J1550-564 is different in several intriguing ways. First, the eastern component, although fading, remains detectable more than 3 years after the initial explosion. The radio emission is synchrotron, produced by relativistic particles spiraling around strong magnetic fields, which together exert an enormous pressure. The mere fact that we still see this emission after such a long time requires some way of confining those particles and fields.

Second, this same eastern component has decelerated considerably, with the velocity declining by at least a factor of 2 between 2000 and 2002. The most obvious explanation is that it is slowing down as it plows through the circumstellar or interstellar medium. The resulting ram pressure may in fact be holding the relativistic particles together. But ram pressure at relativistic speeds is exceptionally strong. The gradual deceleration of this component, without substantial brightening, implies either that it is much heavier than minimum energy arguments would suggest, or that the surrounding medium is exceedingly tenuous.

The western component is very different. At first, it simply faded below detectability in the usual fashion; but it has recently reappeared as a strong radio and x-ray source about 2 light years from the black hole. This source is aligned with the original radio jets, pointing back toward the parent system. It looks very much like the hot spots and lobes of giant radio galaxies.

These observations suggest that we are at last seeing direct evidence of a transient microquasar jet crashing into the surrounding material. If we can measure the density of that material through optical lines or other tracers, then the evolution of this source may allow us to measure the basic physical properties of a jet, such as its total mass and momentum.

Both jet components have been detected with radio and x-ray telescopes. Similar radio emissions have been seen many times in the past, but this is the first time that x-ray emission has been seen in a microquasar so far from the central object. If this emission is synchrotron, like the radio emission, then the required particle energies are enormous, in the tera–electron volt range. Alternatively, the x-rays may result from inverse Compton emission, in which photons from some background field (such as the cosmic microwave background) gain energy from collisions with relativistic particles. Both processes have been observed in extragalactic jets (*7*). It is not clear which dominates here, because the current optical/infrared data are not sensitive enough to determine whether a single synchrotron power law connects the radio emission to the x-ray emission.

These observations leave a number of puzzles. Why do the jets become visible again a couple of light years from the parent system? Does this observation imply an evacuated cavity, and if so, how did it form? Perhaps the supernova explosion that made the black hole also carved out a vacuum for the jets, or the jets themselves inflated a bubble. More generally, why do we see so few decelerating jets and so few terminal shocks? And given that at least a few microquasars do dump their energy at large distances from the binary system, how effective are these jets in stirring up the interstellar medium?

We have known about the jets and lobes of radio galaxies for decades, but those are very long-lived, and we effectively see only a single snapshot of their structure. By contrast, microquasars are ephemeral: The initial explosion that gives rise to the jets may last a few days, and the jets themselves quickly escape their parent system. The observations of Corbel *et al.* (*5*) illustrate the final stages of this rapid evolution, as the jets crash into the interstellar medium, expiring in a blaze of glory only a few years after their birth.

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4. The most famous is SS433 (*9*), where the jets clearly interact with the remnant W50 some ~80 light years away. But whereas the jets of most microquasars are present only occasionally and for a brief time (days to weeks), those in SS433 seem to have been "on" almost continuously for thousands of years. The question is what happens to the energy released in the more common, transient events.
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PERSPECTIVES: PARASITOLOGY

A Requiem for Chloroquine

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Chloroquine (CQ) has historically been the mainstay of malaria treatment, particularly in the worst affected regions of sub-Saharan Africa. The recent development of widespread CQ resistance in *Plasmodium falciparum*, the most dangerous of the four malaria parasite species, has contributed significantly to escalating mortality rates in Africa (*1*) and to the resurgence of malaria as an immediate public health priority (*2*). Several pressing scientific questions have emerged within the context of this humanitarian disaster: What is the molecular basis for CQ resistance, and how has this influenced the dynamics of resistance? Why did CQ remain effective for 20 years, yet its immediate replacement sulfadoxine-pyrimethamine (SP) last less than 5 years? Has the widespread deployment of CQ jeopardized the use of other drugs targeting the same parasite biochemical pathways? As reported on page 210 of this issue, Sidhu *et al.* (*3*) have obtained data relevant to all three questions by creatively exploiting the *pfcrt* gene, which encodes a putative transporter protein in the digestive vacuole membrane of the malaria parasite. They replaced the endogenous *pfcrt* gene in a CQ-sensitive strain of *P. falciparum* with a *pfcrt* gene from each of three CQ-resistant strains. All such replacement strains ("constructs") showed CQ resistance in vitro, demonstrating that *pfcrt* mutations are sufficient, within their selected genetic background, to encode resistance. Reduced levels of *pfcrt* gene expression in the constructs also showed that up-regulation of *pfcrt* is not required for resistance. Next, the authors investigated cross-resistance between CQ and other antimalarial drugs.

