Chapter 1
of

Contrary Life and the Technical Fix
from malaria vaccine to hormone contraceptive

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Chapter 1
Malaria: set a vaccine to catch a parasite

A child floats on waves of drowsiness, dipping in and out of sleep as if subconsciously aware of some danger on the night air. Laden with vapours, the air spreads moisture from the recent rains and wafts odours of cooking and wood smoke through a pervading blossom scent and the fustiness of rot. Vibrating on the air are the incessant croakings of frogs, bursts of laughter, scraps of music from a radio. The people of the village attend their evening chores, social business and pleasures. Thunder clashes roll southward from beyond the nearby border, somewhere beyond the edge of the Congo River basin.

The child tries to sleep under the bednet she will later share with her mother. At four years old she already understands to keep to one side of the bed, but there her arm presses against the net's fine meshwork. The bed, pushed up against the wall, makes room for the charcoal cooker, boxes of food and clothes, a table, two chairs and a radio: her home. Up the wall wafts a tremulous plume of air slightly warmer and moister than the rest, the child's breath and sweatiness. The plume oozes between the top of the mud wall and the thatch of the roof to join all the other smells of the night.

On the wing nearby is a mosquito. Sensing extra carbon dioxide in the plume, the mosquito follows the clue up through gaps in the tattered thatch, then down around the plume with its shimmer of warm odours rising through the bednet. Blood is what the mosquito seeks, another meal after laying its third batch of eggs onto a nearby puddle. Hunger for nutrients to supply another batch of eggs drives the mosquito before it dies of exhaustion, or is eaten by a bat. Only blood will do, particularly that of a human, and the child's skin against the net is little barrier to the finely flexible tube of the bloodsucker's mouthparts. As the mosquito thrusts and probes around in the skin, seeking a minute capillary blood vessel just below the surface, it squeezes out saliva to ease the probing and prevent clotting.

The hint of danger on the air manifests here, in that dribble of insect saliva in the child's skin. Microbes, single celled protozoan animals of the plasmodia kind, slither away from the pool of saliva and out between the fibres and cells of the skin. These plasmodia also seek blood, but instead of the cost of minor irritation from the bite of a mosquito, this bunch of just eighteen plasmodia can multiply over and over again to vast numbers of blood seekers teeming within the child.

Within the mosquito, in its gut then salivary glands, the plasmodia have developed to their stage called sporozoites. Miniscule threads of living matter, they have a central nucleus containing the genetic information for the rest of their development, and an array of organelles at the forward end called an apical complex for penetration into individual cells of the child. Moreover, they contain a mitochondrion organelle for energy transformations, and another organelle called an apicoplast which, like the mitochondria, is derived from an ancient microbe that came to live together with another precursor cell. Altogether a new and sinister

combination cell that now seeks to live within yet more cells. Live off them, at their expense.

Even the containing membrane of the sporozoite, the outermost layer of the cell, is more like an organelle than a wrapper. First it acts as a molecular conveyor belt, using molecules of a protein related to thrombospondin that move along the level of the membrane in one direction, gliding the sporozoite forward in the opposite direction over tissues within the skin. These special molecules connect to an internal arrangement of molecules that ratchet back and forth upon a sort of chain, connected in turn to the gliding molecules – a molecular clockwork. The sporozoites seek a blood capillary in the skin. Contact with one is detected by interaction between another type of molecule in the outer membrane, the circumsporozoite protein, and characteristic molecules on the surface of these minute blood vessels. Once gliding along the outside of the vessel, the sporozoite activates its penetration apparatus to invade rapidly the blood within the capillaries. In less than a minute the sporozoite sets out toward its first important home: the liver.

Eventually the child drifts into deep sleep, leaving the sentinels within her body to sound warning of the invaders. Speed becomes crucial for the plasmodia. Already some of the original group of sporozoites seem to have lost their way in the barrier of the skin, with its dense packing of collagen fibres. Between the fibres is a sparse film of liquid, the lymph that spreads from the plasma of the blood, past the cells and tissues of the body and drains into the ducts and nodes of the lymphatic system. Here is the diffuse address of the child's immune system, distributed all over her body and already bearing traces of its defences against previous infections with this *Plasmodium falciparum*.

There are molecules of antibody specifically dedicated to this and only this species of parasite. They will bind and coat the invaders with a molecular layer that can tighten around the sporozoites and cause them to falter and shrivel. A dying invader like that is spotted easily by scavenger cells, the macrophages, within the skin and deeper within the lymphatic system and blood. These cells are equipped with wide veils of their outer membrane that spread out and around such invaders as sporozoites, sensing them by their coating of antibody as not being of their own kind, not the self of the whole child's body. Antibody and macrophages see the sporozoites as antigenic, consisting of molecules that are non-self. Sporozoites are also seen that way by sentinel cells described as dendritic because of their long tentacles covered with a range of molecules sensing, antenna like, for invaders.

The macrophages and dendritic cells are sensitive to the minutest variations in the antigenic character of invaders, and although these sporozoites are *Plasmodium falciparum* they are not the usual variety, the kind that has been circulating between the people of the village for years. These sporozoites seem to be something new. Any of the skin's macrophages and dendritic cells that have contacted this group of sporozoites will become activated to detach from their posting and migrate through the lymph ducts into the nearest lymph node, the one in the child's armpit. There they start urgent communications between immune

mechanisms, presenting molecular evidence that this infection is not another usual one of the local plasmodia. In contrast, it is something to which a new response is urgently needed, adapted specifically to this invader.

The ability of the child's immunity to coat a few of the sporozoites with antibody resides in memory cells deep within her lymph nodes and spleen. These are the archive keepers, the recorders of previous battles with this parasite. They came from a small population of cells, white cells of the blood called lymphocytes. These originated from just a particular type of cell, a stem cell for lymphocytes, deep within the marrow of the child's long bones. They are a clone that had proliferated vastly when that one stem cell, one of trillions of its kind, happened to match precisely to the character of this new antigen. The match was molecular, derived from a precise mechanism that generates random mixing of genetic elements. Once the lymphocyte's lock encountered and engaged its antigen key, the lymphocyte became activated to divide and divide into an army of immune defenders. After the battle they settled down to a stable small reserve of cells with the memory of how to multiply again to fight the same battle. An immune capacity acquired specifically by exposure to infection.

Are these plasmodia new to the village, recognizable by subtly different forms of their clothing? Some of the villagers earn a living working in the mines away in the east. On their journeys home they often have to spend the night in poor accommodations, exposed to the mosquito species *Anopheles gambiae*. A bloodsucker parasitized in turn by plasmodia which themselves will feed on blood in their favourite and most secure home – inside the red cells of human blood. The miners stay for a while in their home village and in their blood may hide a new form of plasmodia, one that has adapted to different conditions, perhaps to survive treatment with different drugs.

