Towards a Malaria Vaccine – A Discussion Paper for RAM

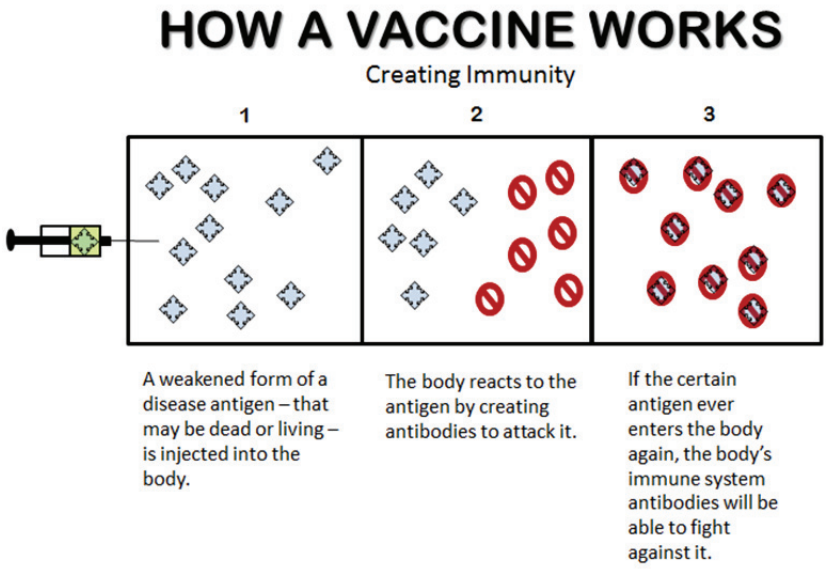
Executive Summary

This Discussion Paper has been prepared with a view to briefing Rotarians on the current state of development of vaccines directed against malaria. In essence, whilst there are some promising vaccine candidates under development, Rotarians Against Malaria (RAM) efforts to eliminate malaria in the countries within which we work (Timor Leste, Solomon Islands & Papua New Guinea) will not enjoy the benefit of a vaccine in the near future and we will continue to rely on traditional approaches such as residual insecticide spraying, long-lasting insecticide-treated bed nets, and the healthy villages program. Within its limited means, it is appropriate for RAM to provide resources to local researchers to undertake basic research, and where appropriate, provide funds to kick-start pre-clinical and Phase-I testing of home-grown vaccine candidates such as the PlasProtect vaccine under development at the Institute for Glycomics at Griffith University and various products being developed at the Walter and Eliza Hall Institute (WEHI).

Preamble (paraphrased in part from https://en.wikipedia.org/wiki/Antigenic_variation)