Previous work from this and other groups has implicated eight or nine different *pfcrt* mutations in the development of CQ resistance (*4*). The sequential accumulation of these mutations plausibly explains the observed genetics and epidemiology of CQ resistance (see the figure). So why did CQ last so much longer than SP as a frontline antimalarial? First, four sequential mutations in the *dhfr* gene—which encodes dihydrofolate reductase, an enzyme essential for parasite folate metabolism and targeted by the drug pyrimethamine—appear sufficient for SP resistance (*5*). These four mutations accumulate much faster than the nine required for CQ resistance. Second, CQ persists at therapeutically useful concentrations for a much shorter period than SP, leading to lower selection pressures for resistance (*6*). Third, CQ resistance may involve genes other than *pfcrt*, such that sexual recombination during the malaria life cycle breaks down genetic combinations, slowing resistance (*7, 8*). The putative involvement of other genes remains controversial. Sidhu *et al.* show that *pfcrt* alone

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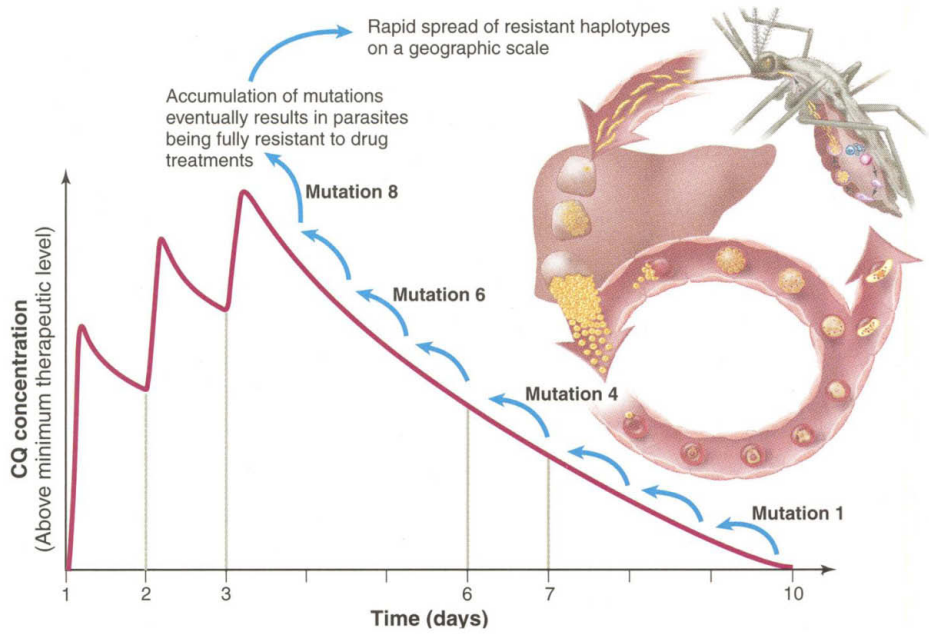
is sufficient to encode resistance, but this only holds for the genetic background of their selected host strain (50% of which was derived from a CQ-resistant strain). Field studies are similarly inconclusive: *pfcrt* Lys[76] → Thr (K76T) mutations appear to be a prerequisite for parasites to survive drug treatment, but many *pfcrt* K76T parasite infections disappear after drug treatment. The interpretation of this simple observation remains unclear: It may implicate other genes, or it may simply represent human immunity eradicating truly drug-resistant infections. The Sidhu *et al.* paper contains the technology required to perform the definitive test of introducing an identical *pfcrt* CQ-resistant construct into a series of different genetic backgrounds; such results are eagerly anticipated.

