

# A conversation with Micah Manary on August 29 and September 30, 2014

## Participants

- Micah Manary – MD/PhD Student, University of California, San Diego (scientific advisor to GiveWell)
- Holden Karnofsky – Co-Founder, GiveWell

**Note:** This set of notes was compiled by the Open Philanthropy Project and gives an overview of the major points made by Mr. Manary.

## Summary

GiveWell spoke to Mr. Manary as part of an Open Philanthropy Project investigation of Research and Development (R&D) for malaria treatment, prevention, and control. Conversation topics included vaccine development, diagnostics, and vector control, but focused most on drug discovery and development.

## Malaria life cycle

Malaria is a mosquito-borne infection in humans caused by a parasite known as "plasmodium." For an introduction to the life cycle of malaria, see:

1. This video: [https://www.youtube.com/watch?v=qMNmOsl5\\_e4](https://www.youtube.com/watch?v=qMNmOsl5_e4)
2. This page: <http://www.niaid.nih.gov/topics/Malaria/Pages/lifecycle.aspx>

Some key points regarding the malaria life cycle are:

- When a mosquito bites a human, it injects saliva. If the mosquito carries the plasmodium parasite, the saliva can carry plasmodium cells known as "sporozoites" into the human. These sporozoites travel to the liver, where they spend a couple weeks reproducing asexually.
  - This is important because salivary glands from mosquitos carrying plasmodium are often removed from mosquitos during dissection in order to be used to infect human cells for purposes of testing possible anti-malarial drugs.
  - The liver is an interesting target for drug and vaccine development because most drugs currently work only in the blood. If plasmodium could be killed in the liver, it might eventually help with eradication.
  - Finally, one plan for developing a vaccine against mosquitos involves injecting frozen sporozoites into the bloodstream.
- After growing in number and differentiating, plasmodium cells travel from the liver to the blood stream.

- These cells produce malaria symptoms by rupturing red blood cells and releasing toxins into the blood stream.
- In the blood, reproductive cells that come in both male and female forms (known as "gametocytes") develop. This happens well into the infection of a human, usually when symptoms are already showing.
- In order for plasmodium to transmit from a person to a mosquito and then to another person, a mosquito must to draw both male and female gametocytes from someone's blood, grow new sporozoites, and then inject its sporozoites into someone else.
  - This is important because one idea for drug and vaccine development is to find a drug that prevents either male or female gametocytes from growing. If successful, this would prevent the transmission of malaria from the person treated with the drug to other people.

## Overview of options for malaria treatment, prevention, and control

The following categories of tools for fighting malaria are potential targets for research and development:

- **Diagnostics** can be used to test blood samples for the presence of plasmodium in red blood cells, so that we can respond with other tools listed below. The most reliable diagnostic technique involves examining blood cells under a microscope, though rapid diagnostic tests (RDTs) are also effective. RDTs are less reliable, but require less skill and lab equipment to use.
- **Drugs** can be administered once malaria cases are identified. As mentioned above, drugs can kill plasmodium while in the blood, but researchers have not yet developed drugs capable of killing plasmodium while in the liver.
- **Vector control**, such as insecticide-spraying or insecticide-treated nets (ITNs), can prevent mosquitos from delivering sporozoites to humans, thereby preventing infections.
- **Vaccines** can enable the human immune system to create antibodies that respond to plasmodium. Currently, the most effective vaccine is RTS,S. RTS,S is roughly 50% effective for 18 months. In Mr. Manary's view, RTS,S is not yet effective enough to be a strong alternative to ITNs.

Plasmodium is a complex organism in comparison with bacterial diseases such as tuberculosis or viral diseases such as HIV. One comparative indicator of this complexity is that HIV has only about nine genes, whereas plasmodium has approximately 5,000 genes. This complexity makes malaria hard to understand and poses special challenges for research and development. For example:

- Plasmodium has more redundant genes in the sense that any one protein is less likely to be essential for the functioning of the plasmodium than any one protein is for HIV. This means that finding a way of making

antibodies that successfully bind to a plasmodium protein is less likely to reliably kill plasmodium, tag it for destruction by the immune system, or otherwise neutralize it.

