Institution: Liverpool School of Tropical Medicine (LSTM)

Unit of Assessment: UOA1

Title of case study: Reducing Malaria Prevalence in Africa Through New Classes of Insecticide-Treated Nets.

Period when the underpinning research was undertaken: 2001-2020

Details of staff conducting the underpinning research from the submitting unit:

Name(s):	Role(s) (e.g. job title):	Period(s) employed by submitting HEI:
Mike Coleman	Senior Lecturer, Medical Entomology	2002 –
Martin Donnelly	Professor, Evolutionary Genetics	2001 –
Janet Hemingway	Professor, Vector Biology	2001 –
Gareth Lycett	Lecturer, Vector Genetics	2006 –
Philip McCall	Professor, Medical Entomology	1998 –
Mark Paine	Reader, Biochemistry	2006 –
Hilary Ranson	Professor, Medical Entomology	2001 –
David Weetman	Reader, Evolutionary Genetics	2006 –
Charles Wondji	Professor, Genetics	2004 –

Period when the claimed impact occurred: 2016 – 2020

Is this case study continued from a case study submitted in 2014? Y/**N** **NO**

1. Summary of the impact (indicative maximum 100 words)

Insecticide Treated Nets (ITNs) are the main malaria prevention tool in Africa, but their efficacy is being eroded by pyrethroid resistance in the mosquito vectors. LSTM's research determining the underlying molecular mechanisms led to new classes of ITNs that can control resistant mosquito populations. Our research and advocacy led to the founding of the Product Development Partnership IVCC and has generated the evidence leading to deployment of new classes of ITNs. By 2020, 13 of the 23 malaria endemic countries in Africa included these new classes of nets in their national distribution campaign, protecting over 35,000,000 individuals. A large-scale trial in Uganda found that use of this new net class reduced malaria prevalence by 27%.

2. Underpinning research (indicative maximum 500 words)

Since 2000 the global burden of malaria, especially in Africa, has reduced. Clinical incidence of malaria across the continent decreased by over 50% between 2000 and 2018, and annual deaths decreased from 596,000 to 405,000. The decline has been largely attributed to the distribution of over 2,000,000,000 pyrethroid insecticide-treated bednets (ITNs) protecting at-risk populations from bites from malaria mosquitoes that typically feed indoors and at night. However, the success of ITNs is threatened by the rapid increase in pyrethroid resistance in mosquitoes. All ITNs contain pyrethroids, and resistance is putting the entire global fight to reduce, and ultimately eliminate, malaria at risk. Historically, malaria resurges rapidly if effective interventions are not maintained at an adequate level of coverage and effectiveness. Reversing this trend is difficult and imposes major economic and public health burdens on the disease endemic countries.

LSTM has pioneered research into the causes, consequences and rapid spread of pyrethroid resistance in African malaria vectors and provided evidence-based solutions to combat the problem.

Mosquito genetics and behaviour

Understanding the genetic basis of resistance in mosquitoes has driven the development of strategies to lessen its impact. Since 2003, LSTM researchers (Donnelly, Paine, Lycett, Ranson, Weetman, Wondji) used a variety of genomic and biochemical approaches to identify the key genetic changes associated with pyrethroid resistance. We found the most potent mechanism present in African malaria vectors to be increased rates of insecticide detoxification, caused by elevated production of a small number of cytochrome P450 enzymes that are very efficient at metabolizing this insecticide class [1]. We developed *in vitro* and *in vivo* platforms for screening new insecticides for potential cross-resistance, which have been widely adopted by agrochemical companies [2]. Novel video techniques (developed by McCall) to observe and measure mosquito behaviour at bed nets demonstrated that the vast majority of activity occurs at and above the roof of the net [3], a seminal discovery that had major impacts for bednet design; for example, in trials in Liverpool and Burkina Faso, McCall has shown that small net panels, or barriers, attached outside the top of a bed net, improve the net's performance, while using less insecticide.

Impact of resistance on malaria control

LSTM has been monitoring and mapping insecticide resistance spread in Africa for over 20 years and has supported World Health Organization (WHO) in the generation, analysis and dissemination of this data via lead author contributions to the *Global Plan for Insecticide Resistance in Malaria Vectors* (2012) (Coleman, Ranson and Hemingway) and the *Global Report on Insecticide Resistance in Malaria Vectors; 2010-2016* (2018) (Coleman). We have developed new bioassay approaches for pyrethroids (and new insecticides with novel modes of action) and developed a comprehensive suite of molecular markers to track the spread of resistance through the application of genomic technologies (Ranson, Weetman, Wondji, Donnelly). These tools have enabled the impact of resistance on malaria transmission to be directly assessed [4] and are being incorporated into WHO guidelines.

