evotec

DDup

DRUG DISCOVERY UPDATES #5

ANTIBIOTICS
BREAKING DOWN THE CELL WALL

ANTIBIOTIC RESISTANCE
Global threat to patients and healthcare systems

EUPROTEC ACQUISITION
Adds biology expertise and capabilities in anti-infectives

CONTENT

Dr Werner Lanthaler, CEO

WELCOME TO DDUP!

DEAR FRIENDS OF EVOTEC,
You will be well aware that the rapid emergence of antibiotic resistance is a threat on a global scale with the potential to undermine the advances that have been made in medicine over the last 100 years. The potential crisis that the world faces has been compounded both by the innate difficulty in the discovery of new antibiotics and the exit of pharmaceutical companies from antibacterial research for commercial reasons.

Over the last decade at Evotec we have collaborated with a variety of Biotech and Pharma companies on antibiotic drug discovery and recently we have seen increased interest from our collaborators in this therapeutic area. In the last year we have grown Evotec's expertise in antibacterial research and leveraged our capabilities in concert with expert academic groups to deliver new options for therapeutic intervention for resistant bacterial infections of urgent and serious medical need.

In this, the fifth, issue of DDup we introduce our new capabilities in microbiology that result from the integration of Euprotec into the Evotec family and also the first internal antibacterial project ("Target*PGB*") which is being conducted in collaboration with Professors Dan Kahne and Suzanne Walker at Harvard University to identify inhibitors of peptidoglycan biosynthesis.

The Euprotec team brings to Evotec an extensive and innovative portfolio of assays and disease models in infectious disease as well as significant expertise gained over many years in the area. The Kahne and Walker research groups have decades of experience in fundamental research on bacterial biosynthesis pathways and their inhibition and have developed an extensive toolbox of complex reagents and protocols that unlock a family of enzymes referred to as peptidoglycan transferases ("PGTs") pivotal in bacterial cell wall synthesis. The enzymatic reaction catalysed by PGTs is essential for bacterial cell wall synthesis and hence survival yet has not been previously addressed by any systemically available antibiotic. Building on the important findings of the Kahne and Walker research groups, Evotec is applying its enhanced antibacterial drug discovery platform to industrialise the discovery and optimisation of inhibitors of peptidoglycan transferases as a powerful class of new antibiotic agents.

We see the antibacterial field as being critical to our strategy moving forward and we are passionate about finding effective ways in which to discover and progress new agents to the clinic. We seek fruitful discourse with academic specialists, industrial collaborators and national research agencies to achieve the crucial advances needed to address the treatment of resistant bacterial infections. I hope that you find this latest edition of DDup of particular interest and as always please don't hesitate to contact us.

Yours sincerely
Werner Lanthaler

ANTIBIOTICS

Resistance to antibiotics has become a critical health issue for patients and payers. It has been known for many years that bacteria develop, to varying extents, resistance to each antibiotic over time. Indeed in his 1945 Nobel Prize Lecture Alexander Fleming who discovered penicillin stated "it is not difficult to make microbes resistant to penicillin in the laboratory by exposing them to concentrations not sufficient to kill them, and the same thing has occasionally happened in the body." Over the years antibiotic resistance was addressed by the introduction of new classes of antimicrobials that possess different mechanisms of action or by protecting existing antimicrobials against the resistance mechanism.

However, in recent years many pharmaceutical companies, who were once pre-eminent in the field, have curtailed their antibiotic drug discovery activities. There are two key factors that have contributed to this situation. Firstly, the financial rewards were perceived to be less attractive to pharmaceutical companies in comparison to other therapeutic areas, such as oncology and diabetes, which require chronic dosing. This was a result of rising development costs, as more stringent regulatory hurdles were introduced, and concerns that revenues would be limited by both the low established pricing and by new compounds being held in reserve for the treatment of severe resistant infections.

Secondly, it became increasingly difficult to discover new antibacterial compounds as the "low hanging fruit" of tractable drug targets addressed by natural products was exhausted. Attempts to address this lack of productivity in antibiotic research have for the most part failed and have not been helped by the closure of expert research groups within the pharmaceutical industry.

The concern is that if the challenge of antimicrobial resistance is not addressed, medicine could revert to a pre-antibiotic era whereby

patients undergoing routine surgical procedures die of infection because no effective antibiotics are available. In its recent report entitled "Antibiotic resistance threats in the United States, 2013" the Centres for Disease Control and Prevention ("CDC") estimate that in the US more than two million people contract antibiotic-resistant infections each year, with at least 23,000 dying per annum as a result. Similarly, the European Centre for Disease Prevention and Control has estimated that 25,000 deaths each year in the EU result from infection by resistant organisms.

The CDC report identifies three hazard levels for antibiotic resistance: Urgent, Serious & Concerning. Whilst some pathogens, such as MRSA ("Methicillin-resistant Staphylococcus aureus:- a Gram-positive organism associated with serious hospital infections"), are considered a serious threat others such as CRE ("carbapenem-resistant Enterobacteriaceae"), Klebsiella and E. coli have rapidly emerged as an urgent threat resistant to all or nearly all the antibiotics we have today.

To avert the global threat to public health will require a concerted scientific and political effort both to ensure effective stewardship of antibiotic therapies and incentives for academia and pharmaceutical companies to engage in antibiotic discovery on a sufficient scale to tackle the current problem. The first steps are being made to establish a new environment for antimicrobial research and a key example is the passing in the United States of the GAIN ("Generating Antibiotic Incentives Now") Act. This provides companies with 5 years of additional market protection for a new antibiotic and 6 months of additional exclusivity, if there is a companion diagnostic test, as well as prioritised review by the FDA.

Recognising the urgent medical need to address antibiotic resistance, Evotec recently entered into a collaborative partnership with Professors Dan Kahne and Suzanne Walker on the discovery of novel bacterial cell wall inhibitors. Their combined knowledge, expertise and capabilities will enable bacterial cell wall inhibitor drug discovery. In particular, the scientists engaged in the collaboration will benefit from a leading technology platform and robust assay systems including extensive *in vitro* and *in vivo* microbiology capabilities from the integration of Euprotec to advance the optimisation of effective antibacterial agents.

DISCOVERY AND DEVELOPMENT OF ANTIBACTERIAL DRUGS

The discoveries of many of the major classes of antibacterial drugs have been achieved through empirical screening of fermentation products or chemicals for inhibition of bacterial growth. In recent years it has proven easier to make incremental improvements (activity against bacteria resistant to the parent molecule, increasing bacterial spectrum, improving safety and simplifying dosing regimen) to existing antibacterial classes than to discover new drug classes. However, the previous FDA regulatory hurdle of proving non-inferiority has resulted in a number of otherwise valuable antibiotics with improved activity against resistant pathogens not being introduced to the market. Recognising this issue the FDA and EMA in 2013 introduced draft guidelines for the streamlined development of new antibiotics (EMA guidelines accepted May 2014). When fully implemented these should reduce the costs and shorten timelines of introducing new antibiotics effective against resistant bacteria.