- In addition, it is easier for drug resistance to develop because plasmodium can mutate substantially and still be viable. New malaria drugs only last a few years before becoming ineffective due to resistance. So drug developers must keep a steady stream of new antimalarial drugs in the development pipeline.
- The complexity and redundancy of plasmodium can also disrupt the available diagnostic tools. RDTs test for the presence of specific proteins from plasmodium, but some strains of plasmodium no longer carry the proteins that RDTs screen for. Thus, RDTs are beginning to fail to detect malaria. This would be significantly less likely to happen if, for example, the RDT was testing for one of the small number of proteins associated with genes in HIV.

Mr. Manary estimates that aggregate funding for malaria R&D from the National Institutes of Health (NIH) and the Gates Foundation (which provide the bulk of funding to improve these tools for fighting malaria) is under \$10 million per year.

## **Malaria drugs and drug development**

### **Drug resistance as the main challenge for malaria drug developers**

Malaria quickly and consistently develops resistance to new antimalarial drugs, in large part due to the complexity and redundancy in plasmodium discussed above. This process takes several years, but it also takes several years to develop, test, and deliver a new antimalarial drug. The central challenge for malaria drug developers is to keep new malaria drugs flowing through the development pipeline quickly enough to keep up with drug resistance.

Malaria drug resistance almost always starts in Southeast Asia. One explanation of this is that:

1. A lot of Malaria drugs are widely used in Southeast Asia.
2. Many drugs are given as monotherapies (rather than more effective combination therapies) because it is cheaper and the monotherapies are made in Southeast Asia.
3. Because these monotherapies are less effective, parasites in Southeast Asia encounter many drugs, but the drugs don't always kill them.
4. If a drug is unsuccessful in killing malaria when used, that tends to build drug resistance.

In Africa, drugs are not used as frequently to treat malaria, and resistance takes longer to develop.

Due to increasing drug resistance, the effectiveness of malaria treatments peaked in 2011 and has since been declining. Resistance to artemisinin (a highly effective antimalarial drug) was first reported in Southeast Asia in 2008. Artemisinin combination therapies (ACTs)—which combine artemisinin with other antimalarial drugs—are now the standard recommended treatment for malaria. There are approximately 30 ACTs on the market. ACTs are more effective than artemisinin alone, and it is harder to develop resistance to them. However, resistance to ACTs was first reported in 2012, again in Southeast Asia. Other drugs need to be ready when resistance to ACTs becomes more widespread.

The time from drug introduction to the development of drug resistance has been decreasing over time, likely because the number of people in the world being treated for malaria has been increasing.

### **The process for replacing drugs when resistance develops**

In order to become a frontline antimalarial treatment and replace ACTs, a drug must pass through the following stages:

1. **Pre-clinical work (including drug discovery and animal trials)** – Large libraries of chemical compounds are screened in order to identify compounds capable of killing malaria without damaging human cells. A small number of the most successful compounds are selected as drug candidates and tested in mice and then chimpanzees to see if they can kill malaria without overly adverse side effects. When a drug is successful in mice, it can almost always be made to work in chimpanzees. The Gates Foundation funds most of this work. Mr. Manary anticipates that it will take several years to get the next drug candidate through this process. These notes discuss this part of the process at greater length below.
2. **Human trials** – Drugs must pass through three phases of clinical trials in order to ensure that they are safe and effective in humans. This takes 4-5 years. For malaria drugs (though not necessarily for drugs treating other conditions), in almost every case where a drug is successful in chimpanzee trials, it is successful in human trials. Drug companies fund most of this kind of work.
3. **World Health Organization (WHO) recommendation** – Drugs that reach the market must be used for a while and then pass through different stages of recommendation from the WHO ("a recommended treatment," "the recommended treatment," and "the primary recommended treatment"). This process can take years.