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The new sporozoites that remain in the child now plunge into a stormy world of blood. A thick liquid pumped insistently to squeeze along the meandering channels of the capillaries, back into veins, then heart and arteries leading to organ and tissue. A wild and dangerous ride for the sporozoites, now surrounded by traces of antibody that might bind to them, dodging the circulating macrophages, and avoiding capture in the immune filters of the lymph nodes and spleen.

Only the liver will provide respite for the sporozoites. This large organ is dedicated to processing both blood for its load of nutrients from the intestines and also as filter against any product of digestion that is unsafe, from miscellaneous microbes to the alcohol in a villager's brew of beer. The liver consists mostly of biochemical factory cells, or hepatocytes. The blood gets to the hepatocytes along passages in the liver called sinusoids. The first sporozoites arrive here within a few minutes of escaping from the skin. They are close now to a good home, a place where they can settle down to mature and produce offspring. But first they have to run the gauntlet of a fearsome array of more macrophages. These ones are so particular to the liver they go by the name of the scientist who first discovered them – Kupffer cells. Like

any good sentinel cell they extend their protean forms out into the blood channel of the sinusoids to catch particles that may be a threat as they float by in the blood. The lining of the sinusoid is studded densely with these Kupffer cells, seemingly a stout barricade between the sporozoites and the homely liver cells.

As soon as the sporozoites enter the liver sinusoids the circumsporozoite and thrombospondin type molecules on their surface recognize a type of molecule, particular to the sinusoids, which coats the structural cells of these passages. The sporozoites promptly land here and start to glide again. Once moving this way they become poised for another traverse manoeuvre, using their penetration apparatus. And what do they traverse but the very cells that should be trapping and engulfing them – the Kupffer macrophage cells. This audacious subterfuge takes them directly down beneath the lining of the sinusoids.

When they emerge beyond the inner side of the Kupffer cells they continue their penetration into liver cells. They have just bored unharmed through a type of cell dedicated to killing them. Worse is to come. As the sporozoites traverse the Kupffer cells they shed much of their outer molecular coats. These coats then suppress the ability of the Kupffer cells to send out warning signals to the rest of the organs of immunity. The Kupffer cells may secrete into the blood and lymph one of their possible signals, a soluble material called interleukin-12, but only weakly now. So the assaulted Kupffer cells lapse into a state of immune dampening.

Like much of the mechanics of immunity, with its multiple interlinked, overlapping parts of many origins, with its redundancies and even competitions, the liver helps maintain a balance within the various functions of immunity. The balance is delicately dynamic, but handled well by the multi-tasking liver. This organ controls much of the base level of inflammation resulting from exposure to so many threats of parasites and toxins from the intestines.

Having evaded or switched off some cells of this formidable Kupffer barrier, the sporozoites now seek an individual liver cell that best suits their need for a safe hiding place. They traverse several liver cells, hepatocytes, in quick succession, with little damage left in evidence. But something about the surface coat of a particular cell changes the behaviour of the sporozoite. No longer does the sporozoite crudely pierce through the outer membrane of this cell. Instead it precisely aligns its penetration apparatus against the cell's membrane to trigger formation of a hollow in the cell membrane. The hollow deepens into a cup, welcoming the sporozoite. From the penetration apparatus are secreted molecules that add to the liver cell's membrane, and the two different outer membranes zipper up tightly together, engulfing the sporozoite within the liver cell. As the sporozoite descends, the membrane of the liver cell folds over its tail, the cup closes over, leaving all smooth again. Inside the cell is now a balloon, a parasitophorous vesicle, made of its own outer membrane, within which sits the sporozoite.

For the sporozoite this could be a dangerous place to be. There are other small vesicles in the ground substance of the cell, its cytoplasm, that can fuse with such a

vesicle containing an invader to release their loads of digestive enzymes. This invader renders such vesicles futile – unable to fuse. So now the sporozoite hides securely by means of two layers of cell membranes that are impenetrable to molecules of antibody. Rounding up into a ball, the sporozoite transmogrifies into something more sinister still.

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Outwardly the child shows no sign of the new infection that any doctor could detect. Immune memories of her older infections with the usual local strain of plasmodia remain deep within her body, readying for a fight. Now this new infection consists of just nine of the original batch from the mosquito. Nine minute specks of living matter on the massive scale of the liver, but the scale that counts for health and disease now is measured in molecules: huge ones as proteins like enzymes, tiny ones like simple compounds of oxygen plus nitrogen atoms. The sporozoites traversing the liver cells were safe from antibody, the molecules of which are too large to diffuse through the outer membranes of cells. However, they are not safe from other defences that are well developed within dedicated killers of parasites such as macrophages and also reside in the highly competent hepatocytes of the liver. The sporozoites are not sufficiently safe merely to hide. They need to reproduce fast enough into another form with the ability to invade an even safer type of cell, the red cell of the blood.

The nucleus of the sporozoite, so far more of a manager on the side-lines rather than a forward player, now takes over completely. It divides in two and both in two again, repeatedly and exponentially. This forces the daughter nuclei outward to fill the cytoplasm that itself expands to support these replications. The parasite cell remains a coherent whole, a sphere called a schizont, but comprising many splitting nuclei. The liver cell grows around the schizont, not merely stretching but accommodating the invader with a massive supply of nutrients. The divisions continue until about thirty thousand nuclei have been created from the original one. Then cytoplasm of the parasitic schizont gathers itself around each nucleus, and an outer membrane forms to define each new individual parasite. These new forms of plasmodia, now called merozoites, remain within the original vesicle made of the membranes of their host liver cell.

Within this disguise, the cloaking vesicle or merosome, thousands of new parasites squeeze out of the now dying liver cell. The liver cell might be dying but the usual molecular signal that activates phagocytic cells to dispose of such a cell is inhibited by the merozoites. The vesicle pushes past the cells lining the sinusoid of the liver, past the Kupffer cells, and on reaching the blood stream is rapidly transported far away. Reaching other organs the merosome finally bursts open to spew its cargo of invaders amongst the red cells of blood.

Antibody cannot reach the developing schizonts but more complex weapons and tactics are deployed despite having been trained against the usual village strain of *Plasmodium falciparum* and uncertain against this newcomer. As a schizont grows, its own biochemical processing of nutrients provided by the liver cell inevitably

produces by-products and wastes that are detectable in the cytoplasm of the liver cell. These signals of infection alert the nucleus of the liver cell to activate a processing of these foreign molecules with an array of what are known as histo-compatibility complexes. These engage the foreigners and together the complexes move to the outer membrane of the liver cells.

There the complexes become molecule sized flags, waving to wider immunity that precisely here in this cell is a parasite that must be killed, whatever it takes. Away in the lymph nodes and spleen, in response to the first faint signals from the sentinel macrophages and dendritic cells, special white cells, T-lymphocytes of the blood, have finally matured for action. These are of a type positive for a character known as CD8. Like the B-lymphocytes that generate antibody, these T lymphocytes originated as a clone from stem cells. They developed with receptors that exactly matched this new invading strain of parasite. These T cells contain tiny vesicles within their cytoplasm that contain powerful enzymic proteins that can perforate the outer membrane of an infected cell and similar ones that once inside the infected cell will digest it.