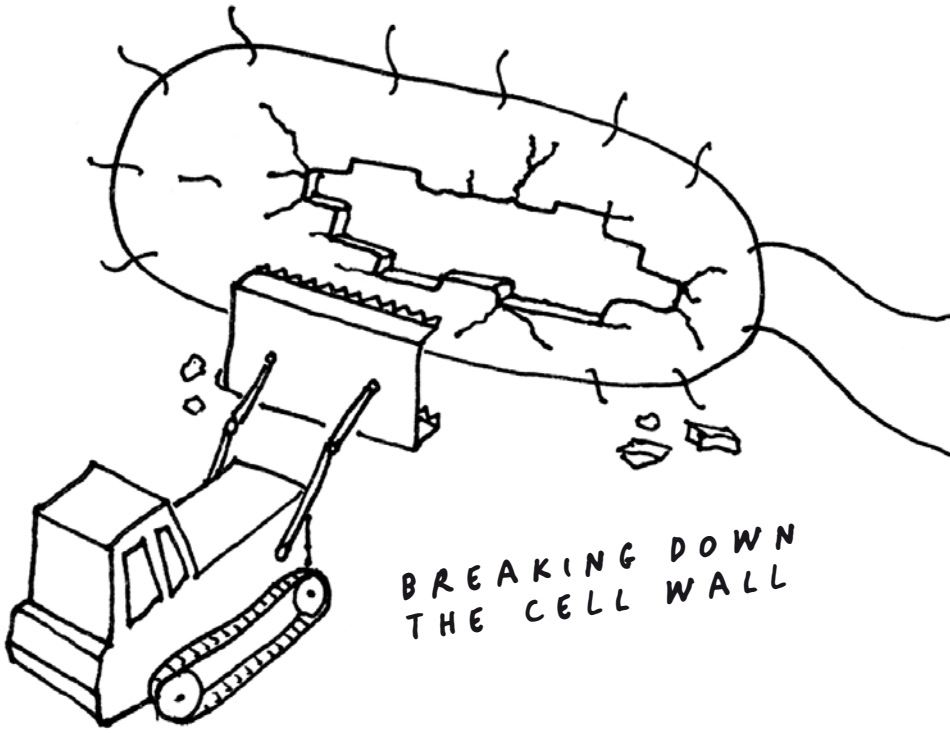
THE COLLABORATION WITH HARVARD UNIVERSITY

Harvard University became a centre of excellence for medical research soon after the Medical School was established in 1782. It is at present ranked the number one research medical school in the USA. Anti-infectives research has been a key activity since 1800 when Harvard Harvard Professor Benjamin Waterhouse introduced the first smallpox vaccine to the United States. The work of Professors Dan Kahne and Suzanne Walker focuses on the study of pathways of cell wall biosynthesis and applications to antibacterial drug discovery. The Walker group studies metabolic pathways involved in microbial survival and pathogenesis with the aim of developing new approaches to overcome resistant microorganisms and the Kahne group peptidoglycan biosynthesis and outer membrane assembly.

The partnership between Evotec and Harvard University is based on a new collaborative model that completely aligns the interests of both partners, and also merges Harvard's strengths in the biology and chemistry of peptidoglycan biosynthesis and Evotec's strengths in drug discovery. Evotec's industrialised drug screening and biophysical characterisation assays, structural biology and medicinal chemistry experience will be particularly important in discovering candidate compounds for advanced biological studies.

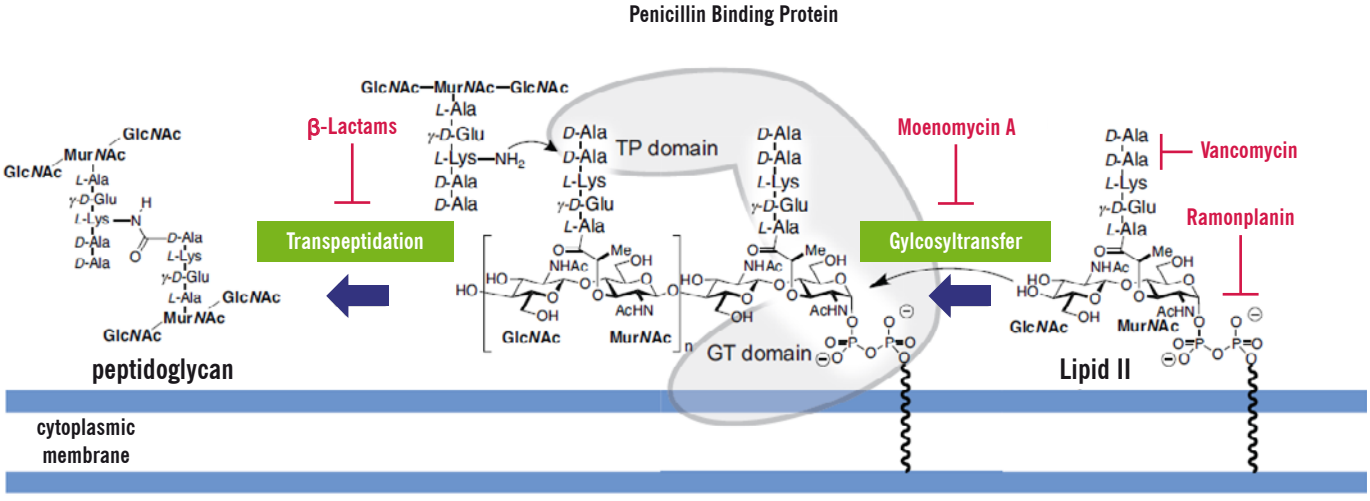
A distinguishing feature of the cell wall of bacteria is a peptidoglycan mesh located outside of the cytoplasmic membrane. This peptidoglycan mesh provides rigidity and shape to bacteria cells and is required for survival due to their high internal osmotic pressure. The cell wall structure of different species of bacteria fall into two main classes. The Gram-positive class of bacteria feature a thick cell wall consisting of many layers of peptidoglycan whereas the cell wall of the Gram-negative class of bacteria has fewer layers of peptidoglycan but features a surrounding second outer phospholipid membrane layer. The peptidoglycan mesh for both classes is made up of a polysaccharide backbone cross linked by short peptides. Penicillin and other β-Lactam antibiotics inhibit the formation of these peptidic cross links and so lead to cell wall rupture and death of growing bacteria due to their high internal osmotic pressure. This important class of antibiotics does not affect human cells in the same way as human cells just have a cell membrane and do not have a peptidoglycan cell wall.

Professors Dan Kahne and Suzanne Walker at Harvard for many years have been studying bacterial cell wall biosynthesis which involves the concerted activity of more than ten different enzyme families each of which could be a potential antibiotic target. In particular, they have identified the penultimate step in the synthesis of the bacterial cell wall as a target of particular interest. This is the transglycosylase step which is responsible for forming the polysaccharide backbone of peptidoglycan.