Drugs currently under development that could plausibly replace ACTs as resistance develops include:

- KD707

- KAE609
- KAF156
- OZ439

Other anti-malaria drugs are under development, but they are, in Mr. Manary's view, unlikely to become frontline treatments.

*When will future generations of drugs be needed?*

Though resistance to ACTs is developing, other drugs are on the way. KD707 recently finished its phase III trials, and should soon be on the market. The other drugs will reach markets years later, and it's likely that resistance to them will develop after resistance to KD707 develops. KAE609 should be introduced in 2-3 years and become a frontline treatment in about 8 years. Resistance is likely to develop 3-5 years after it is introduced, which is much faster than resistance to artemisinin developed. KAF156 is a separate drug that works the same way as KAE609, so resistance will probably develop around the same time.

After that, the next likely candidate is OZ439. It is currently in phase II trials. Assuming it passes through human trials—as seems likely—it will reach markets in 4-5 years. After that, it will probably take roughly 5 years for resistance to OZ439 to develop.

*The expected timeframe for new drug development*

After OZ439, there isn't a likely candidate for the next frontline antimalarial, so it is hard to estimate when it will be developed.

Because the drug screening process is essentially random, the time to identify a candidate ready for trials in chimpanzees is likely to be roughly a linear function of the rate at which new compounds are screened. We're currently screening 250,000-500,000 compounds per year. Roughly speaking, about 1 compound in 250,000 screened would be suitable as a malaria drug, and about 1 in 5 of those are sufficiently different from existing treatments to be useful as new drugs. So roughly 1.25 million compounds need to be screen in order to find a useful new malaria drug. At this rate, researchers should find a useful new antimalarial drug in the next 3-5 years.

However, Mr. Manary anticipates that the rate at which compounds are screened will slow down because the labs screening compounds are not getting enough new libraries of compounds to screen. He expects that only about 500,000 compounds will be screened over the next three years, and that it will take 8-10 years to find a malaria drug that could be the successor of OZ439. It will then take an additional 4-5 years for this drug to pass through clinical trials. So Mr. Manary estimates that researchers are 12-15 years away from having a successor to OZ439 on the market.

**Pre-clinical work in greater detail**

The key steps in drug discovery and screening include:

1. Acquiring plasmodium, usually by importing mosquitos carrying malaria from Peru.
2. Preparing many samples of blood or liver cells infected with malaria. This requires infecting human cells with malaria, usually by dissecting mosquitos carrying malaria and inserting their salivary glands (which carry sporozoites) into populations of human cells in petri dishes.
3. Testing hundreds of thousands of compounds on human cells infected with malaria to determine which can kill the malaria without killing the human cells. Theoretical considerations provide little basis for distinguishing some of these compounds as significantly more likely to work as malaria drugs than others. So large quantities of essentially random compounds must be tested.
4. Iteratively making many chemical variations on compounds that do well at the stage above, screening them for effectiveness, chemically varying the more successful compounds, and screening them for effectiveness.
5. Testing the compounds that come out of this process against flasks of malaria to determine appropriate dosage for tests in mice.

As stated above, laboratories currently screen about 250,000-500,000 compounds per year. This relies on \$25 million of capital, and costs a couple million dollars per year. There are 15 labs in the US and 5 labs in Australia that develop drugs for malaria treatment. The Gates Foundation, Novartis, and Glaxo-Smith-Kline (GSK) are the primary funders for this work. Mr. Manary's impression is that the Gates Foundation provides more than 50%, but less than 75%, of this funding. Usually, an individual lab does not receive funding from more than one drug company.