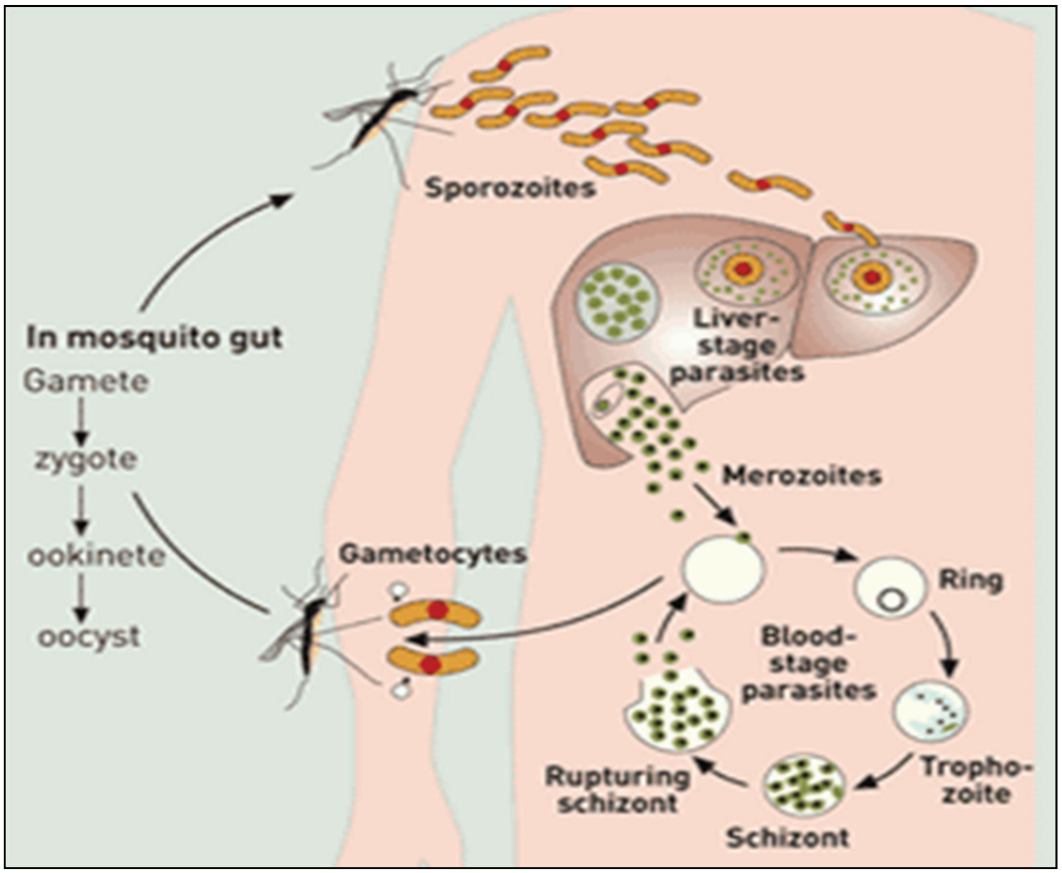
The problem of antigenic variation, and the complex life-cycle of the malarial parasite within both humans and mosquito, has forced the development of vaccine strategies far more complex than those used for traditional virus and bacterial diseases such as polio, hepatitis, measles, whooping cough, diphtheria and tuberculosis.

Antigenic Variation is a mechanism by which infectious agents alter their surface structures (antigens) in order to evade host immune responses. When Humans are exposed to a particular antigen (for example a protein on the surface of a pathogen such as the malaria parasite an immune response is stimulated and antibodies are generated to target that specific antigen. The immune system will then "remember" that particular antigen, and defences aimed at that antigen become part of the immune system's acquired immune response. If the same pathogen tries to re-infect the same host the antibodies will act rapidly to target the pathogen for destruction. A simple vaccination strategy is shown in the diagram below:



However, if the pathogen can alter its surface antigens, it can evade the host's acquired immune system. This will allow the pathogen to re-infect the host while the immune system generates new antibodies to target the newly identified antigen, and then the parasite can simply alter the surface antigens again, always staying one step ahead of the host immune responses.

Plasmodium falciparum (Pf), the major causative agent of human malaria, has a very complex life cycle that occurs in both humans and mosquitoes. A schematic diagram of the life-cycle stages occurring in humans and in the mosquito is shown below (Diagram courtesy of Dr. Danielle Stanisic, Institute of Glycomics, Griffith University):



While in the human host, the parasite spends most of its life cycle within liver cells and red blood cells. In order to survive the parasite must modify the parasite-specific proteins displayed on the surface of the infected cells to prevent those cells from being destroyed by the host immune defences, including antibodies and various white blood cells of the immune system. By way of example, one very important protein in *Plasmodium* species is a protein (PfEMP1) which saves infected red blood cells from being eliminated in the spleen by causing the infected cells to adhere to endothelium surfaces. Endothelium consists of single layers of smooth, thin cells that line the heart, blood vessels, the lymphatic system and the cavities within many bodily organs. The genes controlling production of PfEMP1 number around 60 genes in all, and there are other genetic mechanisms by which even more distinct versions of PfEMP1 can be displayed. The parasite is able to evade host defence mechanisms by changing the gene used to code the PfEMP1 protein to one which is not (yet) recognised by the host. Any candidate vaccine targeting proteins such as PfEMP1 would need to generate immunity in vaccines to all possible forms of PfEMP1 – an impossible task!