A key observation that has been built upon by Dan and Suzanne is that this step is inhibited by the complex natural product antibiotic moenomycin. Whilst this molecule is used as growth promoter in animal feed its properties make it unsuitable for use as an antibiotic in humans. However, through complex synthetic chemistry in Dan's lab and structural studies performed in Suzanne's lab, they have been able to develop an understanding of the features of moenomycin required for binding to the peptidoglycan glycosyltransferase enzymes. Furthermore, based on this understanding they have designed and synthesised a fluorescently labelled derivative of moenomycin to enable a high throughput screen to be conducted to identify small molecule inhibitors of binding of this probe molecule to a peptidoglycan glycosyltransferase enzyme.

On learning about the results of an initial screen we were very excited to enter into a collaboration since Suzanne and Dan have put in place all the tools to enable industrialisation of the screening and drug discovery for this essential bacterial target. In a relatively short period of time Evotec has conducted both a high-throughput screen and a fragment screen of a bacterial peptidoglycan glycosyltransferase enzyme and identified hit compounds which in subsequent hit validation experiments have been shown to be antibacterial. Importantly, results from assays performed in Dan and Suzanne's groups are consistent with on target be hyphenated. ●



LEADING THE FIELD IN THE BIOLOGY AND CHEMISTRY OF BACTERIAL WALL BIOSYNTHESIS

Suzanne Walker, Professor of Microbiology and Immunobiology at Harvard Medical School

Dan Kahne, Professor of Chemistry and Chemical Biology and Professor of Biological Chemistry and Molecular Pharmacology at Harvard

Suzanne Walker has expertise in studying bacterial cell wall biosynthesis and its inhibition. She has developed chemical approaches to study the membrane-linked steps of peptidoglycan and teichoic acid biosynthesis, and has made fundamental contributions to understanding the enzymes involved in these processes and the mechanisms of action of antibiotics that inhibit them.

Suzanne works on Gram-positive organisms, including the pathogens *Staphylococcus aureus* and *Enterococcus faecalis*, and a major research focus includes exploring novel strategies to overcome antibiotic resistant microorganisms. She also works on an unusual eukaryotic glycosyltransferase, O-GlcNAc transferase ("OGT"), which is essential for embryonic development and continues to play critical roles in various signalling pathways throughout life. Suzanne's research program combines organic chemistry, enzymology, high throughput screening, biophysics, cell biology, and bacterial genetics to address problems of interest.

Dan Kahne started his career as a synthetic chemist where he developed a novel method to make glycosidic linkages for use in complex carbohydrate synthesis. He was the first to develop methods to change the natural carbohydrates attached to vancomycin allowing him to make derivatives of vancomycin that kill vancomycin-resistant bacteria.

For the last fifteen years, Dan has focused on understanding the biogenesis of the cell envelope of Gram-negative bacteria, in particular peptidoglycan biosynthesis and outer membrane assembly. The assembly of this organellar membrane must be accomplished outside the cell in the absence of an obvious energy source. Dan's research focuses on identifying and understanding the machinery necessary for proper assembly of this membrane barrier, as well as the mechanisms that lead to defects. Because the outer membrane creates an effective permeability barrier to most antibiotics, understanding how to interfere with its assembly could provide new targets for antibiotic discovery.

5 MINUTES

WITH DAN KAHNE AND SUZANNE WALKER ON PEPTIDOGLYCAN BIOSYNTHESIS

Cord Dohrmann, CSO of Evotec ("CD"): The cell wall is essential to bacterial survival. Your research groups have worked to unravel critical biosynthetic pathways and give new insight into antibiotic discovery. What led you to focus on bacterial cell wall biosynthesis rather than other essential pathways?

SW: This pathway attracted us because there was an obvious problem we thought we could solve – namely, how to study the enzymes involved in the latter half of the pathway. Because peptidoglycan is a target for some very important antibiotics, we were quite surprised to learn that almost nothing was known about several of the enzymes in the pathway. And that was because it was almost impossible to isolate the necessary substrates. So we decided to work out methods to make them.

DK: I think part of the interest derived from having this unique approach. The other thing I would say is that there are a lot of interesting issues in this pathway. Even now there is a lot that is not well understood about how the cell wall is made and modified, and there are a lot of challenges in studying it. From an academic perspective, it's an interesting (hard) problem.

CD: The synthesis of the bacterial cell wall is a complicated multistep process. What benefits or advantages do you see in targeting peptidoglycan transferases over alternative steps in bacterial cell wall biosynthesis?

DK: Obviously, the pathway is essential and some steps have been validated with very important drugs such as the β-lactams and glycopeptides. In general, our perspective on antibiotics is that not all essential targets are suitable for antibiotic intervention, but one criterion one can use to determine if a target is good is if there is a natural product that targets it. That's an indication that you can kill cells by blocking that step. In the case of the PGTs, there is a potent natural product inhibitor: moenomycin.
At the same time, it is attractive that this target hasn't been clinically validated because it means there is a big opportunity. We could get a first-in-class antibiotic with a novel mechanism of action – and of course, new composition of matter. I think that's pretty attractive. The academic always says, if people haven't already exploited a target, there's an opportunity to develop new basic stuff, so we like that.

SW: That PGTs are on the extracytoplasmic surface of the membrane is also attractive as it means that gaining access to the target is easier – at least for Gram-positive organisms.

CD: For peptidoglycan transferases the complexity of both substrates and products is particularly challenging. What do you consider are the key advances that you and your research groups have made to unlock the discovery of inhibitors of this target class?

DK: One of the things that we would say is that the ability to synthesize the substrates, and in particular, to be able to make discrete chain length polymers of cell wall, is a huge advantage and uniqueness, I think, to our groups. Then taking those, being able to obtain well-defined substrates, allowed us to develop enzymatic tools, and obviously the development of enzymatic tools eventually led to the development of a high throughput screen for PGT inhibitors.

SW: Another point is that we spent a great deal of time trying to get structural information on the targets. The ability to get structures complexed with the natural product moenomycin has given us a unique perspective on where we're trying to target the inhibitors in the active site of these enzymes. Obviously the one thing we're lacking at the moment – no one has a structure of a non-carbohydrate-based inhibitor bound to any of these targets. But that would be a huge step towards making something that could be a lead compound. We are optimistic that that will happen through the Evotec collaboration.

CD: An ideal antibiotic exhibits broad spectrum activity across different bacterial species allied to a low frequency of spontaneous resistance. Can you comment on your expectations on the potential for inhibitors of peptidoglycan transferases as antibiotics?

SW: The catalytic machinery of the PGTs is highly conserved across all species – both Gram-negative and Gram-positive – and that suggests there is potential to make a broad spectrum antibiotic. In fact, the natural product that we study, moenomycin, inhibits PGTs from Gram-positive and Gram-negative bacteria. However, moenomycin is not a good lead compound because of limited solubility. It can't penetrate the outer membrane of Gram-negative bacteria such as E. coli, and because of poor physical properties it's not really a good starting point even for treatment of Gram-positive infections. So that's why we would like to find molecules that target the most conserved region of the active site but don't have the same problems as moenomycin.