The libraries of chemical compounds used for testing come from:

- Nineteenth-century German textile factories and their dyes. The modern pharmaceutical industry has its roots in chemical techniques that these factories used to produce dyes, and many of the compounds we screen were first collected, created, and organized during this period.
- Novartis's library of approximately a million compounds used for cancer testing, which can also be used for malaria drug screening.
- Medicines for Malaria Venture's (MMV) large private portfolio of compounds produced in Japan and South Africa.

The same compounds are often screened when developing drugs for other diseases. These libraries contain the range of reasonably random compounds that could be used for screening.

In still greater detail, a typical drug screening and development process might proceed as follows:

1. Many samples of infected blood cells are prepared in test tubes. A very large number of compounds are tested by seeing if adding the compound to the well kills the malaria.
2. For samples corresponding to about 100 compounds that pass this first test, a chemist will make 20 variations on each.
3. The resulting 2,000 compounds are tested on blood samples for whether the samples have (a) all live parasites gone and (b) all red blood cells intact.
4. About 10 compounds satisfy both (a) and (b). Usually, these are from one family of drugs that shares a common chemical structure (aka a "scaffold").
5. A chemist makes 100 variations on that scaffold. These 100 compounds are tested against larger amounts of malaria in flasks, such as 10 mL.
6. Of these, the 10 that clear the malaria the fastest and most thoroughly are identified.
7. These compounds are tested at various levels of concentration against flasks of malaria to identify appropriate levels of concentration.
8. These 10 or so compounds, at appropriate levels of concentration, are tested in mice.
9. The three or so compounds that perform the best in mice trials are identified.
10. A chemist makes 20-100 variations on these three.
11. Among these, drug developers optimize for a variety of criteria, such as toxicity to humans, effectiveness at killing malaria, solubility, and patentability.
12. When a compound is effective against plasmodium in mice, it is nearly always possible to eventually get a version of the compound to work in chimpanzees, and then the drug nearly always passes human trials. Mr. Manary has never heard of a malaria drug that reached trials in chimpanzees but was never approved for use with humans.

At each stage, a compound can pass the screening for a variety of reasons, not all related to the effectiveness of the compound as a malaria drug (e.g. placement of sample on the plate, if a plate was spun in a different direction). This is one reason that many compounds that are not effective malaria drugs pass through early stages of the screening process.

At Mr. Manary's lab, the Gates Foundation funds the work until step 9, when the three or so compounds that work best in mice are identified. Drug companies usually fund subsequent research. At Mr. Manary's lab, this work is typically funded by Novartis.

When testing promising compounds, drug companies sometimes also test similar "open source" drugs, such as cancer drugs that are no longer under patent. Head-to-head comparisons of the drug company's drug and the open source alternatives

sometimes show that both drugs work well. The drug company will do such tests because:

- Ethical guidelines require testing a drug in the development pipeline against the most similar alternative treatment.
- When the patent on the drug under development runs out in 7-14 years, early experience with the open source alternative can lead to an advantage in working with the open source alternative.

Sometimes, drug companies will seek to find ways to extend the patents on drugs they develop beyond the initial 7-14 year period on grounds that can be detrimental to treatment, e.g. by patenting the use of the original compound in babies. In this example, the patent extension would be based on limited innovation and reward the drug company at the expense of consumers.

### **Historical contributions of basic science to malaria drug development**

Major contributions of fundamental science to malaria drug discovery and development include:

- Gene regulation, especially gene regulation of large cellular processes. Specific genes in human DNA are not used to make proteins in all cells all of the time, and "gene regulation" is the general term for the set of processes that determine which genes are expressed in which cells at which times. In the past year, people have used this knowledge to develop compounds that might prevent plasmodium from producing gametocytes, which may eventually help to create drugs or vaccines that prevent the transmission of malaria.
- The ability to grow human liver cells in the lab, combined with the ability to make severe combined immunodeficiency (SCID) mice, has allowed for more realistic tests of malaria treatments in mice. Ordinary mice are often used as model organisms in biology and medicine, but they are imperfect models of humans because they have different cells and tissues. If human cells and tissues were introduced into the body of an ordinary mouse, the mouse's immune system would attack those cells and tissues. SCID mice have a rare recessive mutation that compromises their immune system, making it possible for scientists to grow human livers and bone marrow inside of them. These mice are therefore better proxies for humans in drug development and other biomedical research.
- Basic science work on sea urchins and *c. elegans* has contributed to the understanding of malaria metabolism, contributing to the development of two of the most promising antimalarial drug candidates (OZ439 and KAE609).
- The ability to make liver cells in the lab and to make those cells smaller has made screening of drugs targeting liver-stage malaria more efficient. In order to infect liver cells with human malaria, mosquitos are imported



