Institution: University of Dundee		
Unit of Assessment: UoA5 Biological Sciences		
Title of case study: Adoption of a new rapid diagnostic test for Human African Trypanosomiasis		
Period when the underpinning research was undertaken: 2008-2014		
Details of staff conducting the underpinning research from the submitting unit:		
Name(s): Prof Sir Mike Ferguson CBE	**Role(s) (e.g. job title):** Regius Professor of Life Sciences	**Period(s) employed by submitting HEI:** 1988-present
Period when the claimed impact occurred: 2013-present		
Is this case study continued from a case study submitted in 2014? N		

1. Summary of the impact

In 2012, the WHO declared its intent to eliminate Human African Trypanosomiasis (HAT) through a 'test-and-treat' strategy, requiring new screening tools that are accurate, easy to use and can be deployed at all levels of the health system. Ferguson's team identified a protein from the trypanosome surface (Invariant Surface Glycoprotein-65 or ISG65) as a superior diagnostic antigen for HAT. This directly led to the development of a new Rapid Diagnostic Test for HAT incorporating this antigen. The test has been adopted by the global not-for-profit Foundation for Innovative New Diagnostics as their preferred test for HAT screening. Further, in anticipation of post-elimination surveillance, they are using ISG65 in a combined test for HAT screening and malaria diagnosis.

2. Underpinning research

Human African Trypanosomiasis (HAT), or sleeping sickness, is caused by subspecies of the parasite *Trypanosoma brucei* transmitted by infected tsetse flies. It is generally fatal if untreated. Although the prevalence of HAT decreased dramatically over the past 20 years due to strict control measures, history has shown that it can quickly re-establish to epidemic proportions if surveillance is relaxed. It is endemic in 36 sub-Saharan African countries with 57 million people at risk. The WHO has targeted HAT for elimination of community transmission by 2030. Accurate testing to identify infected individuals is critical to enable timely treatment and for active surveillance to identify reemergence in areas of previous disease elimination. When Ferguson's team began the research, HAT screening used the relatively primitive CATT (Card Agglutination Test for Trypanosomiasis) test where trypanosomes fixed to a card are used to test patient's serum for the presence of antibodies. The CATT needs a refrigerated 'cold-chain' during distribution, and trained personnel with electrical lab equipment for patient testing. It has severe limitations for large-scale use for community screening in remote locations.

Ferguson recognised that new approaches were needed. In research published in 2013 **[R1]**, he identified improved diagnostic antigens and a more suitable platform for inexpensive field-based testing. Taking an unbiased approach, he used antibodies from the blood of HAT patients to probe a mixture of trypanosome proteins and see which proteins they bound to. This identified a trypanosome surface protein, the Invariant Surface Glycoprotein ISG65, as the best diagnostic antigen. Ferguson obtained experimental tools from Prof Mark Carrington at the University of Cambridge and subsequently produced ISG65 in an engineered bacterium (recombinant ISG65) ensuring unlimited future supply without the need to grow parasites. To progress this research towards a usable diagnostic, Ferguson and colleagues developed a prototype lateral flow Rapid Diagnostic Test (RDT) in collaboration with Dundee-based immunoassay development and manufacturing company BBI Solutions. The performance of this recombinant antigen prototype RDT **[R1]**, and a subsequent version incorporating a second native antigen also discovered in

the Ferguson lab **[R2]**, fulfilled the desired characteristics of a priority diagnostic test for FIND, the Foundation for Innovative New Diagnostics, a global not-for-profit product development partnership accelerating the development of diagnostic tests for poverty-related diseases.

Prior to Ferguson's involvement, the first-generation RTD commissioned by FIND for HAT used *native* antigens purified from live parasites. This has several disadvantages; few laboratories have the specialized facilities and expertise needed to grow the parasites and purify the antigens, and batch-to-batch quality issues can interrupt production of tests. WHO and FIND therefore required second-generation RTDs to be developed using *recombinant* antigens **[R1]**. At the suggestion of Ferguson, FIND commissioned an independent (blinded) three-way side-by-side comparison of the existing dual *native* antigen test (SD BIOLINE HAT), a dual *recombinant* prototype of the same antigens, and the Dundee dual antigen prototype. Statistical analysis of the data showed that one of the two best recombinant diagnostic antigens was Dundee's ISG65 **[R3]**. All of Dundee's knowhow and reagents were transferred free of charge to FIND who contracted Abbott (Standard Diagnostics Inc, South Korea, hereafter SD Inc) to manufacture a second-generation RDT (BIOLINE HAT 2.0) for front-line use in Africa with recombinant ISG65 as one of its two diagnostic antigens. The new kit is cheap, can be stored at room temperature, is easy to use even in remote resource-poor settings and will give rapid results in 15 minutes from a pin-prick of blood.