Evaluation of new vector control tools to combat insecticide resistance

The Liverpool Insect Testing Establishment (LITE), established by Ranson and housed within LSTM's Vector Biology Department, has developed a screening pipeline to evaluate new insecticides and formulations against fully characterized resistant mosquito colonies [2]. All the major insecticide and ITN manufacturers have utilized LITE's services to assess performance of new insecticides and formulated products against our mosquito colonies, and many of these industry partners have also enlisted LSTM's expertise for field trials. For example, an LSTM-led consortium conducted the first clinical trial of a dual insecticide bednet (Olyset Duo®) in Burkina Faso, which demonstrated a significant public health benefit of these nets in areas of high pyrethroid resistance (decrease of 12% in malaria incidence, $p=0.04$) [5]. Hemingway and Donnelly led the evaluation of the first operational deployment of PBO-pyrethroid nets in Uganda; 12 months post deployment, PBO-pyrethroid nets reduced malaria infection prevalence by 27% ($p=0.0001$) and malaria mosquito density in houses by 87% ($p<0.0001$) relative to conventional nets [6].

3. References to the research (indicative maximum of six references)

1. Mitchell SN, Stevenson BJ, Müller P, Wilding CS, Egyir-Yawson A, Field SG, **Hemingway J**, **Paine MJ**, **Ranson H**, **Donnelly MJ**. Identification and validation of a gene causing cross-resistance between insecticide classes in *Anopheles gambiae* from Ghana. Proc Natl Acad Sci U S A. 2012. DOI: 10.1073/pnas.1203452109
2. Lees RS, Ismail HM, Logan RAE, Malone D, Davies R, Anthousi A, Adolfi A, **Lycett GJ**, **Paine MJ**. New insecticide screening platforms indicate that Mitochondrial Complex I inhibitors are susceptible to cross-resistance by mosquito P450s that metabolise pyrethroids. Sci Rep. 2020. DOI: 10.1038/s41598-020-73267-x

3. Parker JE, Angarita-Jaimes N, Abe M, Towers CE, Towers D, **McCall PJ**. Infrared video tracking of *Anopheles gambiae* at insecticide-treated bed nets reveals rapid decisive impact after brief localised net contact. Sci Rep. 2015. DOI: 10.1038/srep13392
4. Barnes KG, Weedall GD, Ndula M, Irving H, Mzihalowa T, **Hemingway J**, **Wondji CS**. Genomic Footprints of Selective Sweeps from Metabolic Resistance to Pyrethroids in African Malaria Vectors Are Driven by Scale up of Insecticide-Based Vector Control. PLoS Genet. 2017. DOI: 10.1371/journal.pgen.1006539
5. Tiono AB, Ouédraogo A, Ouattara D, Bougouma EC, Coulibaly S, Diarra A, Faragher B, Guelbeogo MW, Grisales N, Ouédraogo IN, Ouédraogo ZA, Pinder M, Sanon S, Smith T, Vanobberghen F, Sagnon N, **Ranson H**, Lindsay SW. Efficacy of Olyset Duo, a bednet containing pyriproxyfen and permethrin, versus a permethrin-only net against clinical malaria in an area with highly pyrethroid-resistant vectors in rural Burkina Faso: a cluster-randomised controlled trial. Lancet. 2018. DOI: 10.1016/S0140-6736(18)31711-2
6. Staedke SG, Gonahasa S, Dorsey G, Kamya MR, Maiteki-Sebuguzi C, Lynd A, Katureebe A, Kyohere M, Mutungi P, Kigozi SP, Opigo J, Hemingway J, Donnelly MJ. Effect of long-lasting insecticidal nets with and without piperonyl butoxide on malaria indicators in Uganda (LLINEUP): a pragmatic, cluster-randomised trial embedded in a national LLIN distribution campaign. Lancet. 2020. DOI: 10.1016/S0140-6736(20)30214-2

4. Details of the impact (indicative maximum 750 words)

The beneficiaries of two decades of research at LSTM on the biology and behaviour of malaria mosquitoes include: (1) bednet manufacturers, who have translated our research into new ‘resistance-breaking’ nets that now make up a significant share of total net sales; (2) communities in malaria-endemic regions of Africa, who are now at reduced risk of malaria; (3) global policy makers and implementers, who have a strengthened evidence base on which to select malaria-prevention tools; and (4) national malaria control programmes, who have an improved toolset to mitigate against insecticide resistance.

