologist at the **Public Health Laboratory, Kennemerland**, collaborated with Micreos to test whether lysins were prone to resistance. His team recently showed that clinical isolates of *S. aureus* exposed to suboptimal treatment levels of the antibiotic Centany mupirocin or Staphefekt developed resistance to the antibiotic but not to Staphefekt.[2]

Centany is marketed by **Medimetriks Pharmaceuticals Inc.** as a topical antibiotic for SSSIs.

Herpers said that he and Micreos are collaborating with the **Association of Dutch Burn Centres** to see whether Staphefekt can be used in burn wounds. He said that the local treatment of open wounds and surgical sites with lysins could become available in one or two years.

Synergies with antibiotics

A conservative approach for companies in early clinical testing of lysins could be to add them on antibiotic regimes.

According to Fischetti, combination therapy may be one of the best uses of lysins as the two types of anti-infectives could work synergistically. "You can mix them to get an enhanced effect," he said. "Therefore, you can use less antibiotic and less lysin."

In Fischetti's studies on the mouse bacteremia model, combinations of CF-301 and minimally efficacious doses of Vancocin vancomycin or Cubicin significantly increased survival compared with antibiotics or CF-301 alone.[1] Vancocin is marketed by **Eli Lilly and Co.** for infectious diarrhea.

Wittekind told *SciBX*, "Lysin therapy combined with antibiotics has the potential to reduce treatment times, improve patient outcomes and shorten hospital stays while not contributing to the global crisis of drug resistance."

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2. Herpers, B.L. *et al.* Specific lysis of *Staphylococcus aureus* by the endolysin Staphefekt SA.100: *in vitro* studies and human case series. Presented at the Antibiotic Alternatives for the New Millennium Conference, Nov. 5–7, 2014

COMPANIES AND INSTITUTIONS MENTIONED

Association of Dutch Burn Centres, Beverwijk, the Netherlands
ContraFect Corp. (NASDAQ:CFRX), Yonkers, N.Y.
Cubist Pharmaceuticals Inc. (NASDAQ:CBST), Lexington, Mass.
Eli Lilly and Co. (NYSE:LLY), Indianapolis, Ind.
Medimetriks Pharmaceuticals Inc., Fairfield, N.J.
Merck & Co. Inc. (NYSE:MRK), Whitehouse Station, N.J.
Micreos B.V., Wageningen, the Netherlands
Public Health Laboratory, Kennemerland, the Netherlands
The Rockefeller University, New York, N.Y.
The University of Edinburgh, Edinburgh, U.K.