Once one of these T cells has docked onto the warning flags of the histo-compatibility system it orients its internal composition so that a bunch of toxic vesicles comes to the contact surface, bursts out and floods over the infected cell. The liver cell dies and any developing schizont will succumb with it. The T cell closes up its outer membrane, reorganizes itself and continues its search.

There are other defenders on the way. These are T-lymphocytes of the CD4 variety, helper cells that also detect the warning flags, orient against the cell and respond by releasing a pulse of a specific chemical messenger, gamma interferon. This small labile molecule penetrates into an infected liver cell where it stimulates two types of enzyme of the cell.

One enzyme catalyzes the production of nitric oxide, that is the molecule also known as nitrogen monoxide. The other enzyme catalyzes the production of the superoxide of oxygen. This is one of the intermediates in the normal pathway of energy generation in the cell when oxygen is converted into water during a series of steps involving transfer of electrons and protons. These tiny molecules, just two atoms each, are potent and vital parts of the normal workings of any cell. But now they combine into a more stealthily fatal oxidizing agent called peroxynitrite. This destroys the very cell that produced it. The sudden burst of oxidation causes a combination of ordinary tissue damage, and it triggers a mechanism of natural cell suicide, called apoptosis. Death of the host cell at this stage of the plasmodial cycle kills the parasite.

The liver, however, is a big place to hide. Somewhere within its bulk there started out nine sporozoites that managed to turn into small schizonts. For the hunting lymphocytes to chance upon the flags warning of infection before the merozoites are mature and ready for the next phase is a race. Chance is on the side of this swarm of invaders now explosively replicating. Only five of the schizonts survived

to produce its battalion of merozoites, but five doses of thirty thousand each might be enough to overwhelm the proliferating white cells.

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Blood is thicker than water – packed with red cells, or erythrocytes. Simple cells, so dedicated to their one task of carrying oxygen deep into every organ that, as they mature from their stem cells, they each dispense with their nucleus. A great overall efficiency, but burdened with a dangerous weakness. They become ideal as the next home for plasmodia. As a merozoite floats in the plasma, colliding with the densely packed red cells, the merozoite at one micrometre across (one thousandth of a millimetre) is attracted electrostatically to the ten times larger disc of the red cell. A red cell is easily seen under the highest magnification of an ordinary microscope. In contrast, a merozoite, when suitable stained, is near the limit of what can be resolved because the wavelength of visible light is only just short enough.

A merozoite packs a lot into a minute space. Slightly pear shaped, with its nucleus occupying much of the blunt end and the penetration apparatus poised at the sharper end. There are two organelles – the mitochondrion and the apicoplast. At the penetrating end there is a complex comprising a pair of rhoptries and ten micronemes, all bound round with three polar rings which in turn are anchored to the depth of the cell by microtubules. The remaining space is packed with sundry granules. Everything works together for invasion and reproduction whilst evading the child's immunity.

Now the penetration apparatus becomes activated. Molecules protruding from the outer membrane of the merozoite match up with specific molecules on the red cell. The merozoite orients to place its sharp end against the red cell's surface and an intimately tight junction promptly forms between the two surfaces as their molecular surfaces mutually attract. Suddenly the red cell surface convulses and collapses as a deepening cup. Tubes in the merozoite contract to push the penetration apparatus deeper in. As the merozoite penetrates, its surface coat is shed into the plasma of the blood. All is done within thirty seconds as the cup in the red cell envelops the whole merozoite. Finally the lip of the cup closes in and coalesces to form a membranous vesicle around the merozoite.

There now sits the merozoite, within a membrane, the parasitophorous vesicle, which in turn is within the outer membrane of a red blood cell. This red cell, without a nucleus, has lost its ability to process the antigenic proteins of an invading parasite and deploy them on its outer surface as distress signals crying out: any passing competent white cell must eat and digest me and the parasite infecting me. Instead, the merozoite sinks below the scanning horizon of the child's immunity. The quicker the better to avoid antibodies that will soon be circulating in blood in increasing concentration. Antibodies that will accurately detect and latch onto the particular proteins on the outer surface of merozoites as they briefly float naked in the blood plasma. Any such coating with antibody will render a merozoite both incapable of invading a red cell and an immediate target for engulfment and

phagocytosis by the macrophage cells that circulate in the blood stream and line the ducts of the lymph nodes, spleen and liver.

The merozoite engulfs droplets of the material making up the red into its digestive vacuole. The food for the merozoites reproduction is mostly haemoglobin – the oxygen carrying protein of blood – and this is digested with the aid of enzymes. The merozoite grows and starts to divide, as a second type of schizont, to form daughter merozoites. Two days are spent going through four series of divisions, stretching the red cell until it bursts with release of sixteen new merozoites into the plasma. Soon each of them in turn will be either infecting another red cell, or succumbing to antibody and macrophages. They are small, but the volume of blood is large and the number of red cells that are free to be infected is enormous, so there is some time to go before anyone with a microscope is likely to be able to detect a red cell infected with a merozoite. For a while – but all too soon the child will become aware of sickness within.

The merozoite takes in much haemoglobin and has to process this molecule to rid itself of the potentially toxic part of this protein that contains iron. Crucial to the function of haemoglobin as the carrier of oxygen, this iron lies safely at the centre of the huge molecule. As digestion proceeds the iron is exposed and oxidized, stimulating the release of the superoxide form of oxygen that could kill the merozoite. Instead the merozoite commandeers a specific enzyme to deactivate the superoxide. The merozoite's digestion of haemoglobin results in the eventual formation of a much simpler molecule called hemozoin. This poison is discarded into the blood plasma when the red cell bursts apart. Not simply poisonous, the hemozoin suppresses the activity of immune sentinel cells as the first line of defence against plasmodia and other invaders.

The child starts to feel ill: initially a vague malaise, rapidly followed by fever. The sequence of infection of red cells followed by replication of the merozoites goes in a two day cycle. With every cycle the number of merozoites in the blood increases ten-fold and the child's fever rages when hemozoin is released. Her immunity is now working all out, exhausting her reserves of energy. A cascade of interconnecting inflammatory reactions to all these parasites flooding directly into her blood plasma brings on fulminating malaria with bizarre symptoms – paradoxical sensations of shivering coldness follow each burning fever.