3. References to the research

[R1] Sullivan, L, Wall, SJ, Carrington, M & **Ferguson, MAJ (**2013) 'Proteomic selection of immunodiagnostic antigens for human African trypanosomiasis and generation of a prototype lateral flow immunodiagnostic device', *PLoS Neglected Tropical Diseases*, vol. 7, no. 2, e2087. DOI: 10.1371/journal.pntd.0002087

[R2] Sullivan, L, **Fleming, J**, **Sastry, L**, **Mehlert, A,** Wall, SJ & **Ferguson, MAJ (**2014) 'Identification of sVSG117 as an immunodiagnostic antigen and evaluation of a dual-antigen lateral flow test for the diagnosis of human african trypanosomiasis', *PLoS Neglected Tropical Diseases*, vol. 8, no. 7, pp. e2976. DOI: 10.1371/journal.pntd.0002976

[R3] Sternberg, JM, **Gierlinski, M**, Biéler, S, **Ferguson, MAJ** & Ndung'u, JM (2014) 'Evaluation of the diagnostic accuracy of prototype rapid tests for human African trypanosomiasis', *PLoS Neglected Tropical Diseases*, vol. 8, no. 12, pp. e3373. DOI: 10.1371/journal.pntd.0003373

Key research grants relevant to this case study:

1. **Ferguson, MAJ.** The biosynthesis of glycoproteins in *Trypanosoma brucei*: basic and translational research. Wellcome. (2008-2013). Value: GBP1,707,314.

2. **Ferguson, MAJ.** Protein glycosylation in trypanosomes: defining and exploiting a biological system. Wellcome (2013-2020). Value: GBP2,360,250.

4. Details of the impact

Impacts:

(a) Partnership with FIND and Abbott to accelerate HAT diagnostic test deployment
(b) Development of improved, more cost-effective diagnostic, BIOLINE HAT 2.0
(c) Successful clinical study of diagnostic BIOLINE HAT 2.0
(d) Adoption and implementation of the new, supply chain-resilient HAT diagnostic
(e) Commissioning of a combined RDT prototype for both HAT and malaria

a) Partnership with FIND and Abbott (SD Inc) to accelerate HAT test deployment

Ferguson identified FIND as the ideal partner to help translate his research into a product for clinical use. In 2014, FIND and the University of Dundee entered a formal partnership, signing a

License and Material Transfer Agreement under which reagents developed by Ferguson were made available free of charge for the development of an improved RDT for HAT **[E1]**. FIND provided the bridge to the private sector and commissioned Abbott (SD Inc) to produce BIOLINE HAT 2.0 using Dundee's recombinant ISG65. Dundee provided the ISG65 expression clone, optimised protocols, and consultancy advice to enable scale-up of rISG65 production by Abbott **[E2]**. Both FIND and Abbott benefited from the partnership with Dundee. Abbott benefit through for-profit production of the RDT, while FIND Senior Project Manager says **[E2]**:

> "*FIND is grateful for Professor Ferguson's continued engagement and assistance in technology transfer to Standard Diagnostics and Abbott… we acknowledge the University of Dundee's significant research contributions to achieving FIND's goals in supporting the elimination of HAT.*"

b) Development of improved, more cost-effective rapid diagnostic test BIOLINE HAT 2.0

By providing antigen ISG65 and prototype technology, Dundee initiated a workflow at FIND that included evaluations on affordability, performance, market analysis, and risk assessment **[E2]**. FIND Senior Project Manager describes their conclusions and confirms that Dundee research:

> "*had a significant impact, prompting FIND's development of their second-generation 'all-recombinant' RDT for HAT…The removal of reliance on the supply of native antigens, which can be laborious and dangerous to produce, provided the opportunity for a step change in the robustness of HAT screening…The all-recombinant nature of this second-generation device is critical to RDT production resilience, quality and cost control. The Bioline HAT 2.0 is affordable at US$ 0.50/test and stable for up to 2 years at temperatures up to 40°C… Packaging of HAT 2.0 will be smaller… giving distribution cost savings. The RDT format also provides several advantages over the traditional CATT test, including no requirements for electrical supply, ancillary equipment, a cold-chain or highly-trained operators. All of these factors are important for deploying HAT screening programmes in sub-Saharan Africa"* **[E2]**.

c) Successful clinical study of rapid diagnostic test BIOLINE HAT 2.0

In 2015, FIND began a multi-centre clinical study in the Democratic Republic of the Congo to compare the performances, under field conditions, of the all-recombinant BIOLINE HAT 2.0, the native antigen SD BIOLINE HAT, and the traditional 'standard of care' CATT test **[E3]**. A total of 57,632 people were screened, and 260 HAT cases were confirmed by microscopy with benefit to the individuals involved who received treatment. The outcome was that the new BIOLINE HAT 2.0 device was more sensitive than SD BIOLINE HAT and CATT (71.2% versus 59.0% and 62.5%, respectively) while all three tests had a specificity above 98% **[E3]**.

