Taking aim at malaria:

a target-based approach to drug design

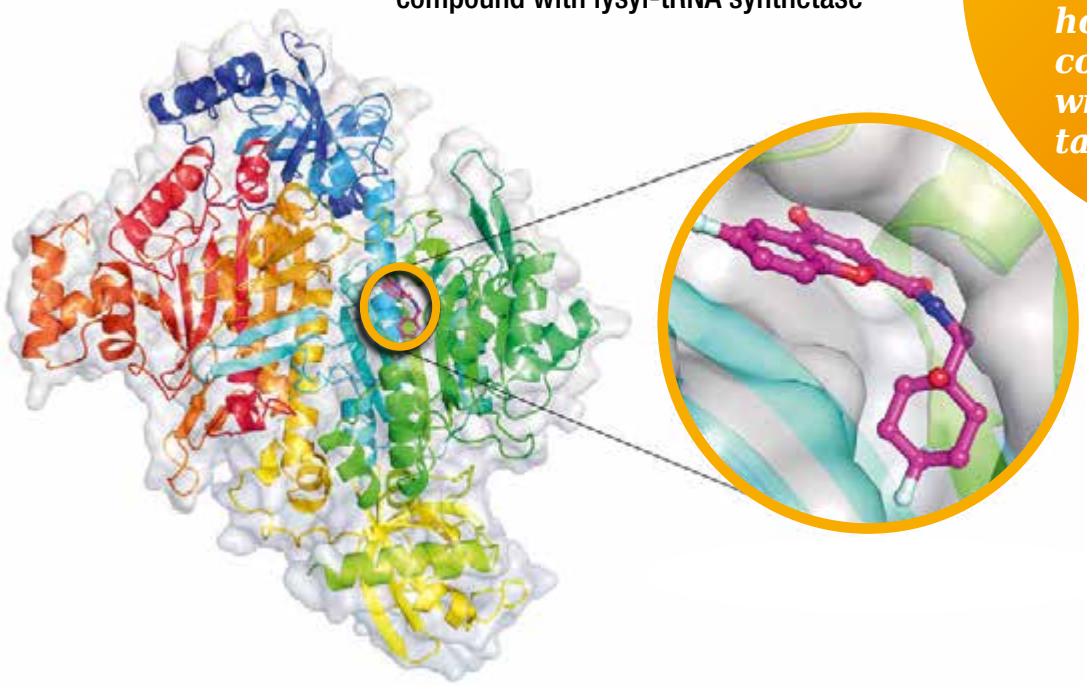
MMV's Project of the Year 2018 is awarded to a discovery team led by **Prof. Ian Gilbert**, **Prof. Kevin Read** and **Dr Beatriz Baragaña** at the Drug Discovery Unit (DDU), University of Dundee, UK. Working alongside **Dr Paul Willis** and **Delphine Baud** at MMV, as well as **Sir Simon Campbell**, of MMV's Expert Scientific Advisory Committee,[1] the project team have identified an exciting new compound series, active against a novel biological target – *Plasmodium falciparum*'s enzyme lysyl-tRNA synthetase (*Pf*KRS1).

In the early phases of malaria drug discovery, scientists use validated assays to perform two types of screening – phenotypic or target-based. In phenotypic screening, the aim is simply to identify compound series that kill the parasite, even if the exact mechanism of action is unknown. Over the last decade, MMV has identified the majority of its compounds this way. A more focused approach, however, is target-based screening, which aims to identify compound series that can inhibit specific biological processes or molecules that are known to be effective treatment targets. This approach, which has proved successful in many therapy areas, brings together complementary methodologies (including structural biology, computational chemistry, molecular biology and biochemistry). It also builds on existing knowledge, where available – such as the structure of a binding site – to inform and expedite drug design.[2] The initial challenge with the target-based approach, however, is identifying a good drug target.

At present, there are relatively few validated targets in the malaria parasite. The discovery and validation of *Pf*KRS1 as a novel biological target is therefore an important and exciting development. Representatives from the team at DDU and MMV tell us more.

> ***"[The technique]... allows scientists to use 3D computational models to visualize how specific compounds interact with their biological target."***

Figure 2: A co-crystal structure of the early lead compound with lysyl-tRNA synthetase



1 Expert Scientific Advisory Committee: an external body of experts that helps to identify the best projects worthy of inclusion in MMV's portfolio and continues to monitor progress through an annual review of all projects.
2 van Montfort R, & Workman P. *Essays in Biochemistry* 61:431-437 (2017).
3 Cladosporin was discovered in a phenotypic screen carried out by the Novartis Institute for Tropical Diseases (NITD) in collaboration with the Genomics Institute of the Novartis Research Foundation (GNF).
4 Baragaña B *et al.* "Lysyl-tRNA synthetase as a drug target in malaria and cryptosporidiosis". *PNAS*. 116(14):7015–7020 (2019).
5 SCID model: the laboratory model of malaria that provides the most accurate prediction of drug response in humans.
6 Selective activity against enzymes in the malaria parasite, without any cross-reactivity to the equivalent human enzymes.

