

Malaria: Marking Milestones

It is our hope and expectation that researchers around the globe will use the information and biological insights provided by complete genome sequences to accelerate the search for solutions to diseases affecting the most vulnerable of the world's population.

M. J. Gardner *et al.*, *Nature*, 2002, **419**, 498

For his work on malaria by which he has shown how it enters the organism and thereby has laid the foundation for successful research on the disease and the methods of combating it.

Citation, 1902, Nobel Prize for Physiology or Medicine awarded to Ronald Ross

A 105 years have passed since Ronald Ross published his famous paper 'On some peculiar pigmented cells found in two mosquitoes fed on malarial blood' (*Br. Med. J.*, 1897, **2**, 1786). Ross made his epoch-making observations in Secunderabad on 20 August 1897. The human malarial parasites *Plasmodium vivax* and *Plasmodium malariae* had been identified in Rome by Camillo Golgi in 1886. In 1890 Italian physicians described *P. falciparum*, the most lethal of the parasites causing human malaria. Ross conclusively established that the mode of transmission of the disease required the intermediacy of the *Anopheles* mosquito. He decisively identified the presence of the parasite in the guts of mosquitoes fed on malarial blood of a patient, Husein Khan, who was paid one anna for each bite. As late as 14 August 1897, Ross had reported little success, confessing in a letter to his wife: '... perhaps (I) am not using the proper kind of mosquito'. In an historical analysis, K. Rajakumar and M. Weisse note: 'He hired three native men to help find mosquitoes, adults and larvae. As if the Lord were answering his plea, an 'angel of fate' put into the hands of one of his 'mosquito men' some larvae that hatched out a different dappled-winged mosquito, with which he began experimenting, hoping for success (*Southern Medical Journal*, 1999, **92**, 567). The 'hatched-dappled' mosquitoes, *Anopheles*, were the vectors which harboured, nurtured and transmitted the malarial parasite. Ross moved to Calcutta in 1898 and established the cycle of avian malaria. The first major step in understanding a disease that has been with humans for thousands of years

had been completed. Ross' discovery, which has been repeatedly chronicled, provides a fascinating example of the early successes of tropical disease research, in a historical setting, where the imperatives of empire building and colonial expansion provided a compelling rationale for the studies on malaria. In introducing Ronald Ross at the Nobel ceremonies in December 1902, the Rector of the Royal Caroline Institute, K. A. H. Morner noted that, of the 178,000 men of the British Army in India, 76,000 suffered from malaria in 1897, the year of Ross' success. The estimated mortality in the civilian population was, of course, much higher. There was the inevitable element of serendipity in Ross' encounter with the *Anopheles* mosquito; his 'native' assistants might have been acute observers, a thought that has been played out as fantasy in Amitav Ghosh's novel, *The Calcutta Chromosome* (Ravi Dayal Publisher, Delhi, 1996).

Exactly a century after the award of the Nobel Prize for Ross' work establishing the integral connectivity between the mosquito, the parasite and the human, come the announcements of the completion of the sequencing of the genomes of the parasite, *P. falciparum*, and the mosquito, *Anopheles gambiae*. The parasite genome and related studies appear in the October 3 issue of *Nature*, while the work on the mosquito is described in the October 4 issue of *Science*. As in the case of the report on the human genome, the conquest of the parasite and vector genomes has been a media event, with both journals orchestrating a publicity blitz, which once again focuses public attention on malaria. Ross would be surprised by the fact that over a century after his work the disease still flourishes. As the authors of the *P. falciparum* genome sequence paper emphasize: 'Approximately 40% of the world's population lives in areas where malaria is transmitted. There are an estimated 300–500 million cases and up to 2.7 million deaths from malaria each year. The mortality levels are greatest in sub-Saharan Africa, where children under 5 years of age account for 90% of all deaths due to malaria' (M. Gardner *et al.*, *Nature*, 2002, **419**, 498). In the long battle against malaria the first signs of a possible treatment appeared in the 17th century, when Jesuit priests brought back to Europe the knowledge that the bark of the cinchona tree appeared to alleviate the symptoms of the disease. The alkaloid quinine, was the first widely used antimalarial. Chloroquine,