The red cells are not, however, an impregnable bunker. Despite their lack of nuclei they retain a trace of the capacity that normal nucleated cells, such as hepatocytes, have for processing the antigens of invaders and to place them as warning flags on the surface of the cell. So some of the infected red cells get a coating of specific antibody and this enables macrophages to engulf them. Moreover, as any red cell is infected by a merozoite the process of passing into the red cell produces molecules from the merozoite out onto the surface of the red cell. There are various types of molecule involved, such as the one called erythrocyte membrane protein. Out on the surface of the red cell, such an antigen from *Plasmodium falciparum* will identify to immunity that this cell is infected. But this identification has to be exact; it will only work if there is a precise match with receptor molecules on the outer

surface of lymphocytes and macrophages. These immune defences have evolved to kill invaders only, if they deploy their deadly granules and secretions and tentacles too freely the whole healthy body will suffer. The immune antibody and cells react immediately with what they or their parents have recognized before as foreign threats.

For the child, her immunity struggles with this plasmodial protein coating the surface of her red cells. Has her body acquired immunity to it before or is it of a slightly different form? There are sixty different genes for this protein, each of which can code for another variant. The merozoites slowly cycle through their repertoire, every slight variation another disguising cloak for the plasmodia to hide behind. If the erythrocyte membrane protein is new then several weeks will elapse before the child's immunity acquires a fully effective response to it. And so the fight blunders on with an advance here, a retreat there. Ever more evasions, disguises and decoys are deployed by the plasmodia against the weapons in the multifarious immunity of the child.

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Her mother has witnessed these fevers before and she knows them from bitter personal experience. But now she sees something new and strange happening – her daughter seems to be drifting away to a place she has never travelled before. The mother calls her nephew to seek somebody in the village with an automobile or truck who can take them to the hospital in Kalene. Not far away, but a struggle along a road now turned to clinging black mud in this season of rains. The admissions nurse recognizes yet another acute case requiring an immediate blood sample for diagnosis.

In less than an hour the laboratory technician has smeared a drop of the child's blood thinly onto a glass slide, stained it deep blue and purple and peered within the red blood cells under a microscope. Just a few, minute but unmistakable, the plasmodia: blue, purple, and dead within the grey shadow of the red cell discs. He sees not just the ordinary stage in the blood, the merozoites. Under the microscope are the elongated forms of the parasite bent into crescent shapes tight within the confines of the red cells. Sexual stages of the plasmodia, from genetically female and male cells that have developed as an offshoot of the regular cycles of asexual replication of the merozoites. The technician knows the warning – this infection is well advanced, crisis is impending. By now there could be tens of thousands of plasmodia in every millilitre of blood, in every quarter teaspoonful.

Not just dangerous to this child – now to those around her as well. Once any sexual forms of the plasmodia get back into an *Anopheles* mosquito that might feed on the child, they will fuse as an egg stage, a zygote. The genetic mixing that follows will add more possibilities for variant forms of the plasmodia that better evade immunity of people. Similarly in humans, reproduction with two sexes – rather than virgin-birth or similar – has a matching function of enabling a vast variety of possible immune defences, of molecular locks to match the foreign antigenic keys.

An endless battle of attack and counter-attack, of camouflage, subterfuge, calibres of weapon and multiple redundancies.

The duty physician prescribes the usual course of drug against this parasite. One of the original anti-malarials and still the favoured choice: quinine, extracted from the bark of the cinchona tree. The last chance treatment because now the plasmodia are progressed toward their most extreme tactic. When a merozoite infects a red cell, that cell loses much of its flexibility and slipperiness that is essential to its ability to be squeezed by pressure from the heart along the minutely narrow passages of the capillaries that supply deep through every tissue and organ.

The brain is always ravenously hungry for the blood's supply of oxygen and sugar. When infected red cells stiffly jostle deep in the brain, some of them jam there. They not only jam like bricks but they stick to each other and to the inner walls of the capillary. The stickiness derives from the same erythrocyte membrane protein that served the merozoite as a disguising cloak worn by the red cell. Clumps of infected red cells become sequestered, not just in the brain but also in the heart and kidneys, all safe from the immune filtering of the spleen. These sequestered merozoites continue to replicate and maintain a reservoir of infection and so pass to another human.

In her hospital bed the child approaches the crux of her fight with *Plasmodium falciparum*. The clumps of sticky infected red cells deprive spots of her brain of sufficient oxygen. Immune white cells, clustering around these infected red cells, fire off their chemical messages and toxic granules, but this is a dangerously confined space for such powerful weapons. Again, nitric oxide is produced. This molecule is very short lived and active. In minuscule and steady doses it is vital to many functions of the body as a regulator message, including normal working of nerve cells. The groups of macrophages around the sequestered red cells in the brain fire off nitric oxide and the superoxide of oxygen. Again peroxynitrite is formed and if the right amount in the right place is directed well it is a powerful killer of microbes and parasites.

Too much in the confined spots of sequestered plasmodia within the brain damages the ability of nerves to transmit impulses. The child swings in and out of consciousness and her body heaves with the contortions of cerebral malaria. Her blood has lost much haemoglobin making her anaemic and her body tissues have become acidic. Even the quinine has the side effect of reducing the sugars available in her blood, depriving her of the energy needed for this fight.

She wins. With the help of the hospital staff, quinine, and her mother's love she lives on. But her mother will worry for years whether her daughter's battle with this remorseless foe has left her with some diminished ability. How well will she fare when she starts school?

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This story of a malaria parasite and its interaction with a person's immunity is partly true – a composite of knowledge of species of *Plasmodium* in various species of host, but rarely from humans because it is so difficult to study this disease directly in people. My account has some approximation to objective reality as it occurs in the natural world, but no better. It derives from many disparate descriptions of bits and pieces of that reality that have been made accessible by an enormous variety of researchers, rarely in full agreement with each other, working tirelessly and with fantastic ingenuity.

Let me give an impression of a single technique of research, selected for its simple vocabulary. The precise movements of sporozoites as they invade a liver are studied because their passage from the skin to the liver cell exposes them to the effect of a vaccine. How is possible to track them? By video photography! Instead of examining a piece of liver as transparently thin razored slices in which to search with a microscope, the slicing is done by light. The researchers can thereby keep the liver alive and observe the movements of wriggling sporozoites in three dimensions.

But a sporozoite is just a translucent wisp. To render it visible the researchers will have inserted into it the genetic code for a protein that glows green when illuminated. This green fluorescent protein was first found in a species of jellyfish common in coastal waters of America, it self-illuminates small spots around the jellyfish's outer rim. The result is that live sporozoites glow green against the dull background of the liver.

One photon of ultraviolet light is all that is needed to excite one molecule of the protein, but this type of light damages cells. Instead the protein is excited using two photons of light of a longer, safer, wavelength. But two such photons must arrive simultaneously at the protein molecule to do their work and the way to ensure that is to pack the photons together a trillion times more tightly than usual. The laser source of light delivers exceeding short bursts of light and these in turn are focused onto the liver down through the lens of the microscope. The spectacle of a crescent shaped sporozoite gliding along the sinusoid of a liver is captured with a video camera mounted on the microscope.