Impact on bednet manufacturers, agrochemical companies and the communities their products protect

LSTM’s research into the mechanisms of insecticide resistance has directly led to the development of new ITNs. Our evidence that blocking P450-based metabolism with the synergist piperonyl butoxide (PBO) can restore pyrethroid efficacy in malaria mosquitoes led to manufacturers developing a new class of nets that contain pyrethroids plus PBO, with the aim of maintaining ITN efficacy even in the face of pyrethroid resistance. Four manufacturers (Vestergaard (Switzerland), Sumitomo Chemical Co., Ltd (Japan), AKA Polymers (India) and Moon Netting (Pakistan)) now produce PBO-pyrethroid nets. These have all been pre-qualified by WHO (a necessary step before most major donors will procure a new type of net). Over 43,000,000 PBO-pyrethroid nets were distributed in Africa in 2020, protecting over 95,000,000 people. Since 2020, 27 of the 40 African national ITN distribution programmes included PBO-pyrethroid nets, with PBO-nets constituting 21% of the market share of nets delivered to Africa for malaria prevention in 2020 [1].

The discovery that malaria vector mosquitoes preferentially contact the top of a net when seeking a bloodmeal led one major net manufacturer (Vestergaard) to incorporate PBO only into the roof panel in their market leading PermaNet 3.0 ®. Since 2017, LSTM have been working with Vestergaard to develop novel bednet prototypes exploiting this aspect of mosquito behaviour. Limiting insecticides to small external net barriers improves the net’s performance, while using less insecticide thereby reducing manufacturers costs, and limiting the risk of adverse effects caused by insecticide exposure [2, 3].

Our clinical trial of the pyriproxyfen-pyrethroid net, Olyset Duo®, demonstrated the potential of insect sterilizing agents to reduce malaria transmission when combined with pyrethroid insecticides in bednets. Although Olyset Duo® is not currently being deployed, the positive results of our trial of ITNs with this mode of action led to a very similar competitor product (Royal Guard, DCT USA that was listed by WHO in 2019) being deployed in Mozambique and Nigeria since 2020 (1,000,000 nets distributed in 2020) [1, 4].

Hemingway established the Innovative Vector Control Consortium (IVCC) in 2005 at LSTM. IVCC is the leading global agency for the development of new insecticides and vector control tools. The consortium has re-formulated two classes of agricultural insecticide that have been in use for indoor residual spraying, a very effective alternative to ITNs for malaria control, since 2014. Rotation of these chemistries has enabled the implementation of resistance management strategies, and these new formulations have been deployed in indoor residual spraying in 30 malaria endemic countries, averting over 4,800,000 malaria cases between 2016 and 2019 [5]. In addition, the work of the Liverpool Insect Testing Establishment (LITE), plus the suite of tools developed by LSTM including the P450 *in vitro* screen and transgenic mosquito lines, has streamlined the insecticide development pathway, terminating investment in chemistries exhibiting potential resistance liabilities and accelerating the most promising candidates. For example, the multi-national agrochemical company Syngenta (Switzerland) used the LSTM testing pipeline as a critical differentiator to drive candidate selection and investment decisions [6].

Impact on global policy makers, implementation agencies and national malaria programmes

LSTM led the evaluation of PBO-pyrethroid classes of nets pre and post deployment. Our meta analyses of the performance of PBO-pyrethroid versus standard ITNs on mosquito populations, which Ranson presented at Evidence Review Groups on PBO-pyrethroid nets at WHO in 2015 and 2017, led to policy recommendations on use of this net class (WHO 2015, 2017) [7]. We conducted one of only two clinical trials that have demonstrated significant improved public health benefit of PBO-pyrethroid nets in reducing malaria transmission compared to standard ITNs; the scale of this trial was unprecedented and enabled 10,700,000 Ugandans to benefit from the increased protection afforded by this new net class (reduction of 27% in malaria prevalence) whilst the evaluation was ongoing [8].