DK: Resistance also does not develop readily to this target. *In vitro* target resistance frequencies to moenomycin are very low. And although moenomycin has been used as a feed additive for a long time, resistance does not seem to be a problem. Maybe this is because bacteria utilize multiple variants of these PGTs in order to build and modify the cell wall, and a resistance mutation in one of them doesn't confer high level resistance. I think the ability to hit multiple targets is a feature that has made the β-lactams such an effective class of antibiotics for so many decades. We're interested to see whether we can create compounds which are difficult to develop resistance against – and there is some expectation that we might be able to if we can bind the conserved active site residues.

> "We could get a first-in-class antibiotic with a novel mechanism of action"

The other comment is, I think we are still interested in broad spectrum antibiotics. There will be a point in time when people will go for narrow spectrum antibiotics. I don't think we're there yet. I do think that the question about resistance is something that I would worry a great deal about with a narrow spectrum antibiotic – if resistance is possible to develop, narrow spectrum antibiotics really lose their potential commercial value fast, and that could be worrisome.

CD: Whilst the exit of pharmaceutical companies from antibacterial research has in part been driven by financial considerations I presume an additional factor has been difficulty in discovering new classes of antibiotics. What do you see as the future challenges for antibacterial discovery?

DK: There are certainly regulatory barriers that make development of antibiotics challenging. But there are always going to be opportunities when you have new chemical matter, and also have a new target with a distinct mechanism of action. There are clear arguments to be made for the PGTs as really good targets. The challenge then is the chemical matter. We need compounds that have a good safety profile that are cell-permeable. Obviously, if we could find chemical matter that could penetrate the outer membrane, the possibility of a truly broad spectrum, antibiotic that can target Gram-positive and Gram-negative would make for a blockbuster drug. Everything depends on what we're going to find in this collaboration with Evotec, in terms of the chemical matter, but that's what makes it exciting and a nice collaboration.

CD: Thank you for your time.

Dr Cord Dohrmann is Chief Scientific Officer and Member of the Management Board of Evotec. Dr Dohrmann has spent over 20 years in biomedical research at leading academic institutions and in the biotech industry.

FACTS & FIGURES

ANTIBIOTICS

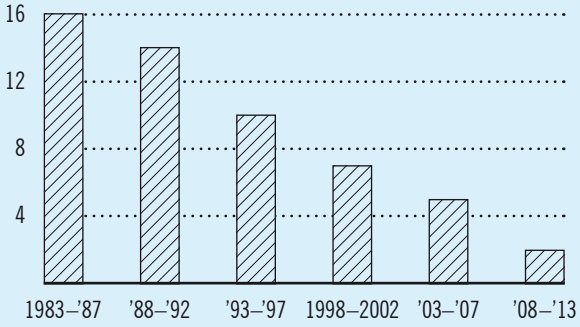
- Between 1945 and 1968, drug companies invented 13 new categories of antibiotics, but **only 2** in the next 2 decades and **none** since 1987.
- According to the Infectious Diseases Society of America, just a handful of large multinational Pharma companies are researching new antibiotics
- Currently less than 10 drugs are in Phase II or III development for treatment of infections caused by resistant gram-negative bacilli

SALES STATISTICS

Best selling antibiotic drugs are generic yet (global sales in 2011):

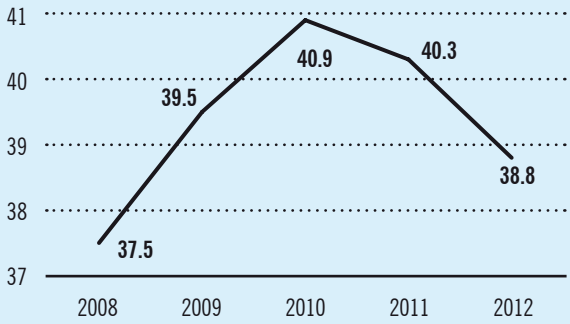
- Amoxicillin - $ 2.41 bn
- Augmentin - $ 1.63 bn

NEW SYSTEMIC ANTIBACTERIAL AGENTS APPROVED BY THE FDA THROUGH 2013



- Drastic decrease of approvals in the past 3 decades
- Only 4 drugs approved with novel mechanism of action since 1998
- Less approvals from 1998 to 2013, than in the period 1983 to 1987

ANTIBIOTICS – GLOBAL SALES IN BN



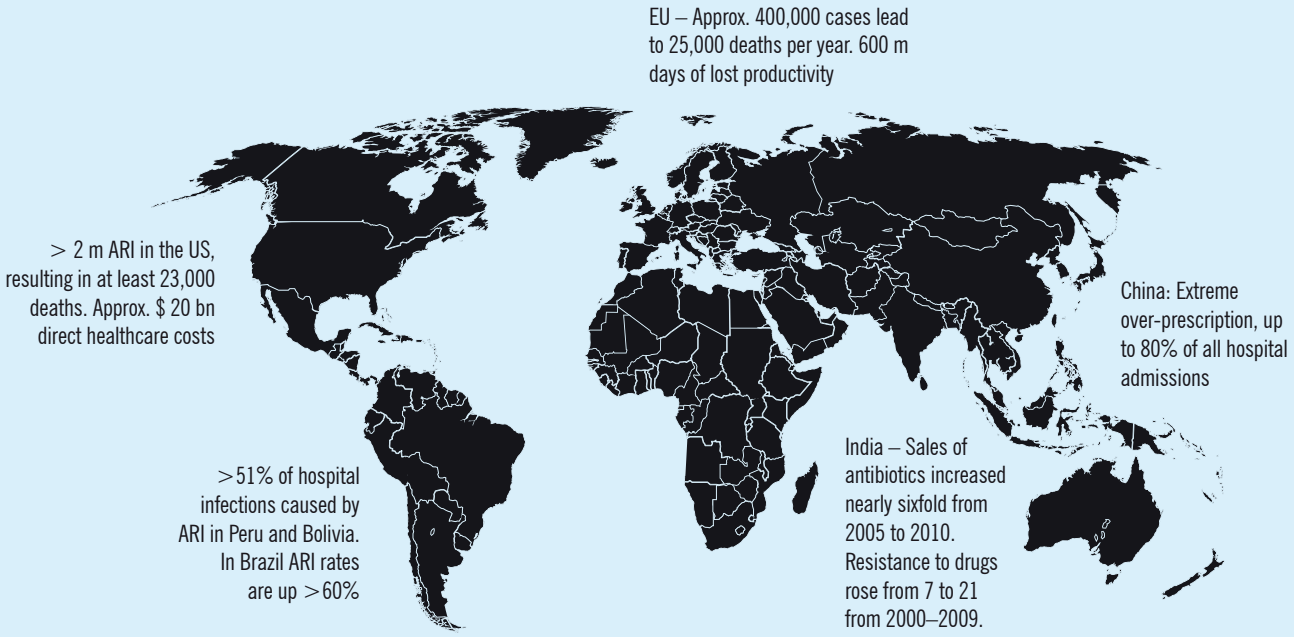
- Little change in total sales over the last 5 years
- Daptomycin, approved in 2003, reached global sales of > $ 1 bn in 2012, Ceftaroline, approved in 2010, $ 44 m.