Obviously this story of malaria is an approximation because so simplified and compressed; all the information written about malaria would overflow the libraries of a university. Worse than that, such information is doomed to be an approximation because the deeper researchers observe and experiment within the complexities of the molecular tricks and contortions of *Plasmodium* and the intricate maze of the human immunity the less it is possible to give any single summary account.

None of this is a sufficiently severe impediment to continuing on to our technical fix. This is one fix of many available now, and of a large array of all manner of solutions tried in the past with various degrees of success, from the attempt started in the 1950s to eradicate malaria by killing mosquitoes with DDT insecticide, to the invention of chloroquine as a cheap, safe and effective synthetic version of

natural quinine. Both of those fixes succumbed partially to the ability of mosquitoes and *Plasmodium* respectively, to mutate into resistant strains. Only partially: these chemicals remain in some specially limited use and alternative versions continue to be invented and deployed despite repeated predictions of impending failure to maintain the rate of invention.

A killer of children on a global scale as *Plasmodium* deserves the attention devoted to killers and maimers like the viruses of smallpox and polio; the first truly eradicated from the Earth (except for two samples stored in USA and Russia) and the second soon to be. Both were eradicated by mass vaccination. However, although some viruses and many bacteria have proved easily susceptible to vaccines, others such as the influenza virus easily slip past the protection provided by vaccines. As for protozoans, and for parasites made up of many cells, it seems the more complex they are the more difficult it is to devise a vaccine against them and the more difficult to ensure the vaccine is sufficiently comprehensive and efficacious.

So talk of eradication is not a suitable way to promote vaccination against malaria. But the modern ability to manipulate molecules combined, with the deep collective understanding of malaria, now encourages more funding and research on a vaccine to protect humans from malaria. Hopefully the advantage of an effective vaccine over the insecticides to kill mosquitoes and drugs to kill plasmodia is that the problem of acquisition of resistance will be avoided sufficiently. Furthermore, when vaccines work well they do not have to be delivered year after year, a primary shot followed by some boosters often suffice. This should be easier to deliver than heavy combined doses of anti-malarial drugs. These need to be taken almost continuously for full protection because of the frightening ability of plasmodia to acquire resistance to the drugs if there are any lapses in levels of the drugs.

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A doctor developing an early vaccine against malaria, W. Ripley Ballou, came down with the disease. He was shocked, despite having been exposed to the plasmodia. After all, he had been vaccinated and was eagerly anticipating exiting developments along the way to an anti-malaria vaccine. He was then, 1987, working as an officer in the Walter Reed Army Institute for Research, just north of Washington DC – a laboratory with a long history of battles with malaria. One of the many dismal facts of warfare is that in many campaigns more soldiers have been killed by agents of infectious diseases than by enemy combatants. Army and naval laboratories to study and prevent these losses have made many contributions to protect their own forces, and these improvements spill over into civilian medicine.

Doctor Ballou was a volunteer to receive a trial anti-malarial vaccine, then be exposed to plasmodia from bites of infected mosquitoes. And so, when remaining healthy, demonstrate the vaccine worked. This was an early vaccine of a new type, less effective than he hoped, but he was cured with drugs according to the

emergency plan. Volunteering yourself as a test animal is a well established tradition in the infectious disease business.

Ballou, more determined than ever from his salutary experience, went on to become one of the leaders of research that has produced, at the time of writing, the type of vaccine against malaria with the largest and widest clinical testing. Aimed specifically at infants, it has been tested widely and for many years in Africa south of the Sahara, where malaria caused by *Plasmodium falciparum* is more intense and devastating than anywhere else in the world. Named cumbrously as RTS,S/AS01, a vaccine abbreviation that I will explain later. Hopefully it will soon have a catchy title that journalists can publicize; for now calling it 'sporozoite-vaccine' will suffice. This is the technical fix most appropriate to explore further with you. First, we need some history.

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A vaccine against malaria was tested about 1941: it gave some protection to birds. Yes, birds: they suffer from their own type of malaria. One of the discoveries of how malarial parasites are transmitted by mosquitoes was made by Ronald Ross working in Kolkata, India, in 1898 with a species of *Plasmodium* of sparrows. They were easier to manage than Ross's human patients.

That vaccine was tested in chickens, and although funded by the Rockefeller Foundation and developed in a malaria laboratory in India, was crude even by the standards of its day. The researchers ground up parts of infected mosquitoes – the thorax containing the salivary glands and their load of malarial sporozoites. Crude though it was, partially effective and only in chickens, it was the first tentative step on a journey that led, haltingly and tortuously to our current sporozoite-vaccine. The stage of the malaria parasite those researchers in India targeted was the sporozoite; hoping to halt the disease before it became established deeper in the body.

Research on malaria during the 1940s concentrated on the urgent need for a synthetic substitute for natural quinine, then scarce because of the disruption of wartime. Chloroquine was discovered in 1934 and similar chemical drugs were found by 1945 amongst fourteen thousand compounds screened. By 1939 the powerful insecticide DDT was found amongst a ragbag of synthesized chemical oddities. It was a vast improvement on the general poison arsenic, which had been used in controlling malarious mosquitoes. This new material was a genuine insecticide, specifically and acutely poisonous for insects. So now the plasmodia could be killed in the blood by regular, prophylactic, doses of cheap and effective drugs whilst the habitats of mosquitoes were sprayed with DDT.

The eradication of malaria suddenly rose high up the political agenda and the Fourteenth World Health Assembly in 1955 adopted this as a principle. Two years later the World Health Organization started a campaign to eradicate malaria. The plan, however, was vague, lacking even definition of whether it was to eradicate the disease or the plasmodia that cause the disease. Also it lacked focus on the most

severe endemic continent of Africa. The main means was by mosquito control. Implementation of the campaign over the next decade competed with the effective campaign to eradicate the virus of smallpox, also run by the WHO. Nevertheless, with massive effort and money going into eradication, who might be bold enough to investigate a vaccine against malaria?

Ruth S. Nussenzweig, working in the medical school of New York University, understood from her upbringing and medical education in Brazil that there remained a future for a vaccine. In Brazil she had learnt of the confounding complexities of a mosquito eradication campaign that had been run in part of that vast country. Moreover, support for vaccine research became easier to obtain as resistance was acquired by both plasmodia and mosquitoes to the new chemicals deployed against them by the 1950s. The US Army provided funds for a small project in her laboratory on First Avenue, New York, to test a way of vaccinating mice against a species of *Plasmodium* they naturally suffer from.