What is the role of *Pf*KRS1 in the malaria parasite? What makes it a good drug target?

BB. KRS1 is a key enzyme involved in protein synthesis. Without it, the parasite cannot grow and therefore cannot survive. Unusually, *Pf*KRS1 is present across all stages of the parasite lifecycle – both asexual and sexual blood stages, as well as the liver stage – making it an attractive biological target that can address multiple target candidate profiles (p. 8). KRS1 is also present in other pathogens, such as the parasite that causes cryptosporidiosis. Compounds active against *Pf*KRS1 therefore have the potential to treat not only malaria, but also other neglected tropical diseases.

How was *Pf*KRS1 discovered and validated?

IG. The discovery and validation of *Pf*KRS1 speaks to the power of collaboration in malaria research. The first step was the discovery of a natural product (cladosporin) active against the malaria parasite, and the subsequent identification of its target (*Pf*KRS1) by Prof. Elizabeth Winzeler, one of MMV's long-term collaborators at the University of California San Diego.[3] Cladosporin was not developable as a drug candidate; however, working through the Structure-guided Drug Discovery Coalition, funded by the Bill & Melinda Gates Foundation, we carried out a project with the specific aim of exploring the potential of *Pf*KRS1 as a novel therapeutic target.

Working in collaboration with Prof. Wes van Voorhis at the University of Washington, we identified an inhibitor from a compound screen against *Pf*KRS1. This led to the screening of further compound libraries, generating more chemical series for optimization at DDU. We were also able to validate *Pf*KRS1 as a drug target in the SCID model of malaria[4,5] – the first time an animal model has been used to validate this target in the malaria parasite.

Structure-based drug design was also used in this project. Why was this important?

PW. Traditionally, the malaria drug discovery community has relied primarily on phenotypic screening to identify new compounds, but since this approach lacks an understanding of how the compound works in humans, it can make the optimization of a series to deliver a drug candidate more challenging. Structure-based drug design is a more sophisticated (though less commonly used) technique that allows scientists to use 3D computational models to visualize how specific compounds interact with their biological target (Figure 2). On this project, the insights gained from structure-based drug design have enabled us to optimize the potency and selectivity[6] of compounds active against *Pf*KRS1.

How has team collaboration contributed to the success of this project?

KR. The team synergy has been fantastic. MMV and DDU have enjoyed a successful working relationship for almost 10 years now, starting in 2010/11 with a project that ultimately led to the delivery of a new drug candidate – currently in Phase I testing (M5717 – p. 18). MMV has an in-depth understanding of malaria and an extensive network of global collaborators, which really helps to move compounds along the discovery pathway. At all stages, DDU has benefited enormously from MMV's advice and mentorship.

DB. Team collaboration has played a fundamental role in the success of this project. Testing new compound series is a logistically complex process as the validated assays for studying different stages of the parasite lifecycle are based all over the world. Without the strong partnerships we have with a wide range of centres – such as Imperial College (London, UK) and TropIQ (the Netherlands) for transmission-blocking assays; DDU (Dundee, UK) and GSK (Tres Cantos, Spain) for asexual blood-stage testing; and centres in Cambodia, Thailand and the USA for liver-stage testing – this work would simply not be possible.

How does it feel to receive MMV's Project of the Year award?

BB. It's a great endorsement for the team. The drug discovery pathway is a long one, and we first started working on this project back in 2014. This award recognizes the consistently hard work of the project team and its collaborators, and also motivates us to keep pushing forward. We are particularly proud given the many other competitive drug discovery projects currently active in MMV's portfolio.

What are the next steps for this project?

DB. Our focus now is on improving the properties of the compound series to deliver a drug candidate that can pass all the safety milestones required to advance to clinical development. Because *Pf*KRS1 is an enzyme essential for protein synthesis, we know that if we can successfully develop a drug candidate, it could kill the parasite at several different stages in the lifecycle, giving rise to a compound with good therapeutic potential for malaria. Going forward, we will continue to ensure that the goals of the project align with MMV's target candidate profiles and strategic imperatives, as well as the global elimination and eradication agenda for malaria.

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