ANTIBIOTIC RESISTANCE

"New forms of antibiotic resistance can cross international boundaries and spread between continents with ease."

Quotation from the 2013 CDC AR report

- HIGH DEMAND VS FEW APPROVALS AND NEW DEVELOPMENTS
- ANTIBIOTIC RESISTANCE, AN INCREASINGLY SIGNIFICANT THREAT FOR HEALTHCARE SYSTEMS WORLDWIDE
- DEVELOPMENT OF NEW CLASSES OF ANTIBIOTIC DRUGS NEEDED



According to the CDC, the US figures are likely minimum estimates. In addition, many more die from other conditions that were complicated by an antibiotic-resistant infection. Examples of key antibiotic resistance events:

- 1962: Methicillin-R *Staphylococcus* – Approval in 1960
- 1987: Ceftazidime-R Enterobacteriaceae – Approval in 1985
- 1996: Levofloxacin-R *Streptococcus pneumoniae* – Approval in 1996
- 2011: Ceftaroline-R *Staphylococcus* – Approval in 2010

CURRENT ANTIBIOTIC RESISTANCE THREATS IN THE US ACCORDING TO CDC

Microorganism	Infections/year	Deaths/year	Remarks
Clostridium difficile	250,000	14,000	Deaths related to *C. difficile* increased 400% from 2000 to 2007
Carbapenem-resistant Enterobacteriaceae	9,000	600	CRE have become resistant to nearly all available antibiotics
Drug-resistant *Neisseria gonorrhoeae*	246,000	Figure not available	CDC estimates more than 800,000 gonococcal infections per year
Vancomycin-resistant *Enterococcus*	20,000	1,300	Few or no treatment options left due to Vancomycin resistance
Methicillin-resistant *S. aureus*	80,400	11,285	Leading cause of healthcare-associated infections
Drug-resistant *S. pneumoniae*	1,200,000	7,000	Leading cause of bacterial pneumonia and meningitis in the US

EVOTEC'S NEW CAPABILITIES IN ANTI-INFECTIVE RESEARCH

Evotec's specialist group in the infectious disease therapeutic area boasts state-of-the-art microbiology facilities including a unique and highly characterised strain bank. The group was established in 2008 as Euprotec and was acquired by Evotec in May 2014. The group provides bespoke anti-infective drug discovery and development services to a growing number of global clients and brings to Evotec an established track record in collaborating to discover and develop therapies and vaccines to treat and prevent serious and life-threatening infections resulting from multi-drug resistant bacteria and fungi including MRSA, Pseudomonas, and Clostridium difficile.

In 2012, Euprotec was named the Bionow "Biomedical Service Provider" of the Year in recognition of delivering scientific excellence and the very best in customer service to all of its clients. The group's offering is fully integrated with the wider discovery platform at Evotec to enable us to offer either standalone microbiology services or conduct fully integrated anti-infective drug discovery projects. Key capabilities of Evotec's microbiology group fall into the four areas of StrainBank, Microbiology, Pharmacology and ADME/PK/PD as follows:

STRAINBANK

StrainBank is a unique collection of clinical isolates that can be used establish the activity profile of lead compounds and candidates. A key feature is that the isolates are highly characterised and, in many cases, mechanisms of resistance defined. Our StrainBank collection contains an extensive range of geographically diverse human bacterial and fungal pathogens that cover isolates susceptible and resistant to current antimicrobial drugs.

MICROBIOLOGY

We employ industry-standard methods including CLSI, EUCAST and BSAC to test compounds for antimicrobial activity against organisms from our extensive collection, StrainBank, or strains provided by our collaborators. This includes the ability to conduct whole-cell screening for antimicrobial activity for hit identification, MIC, MBC/MFC, time-kill and PAE studies using single or combination agents, hollow fibre PK/PD or bioreactor human cell systems for detailed profiling for characterisation of novel anti-infective agents and compound/drug combination studies for assessment of synergistic, antagonistic and additive effects.

Bespoke methods for susceptibility profiling can be developed for testing novel agents where standardized methods may not be appropriate. In parallel mechanism of action and resistance frequency assays can be performed.

PHARMACOLOGY

We specialise in assessing the efficacy of lead and candidate compounds in highly relevant and validated disease models. Our extensive range of established models are well-suited to the development of multiple classes of agent including small molecules, natural products, peptides, antibodies, other biologics and vaccines. We provide a highly bespoke service, customising studies to meet the exact requirements based on programme needs and parameters/endpoints of interest.

For agent efficacy assessment against bacterial and fungal infection our models can address different sites of infection (localised and systemic) by Gram-positive (including *Staphylococcus aureus*, *Streptococcus pneumoniae*, and others), Gram-negative (including *Pseudomonas aeruginosa*, *Escherichia coli*, *Acinetobacter baumanii* and others), anaerobes (including *Clostridium difficile*) or fungal species (including *Candida* sp., *Aspergillus* sp., Mucorales, *Malassezia* sp.).

ADME/PK

Our existing extensive ADME/PK offering is now enhanced with the ability to address anti-infective specific capabilities. In addition to high quality pharmacokinetic profiling and bioanalysis these include:

a) Bioassay/bioactivity drug quantification
b) Development of bespoke PK models and assays with drug quantification at multiple sites
c) Translation of PK data into efficacy models to optimise outcomes
d) Tolerability assessment utilising multiple endpoints
e) Early assessment of cytotoxicity potential against multiple mammalian cell lines.

In particular, Euprotec specialises in PK/PD profiling and modelling in multiple disease models.

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TECHNOLOGY OVERVIEW

EVOTEC'S INTEGRATED ANTIBIOTIC DRUG DISCOVERY PLATFORM

With the incorporation of microbiology into its drug discovery platform Evotec is able to offer integrated antibacterial drug discovery from screening through to candidate identification. This platform is being applied in the collaboration with Harvard. The team that has joined from Euprotec has great experience of working across a wide range of biological targets and with multiple classes of drug including small molecules, natural products, peptides and biologics.

