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Is there a future for FabI inhibitors as antibacterial agents?

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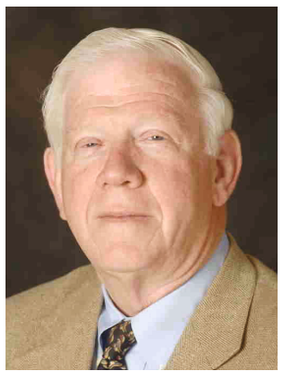

"The clinical role of FabI inhibitors will become clearer over time. They definitely represent a novel and important antibacterial platform that has the potential to address current bacterial resistance problems."

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The bacterial target FabI, enoyl-acyl carrier protein reductase (ENR), has stimulated considerable interest in the anti-infectives arena as a new fully validated target in all strains of *Staphylococcus* and the target for several potential antimicrobial agents that may provide specific therapy for a very common and significant bacterial pathogen, *Staphylococcus aureus*, including MRSA.

The widespread use of broad-spectrum antibiotics, such as cephalosporins, aminoglycosides and fluoroquinolones over the last 20 years has given rise to significant rates of antibiotic resistance among a broad range of bacterial pathogens [1]. Alarmingly, many of these pathogens have developed resistance to multiple classes of antibiotics. Particular examples are found among the Enterobacteriaceae*: Pseudomonas aeruginosa*, *Acinetobacter baumanni* and *Staphylococcus spp.* specifically MRSA [2,3]. Treatment of these pathogens has become so difficult that physicians are calling for new antibacterial agents with novel modes of action, thereby avoiding existing resistance mechanisms. Ideally, such agents would incorporate other features such as good safety profiles and flexibility in dosage forms and regimens.

Recently, there has been growing pressure for effective and widespread implementation of antibiotic stewardship programs to slow or limit the emergence of further bacterial resistance. Experts have advocated the development of narrow-spectrum agents to target specific bacterial species in an effort to reduce off-target selection pressures on the human microbiome [4]. These agents would be more targeted than even vancomycin or linezolid, the use of which led to the development of vancomycin- and/or linezolid-resistant enterococci as well as reduced susceptibility or resistance in MRSA. This targeted approach will likely have the additional benefit of causing less disturbance of the commensal flora, resulting in a decrease in such conditions as antibiotic-induced colitis, including *Clostridium difficile* enterocolitis or candidiasis.

Unfortunately, many pharmaceutical companies have withdrawn from antibacterial drug development. Reasons include high-development costs relative to the low sales potential of drugs that are typically used in only short-term treatment regimens, reduced product life due to rapid resistance emergence, and the low probability of identifying a new chemical target that is not compromised by existing resistance mechanisms. The concept of narrow-spectrum agents seems even less appealing due to reduced market potential relative to broad-spectrum agents, despite the potential for a longer window of utility due to reduced emergence of resistance.

Genomics-based discovery programs in the early 2000s identified the bacterial fatty-acid (FASII) biosynthetic pathway and the component Fab enzymes (FabA to FabX) as relatively unexploited bacterial targets for antimicrobial agents [5–9]. The Fab family of enzymes are found in all bacteria, with each enzyme catalyzing a step

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in the essential fatty acid biosynthesis pathway. That said, additional work in this area has revealed significant complexity in bacterial fatty acid metabolism, and although many bacteria have a specific kind of ENR enzyme called FabI, they may have markedly different primary structures. Other bacteria utilize ENR classified not as a FabI, but as FabK, FabV or FabL enzymes; and others may have redundancy with FabI and FabK such as *Enterococcus faecalis*. As a consequence of this diversity, a specific FabI inhibitor is assured to have a relatively narrow spectrum of activity.

Mammals also possess a fatty acid biosynthetic pathway (FASI); however the various enzymatic functions are catalyzed by a single dimeric enzyme [10]. This organizational and structural difference between bacteria and mammals provides an *a priori* reason why inhibitors of bacterial fatty acid biosynthesis should be selective and safe for use in humans.