in many respects a wonder drug, appeared much later, but in 20 years after its introduction, resistant strains of the parasite began to emerge. Intriguingly, chloroquine's successors sulfadoxine-pyrimethamine were universally effective in the field for a shorter period; the parasite acquired resistance in less than 5 years. Coincidentally, the issue of *Science* that reports the genome sequence of the *Anopheles* mosquito also carries the results of an important study that probes the dynamics of drug resistance in the parasite (A. B. S. Sidhu *et al.*, *Science*, 2002, **298**, 210). In a *Perspectives* essay entitled 'A Requiem for Chloroquine' I. M. Hastings *et al.* note: 'Now it appears that the application of modern genetic technology may enable chloroquine to leave one more valuable legacy – a detailed genetic, clinical and epidemiological epitaph that can be used to inform the deployment of its successors' (*Science*, 2002, **298**, 75). There are, in fact, few new drugs on the horizon, which will effectively target all strains of the parasite, including those that have learned to withstand the onslaught of chloroquine and its successors. Ross, however, firmly held the view that the key to malaria control and eradication lay in the ability to destroy the vector, the *Anopheles* mosquito. For a while it appeared that the insecticide DDT would win the war against the mosquito. But, DDT-resistant strains surfaced quite soon. Clearly, both the parasite and its mosquito vector were more than a match for the weapons produced by science.

In this background we might well ask: 'What do the genome sequences of *P. falciparum* and *A. gambiae* offer in the fight against malaria?' The answer seems to come from the sequencers themselves: 'In the short term, however, the genome sequences alone provide little relief to those suffering from malaria. The work reported here and elsewhere needs to be accompanied by larger efforts to develop new methods of control, including new drugs and vaccines, improved diagnostics and effective vector control techniques. Much remains to be done' (M. J. Gardner *et al.*, *Nature*, 2002, **419**, 498). Despite this sombre assessment, the completion of the genome sequences of the parasite and its mosquito vector marks a milestone in the long struggle against malaria. The genome sequences are a remarkable resource for biologists attempting to unravel the details of parasite biology and biochemistry, which may be essential for developing rational methods of control. In war, it is essential to understand the enemy; *Plasmodium* has proved a sophisticated and devious foe. The life cycle of the parasite requires two distinct hosts, the mosquito and the human. Even in the human, the parasite resides at different stages in two distinct cells, the hepatocytes in the liver and the erythrocytes in the blood. The parasite reveals itself only fleetingly in the bloodstream, possessing an uncanny ability to vary its exterior; a skill invaluable in evading immune surveillance. *Plasmodium* also prefers to multiply in the confines of the erythrocyte, largely hidden from a potentially hostile environment. It is this remarkable biological sophistication that has made the development of diagnostics and vaccines so difficult. The para-

site's ability to learn to defuse drugs directed against its metabolic machinery makes it a truly formidable enemy. The genome sequence of *P. falciparum*, translates into about 5300 predicted proteins. At present about 60% of these do not bear 'sufficient similarity to proteins in other organisms to justify provision of functional assignments'. Put simply, our ignorance of the details of parasite biochemistry is substantial. But gene sequences are only the tip of the iceberg. It is essential to identify the protein products of expressed genes, at every stage of parasite development. Here the emerging tools of proteomics, high resolution protein separation coupled with mass spectrometric analysis of sequences, hold the key. The first major steps in analysing the *Plasmodium* proteome, which may eventually provide valuable clues on antigen variation, have just been reported (L. Florens *et al.*, *Nature*, 2002, **419**, 502; E. Lasonder *et al.*, *ibid*, 2002, **419**, 537).

The publicity over the announcement of the genome sequences of the parasite and the mosquito has turned the spotlight, albeit briefly, on malaria. Research on the disease is, however, poorly funded with an annual worldwide investment of \$200 million per year; an amount unlikely to permit major advances based on the bridgehead of the genome sequences. Support for malaria research has largely come from charities like the Wellcome Trust and, more recently, the Gates Foundation. The disease is not a subject of major interest for the giant pharmaceutical companies, who prefer to invest their research resources on more lucrative areas. The estimated market for a malaria vaccine is small, less than half a billion dollars annually. This figure may be unattractive for companies which are loath to enter a race for products that have a potential market of less than a billion dollars, annually. Malaria, is truly an 'orphan disease' with potential drugs and vaccines aimed at the poorest populations in the world. With the departure of colonizing armies, the incentives for major investments by the developed countries on the fight against malaria are not readily apparent. Ronald Ross was moved to poetry when he saw the malarial parasite in the stomach wall of the mosquito. His verse has been widely quoted.

*This day relenting God
Hath placed within my hand
A wondrous thing; and God
Be praised. At his command
Seeking his secret deeds
With tears and toiling breath
I find thy cunning seeds
O million-murdering Death.
I know this little thing
A myriad men will save.
O death, where is thy sting
Thy victory, O Grave?*

After the blaze of publicity over the genomes dies down, present-day malaria researchers may be more circumspect.

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