1. PROPRIETARY EVOSCREEN™ AND EVOLUTION™ HIT IDENTIFICATION PLATFORMS

- 400,000 member screening collection
- 24,000 fragment library
- Orthogonal screening with NMR, SPR and RapidFire™

2. EXPERIENCED STRUCTURE-BASED DRUG DESIGN PLATFORM

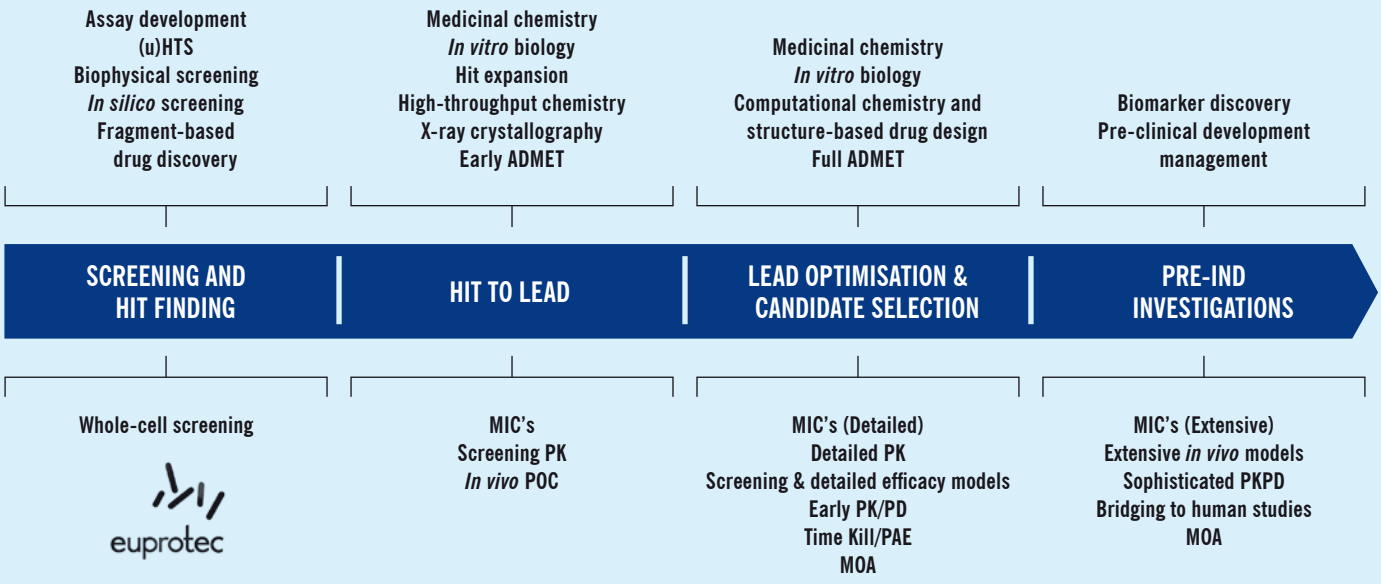
- Computational support for structure and ligand-based approaches
- Rapid generation of crystal systems
- Integrated medicinal chemistry teams

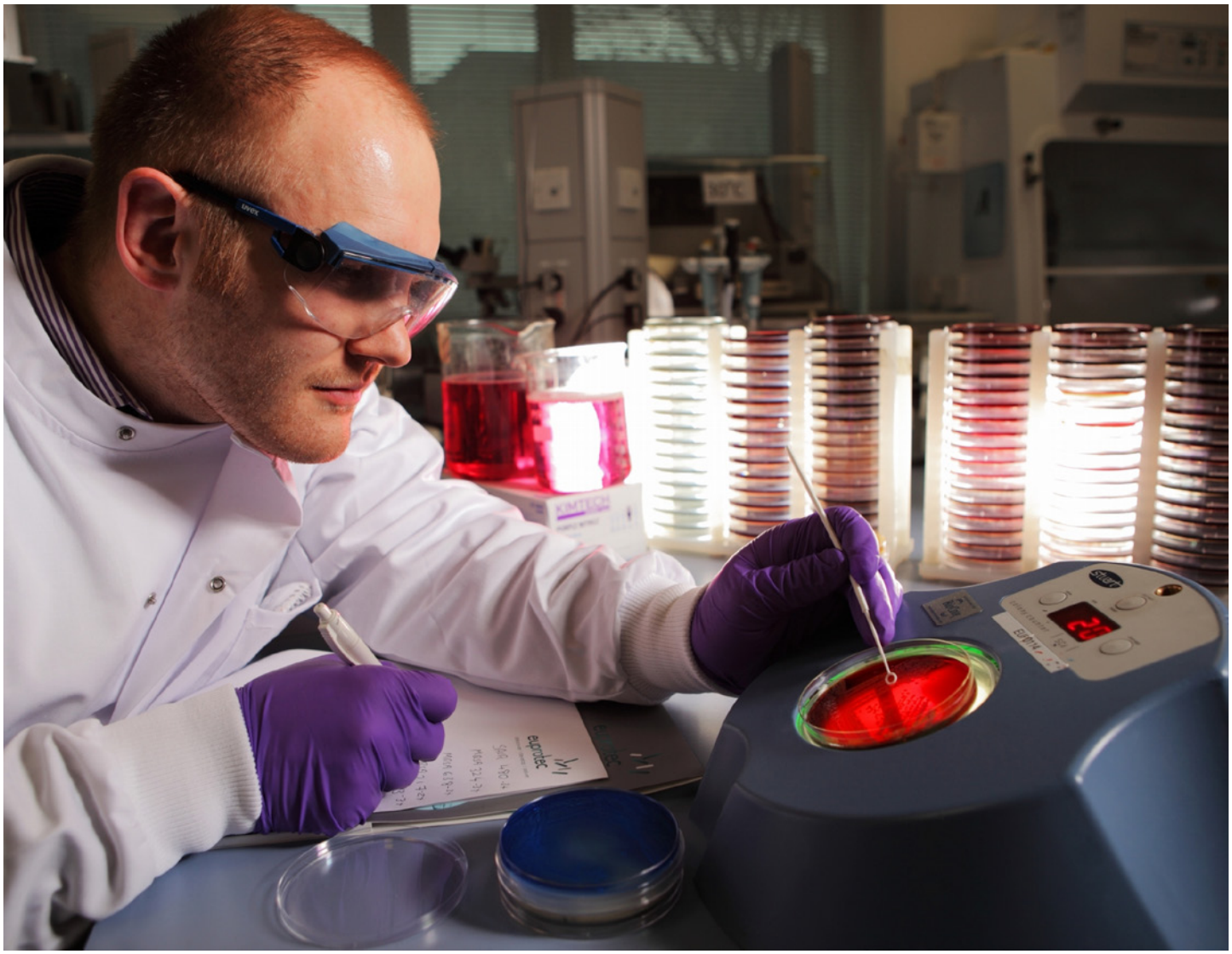
3. BROAD DRUG DISCOVERY PLATFORM FOR ANTIBACTERIALS

- Biochemical and biophysical screening
- Whole cell screening of compound collections
- Characterisation of binding dynamics
- Microbiology and susceptibility profiling
- Mechanism of action determination

4. SOPHISTICATED ANTIBIOTIC DRUG EVALUATION PLATFORM

- Mechanism of action determination
- Efficacy model development
- ADMET and PK
- PKPD profiling and mathematical modelling





In the Target*PGB* collaboration with Harvard this integrated antibiotics drug discovery platform will be applied in concert with unique capabilities available within the research groups of Kahne and Walker.