from Peru and brought to research labs. The mosquitos are dissected and their salivary glands are used to infect liver cells. The smaller the liver cells, the smaller the number of mosquitos that need to be dissected.

### **Options for funding malaria drug development**

A funder could speed up malaria drug development by paying for equipment and people to screen more compounds. To screen an additional 300,000-400,000 compounds per year would require expenditures of roughly:

- A one-time cost of \$5 million for machinery
- Annual costs of:
  - Five full-time researchers at \$50,000-\$100,000 per year each
  - Five senior part-time researchers, each with a full-time-equivalent salary of >\$100,000 per year each
  - \$500,000 per year for maintenance of machines
  - \$1 million for overhead for joining an existing research institution

Mr. Manary has a contact that is starting a lab dedicated to malaria drug development under the California Institute for Biomedical Research (CALIBR).

A funder who wanted to improve the general process of drug discovery and development could support:

- **Efforts to figure out how to get mosquitos to more effectively draw blood through plastic film in a lab setting in order to make it easier to get malaria samples.** In order to get samples of malaria for drug screening, researchers often import mosquitos from Peru. It would be more efficient if, instead, they could simply get mosquitos to draw blood from petri dishes covered with a thin plastic film in a lab setting. However, researchers currently have difficulty making mosquitos attracted to such blood samples. Researchers could investigate what attracts mosquitos to blood (e.g. smell, CO<sub>2</sub> gradient, etc.) and determine how thin the plastic film should be. Very little work has been done on this problem, and, for a few hundred thousand dollars, it's very likely that someone could solve it. Success might increase yield from its current range of 1-5% to perhaps as high as 50%, meaning only 10% as many mosquitos would be required for drug screening. Mr. Manary roughly estimates that this would increase throughput of the drug screening process by a factor of three or so.
- **Research related to protein small target modeling.** Drugs are often small molecules that bind to larger proteins. If scientists know how a drug candidate could bind to a protein, it can help with drug development. More work could be done to better answer questions like, "Given a protein with a known structure and a small molecule, how can

we determine where the small molecule could bind with the protein?" This is a basic research question with many other possible applications.

- **Efforts to grow artificial livers in a lab in order to make drug screening easier and more realistic.** To test drugs that target the liver, drug developers need to infect liver cells with malaria. Mosquitos can't directly bite liver cells to infect them, so drug developers infect liver cells by dissecting mosquitos, taking their salivary glands out, and putting the salivary glands in liver cells. But infecting liver cells this way is laborious and imprecise, with a yield on the order of 0.1%. It also may not be analogous to just putting sporozoites into the liver, since other parts of the mosquito often get into samples of liver cells as well. If scientists could grow functional livers and connect them to a bloodstream, mosquitos could infect liver cells with malaria just by drawing blood, streamlining the drug discovery and development process and making screening more realistic.
- **Research related to sex-determination in plasmodium in order to help develop drugs that would prevent the transmission of malaria.** In order for malaria to transmit from a person to a mosquito to another person, a mosquito needs to ingest malaria gametocytes of both sexes from human blood. Otherwise, sexual reproduction can't take place inside of the new mosquito, and that mosquito cannot deliver sporozoites to other people. If a drug could affect sex-determination so that malaria could only produce one sex of gametocytes (or produce one sex in substantially decreased abundance), it would decrease the spread of malaria. Which genes determine sex in plasmodium is unknown, and knowing this could help with developing such a drug. Researchers could seek to identify genes that are active when and only when plasmodium is becoming sexualized. Knowledge of one such gene could help to identify other genes important in this process, and then identify targets for drug development. Mr. Manary knows of two labs that are working on this problem. In Mr. Manary's view, a couple million dollars in grants might significantly increase progress toward this goal.
- **Basic genetic research on malaria, such as long-range sequencing and structural genetics.** Currently, almost all genetic research is done in humans and adapted to other organisms. Genetic knowledge might be useful in drug development, as it could be in the case of understanding how plasmodium is sexualized.
- **Expanding the services of MR4.** The Malaria Research and Reference Reagent Resource Center (MR4) is a database for malaria biosamples (such as mosquitos, nucleic acids, plasmodium, and monoclonal antibodies) that supports researchers working on malaria drug discovery and screening. Additional funding could help MR4 to carry more strains of plasmodium or antibodies. Mr. Manary's understanding is that MR4 only has two staff.