Again, sporozoites were the focus, but this time with a combination of delicacy and power appropriate to such a formidable microbe. Mosquitoes bred in the lab were infected. When the infection matured, the mosquitoes, one by one, were pulled apart. Gently, so that their large paired salivary glands emerged from the thorax as the head slid off, all in a drop of saline solution. The glands were disintegrated to release the sporozoites which were then bombarded with X-rays. The notorious effect of these rays on the genetic capacity of living things, as high energy photons collide with genes along the DNA molecule, was anticipated to prevent the live sporozoites from developing through their life cycle. Nussenzweig and her colleagues then delivered these irradiated sporozoites, massive numbers of them, as a vaccine. They recorded some protection against further infection in the mice, and were encouraged to extend the project to human malaria.

Ruth and Victor Nussenzweig, with an extensive series of collaborators and volunteer vaccinees, achieved high levels of immunity in people when the irradiation of the sporozoites was done whilst the plasmodia were still in the mosquitoes. Where to take this method next became an increasing problem. This was few people's idea of how to produce material for mass vaccination. Contrasting with all the talk about rational design of vaccines as the modern improvement on guesswork, trial and much error, this was worryingly low-tech and a poor way to inspire investment. Rationality was hampered not only by lack of knowledge of the precise immune mechanisms working against sporozoites under conditions of natural infection. There were no other methods to culture or replicate the sporozoites and their antigenic proteins that surely were inducing the immunity.

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Help for vaccinologists was soon to hand from a completely unexpected direction. In 1984 news of the invention of a method to synthesize insulin spread fast through medical schools and clinics. Insulin, for the treatment and management of diabetes, was then being harvested from the pancreas glands of cattle and pigs cut out at the slaughterhouse. Demand was expanding steadily and supplies were predicted to be

inadequate. Following a direct line of basic research to applied research to pure market driven invention, a route had been carved out from the discovery by James Watson and Francis Crick of the structure and function of DNA through to the invention of synthetic insulin by Herb Boyer and Sidney Cohen. The inventors described in their patent how to manufacture individual proteins by manipulating DNA so that the protein could be mass-produced in ordinary cultures of bacteria or yeast cells. That is another story, not only of a superbly successful technical fix, but of a true revolution in science and the birth of the industry of biotechnology (see Chapter 2).

For the Nussenzweigs, for Ballou, and for everybody else groping in the dark maze of the immunology and vaccinology of malaria a door swung open onto bright new vistas. By the early 1980s the techniques for working out the sequence of the sub-units of DNA that make up the gene carrying the code for a specific protein were applied to malaria. Fidel Zavala, with the Nussenzweigs, described a protein that forms a distinct coating all over the outer surface of a sporozoite. If sporozoites were exposed in the laboratory to solutions of antibodies previously raised against sporozoites, the antibody bound so tightly to the coat that it neutralized the sporozoite – stopped it from functioning, killed it. Zavala named the coat circumsporozoite protein. Surely this was the one to focus on – already it was well known that whole sporozoites could constitute a vaccine. Highly specific antibodies could react with it and so kill the sporozoites.

John B. Dame and colleagues at the National Institute of Allergy and Infectious Diseases in Bethesda, Maryland, then wrote out the entire sequence coding for the gene of the circumsporozoite protein. Finally, in this rush of conversion of malaria vaccinology from grinding up parasites to genetic engineering, James F. Young published a paper describing how to produce a synthetic version of circumsporozoite protein. He was with a large team spread across the Walter Reed institute, the Naval Medical Research Institute in Maryland, and the pharmaceutical company then known as Smith Kline & French Laboratories.

By 1985 there were two routes open to producing a synthetic mimic of those parts of the circumsporozoite protein known to be most active at inducing a strong immune response. One route, a chemist's style, was to assemble the immunologically relevant sub-units of the protein, in the form of amino acids, to make what are called synthetic peptides. The other route, more biological in style, was to trick microorganisms into producing the required segment of the circumsporozoite protein. The result is called a recombinant protein, named after the subtle action of an enzyme in the manipulation of the DNA (the root word recombine has a meaning separate from its use in the genetics of sexual reproduction). The team in New York took the synthetic peptide route whilst the Smith Kline & French and Walter Reed workers formed a formal collaboration for the recombinant protein approach.

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The vaccine of our story, the sporozoite-vaccine is formulated with a recombinant protein. It is complex as you might guess from its gnomic abbreviation and to explain it I will need to divert into a sub-plot. This is another technical fix story in itself, but not one for here because the disease causing organism of that story is a virus, amenable to the attentions of vaccinologists.

Within RTS,S/AS01 the R stands for repeat – of a row of identical sequence codes along about one third of the string of sequences for amino acids as represented in a line diagram of the protein, or its primary structure. These were selected for their ability to induce good antibody. They also induce good cellular immune responses, hence the T for T-lymphocyte. Neither S is for sporozoite however, but for what is known to vaccinologists as the S-antigen. This is from the surface coat of the virus that causes liver disease of the type called hepatitis B. The second S in the abbreviation is more of the same material, but coupled separately as a major proportion to the rest of the construction. AS stands for adjuvant system – more of that later, whilst 01 is serial code for a version. So the RTS is a hybrid synthetic protein derived from genetic information of two disparate parasitic organisms, a protozoan and a virus.

Vaccinologists sometimes call such combination a chimera. They could be tempting fate: a fabulous beast made up of parts from various animals, or a wild and unrealistic dream? The S-antigen has the inherent property of spontaneously folding up into spheres, about the size and shape of a virus. Without a genetic core it cannot replicate but it appears to human immunity like an invading agent of infection. Immune sentinel cells are alerted by it.

Where and how did a vaccine against malaria get into bed with an imitation of a hepatitis virus? This story starts with Baruch S. Blumberg, doing medical research from 1964 at what was then called the Institute for Cancer Research, Philadelphia. Rather than tackle cancer head on Blumberg typically continued with the highly individual approach he had cultivated since being a medical student and then consolidated whilst first doing formal research in the University of Oxford. He sought markers of diseases to be found in blood, in the innumerable variations of groups and immune characters of blood from different people and countries.

One of Blumberg's colleagues found a most unusual protein in a sample of blood from an Australian. The protein gained the lab nickname of Australia antigen. There it remained, a half-remembered oddity in the freezer. For a while the team thought they were on the track of another marker for leukaemia, but instead found more Australia antigen sufficiently frequently for the data to resolve into vague patterns of the sort that fascinate epidemiologists. Could this be a marker for an infectious disease? The patterns hinted at a virus; confirmed by finding some under an electron microscope. Further tests revealed it was identical with the virus causing hepatitis B. Australia antigen soon was revealed as more than a marker for the virus, it was the outer coat of protein normally wrapping around the core genetic material of the virus. This protein was found, by itself, floating in prodigious quantities within the blood of people suffering long-term infection with the virus.

The team's collective imagination stirred and the idea of trying to produce a vaccine crystallized. Their technical problem was how to produce this S-antigen as they now called it, in a form suitable for manufacturing a vaccine. Because it formed naturally into discrete spheres, they found it simple to separate the protein from the liquid portion of blood, the plasma, by spinning the plasma in a centrifuge. Plasma was obtained from suitable donors at blood transfusion clinics, where the clinicians needed to detect this virus to improve infection control. Early experiments proved the S-antigen produced strong antibody responses that seemed to be protective: the antibody could kill invading virus.