ASSAY TOOLBOX	COMPOUND SCREENING	HIT VALIDATION	HIT CHARACTERISATION	DRUG DISCOVERY
HARVARD ▶ PGT/PBP expression protocols ▶ Moenomycin-based fluorescent probe ▶ Demo HTS & 1st hit series	**EVOTEC** ▶ Diverse lead-like and fragment compounds collections ▶ Miniaturised HTS	**EVOTEC & HARVARD** ▶ SPR assays ▶ MIC assays ▶ MOA studies ▶ Cellular reporter gene assay ▶ Transposon assays ▶ Lipid II polymerisation assay	**EVOTEC & HARVARD** ▶ Detailed microbiology ▶ X-ray crystallography	**EVOTEC & HARVARD** ▶ Structure-based drug discovery ▶ Microbiology ▶ Animal models

ADVANCING THE DISCOVERY OF NEW ANTI-INFECTIVES

Peter Warn
SVP Anti-infectives Discovery

Before co-founding Euprotec, Peter was a senior lecturer at the University of Manchester's School of Translational Medicine, running a research group investigating the pathogenesis and treatment of severe infections. Peter has published over 60 peer reviewed articles on the development of antimicrobial agents. He has been awarded 14 years' continuous funding from the NIH, as well as from MRC, BBSRC and Pharma.

Four novel antifungal agents that he has assisted in developing are now either in clinical use or phase III clinical trials. Peter was previously at the University of Oxford's Tropical Infectious Diseases Unit where he studied severe malaria and bacterial infections in the paediatric population of coastal Kenya. In this role Peter ran the diagnostic laboratory at Kilifi-KEMRI (publications in Nature and New England Journal of Medicine) and assisted in the development of three novel anti-malarial drugs that

Lloyd Payne
SVP Anti-infectives Operations

are now widely available. Peter has also held posts as a senior clinical scientist within the HPA and NHS. These roles involved diagnostic microbiology from cases of human clinical disease in both hospital and community patients.

Prior to co-founding Euprotec Lloyd spent nearly 20 years in the biopharmaceutical and CRO sectors where he directed a number of drug discovery programmes in the infectious disease, oncology and inflammation therapeutic areas.

Lloyd was previously CSO of a UK-based biotech company focused on the development of novel classes of drug to treat life-threatening fungal infections. He was also a senior scientist at Millennium Pharmaceuticals (Takeda Millennium) where he led multidisciplinary programmes based in the UK and US. Lloyd has also held senior scientific and management positions within drug discovery CRO's Cambridge Discovery Chemistry and NCE Discovery (latterly Domainex) where he was responsible for programmes working with organisations across Europe, Japan/Asia, and the U.S. He has presented at a number of international conferences, published in peer reviewed journals, and is a co-inventor on patents related to drug discovery technologies as well as novel agents for the treatment of cancer, inflammation and infection.

PETER WARN AND LLOYD PAYNE ON ANTIBIOTIC DRUG DISCOVERY

Mario Polywka, COO of Evotec ("MP"): We are delighted to welcome you and your colleagues at Euprotec as part of Evotec. What led you to come together in 2008 to found Euprotec as a new provider of anti-infective drug discovery services?

LP: Both Peter and I have been completely committed to the infectious disease therapeutic area for a long time. After spending considerable time working together it became quite apparent to us that something was missing in the marketplace in terms of a CRO delivering the specialised biology capability required to discover and develop new antimicrobial agents. This, combined with the ever increasing trend for Pharma and biotech to outsource R&D, led us to bring together our industry and academic experience and incorporate Euprotec.

PW: Being passionate about the development of novel antimicrobial agents, we realized there was an urgent need for a specialist CRO focusing on infectious diseases with a portfolio that bridged the entire development pipeline from discovery to patient. Using our combined skills we believed that Euprotec could fill this vital niche to accelerate drug discovery.

MP: The business that you founded has shown good growth over the last few years. What are the trends that you have observed in anti-infective drug discovery during this time?

LP: In the past there was an increasing move by Big Pharma to reduce or cease investment into antibacterial R&D in order to focus on the development of drugs to treat chronic diseases. This has created an almost 'perfect storm' where a dry antibacterial pipeline cannot meet the challenge that antimicrobial resistance has brought to us. However, we believe the change we are seeing has led to the growth in our business. We are seeing examples of Pharma companies moving back into the therapeutic area and a significant number of new biotech companies focused on the discovery of new antimicrobials. This trend has been catalysed by a combination of factors including several Public Private Partnership initiatives as well as a number of new regulatory and economic incentives such as the GAIN act and expedited clinical development pathways. These factors, together with increased multinational government support has led to a greater emphasis in the discovery of new drugs, a trend we expect will continue and drive further significant growth.

PW: One of the major drivers has been the unexpectedly rapid increase in antimicrobial resistance exemplified by the appearance of virtually pan-resistant organisms expressing NDM-1 and the rapid increases of the incidence in CRE. Whilst 6 years ago there was concern within the scientific community about the increase in antimicrobial resistance there is now a clear realisation by governments that this is a major public health concern, and there is now a real drive to develop new antimicrobials supported by WHO, the EU and NIAID amongst others.

MP: Whilst this issue of DDup is focused on Evotec's capabilities in antibacterial drug discovery what other areas of anti-infective research are you involved in?

PW: Protecting the public from infection is multi-faceted and Euprotec has tried to provide services in many areas. Euprotec has developed bespoke models of

infection to assist in the optimisation of vaccines and biologics to prevent or treat bacterial and fungal infections against key pathogens. A key feature of these models is that they are clinically relevant and generate biomarkers that enable rapid progression towards lead identification and optimisation. Euprotec has a wide portfolio of strains and assays to assist in the development of antifungal drugs. Using these assays Euprotec has helped develop an antifungal that has recently completed phase III clinical trials.

LP: Since day one, Euprotec has continually developed its portfolio of assays and models to accelerate the development of new drugs to treat infection. One particular interesting area of research has its roots in the development of new drugs to treat *Clostridium difficile* associated diarrhea ("CDAD"). Much of the focus in this area has been on agents that are exquisitely selective for *C. difficile*, with the goal of sparing or quickly restoring normal gut microflora. This has led us to become involved in the development of pre- and pro-biotics as companions to new drugs. The skills we have developed have resulted in a deeper understanding of the human microbiome and how we are able to analyse and measure the impact of new drugs on resident microflora in models of disease, something which is receiving much attention across multiple therapeutic areas.

MP: Antibacterial drug discovery is a challenging area. What particular challenges do you see in a lot of your collaborations?