LSTM vector biologists play a key role in advising WHO. Donnelly was the lead entomologist on the WHO study on the impact of insecticide resistance on malaria control (between 2012 and 2016) which helped raise the profile of the public health threat posed by pyrethroid resistance. We have been members of the WHO Vector Control Advisory Group (VCAG) since its inception in 2014 (Hemingway, Ranson) advising WHO on trial designs, and evaluating results from trials prior to policy recommendations. Wondji is a member of the WHO pre-qualification team of independent assessors who review data packages submitted by ITN manufacturers to determine whether these meet accepted WHO standards [9]

LSTM has made a major contribution to the development of WHO guidelines on resistance monitoring and management, both from the outputs of our research which has informed best practice, and via our participation in WHO committees. For example, Ranson and Donnelly contributed to the 2016 *WHO guidelines on insecticide resistance monitoring* and Coleman, Hemingway and Ranson were major contributors to the 2012 WHO *Global Report on Insecticide Resistance Management in malaria vectors* [10]. Via long-term partnerships with African Ministries of Health, we have greatly strengthened capacity in resistance monitoring and supported the interpretation of national data to inform the sub national deployment of different insecticide-based strategies (e.g. Zambia and Equatorial Guinea Insecticide Resistance Management Plans and policy briefs for Malawi [11]).

5. Sources to corroborate the impact (indicative maximum of 10 references)

Impact on bednet manufacturers, agrochemical companies and the communities their products protect

1. Database on net procurements for 2020: https://allianceformalariaprevention.com/net-mapping-project/
2. Letter from CEO of Vestergaard Frandsen confirming impact of mosquito behaviour study on net design
3. Murray GPD, Lissenden N, Jones J, Voloshin V, Toé KH, Sherrard-Smith E, Foster GM, Churcher TS, Parker JEA, Towers CE, N'Falé S, Guelbeogo WM, Ranson H, Towers D, McCall PJ. Barrier bednets target malaria vectors and expand the range of usable insecticides. Nat Microbiol. 2020. DOI: 10.1038/s41564-019-0607-2
4. https://www.ivcc.com/market-access/new-nets-project/ (Evidence Base for New Dual-AI Nets download).
5. Data available on NGENIRs website: https://www.ivcc.com/market-access/ngenirs/ (see NGENIRS Evidence, NGENIRS project overview)
6. Letter from Syngenta, confirming changes in lead candidate after evaluation using LSTM testing pipeline.

Impact on global policy makers, implementation agencies and national malaria programmes

7. Conditions for deployment of mosquito nets treated with a pyrethroid and piperonyl butoxide: **WHO reference number**: HTM/GMP/2017.17
8. Staedke SG, Gonahasa S, Dorsey G, Kamya MR, Maiteki-Sebuguzi C, Lynd A, Katureebe A, Kyohere M, Mutungi P, Kigozi SP, Opigo J, Hemingway J, Donnelly MJ. Effect of long-lasting insecticidal nets with and without piperonyl butoxide on malaria indicators in Uganda (LLINEUP): a pragmatic, cluster-randomised trial embedded in a national LLIN distribution campaign. Lancet. 2020. DOI: 10.1016/S0140-6736(20)30214-2
9. Letter from WHO Global Malaria Programme, acknowledging LSTM's contribution to WHOs work on ITNs
10. Global Malaria Programme, WHO, Test procedures for insecticide resistance monitoring in malaria vector mosquitoes 2016 (2nd edition) https://www.who.int/malaria/publications/atoz/9789241511575/en/; Global Malaria Programme, WHO, Global Report on Insecticide Resistance Management in malaria vectors 2012 https://www.who.int/malaria/publications/atoz/gpirm/en/
11. Zambia: Chanda E, Thomsen EK, Musapa M, Kamuliwo M, Brogdon WG, Norris DE, Masaninga F, Wirtz R, Sikaala CH, Muleba M, Craig A, Govere JM, Ranson H, Hemingway J, Seyoum A, Macdonald MB, Coleman M. An Operational Framework for Insecticide Resistance Management Planning. Emerg Infect Dis. 2016. DOI: 10.3201/eid2205.150984. Equatorial Guinea: Hemingway J, Vontas J, Poupardin R, Raman J, Lines J, Schwabe C, Matias A, Kleinschmidt I. Country-level operational implementation of the Global Plan for Insecticide Resistance Management. Proc Natl Acad Sci U S A. 2013. DOI: 10.1073/pnas.1307656110. Malawi: https://www.piivec.org/resources/evidence-to-inform-how-new-bed-nets-can-be-used-to-prevent-malaria-in-malawi