d) Adoption and implementation of the new, supply chain-resilient HAT diagnostic

As a result of the definitive outcome of the clinical study, FIND officially adopted BIOLINE HAT 2.0 (Figure 1) as their preferred test for HAT screening and surveillance and launched the commercial product with Alere in September 2017 (Alere subsequently acquired by Abbott), acknowledging the contribution of the University of Dundee **[E4]**. The General Manager of SD Inc. said at the time:

> "*This test is the fruit of several years of joint efforts, and a major achievement resulting from the successful partnership that has been established with FIND to support the development of rapid tests for diseases associated with poverty*" **[E4]**.

The Chief Executive Officer of FIND said the test:

> "*will facilitate diagnosis of sleeping sickness patients in even the most remote corners of affected countries and enhance the prospects of achieving and sustaining elimination of the disease*" **[E4]**.

Figure 1. BIOLINE™ 2.0 Rapid diagnostic test (C = control; 2,1 = test antigens).

Full commercial roll out was anticipated to complete in spring 2020 but has been delayed due to FIND's need to develop and market RDTs for COVID-19. The test is now planned to complete roll out in February 2021 **[E2]**.

e) Commissioning of a combined RDT prototype for both HAT and malaria

Patients with malaria or the early stage of HAT have similar symptoms. A diagnostic test that differentiates between them could inform treatment and control transmission. The success of BIOLINE HAT 2.0 led FIND to commission Abbott to develop a combined RDT incorporating Ferguson's rISG65 recombinant antigen and an antigen from the malaria parasite. The new combined RDT is as accurate as the individual HAT and malaria RDTs under laboratory conditions **[E5]**. FIND has sponsored a new clinical observational study to evaluate the dual RTD in the field to screen for HAT and diagnose malaria **[E6]**.

FIND has declared the transformational impact of the availability of BIOLINE HAT 2.0 and the development of the HAT/malaria combined RDT in their diagnostic strategy pipeline for HAT **[E7]**. The tests are FIND's two prioritized activities for HAT in "*addressing the most critical gaps in the diagnostic landscape*" for screening interventions (the other two tests are for case confirmation) **[E7]**. FIND's HAT screening programme was supported by investment from the Bill & Melinda Gates Foundation, UK aid from the UK Government, and the Swiss Government **[E4]**.

5. Sources to corroborate the impact

[E1] University of Dundee press release confirming signing of the Material Transfer Agreement with FIND whereby Dundee would provide their reagents to FIND for the development on an improved diagnostic for HAT, 22nd April 2014.

[E2] Corroboratory testimonial from Senior Project Manager of the Neglected Tropical Diseases Programme at FIND.

[E3] Clinical study reporting on evaluation of the diagnostic accuracy of BIOLINE HAT 2.0 (previously SD BIOLINE HAT 2.0) acknowledging role of Ferguson and citing publications R1 and R3. Lumbala, C, Biéler, S, Kayembe, S, Makabuza, J, Ongarello, S, Ndung'u, JM (2018) 'Prospective evaluation of a rapid diagnostic test for *Trypanosoma brucei gambiense* infection developed using recombinant antigens'. *PLoS Neglected Tropical Diseases,* vol.12, no. 3, e0006386. DOI: 10.1371/journal.pntd.0006386

[E4] Press release from FIND announcing launch of second-generation rapid diagnostic test for sleeping sickness, 12th September 2017.

[E5] Report on the accuracy of the BIOLINE HAT/Malaria Combined prototype RDT. The Combo RDT uses Dundee rISG65. (SD BIOLINE HAT 2.0 used as a comparison here subsequently renamed BIOLINE HAT 2.0 by Abbott). Lumbala, C, Matovu, E, Sendagire, H, Kazibwe, AJN, Likwela, JL, Muhindo Mavoko, H, Kayembe, S, Lutumba, P, Biéler, S, Van Geertruyden, JP & Ndung'u JM. (2020) 'Performance evaluation of a prototype rapid diagnostic test for combined detection of gambiense human African trypanosomiasis and malaria'. PLoS Neglected Tropical Diseases. 2020, vol.14, no.4, e0008168. DOI: 10.1371/journal.pntd.0008168

[E6] Clinical Study sponsored by FIND employing the BIOLINE HAT 2.0 (previously SD BIOLINE HAT 2.0) and BIOLINE HAT/Malaria Combo RTDs that use the Dundee rISG65. ClinicalTrials.gov (NCT003394976) 9th January 2018.

[E7] 2015-2020 'Strategy for Neglected Tropical Diseases' FIND. pp 12-13.