PW: There are several continuing themes we observe that are related to the complexity of access to the bacterial target site. The first common theme is the whilst many compounds are highly effective against an isolated biological target they are less effective or inactive in whole cell assays; in many cases the lack of efficacy is due to the difficulty in crossing the cell wall of bacterial or fungal cells due to the physical properties of the test articles. Euprotec has been successful in working with its partners to overcome these difficulties using SAR to progress the hits. With much activity in the natural product space, there are commonly small amounts of compound available meaning miniaturized assays are required to maximise data from the available material

LP: There are multiple challenges that we see in antimicrobial drug discovery, many of which are not limited to this particular therapeutic area. It is well documented that the cell wall creates a significant barrier to identification of potential new antimicrobial leads, however one aspect that we are constantly challenged by is getting the drug to the site of infection. With an increase in novel approaches and new types of compound, we often have to adapt our disease models and drug delivery to ensure *in vivo* proof of concept is not missed and to avoid 'losing' potential valuable leads. A good example of this is in the antimicrobial peptide ("AMP") space where drug delivery can be very difficult and compounds are rapidly cleared when first used in animals. In these situations, we have managed to 'tune' our disease models to achieve a meaningful endpoint to allow compounds to be further optimised.

MP: What do you see as the opportunities for growth and development of anti-infectives drug discovery as part of the Evotec group?

LP: We are very excited by the significant number of possibilities; perhaps one of the biggest opportunities arises from the fact that Euprotec is now part of an organisation that has a very highly accomplished drug discovery platform spanning multiple scientific disciplines. Bringing together the depth of expertise in anti-infective drug discovery and the broader Evotec platform, we see real opportunity in being able to strike up significant strategic relationships with Pharma and biotech companies in this therapeutic area. Additionally, the global presence of Evotec's brand and business development team will enable a strong commercial focus to help develop new relationships.

PW: One of the key areas we see is related to the ability to link our capabilities with a much broader platform leading to a deeper understanding of antimicrobial agents. Access to medicinal chemistry is a key component and also bringing to bear the ADME capability, physical assays, and bioanalysis that exist within Evotec. These are all components we are regularly asked for and, as a result of being part of the Evotec group, are now able to deliver.

DR MARK WHITTAKER

SHORT SUMMARY OF SCIENTIFIC CAREER

Dr Mark Whittaker obtained his DPhil in chemistry at the University of York in 1982 and then undertook post-doctoral research first at York University in Toronto and subsequently at the University of Oxford with a focus on asymmetric organic synthesis.

Mark worked at British Biotechnology from 1987 in a number of roles from Group Leader Medicinal Chemistry to Director of Chemistry. During this time he contributed to the discovery of seven molecules that progressed to human clinical trials in oncology, inflammation and antibacterial therapeutic areas.

Mark joined Evotec in 2001 as Director of Drug Discovery where he has assisted with the development of Evotec's service offering including enhancing computational chemistry and introducing structural biology as well as managing multiple drug discovery collaborations. More recently in his current role as SVP Drug Discovery, Mark is assisting with Evotec's early-stage anti-infectives research activities including leading the collaboration with Dan Kahne and Suzanne Walker at Harvard.

1. **What is so special about Evotec's scientific approach to antibacterial drug discovery?**

The integration of the extensive microbiology capabilities of Euprotec with Evotec's drug discovery platform provides an unparalleled capability in antibacterial drug discovery. As with all drug discovery projects the key factor for success is effective target selection and in the case of antibacterials it is important to understand early on not only the essentiality of the target across bacterial species but also the potential for resistance.

These questions are routinely addressed by our new microbiology group. For hit identification we can apply both cellular phenotypic assay methods and target specific biochemical assays and most importantly we have the biophysical tools in hand to address whether observed antibacterial activity is actually due to on target activity. Where possible we apply structure-based techniques for optimisation of binding to conserved target features across bacterial species and our medicinal chemists have an in depth understanding of MIC-based SAR development. In particular where the target is present in both Gram-positive and Gram-negative organisms we understand the design principles that lead to broad spectrum activity. In lead optimisation it is critical to assess candidate compounds in relevant infection models and to understand the relationship between pharmacokinetics and pharmacodynamics that leads to effective pathogen eradication. Our *in vivo* group has a wealth of experience of running such PK/PD studies to aid the selection of the best candidate compound for taking forward into pre-clinical development. In summary, we have a complete offering to address antibacterial drug discovery and most crucially our scientists are passionate about working to discover new antibiotic molecules that address the pending crisis of wide spread resistance to existing therapies.

2. **How will you contribute to finding novel drugs in this challenging area?**

I see my contribution as leading Evotec's global cross-disciplinary antibacterial discovery activities

in our collaboration with Harvard University. Drug discovery is truly a collaborative activity and it certainly 'takes a village' to create a new drug. At Evotec, I am privileged to work in a multidisciplinary environment with unity of mission and focus on the creation of novel antibiotics. A specific challenge is finding compounds with novel mechanisms of antibacterial action. This is highly desirable since it will provide antibiotic drugs that exhibit activity against pathogens resistant to contemporary therapies. Therefore, our key goal is the selection of bacterial targets that are viable for progression in terms of novelty, essentiality, potential for resistance and druggability.

I am confident that within Evotec we have the people and capabilities to progress such targets rapidly and effectively through the early stages of drug discovery to identify tractable hit compounds and then beyond into lead optimisation. The skills and expertise brought by our new microbiology colleagues will enable us to effectively understand the intricacies of pharmacology for each molecule and to use this information effectively in compound optimisation. In the past decade a number of the major pharmaceutical companies withdrew from the arena of antibacterial research with the devastating consequence that significant expertise was lost to the industry. With the recent renewed interest in antibacterial drug discovery, we at Evotec are well placed to help fill the expertise void and I see great opportunities for us to work both with academic and industrial collaborators in the creation of novel antibiotics.

3. How will your collaboration with Harvard University work in practice?

Right from the outset it has been a joy to collaborate with Professors Dan Kahne and Suzanne Walker of Harvard University on Target*PGB*. Their laboratories are renowned internationally for generating new insights into bacterial cell wall biosynthesis. Suzanne and Dan bring to our collaboration not only an unparalleled tool set of reagents, assay protocols and structural biology protocols but also an infectious enthusiasm for conducting high-quality research in pursuit of the discovery of novel antibiotics. Our collaboration to discover inhibitors of the peptidoglycosyl transferases ("PGTs") is structured to play to the special strengths of each party. Using the moenomycin-based probe discovered by Dan and Suzanne, Evotec has conducted a high-throughput screen on the EVOscreen™ platform to generate an initial hit set. These have been triaged through a panel of assays conducted both at Evotec and at Harvard. Surface Plasmon Resonance studies performed at Evotec have confirmed binding to PGT targets whilst at Harvard and Evotec MIC assays have confirmed antibacterial activity. Early results from Harvard's reporter gene and lipid II polymerisation assays are consistent with antibacterial activity being due to target engagement. It is pleasing that in such a relatively short period of time that we have jointly been able to identify a set of novel antibiotic compounds. This has been due to the excellent communication that has been established between the scientists at Evotec and Harvard and our focus on obtaining the best quality data possible.

We are now entering the next stage in our collaboration in which we will be undertaking structure-led optimisation of these promising hit series and we are confident that together we can identify a new class of antibiotics for addressing serious multidrug resistant pathogens.

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