### **Other potential upsides of drug development**

It's possible that a new antimalarial drug could have a variety of good consequences above and beyond replacing current antimalarials as resistance develops. This could happen if:

- **The gene the drug targets is very "conserved,"** meaning that the gene occurs in similar forms across different organisms. It is widely thought that mutations to conserved genes lead to non-viable life forms. Therefore, if a drug targets a conserved gene, it would be hard or impossible for parasites to develop resistance to that drug.
- **The drug is very adaptable.** Drugs tend to fit into small "pockets" of larger proteins they bind to. If the pocket changes slightly, the drug may not work anymore. If the drug fits into a larger pocket or a functional part of the protein, it would be harder for parasites to develop resistance to the drug.
- **The drug works with one dose, one time.** This would be an advance over current drugs, which, in the best case, require one dose per day over a period of four days. Whether a drug works by single-exposure or multiple-exposure depends on the half-life of the drug in the blood, how it is metabolized, and how the lethal dose for a human compares with the lethal dose for malaria.
- **The drug prevents the transmission of malaria,** e.g. by preventing the creation of gametocytes.
- **The drug eliminates malaria in the liver.** To eradicate malaria, we'll need to completely remove it from people's bodies. Current drugs target malaria while it is in the blood, but not in the liver. If a drug could target malaria while it is in the liver, it might be helpful for eradication.
- **The drug might not negatively affect people with G6PD deficiency.** G6PD deficiency is a genetic disorder affecting about 500 million people. It results in underproduction of an enzyme that helps break down carbohydrates, as well as some resistance to malaria. Perhaps due to this advantage, G6PD is more common in areas where malaria is common. Antimalarials sometimes—though rarely—have lethal side effects for people with G6PD deficiency. Perhaps 10 million of the 500 million people with G6PD deficiency are at risk of potentially lethal side effects if they use antimalarials. This effect is not fully understood. It is possible that a new antimalarial drug would not adversely affect these people. Perhaps a new antimalarial would not have side effects for these people.

## **Malaria vaccine development**

### **Challenges of malaria vaccine development**

Malaria is a parasite, and it is very challenging to develop vaccines for parasites. Like some other parasites, plasmodium is very complex and many of its genes are redundant in the sense that it can survive without them. In order to be effective, a

vaccine needs to target multiple proteins or a non-redundant protein, which is a challenging task in plasmodium, not least because its genetics is poorly understood.

People develop a natural immune response to plasmodium, but it is relatively weak and happens very gradually. In areas where malaria is common, roughly 1 in 10 mosquitos carry plasmodium, so children in these areas are probably bitten by mosquitos that carry plasmodium very often. These children get malaria very frequently, but they get it less and less often over time as they get older and immunity develops. Eventually, they get it about once per year, and then about once every five years, and then about once every 10-20 years. However, even adults are not fully immune to malaria, and their immunity is not retained if they leave areas where malaria is common for long enough.