Hepatitis B was then becoming rife amongst gay men in New York City and the first clinical trial was held there amongst those men who volunteered for a shot of either a placebo of saline solution or a shot of what was becoming known as the plasma vaccine. The trial was such a success that the job of manufacturing the S-antigen into a form safe and efficacious for registration and sale needed to be handed over to commercial vaccine makers. At the Merck & Company laboratories in New Jersey the formidable vaccinologist Maurice R. Hilleman rolled up his sleeves and took charge of the project, military fashion as he had learnt whilst at the Walter Reed laboratories. His only source of the S-antigen was plasma from blood donors. People infected chronically with hepatitis B virus, and maybe other microbes. Those were still the innocent days before knowledge of human immunodeficiency virus, or of prions as the cause of brain disintegration, were part of routine medical business. Hilleman's stringently severe purification and sterilization process would have killed the agent of AIDS, but prion infections are almost impossible to avoid in such a setting.

The revolutionary ability to synthesize proteins, in bulk, using knowledge of their genetic coding along the DNA molecule was a rare gift of good timing. The recombinant protein version of hepatitis B vaccine became available just a few years after recombinant insulin. That vaccine was the first synthetically manufactured vaccine registered for humans. One of the firms that manufactured it, now known as GlaxoSmithKline, was an obvious partner to produce recombinant proteins as experimental malaria vaccines in sufficient quantity and quality for large clinical trials. Joe Cohen worked for them and was quick to see the potential of using the self-enveloping S-antigen as a carrier for the proteins of a malaria vaccine, as hopefully a stimulant to immunity, and at the least a collateral vaccine providing protection against the virus of hepatitis B. The entire hybrid protein, the RTS, is grown in ordinary yeast cells as a single recombinant protein, whilst the same cells churn out a larger proportion of the plain S-antigen. From the first recombinant circumsporozoite protein of James Young and colleagues to the proof-of-concept trials with Joe Cohen's vaccine took 12 years. Finally, about 1996 full clinical trials could start.

This sporozoite-vaccine was a very different proposition compared to the recombinant hepatitis vaccine. Early versions of recombinant proteins to mimic parts of the circumsporozoite protein induced modest immune responses in the form of antibodies and variable, fickle activity of T-lymphocytes. The efficacy as

vaccines, measured in small trials, were in the zero to twenty percent range. What effect they did offer was enduringly difficult to explain. How much was antibody able to neutralize the progress of sporozoites on their journey through the skin, blood and liver sinusoids before they penetrated hepatocyte liver cell? When in hepatocytes how vulnerable was the schizont stage to T-lymphocytes and macrophages sensitized to the circumsporozoite protein?

Such questions were asked of what happens deep within the body of a human – the only place where *Plasmodium falciparum* lives the full mammalian segment of its life cycle and the only place where this type of vaccine may stop it. But if all you have, as a window onto what is happening amongst that intricate immunity, is a small tube of blood drawn from an arm vein, then you face severe technical problems. There is far too little that can be done with the serum, lymphocytes and macrophages isolated from the whole body to answer such questions. Human immunity against parasites specific for humans works within the bodies of humans. The ability to replicate and experiment with parts of it in plastic dishes is as limited as studying the behaviour of wolves caged in a zoo. The options for rational design are bleakly sparse: try it and see remains the main way forward. At the very least make sure the vaccine is safe; hence the need for the vast body of in-house expertise that follows along the route starting with the vaccine against hepatitis B.

Adjuvants are there to ease progress; literally assistants, stimulators of more powerful action. An adjuvant for a vaccine will both carry the antigens and create conditions to induce stronger immunity than if the antigens are delivered alone. For the current version of sporozoite-vaccine there is an oily component that encapsulates the protein particles and slowly releases the active component of the vaccine. There is a material that mimics the outer cell wall of an invading bacterium to stimulate generally a frenzy of warning signals from the sentinel cells of immunity. Another component, derived from the soap-bark tree, helps to present the vaccine components in a physical form more potently presentable to the antigen-processing cells. Precisely how the adjuvant mixture is formulated is routinely a proprietary secret and there is little evidence of deep understanding of how such adjuvants work. More art and craft than rational technique surrounds the precision of the hybrid protein at the heart of this vaccine, like a jewel wrapped in newspaper.

Progress, nevertheless, is made. An international consortium of funders of malaria vaccines in 2004 agreed strategic goals. By 2015: 'to develop and license a first-generation malaria vaccine that has a protective efficacy of more than 50% against severe disease and death and lasts longer than one year'. For 2025 the goal is 'for 80% efficacy against clinical disease and lasting longer than four years'. By 2011 the efficacy against severe disease over one year was forty five percent. However, by the time the vaccine, named ‘Mosquirix’, was approved inJuly 2015 for release and normal clinical use, its established efficacy was less than earlier studies had indicated. Funding has come so far from philanthropists – notably the Bill and Melinda Gates Foundation, international aid agencies, private research charities, military departments, funders of university research, and cross-subsidy within pharmaceutical companies. Many researchers have conducted these trials widely,

the one listed here for 2011 in *Sources and notes* involved eleven institutes spread over Burkina Faso, Gabon, Ghana, Kenya, Tanzania, Malawi and Mozambique. Most importantly for the long term future of such a vaccine is support from both the ordinary people and the governments in the endemic countries, for without their willingness to find and create the wealth eventually to pay fully for the vaccine then all this effort will not be sustainable long enough to keep a foe like *Plasmodium falciparum* at bay.

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It is easy to find quotes from influential people about their plans and hopes to eradicate malaria. At best this is the management technique of positive thinking, at worst it is ignorance of the heavy burden of implications that the word eradicate carries. Since the international consensus strategy is for eighty percent reduction of clinical disease, amongst those people who receive the vaccine, there will remain an important proportion of people continually re-infected with threatening numbers of plasmodia into the foreseeable future. Those people will form a reservoir of infection to other people. The understated sub-text of such plans and reviews of research contains an acceptance that this technical fix will contribute to reducing the burden of the disease called falciparum-malaria suffered by people in the endemic countries. Of course, it is expected that the vaccine will be improved, there are many other types that can be incorporated but they are delayed by lack of funds. Moreover, the disease referred to is that variety caused by *Plasmodium falciparum*; the more widely spread and less virulent malarias caused by *Plasmodium vivax* and the other two species infecting humans are rarely mentioned.

