Motherhood and malaria

Multigravid women, who are protected from malaria during pregnancy, possess antibodies that block the binding of infected erythrocytes to chondroitin sulfate A, a placental receptor.

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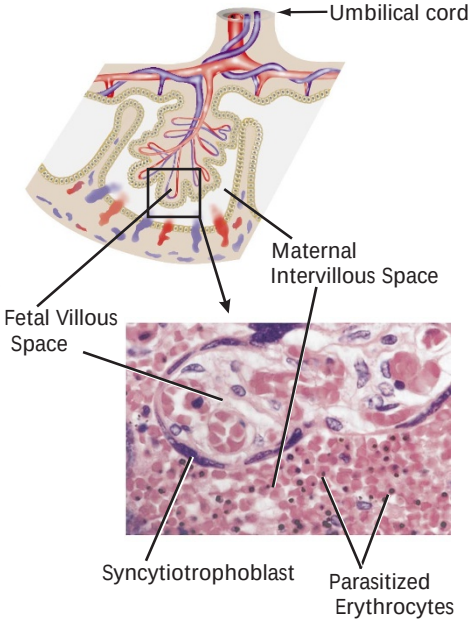
FOR CENTURIES, THE mortality in Africa from infection with *Plasmodium falciparum*, the causative organism of the most severe form of malaria in humans, has equaled mortality from all other causes. Chloroquine and other antimalarial drugs have modified the terrible history of this disease, but recently, with the spread of drug resistance, death from malaria has risen dramatically. The new Director General of the World Health Organization, Gro Harlem Brundtland, has responded appropriately to this emergency with a new program called Roll Back Malaria, which seeks to halve malaria-induced mortality by the year 2010. One component of this program is the search for new drugs, vaccines (none exist today) and other tools to reduce disease and mitigate the cost of malaria control.

A scientific correspondence in *Nature* by Fried *et al.*[1] describes a finding that raises the possibility of a vaccine to reduce the complications from malaria during pregnancy. For a long time it has been known that such complications are less frequent in women who have had multiple pregnancies (multigravid). Fried and colleagues report that multigravid women possess antibodies that block infected erythrocytes from binding to chondroitin sulfate A (chondroitin-4-sulfate, CSA), a receptor that enables the parasites to sequester themselves in the placenta. Sera from multigravid Kenyan women blocked adhesion of infected erythrocytes from pregnant women in different parts of the world suggesting that the antibodies may be strain-independent.

Malaria in Africa is not a single disease. Unlike non-immune individuals from Europe and the United States in whom the first infection can be lethal, many early infections in African children are not severe. There is an age dependency for disease complications[2,3]: severe anemia occurs early in childhood; cerebral malaria, later. After repeated infections, children develop anti-malarial immunity that controls parasite growth and limits disease from subsequent infections. Adults living in endemic regions rarely experience severe malaria morbidity. However, pregnant women, especially those in their first pregnancy (primigravida), are an exception. Although they suffer from high parasitemias and anemia, the main effect of infection is on the fetus and the newborn infant. Infected mothers have low birthweight babies who have an increased risk of dying[4]. The deleterious effects of malaria on fetal development are believed to stem from the large numbers of infected erythrocytes sequestered on the maternal side of the placenta. Ordinarily, the maternal and fetal circulations do not mix, but infection of infant cord blood is more common in mothers whose placentas contain infected red cells at delivery. Fetal infections are rapidly eliminated without treatment[4].



Characteristics of the mature placenta. Maternal blood empties into the intervillous space and flows over syncytiotrophoblasts that line chorionic villi extensions from the fetus. Fetal blood vessels are contained within the villi. The photograph (by Daniel Connor) shows a placenta containing *P. falciparum*-infected erythrocytes . Infection is confined to maternal erythrocytes in the intervillous space and is absent in fetal erythrocytes within the villi. Infected erythrocytes recovered from placenta bind CSA and adhere to syncytiotrophoblasts possibly through thrombomodulin, a proteoglycan that bears CSA. Many infected erythrocytes in the intervillous space do not seem to make close contact with syncytiotrophoblasts. Women who have antibodies that block infected erythrocyte adherence to CSA have fewer parasites in the placenta at delivery than women without these blocking antibodies.

P. falciparum can be distinguished from other *Plasmodium* species that infect humans because only immature, ring-infected erythrocytic forms circulate in the peripheral blood. Mature erythrocytic forms (trophozoites and schizonts) bind to vascular endothelium through 'knobs' (parasite-induced modifications of the red cell surface) that enable them to be sequestered in the venules and to avoid elimination by the spleen. Assays to detect binding of infected red cells to endothelium were developed in the early 1980s and have helped to define several different receptors including CD36, thrombospondin, intracellular adhesion molecule 1 (ICAM1) and others[3].