Moreover, there are some reasons to expect that it may be challenging to provide children with even the imperfect level of natural immunity observed in adults. Children produce more reticulocytes (immature red blood cells) than adults. When plasmodium infects reticulocytes, it thrives for reasons that are poorly understood. This may contribute to immune responses in children being less successful than immune responses in adults.

Finally, many malaria vaccines target proteins that express randomly, making it hard to develop an immune response. When plasmodium is inside of red blood cells, it exports proteins outside of the red blood cell. It is easier for the immune system to identify these proteins than it is to identify proteins from plasmodium inside of the red blood cell, which is one reason that vaccines targeting these proteins are promising. However—perhaps because of this vulnerability—plasmodium has developed a complex system for randomly expressing 40 different genes (known as "var" genes) that produce these proteins. These 40 genes can be expressed one at a time or in combination. The immune system works by reacting to one of these proteins at a time, and it is challenging for the immune system to keep up with all of these changing and varied immune targets. This may help to explain why it takes so long—with many repeated exposures—to develop natural immunity to malaria.

## **RTS,S**

RTS,S is an experimental vaccine for malaria targeting the proteins exported outside of the red blood cell discussed in the section above. RTS,S is the most effective malaria vaccine available, though it is only effective for 18 months, and only about 50% effective on average over that period as a whole. RTS,S targets the proteins produced by the var genes for immune response, with about a dozen different immune responses intended to cover them all. There was a failed vaccine trial with a similar target in the 1980s.

There are some complicated political issues in the background with RTS,S, including overselling the likely benefits of the vaccine. RTS,S reported 100% protection 6 months into its phase II trial on children aged 18-36 months. A year later,

developers were less forthcoming about the data, perhaps because the protection rate had dropped to 50% for one year, and protection had essentially disappeared after 18 months. Mr. Manary believes that RTS,S will be pushed through Phase III trials, but thinks it is will likely have limited value for public health because:

- Glaxo-Smith-Kline (GSK) is not funding RTS,S.
- If an unsuccessful vaccine with the same target as RTS,S were deployed, it would risk making all other vaccines in this class ineffective due to mutations in the antigens used by the vaccine. This is a general issue with vaccines: if a vaccine rollout doesn't work, all future vaccines in that class may become impossible to use effectively.

Though two vaccines in this class have now failed—which suggests to Mr. Manary that this class of vaccines may be unlikely to yield a successful vaccine—many labs continue to work on this class of vaccine because in vitro testing is unusually easy. (I.e., it is easy to inject SCID mice with human stem cells and get them to respond to this class of proteins.)

It's possible that in order to develop lasting immunity, it would be necessary to inject RTS,S much more frequently, such as once per month for a year. It may be that the frequent reinforcement of the immune response caused by exposure to plasmodium through mosquitos is necessary for the development of natural immunity. If this is true, it's possible that much more frequent exposure to RTS,S would substantially increase its effectiveness, though it would also pose a significant public health challenge to distribute a vaccine so frequently.

## **Sanaria**

Sanaria, a biotechnology firm, has a potentially promising approach that involves freezing entire sporozoites and injecting them so that the body can develop an immune response to sporozoites. Successfully delivering such a vaccine would require distributing liquid nitrogen across Africa or developing a version of the vaccine that doesn't require freezing up until a few minutes before injection. A funder could support research addressing those technical challenges.

## **Process of vaccine development**

Many vaccines, such as the Measles, Mumps, and Rubella (MMR) vaccine, work by injecting a weakened version of the disease into people. While Sanaria has a vaccine strategy that involves injecting frozen sporozoites, other researchers developing malaria vaccines seek to target specific genes of plasmodium. For a target gene X and associated protein X', working through a target for malaria involves:

1. Studying how gene X works and where it is expressed.
2. Asking questions such as:
  - a. Can antibodies to X' be synthesized?

