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**New and Improved! A supercharged antibiotic to fight superbugs.**

Mark A.T. Blaskovich, Centre for Superbug Solutions, Institute for Molecular Bioscience, The University of Queensland, Brisbane Qld 4072 Australia. email: m.blaskovich@uq.edu.au

Taken an antibiotic lately? You’re not alone. Almost one-in-two people will have been dosed with an antibiotic over the last year. Unfortunately, all this antibiotic use is causing the bacteria to become more and more resistant to their effects. We are now facing a future where antibiotics may no longer work, and bacterial infections become much more deadly. This problem is made worse because new and improved antibiotics are becoming rare. Our recent publication, ‘Protein-inspired antibiotics active against vancomycin- and daptomycin-resistant bacteria’, describes one approach we’re taking to try to help solve the antibiotic crisis.

Our strategy is based on trying to develop a different way of targeting drugs to where they are needed. Almost all drugs work by interacting specifically with of the two main biological building blocks - either proteins or DNA/RNA. However, there is a third component that is generally overlooked – the lipids that form membranes encasing all cells, both mammalian and bacterial. Membranes are usually only looked at as impediments that prevent drugs from getting to where they are meant to go. However Nature uses variations in membrane compositions between different cell types as a targeting mechanism, employing proteins that bind to specific types of lipids in order to differentiate between various types of cells. Using a similar strategy, we envisioned developing improved versions of antibiotics by functionalizing them with lipid targeting substituents. This would take advantage of the substantial differences between bacterial and mammalian membranes, lead to selective targeting of the antibiotic towards bacteria, and hopefully reduce off-target effects caused by interactions with human cells.

We selected vancomycin as the antibiotic to test this strategy for a number of reasons. It has been, and still is, an important antibiotic for treating Gram-positive infections. While resistance to vancomycin is rising, it has taken longer than for most antibiotics, so a new antibiotic may also retain effectiveness over time. Vancomycin is cheap and readily available, so derivatives could be produced on a large scale. Structural modifications that retain activity are well documented, so we were confident we could attach our desired targeting moiety without destroying its innate activity. Most importantly, the target of vancomycin (Lipid II) is anchored in the cell membrane. A successful membrane-targeting strategy would direct vancomycin right to where it’s needed.

Initially, we started by attaching quite lengthy fragments of proteins that were known to bind to membrane. These “electrostatic effector peptide segments (EEPS)” contained multiple positively charged amino acids (lysine residues) to interact with the overall negative charge of bacterial membrane. The EEPS was capped with a lipophilic group (the “membrane-insertive element”; MIE) that we hoped would embed itself in the membrane. The other end of the EEPS was linked to the *C*-terminal end of vancomycin via a diamino linker group. Some of our initial products, which we collectively termed ‘vancapticins’, showed up to 10-fold greater potency than vancomycin at inhibiting bacterial growth. We soon found that the EEPS could be greatly simplified to only two or three amino acid residues, boosting the potency another 10-fold. The final breakthrough was replacing the original linker, which incorporated a physiologically unstable disulfide group, with linkers that could not be cleaved.

With these modular components in hand, we systematically made hundreds of analogues. Several promising candidates were then progressed through a series of assays typically used to assess potential drug candidates (such as stability to biological degradation, and assessment of concentration over time in mice), and a number of specialized assays specific to antibiotics. These included testing how quickly the vancapticins killed bacteria (similar to vancomycin), and how rapidly bacteria developed resistance (not very!). We also confirmed that the antibiotics worked like vancomycin - preventing cell wall formation - but also had additional activity related to their membrane binding. Finally, and most importantly, we proved that the vancapticins cured infections in mice with greater effectiveness than vancomycin. Unlike mouse models of cancer, these infection models are much more predictive of whether antibiotics will work in humans.

We have continued to work on developing improved versions of the vancapticins, and would like to think that they could help address the looming threat of antimicrobial resistance. As with other antibiotics, the biggest barrier to continuing their development is finding a viable commercial route. This project has involved a many people across numerous institutions for more than a decade. Our main motivation, like the antibiotic pathfinders in the 1940s, has been to help prevent people from dying from bacterial infections.