FabI is the sole form of ENR present in *S. aureus*, *S. epidermidis* and other staphylococci, and catalyses the last step in the bacterial fatty acid biosynthetic pathway. Some publications have cast doubt on the essentiality of FabI, but these concerns have proven unfounded [11–13]. Extensive studies have found no alternative enzyme or rescue pathway for FabI in staphylococci. FabI is essential to cell viability in *Staphylococcus spp.* and FabI inhibition is a potent and targeted potential therapy for all *Staphylococcal spp.*

Considerable research over the years has shown that the FabI enzyme is an appropriate target in a number of pathogens. Isoniazid, diazaboranes, triclosan and other small molecule inhibitors have been widely used in the treatment of bacterial disease.

MUT056399 (FabPharma, Paris, France), an intravenous formulation of a triclosan derivative, has recently completed a Phase I study [14]. Crystal Genomics (Seoul, Korea) have also pursued triclosan derivatives as inhibitors of FabI and an intravenous formulation of the lead compound CG400459 has recently completed a 20-patient Phase II study [101].

AFN-1252, Affinium Pharmaceutical's lead clinical candidate, is perhaps the most advanced FabI inhibitor. This agent is the culmination of a large-scale iterative discovery program with the specific objective of finding a 'magic bullet' for *Staphylococcus spp.*, including MRSA.

Preclinical properties have confirmed a unique mode of action, target specificity, exquisite activity against staphylococci, including MRSA (typical MIC_{90}: 0.015 mg/l) and low resistance potential [15]. Activity of AFN-1252 is not reduced in *Staphylococci* resistant to other classes of antibiotics. Microdosing and Phase I clinical studies indicate high oral bioavailability, a long elimination half-life of 8–11 h and good tolerability [16–18]. A Phase II study has been completed and excellent results have been presented at the European Congress of Clinical Microbiology and Infectious Diseases in Berlin, Germany [19].

AFN-1252 offers considerable potential as a new therapy for the treatment of staphylococcal infections by specifically targeting FabI. The intrinsic species specificity brings considerable benefits and addresses the demand for a specific agent against staphylococci. The benefits include assured antistaphylococcal efficacy, including against MRSA, which may reduce treatment duration, minimize resistance development, safety and cause limited disturbance of the microbiome. The net result should be an enhanced benefit–risk ratio. Additionally, AFN-1252 offers the potential option of intravenous and oral formulations so that patients can be treated within inpatient and outpatient settings with relative ease.

How would a species specific agent such as AFN-1252 be used in the clinic? There are many applications where AFN-1252 could provide benefit and address an unmet need, including:

- Monotherapy, pending microbiological confirmation, in infections where *staphylococcus* is present as a single pathogen infection. As most staphylococcal infections are not polymicrobial, the possibility of effective monotherapy in diseases with defined microbial etiology is attractive;
- Combination therapy for mixed infections, particularly where MRSA is involved (perhaps the highest usage). Vancomycin is the current drug of choice but requires serum monitoring and has become progressively less effective as staphylococcal isolates become less susceptible due to MIC creep and development of hVISA, and the regular need for dose escalation has resulted in increased rates of subsequent nephrotoxicity;
- Ability to de-escalate therapy to a specific targeted drug once resistant *staphylococci* have been confirmed. This may result in AFN-1252 monotherapy or a simpler antibiotic combination, but meets the safety- and resistance-prevention criteria that 'best is least';
- An effective oral agent for use against serious infection with MRSA or VISA, in preference to long-term parenteral therapy with currently available agents;
- Allow safe and long-term oral therapy for bone, and/or joint infections, where long-term safety is paramount

The clinical role of FabI inhibitors will become clearer over time. They definitely represent a novel and important antibacterial platform that has the potential to address current bacterial resistance problems.

Financial & competing interests disclosure

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Website

101 Clinial Trials Database: NCT01593761. www.clinicaltrials.gov/ NCT01593761