PARASITOLOGY

Malaria Strains Appear to Gang Up Against Immune Defenses

The battle between parasites and their host organism's defenses has been likened to a molecular arms race: The evolution of one drives the evolution of the other. Although theoretical studies have supported this view of the parasite-host conflict, direct evidence has been hard to come by, because the interactions are complex and field data are sparse. But new research reported on page 1173 provides evidence of just such a battle being waged in West Africa between the malaria parasite and its human hosts. The work suggests that two strains of the parasites have evolved a surprising tactic to defeat immune defenses: cooperation. "This novel synergism between the two strains is a very exciting result and may shape future vaccine development," says Bryan Grenfell, an epidemiologist at Cambridge University.

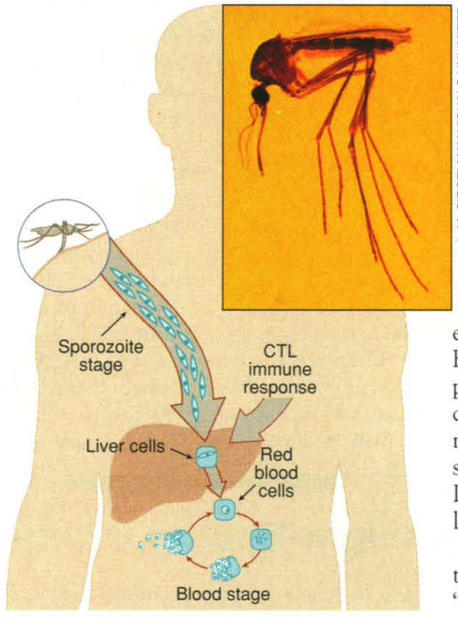
The new work, led by Adrian Hill at Oxford University—with colleagues at Oxford, the U.K. Medical Research Council Laboratories in Fajara, The Gambia, and the London School of Hygiene and Tropical Medicine—studied the dynamics of malaria infections in people living in The Gambia. The parasite that causes the disease, *Plasmodium falciparum*, is endemic in that country, where 20% of the people carry it in their blood. Malaria provides a rare opportunity to study the molecular arms race between parasite and host because researchers can link specific immunological reactions to specific variants of the parasite. This precision is hard to achieve for most parasitic infections, which involve a huge array of immune responses as the parasites progress through often-complex life cycles.

Hill's team focused on the initial immune-system reactions that begin shortly after a mosquito injects the parasites into the bloodstream. The parasites migrate to the host's liver cells, where they appear to trigger a highly specific immune response. Molecules of the host defense system called human leukocyte antigens (HLAs) bind to small fragments of proteins from the parasite; specific HLAs bind specific fragments. This HLA binding then initiates an attack by a population of immune cells called cytotoxic T lymphocytes (CTLs), which latch onto HLA-bound fragments and are then activated to kill the parasites. Some parasites slip through these defenses, however. By studying surviving strains, Hill and his colleagues could infer molecular details of the initial CTL response.

Earlier research had shown that an HLA molecule called HLA-B35 is common in the Gambian population. HLA-B35 binds to a specific fragment from *P. falciparum*'s so-called circumsporozoite protein, provoking CTLs into attacking the parasite. Researchers have found four variants of the parasite in The Gambia that differ in this region of the circumsporozoite protein. Only two of them, called cp26 and cp29, bind with HLA-B35 and hence provoke CTL attack by this route.

In lab studies, Hill's team found, as expected, that CTLs from malaria patients and from people who had not been exposed to malaria could kill cells displaying fragments of these two variant proteins. But they also discovered that if protein fragments from one of these variants were present in the culture, the CTLs were unable to kill the parasites bearing the other variant. The mechanism is a mystery, but Grenfell likens it to a lock and key: "If a key is completely different from the one that opens the lock, then it will have no effect on the lock, but if the key is just slightly different, it can jam the lock," he says. One parasite's proteins appear to jam the lock that unleashes CTLs to destroy the other variant parasite.

This cooperative strategy is highly effective: It works at very low doses, and each variant's protein fragments appear to be equally effective in protecting the other. But the key question is whether this mechanism works to the parasites' advantage in real patients. "There have been other reports of antagonism in lab studies with HIV and hepatitis B virus, but it's not clear what is going on in patients," says Hill. "There's a lack of field data showing that antagonism works within people."



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Teamwork. Malaria strains seem to work together to defeat immune attack in the liver.

To answer that question, the team studied malaria parasite DNA recovered from 800 infected patients. They analyzed which strains were present in patients' blood and assumed that the variants they found were those that had survived the initial attack in the liver. More than 40% of the patients had been infected with more than one strain of the parasite. When Hill and his colleagues compared the distribution of different strains with what would be expected if there were no links, they found the results were highly skewed. "We found a much higher co-occurrence of the two strains containing cp26 and cp29," says Hill. The cooperative approach did indeed appear to help the two strains survive. "It was a striking result," he adds.

The team also compared patients who had the HLA-B35 molecule with those who did not. They found that the cp26 and cp29 variants were more common in the blood of the HLA-B35 group. "These results provide evidence that HLA molecules influence the strains of parasites causing malaria infections," says Hill. It shows the specific interactions between host and parasite molecules you would need for co-evolution, he says. It also shows that particular HLA molecules play a key role in particular parasite strategies, and any change to them could shift the ground in favor of the host. "It's meat on the bones of the [coevolution] idea," says immunologist Jonathan Howard of the Institute of Genetics at the University of Cologne in Germany.

Some researchers say this sort of cooperation may not be limited to malaria parasites. "These are very interesting results, which suggest that the interdependency of the two variants shows that they are gaining a mutual advantage. I would expect to find this phenomenon in other situations," says Howard. "Antagonism has been shown for HIV proteins in the lab, and you'd expect it might also occur in the body."

The results also have sobering implications for vaccine development. "On the face of it, it's bad news," says Hill. A vaccine containing a protein fragment from one strain of malaria parasite could suppress the immune response to a related strain. "Some malaria vaccine programs have been looking at inducing responses to the liver stage of the parasite, but these studies suggest that use of some variant proteins from this stage may be counterproductive," he says. "The work shows just how difficult it may be to manipulate immune responses to our advantage," says Howard.

–Nigel Williams