The realization that the long therapeutic life-span of CQ was likely to be the exception rather than the rule led the World Health Organization to recommend that all new antimalarial treatments be deployed as drug combinations. The efficacy of drug combination strategies has been demonstrated mathematically and by the success of combination therapy for HIV and tuberculosis. A fundamental requirement of combination therapy is that the genetic basis of resistance differs for each drug because even a small amount of cross-resistance dramatically increases the rate at which resistance evolves (*9*), severely limiting the likely therapeutic life-span of the drug combinations. In practice, the dearth of new antimalarial drugs means that existing drugs will be redeployed in combination with the relatively new antimalarial artesunate, a derivative of artemisinin. This has raised fears that cross-reactivity may have evolved because quinolines (such as CQ) and artemisinins both kill the malaria parasite by interfering with its heme detoxification pathway. Reassuringly, Sidhu *et al.* demonstrate that as resistance to CQ evolves, the parasites become more susceptible to artemisinins.

Resistance to CQ probably arises through the sequential accumulation of mutations (see the figure). The first mutations spread because they confer increased tolerance to CQ on parasites, enabling them to infect humans sooner after drug treatment—for example, mutation 4 allows parasites to infect people 6 days after treatment rather than 7 days. The relatively rapid elimination of CQ means that these are rather weak selective forces (*6*) and that the spread of these first mutations will be slow. Eventually, mutation 8 arises, which allows the parasite to survive therapeutic levels of CQ. Once above this threshold, the selective advantage conferred by this mutation becomes enormous and the *pfcrt* haplotype (now containing several sequentially acquired mutations) spreads rapidly across geographic regions where CQ is in common use. This appears to have occurred four times for CQ resistance: twice in South America, once in southeast Asia, and once in Papua New Guinea (see the viewpoint by Wellems on page 124) (*10*). The mutations may not have equal effects: mutations K76T and Ala[220] → Ser (A220S) appear to be the most reliable markers predicting CQ resistance. There are three plausible explanations for this: (i) If the mutations can be acquired in any sequence and K76T and A220S have large effects, then they will have a stronger correlation with resistance; the problem with this argument is that they rarely, if ever, occur alone and invariably occur with other "lesser" *pfcrt* mutations. (ii) Mutation acquisition may follow a set sequence with K76T and A220S near its end. (iii) These are the pharmacologically important mutations. The other mutations are optional—they may have a small effect on CQ tolerance, or compensate for impaired protein activity after the acquisition of the K76T or A220S mutations, or encode resistance during the transmission stages of the malaria life cycle.



Evolutionary dynamics of CQ resistance. The solid line shows the serum concentrations of CQ after treatment of a typical human host. Three consecutive daily doses are given, after which the CQ concentration declines to subtherapeutic levels around day 10 (*11*). Resistance to CQ probably arises through the sequential accumulation of mutations (numbers 1 to 7) encoding gradually increasing drug tolerance. Critically, the malaria parasites remain susceptible to therapy because drug levels after treatment exceed their tolerance. Eventually, a mutation occurs (number 8) that enables the parasites to survive post-therapy drug levels.

In retrospect, CQ was a wonder drug. Cheap, safe, and effective against one of the major human killer diseases, it remained effective for 20 years in Africa amid the chaotic clinical setting of underdosing, noncompliance, and its indiscriminate use in treating all fevers. Hundreds of millions of treatment courses were deployed annually in Africa alone, and in many areas most of the population have detectable circulating CQ. It is hard to envisage any other drug lasting so long under these circumstances, especially since resistance finally arrived from southeast Asia rather than arising in Africa itself. Now it appears that the application of modern genetic technology may enable CQ to leave one more valuable legacy: a detailed genetic, clinical, and epidemiological epitaph that can be used to inform the deployment of its successors.

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11. A subtherapeutic CQ level is defined as a concentration permissive for a fully sensitive parasite, emerging from the liver stage, to establish a viable infection. CQ has a long terminal elimination half-life, enabling it to be detected weeks after ingestion, but its persistence at therapeutic levels is short because the circulating free CQ concentrations achieved post-distribution (after clinical dosing) rapidly fall below that required to kill or exert selective pressure against the parasite. Therapeutic concentrations must be maintained for 6 or 7 days to consistently achieve parasite clearance, but it seems unlikely that CQ therapeutic levels are maintained much beyond this point. The authors used 10 days based on an informed guess, although the qualitative argument remains unchanged even if this period is extended up to 20 days.