The eradication of the virus of smallpox from Earth was feasible for the heroes of that campaign because the virus infects only humans, it is transmitted only by close contact, the killed whole virus delivered as a simple vaccine gave full protection with one shot, and that shot could be delivered by something as simple as a forked needle. Natural survival from a mild infection of smallpox gives life-long immunity: the vaccine mimicked that. The logistics of vaccinating the majority of humanity were colossal, but eased by the simple biology of the disease that affected rich and poor people alike throughout the world. The vaccine to protect against the virus of hepatitis B, given the separately available and revolutionary technology of recombinant protein production, went from concept to licensing and widespread sale in about ten years. The difference from a malaria vaccine is in the biology at the heart of the matter. The peculiarly amenable nature of that hepatitis virus, with its detached S-antigen floating in human blood, could not have been easier to isolate as the original plasma vaccine.

Business people and soldiers going into regions plagued with *Plasmodium falciparum* will be able to afford the full commercial price of a vaccine that is sustainably available. The cost of a drug or vaccine relates to the cost of invention, development, testing and registration: over one billion dollars before a cent of profit has been made. No profits – no companies manufacturing. If the vaccine is only eighty percent effective against acquiring symptoms of the disease, these

customers also will purchase insect repellents, rooms in good hotels and a bednet for the weekend safari. Most of the people at daily and yearly danger from plasmodia find it cruelly hard to decide between the price of bednets for the family or fees for sending their three children to school for a year.

Malaria is a disease arising from poverty whilst malaria increases poverty. This circular relationship has been argued over since the early days of the League of Nations planned anti-malaria campaigns and continues with studies of the economics of the disease, without conclusion about which comes first. As with the conundrum of which came first: the chicken or the egg, an evolutionary perspective helps because eggs evolved many millions of years before chickens.

Parasites like plasmodia evolved in close synchrony with the evolution of blood feeding insects. One of the hosts of blood feeding insects and early plasmodia were monkeys and apes living in warm wet forests. Their living conditions were not poor, since poverty has meaning only in human civilization. But these primate animals got plasmodia into their skin many times each year from biting mosquitoes. The plasmodia thrived where they could pass from animal to animal frequently enough to both flourish in non-immune young livers and blood. All it took for the cycle to keep rolling was sufficient stagnant pools of water for mosquito breeding and easy access to more animals exposed at night as they slept in the tree canopy. The mosquitoes thrived where it was green, humid and warm for sufficient of the year. These primates included our evolutionary ancestors.

The combination of fundamental biological conditions of the focus of *Plasmodium*, *Anopheles* and *Homo*, is known as a nidus, a nest. As an ecologically defined but physical entity, this nidus probably developed at the same time that humans developed as a distinct species – two million years ago. These ecological conditions were little different from the conditions of that variety of human poverty still now found in slums and remote villages in the humid tropics. One dollar per day poverty, scratching at the earth for a living, sheltering in flimsy decrepit houses. Such severe poverty initially increases risk of malaria in the life of any human individual reared in an endemic area. Later, malaria worsens that poverty by weakening vitality: a trap of strong positive feedback.

Will a successful vaccine against malaria help to build better houses, or if not houses then generate the cash to buy bednets? Will it help to manage water sources better? Yes: it will, a little bit, eventually. Should the money that has been spent and will continue to be spent, counting by the billion, on a vaccine be used instead for a combination of better land tenure, property rights and subsidized house construction materials? No: because the world of humanity does not work that way and is unlikely to change in that direction anytime soon. Research and development through to licensing and manufacture of a vaccine against malaria will proceed because it can be done. It will proceed because the ability to provide the wealth and energy can be focused on this singular technical fix. How the vaccine works might never be fully understood, but during that vast endeavour the workers throughout can imagine a small vial of the vaccine material and a syringe in the hands of a nurse. No more T-cell subsets, no more double blind clinical trials, taking of

copious records and ponderous analysis of results. Boxes labelled ‘Malaria Vaccine’, regulated, licensed, internationally approved and supported, shipped out to the endemic regions, or manufactured there.

Then what? Success for a malaria vaccine at the level of the vaccines that truly eradicated the viruses of smallpox of humans and rinderpest of cattle, will not come for any malaria vaccine unless the vaccine can beat the highly flexible and constantly adaptable evasiveness of plasmodia. But hopes and plans for such a cat and mouse chase through the maze of the human immunity miss the context.

‘The way the world works’ is the context. Eliminate poverty as a more direct, thus quicker and cheaper way of eliminating *Plasmodium falciparum*? Reduce relative poverty – possibly, but the prospects are slim. The highly skewed distribution of wealth amongst people was described long ago by the economist Alfredo Pareto in the first formulation of what is now often known as the 80:20 rule. In his home of Italy eighty percent of wealth was in the hands of twenty percent of the people. This type of non-linear distribution, described by power laws, has been found everywhere: from the numbers of times researchers cite the publications of other researchers, to the numbers of intestinal worms that infest individual people in a population. The rule is: most have little, a few have lots. How this distribution comes about is obscure but may relate to flows of energy.

How about reducing absolute poverty? This has been done in terms of available material wealth accessible to everybody and measured on a timescale appropriate to the patterns of malaria infection. Most of humanity used to be at threat from malaria caused by one or more species of *Plasmodium*. The most widespread, *Plasmodium vivax*, was the agent of malaria from Sweden to Argentina, from Canada to Australia via Madagascar. Now this type of malaria is rare in many such countries because of a sequence of agricultural intensification, better housing mostly in cities enabled by the better agriculture and industrialization, taxes to pay for public health controls of mosquitoes, and invention and deployment of cheap insecticides and prophylactic drugs. Vivax malaria was eliminated (short of eradication) from the USA by these various methods and influences without there being any national campaign. As the country intensified its agriculture, industrialized and thereby gained enormous material wealth, so malaria both retreated passively and was actively driven out by many local efforts.

The discrepancy between what can be achieved in reduction of human suffering from malaria and the hopes and expectations placed upon singular technical fixes like drugs, insecticides, drainage, bednets and a vaccine derives from a category error. Human malaria is a small component of a vast conglomerate that operates at a level of complexity that so far defies human comprehension of its workings. How does human wealth get distributed between people? Try asking an economist; then turn to a sociologist, ecologist, politician or historian. To ask why poor people are poor is to hold up a mirror to ourselves that reflects a disconcerting answer.

Any single technical fix can perturb the system. An integrated package of fixes can, through its synergies, perturb the system more strongly. The long term results

of such perturbations are almost impossible to predict because the dynamics of the system operate at the same category of complexity as the weather. The system contains fundamental phenomena and laws of biology currently without explanation of their workings. What can be achieved in a laboratory or a drug factory is a level that usually seems miraculous when viewed in isolation. For any such miracle to work outside the laboratory, in a world where the people who need the cure most are those least able to buy the cure, is a problem in another realm altogether. Sometimes there are technical fixes that transcend these levels, a leap from one category to another, like another smallpox vaccine or penicillin. What has so far kept the technical fix of a malaria vaccine from a transcendental role is the dreadful and beguiling biological contrariness of a tiny single celled invader of our blood.

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