During the search for new receptors, infected erythrocytes were found to bind to CSA (refs. 5,6). Although CD36 is considered the main receptor for the adherence of parasitized red cells to venular endothelium, parasites recovered from the placenta strongly adhere to CSA but not to CD36 (ref. 7, and J. Beeson & S. Rogerson, personal communication). These findings begin to unravel the mystery of the mechanism by which parasites sequester themselves in the placenta. Maternal blood empties into the placental intervillous space that is lined with syncytiotrophoblasts (see Fig.). Whereas the parasitized erythrocytes adhering to venules contain 'knobs' that are in close apposition to the endothelial surface, ultrastructural analysis of the placenta shows that the 'knobs' of infected red cells are not closely associated with syncytiotrophoblasts. Indeed, parasitized erythrocytes in the intervillous space do not seem to make close contact with syncytiotrophoblasts. The syncytiotrophoblast surface bears the proteoglycan thrombomodulin and perhaps other proteoglycans such as betaglycan that display CSA. Infected red cells adhere to thrombomodulin *in vitro*, so it is possible that CSA on this proteoglycan, extending well beyond the syncytiotrophoblast surface, binds infected red cells.

The best-characterized parasite adhesion molecules belong to the PfEMP1 (*P. falciparum* erythrocyte membrane protein 1) family of proteins, which are encoded by *var* genes[8–10]. PfEMP1 proteins are concentrated in the 'knobs' of infected erythrocytes and bind CD36 (ref. 8). There are about 50 copies of the *var* gene per genome and a different set of *var* genes in each *P. falciparum* clone[9]. Thus, there is a large repertoire of adhesion molecules available to the parasite. Antibodies to one PfEMP1 variant kill infected erythrocytes that express this variant. Unfortunately, the malaria parasites constantly switch expression to other versions of PfEMP1. If the immune system has not experienced a particular PfEMP1, then parasite clones expressing this variant are able to expand. During the course of *P. falciparum* infection, children develop antibodies against the variant antigens of the infecting isolate[11]. Presumably, sufficient immunity eventually develops to the full PfEMP1 repertoire to maintain infections at low parasitemias; disease develops in children who lack a full range of antibodies[11].

Why then during the first pregnancy do women develop high parasitemias that are not seen in these same women before pregnancy? It now seems that the placenta may provide a new haven, selecting for CSA-binding parasites. Infected red cells that bind CSA are rare in non-pregnant adults despite the fact that thrombomodulin is present on vascular endothelium. The precise chemistry of CSA in the placenta is unknown, but differences between endothelial and placental CSA may explain parasite adhesion in the placenta and the distance of infected red cells from syncytiotrophoblasts. Furthermore, blood flow within the intervillous space is extremely slow and this might facilitate adhesion of a special placental subset of CSA-binding parasites.

In their study, Fried and co-workers[1] assessed the ability of sera from individuals living in Kenya, Malawi and Thailand to inhibit binding of parasitized red cells to CSA. Anti-CSA adhesion activity was present in multigravid women who had few sequestered parasitized red cells in their placentas, but was absent in primigravid women and adult Kenyan males. Multigravid Kenyan women, especially from high transmission areas, have anti-CSA antibodies that are strain-independent; that is, they block CSA binding of placental parasites collected from pregnant women from Asia and other parts of Africa. The essential question is whether anti-CSA antibodies are directed at a conserved region present on all parasites sequestered in the placenta or whether antibodies have multiple specificities. Two independent studies have shown that *in vitro* selection of infected erythrocytes on CSA leads to enrichment of single *var* mRNAs encoding PfEMP1 (refs. 12,13). It is unknown whether parasites in the placenta bind to CSA through PfEMP1 or some other ligand. If antibodies from pregnant women recognize a conserved epitope, then the CSA-binding domain of the parasite—whether it be in PfEMP1 or some other molecule—becomes a potential target for vaccine development. Definition of the parasite CSA-binding domain will permit more detailed characterization of the conservation of this region in parasites sequestered in the placenta.

The usefulness of PfEMP1 and other parasite adhesion molecules as vaccine candidates has been questioned because they are so highly variable. The Fried study demonstrates how insights into the pathogenesis of malaria may inform and focus vaccine development against *P. falciparum*.

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A happier sequel to *Lorenzo's Oil*?

Pharmacological manipulation of gene expression brings new hope to the treatment of X-linked adrenoleukodystrophy (pages 1261–1268).

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MUCH IS KNOWN about the biochemical and genetic bases of inherited diseases in man, but there has been less progress in the development of suitable therapies. For some inborn errors of metabolism treatment relies on restricted dietary intake of certain food components (for example, phenylketonuria) whereas in others, therapy of any sort is non-existent. Pharmacologically preventing the formation of a harmful compound has shown promise in the treatment of tyrosinemia type I (ref. 1). In other disorders, treatment aims to correct the primary defect directly—a good example is the administration of glucocerebrosidase to Gaucher disease patients[2]. Another strategy uses drugs to increase the expression of a gene that is functionally related to the defective gene causing the disease. This is the approach taken by Kemp *et al*.[3], as reported on page 1261 of this issue, in the treatment of X-linked adrenoleukodystrophy (X-ALD), a disease made famous by the movie *Lorenzo's Oil*. The investigators show that the drug 4-phenylbutyrate (4PBA) promotes the expression of a peroxisomal protein (functionally related to the defective peroxisomal protein in X-ALD), which corrects the metabolism of very-long-chain fatty acids