- b. Can a natural antibody response to X' be produced? (A natural antibody is an antibody that is produced without previous exposure to a pathogen.)
  - c. Can a monoclonal antibody to X' be created? (Antibodies are usually produced by many immune cells and vary in shape. Monoclonal antibodies are all produced by copies of a single immune cell and have the same shape. They are produced in labs and injected directly to people, rather than being produced by stimulating the human immune system in the person receiving the treatment.)
3. Searching for a way to get a mouse or a rabbit to make the antibody to X'.
  4. Searching for a substance that would cause a mouse or a rabbit to produce the antibody that worked against X'.

This process takes an entire lab about five years and costs millions of dollars.

Next, the vaccine must be tested in clinical trials. This costs tens or perhaps hundreds of millions of dollars, and takes 4-5 years. Therefore, the whole process takes about 10 years.

### **Current funding for vaccine development**

Most work on malaria vaccines is being done by European labs, which are working on second-best guesses, but not third-best guesses. Funders are generally not enthusiastic about supporting additional work on malaria vaccine development given that the two targets that have been studied have been unsuccessful, and many funders were told that the failure of the first vaccine was a fluke.

### **Immune response in the liver**

An alternative approach for a funder would be to support research into the immune response to plasmodium in the liver. It takes a relatively long time for plasmodium to get through the liver stage—up to two weeks. If immune cells were ready in the liver in advance of plasmodium reaching the liver, the immune response could be greatly improved. Currently, there are no promising ideas for boosting the immune response while plasmodium is in the liver, but promising ideas may be found as research continues.

One strategy would be to make SCID mice, give them malaria, biopsy their livers, and screen every possible immune response against neutral mice. A challenge with this research project would be distinguishing between cases where a plasmodium injection fails to result in an infection and cases where a plasmodium injection results in an infection, but the immune system fights it off. This kind of research is not funded by the Gates Foundation or the NIH, and is more likely to be funded by European groups.

### **Diagnostics: RDTs**

Malaria Rapid Diagnostic Tests (RDTs) are used to detect malaria parasites in human blood. They are easier to use than examining blood smears under a microscope, but less reliable. RDTs used to have a reliability of 99%, but their reliability has fallen down to 90% (due to false negatives) and will likely continue to decrease. Mr. Manary expects RDT insensitivity to be widespread within the next five years.

Most RDTs work by detecting genes in plasmodium called "HRP2" and "HRP3." These genes are not present in increasing numbers of samples of malaria, which is the cause of the decrease diagnostic effectiveness. A promising approach would be to find some RDT targets that are essential, so that it would be impossible for plasmodium to survive if the gene mutates away. Mr. Manary estimates that this work, together with developing the new RDTs, would cost roughly \$500,000 and take 1-2 years.

It is unclear why these RDTs have become ineffective. Typically, resistance to a treatment develops because disease treatment is natural selection against microbes that aren't resistant to a treatment. An RDT is not a treatment, though it tends to cause a treatment. So it might seem that there could be selection against detection by an RDT. However, if HRP2 or HRP3 are detected in a blood sample, the entire population of plasmodium in the human is affected by the decision to treat with an antimalarial. So selection against detection by RDT would have to be selection against being in a population of plasmodium that is expressing HRP2 or HRP3. This kind of selection would be much weaker than standard forms of antimicrobial selection, making it hard to see how this kind of pressure could result in non-expression of HRP2 and HRP3.

### **Vector control: DDT**

DDT is highly effective for malaria vector control, but isn't used for environmental reasons. Rwanda rejected WHO standards for DDT and successfully used DDT to help control their malaria problems.

A funder could:

- Investigate the suitability of these standards and/or advocate for them to change.
- Support the development of variants of DDT that are less toxic to humans or have more limited environmental consequences.

*All Open Philanthropy Project conversations are available at*  
<http://www.givewell.org/conversations>