This section looks back to some ground-breaking contributions to public health, reproducing them in their original form and adding a commentary on their significance from a modern-day perspective. To complement this month's theme of the *Bulletin*, Robert S. Desowitz reviews the 1948 paper by H.E. Shortt & P.C.C. Garnham on the discovery of the primary tissue phase, the exoerythrocytic cycle, of malaria parasites. The original paper is reproduced from the *British Medical Journal*.

The fate of sporozoites

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In their tribute to Colonel Shortt on his 80th birthday, the renowned malariologists P.G. Shute and Sir Gordon Covell eloquently placed the discovery of the exoerythrocytic (EE) hepatic phase of mammalian malaria parasites in the following historical context (*1*). "Just as the name of Ross will forever be associated with the discovery that mosquitoes transmit malaria, so too, will the names of Shortt and Garnham will be remembered in connection with the primary tissue phase of the parasite." It has been 52 years since Shortt & Garnham published their milestone finding (*2*) of the cyst-like body, filled with thousands of merozoites, in the liver of a rhesus monkey that had been inoculated 102 days before with *Plasmodium cynomolgi* sporozoites from 500 mosquitoes. With their bold experiment they solved a centuries-old mystery — the source of malaria's parasitaemic relapses.

An outstanding parasitological *gaffe* had been the claim by Fritz Schaudinn (best remembered as the co-discoverer of the Treponema causation of syphilis) that the *Plasmodium vivax* sporozoite penetrated the erythrocyte (*3*). Schaudinn's accompanying drawings showed "the theatrical picture of the entry of a malaria sporozoite into a red blood cell", as Knowles commented (*4*). We may now wonder how such an erroneous observation could have been made by so distinguished and expert a protozoologist. We may also wonder at the pervasiveness of Schaudinn's authority, so powerful that it overrode all the failures to substantiate his findings.

According to Shute & Covell (*1*) the first doubts of Schaudinn's theory came from the malariatherapy centres treating paretics. In practices that today would bring down the wrath of hospital patient oversight committees (and a phalanx of lawyers bearing malpractice briefs), malaria, mostly *P. vivax,* was induced either by direct inoculation of infected blood (continental European style) or by inoculating sporozoites by mosquito bites or in isolated salivary glands and ground-up thoraces (British style). The blood-inoculated patients were readily, radically, cured with quinine but the sporozoite-induced infections relapsed after the same therapy. The proof, albeit still circumstantial, that Schaudinn was totally wrong, that there was a missing link in the life cycle of human malaria, came from the remarkable experiment of Sir Neil Hamilton Fairley in Australia (*5*). Fairley showed that the blood of volunteers injected with large numbers of *P. vivax* sporozoites was infectious to other volunteers for only 30 minutes. The blood then became "sterile" until 7 days later when it once again became infectious to volunteers.

Although *P. knowlesi* had been known as a primate malaria since 1932 (*6*), during the first half of the 20th century — until the discovery of *P. berghei* (*7*) in a wild tree rat of the Congo — the avian malarias served as the main experimental models. Distinguished researchers of that period were bird malaria experts, e.g., Huff in the United States, Brumpt in France, Raffaele in Italy, and James in Britain. Bird malarias also relapsed after quinine treatment. Tissue and organ smears from infected birds revealed exoerythrocytic schizonts in reticulo-endothelial and hemoblastic cells (*8–10*). Prediction held that the exoerythrocytic stage of human and primate malarias would also be located in these tissues and the malaria birdmen were chagrined when the site turned out to be the hepatocyte.

Shortt was a traditonalist who held to the importance of lineage in science. He would peer over his half-glasses to issue a stern rebuke to a former student (who might by then be a full professor) for a serious "transgression", such as straying into helminthology: "Remember who your teachers are!" he would say. That line of teachers, in his view, went from the student to Shortt to Rickard Christophers to Ross. In a sense, all present malaria researchers are

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students in the lineage of Shortt (and, of course, Garnham) and it is fitting in this new millennium to be mindful of the far-reaching impact of the discovery of the EE bodies.

Consideration of virtually any aspect of malaria must now make some link with the search for an effective vaccine. Almost sixty years ago the pioneering experiments of Russell, Mulligan & Mohan (*11*) established the principle that each stage of the malaria parasite's life cycle has its own unique antigenic signature. In now devising a stage-specific vaccine, the pre-erythrocytic schizonts have become an important target. An effective vaccine would prevent sporozoites from invading the hepatocyte and/or prevent maturation of exoerythrocytic schizonts to merozoites. Sporozoite ligands binding to hepatocyte receptors and liver stage-specific antigens have now been isolated. As adjuvanted vaccines they induce a cytotoxic T lymphocyte ($CD8^+$)-mediated immunity which will kill the infected hepatocyte. It is also now proposed to use them as DNA vaccines either alone or in a multi-stage formulation (*12–16*).

We do not conventionally associate the awakening knowledge of the EE cycle with the massive, WHO-orchestrated Global Eradication of Malaria campaign (1955–72), but I believe that there was a crucial connection. The programme was proposed as a relatively short campaign after which there would be no continuous "maintenance": with the cessation of transmission and the absence of reinfection, the malarias in a population (with the probable exception of *P. malariae*) would go to self-cure even without chemotherapeutic intervention. Since the discovery of the EE phase and its patterns in the human malarias, the source of the relapses was now known and the strategy, especially in respect of *P. falciparum*, could be applied with confidence. Thus Shortt & Garnham laid the logical foundation for the campaign.

The arguments are now fading with the years, as the graduate students and associates of these two remarkable men also now fade into history. A contention between them has been, "Did Garnham's earlier finding of the liver stage of *Hepatocystis kochi* (*17*) point Shortt in the right direction?" My belief, undoubtedly influenced by being his last graduate student, is that Shortt, who directed the experiment, had no preconceived idea where the hidden parasites might be. The monkey was, literally, taken apart, tissue samples sectioned with the Department's Spencer microtome, stained by the gorgeous Giemsa-colophonium method and scanned under Shortt's new, prized Leitz binocular microscope which eventually revealed the "Eureka!" liver specimen.

My final words are of praise for these two famous men, each a physician-naturalist, each a devoted protozoologist. Shortt, a product of the Indian Medical Service, the relentless pursuer of all quarry — single-celled parasites, tigers, trout and houseflies — was a retiring man and yet generous and caring to his associates and students. Garnham, who came from the East African Medical Service, was a very different man: an aesthete who, as I recall, skied and played the cello. He was as comfortable in discussing the theatre, opera and literature as he was in eruditely explaining the lives and times of the Haemosporidia. And although we here pay tribute to their discovery of the EE cycle we should also be mindful of their many other seminal contributions to medical protozoology. ■

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