Basic Aspects of Malaria Vaccinology

A key limitation for success of malaria vaccines is the difficulty in maintaining durable protection after immunisation, which in part has been ascribed to poorly immunogenic antigens and to the effect of malaria itself in suppressing host responses. Efforts to increase the degree and durability of vaccine protection have included novel adjuvants and platforms as well as altered vaccine schedules, dosages and methods of delivery. However, long-lived protection has not been achieved.

Vaccines Under Development (taken from the report from the 2017 Malaria Vaccine Symposium – see https://www.nature.com/articles/s41541-017-0035-3#Tab1)

The Malaria Vaccine Symposium occurred at Johns Hopkins University in Baltimore, MD, USA on April 25th, 2017, coinciding with World Malaria Day and the WHO announcement that the RTS,S malaria vaccine would begin pilot implementation programs in Ghana, Kenya, and Malawi in 2018. Scientists from several disciplines reported progress on an array of malaria vaccine concepts and product candidates, including:

1) Pre-erythrocytic (red blood cell infection) vaccines that prevent infection,
2) Blood-Stage vaccines that limit infection and disease, and
3) Transmission-blocking vaccines that interrupt the spread of infection.

1) Pre-erythrocytic Vaccines

Pre-erythrocytic vaccines target the sporozoite and liver stages of *Plasmodium*. These vaccines aim to eliminate parasites during early infection and, if highly efficacious (i.e., induce sterilising immunity), will avert disease and interrupt transmission. Whole sporozoite vaccines were shown to provide some protection to humans from *Plasmodium falciparum* in the 1970s, and current whole-organism vaccine approaches include irradiated parasites, genetically-modified parasites, and infection in conjunction with chemoprophylaxis (called chemoprophylaxis vaccination or CVac).

2) Blood Stage Vaccines

Obstacles to blood stage vaccine development including the antigenic variation in both merozoite and infected erythrocyte surface proteins as discussed above in the Preamble. The PlasProtect vaccine under development at Griffith University is an example of a blood stage vaccine involving chemically-attenuated whole blood stage parasites. The chemical attenuation procedure developed at Griffith University entails incubation of blood stage parasites with DNA-binding drugs (Centanamycin or Tafuramycin-A) which affect parasite replication irreversibly.

There are at least two other candidate vaccine strategies being developed to block the invasion of erythrocytes by merozoites and thus prevent blood stage malaria infection.

3) Transmission-Blocking Vaccines (TBVs)

Previous studies have shown that immunity developed within humans can be carried over to act against the sexual stage parasites in the mosquito, and thus prevent onward transmission to new human targets. Antigens (Pfs230 and Pfs25) have been identified which generate immune responses which subsequently interfere with pre- and post-fertilisation processes in the mosquito, and clinical trials have been undertaken. However, the immunogenicity of the antigens is poor and current

research efforts are aimed at improving the immunogenicity by coupling the antigens to immunogenic carrier proteins, or administering them with adjuvants. In another study, a protein (Pfs47) linked to successful infection of mosquitos by malarial parasites has been identified as a potential vaccine candidate. Pfs47 appears to allow the parasite to evade the immune defenses within the mosquito, and antibodies to Pfs47 carried over in the blood after a mosquito bite could possibly be used for transmission blocking.

As outlined above, vaccines aimed at different stages in the *Plasmodium* life cycle are in development, and in the future, successful candidates could be combined together to achieve the greater activity than achievable by individual vaccine candidates alone.

A summary of candidate vaccines under development is provided in Table 1 (below):

Table 1: Current malaria vaccines under preclinical development or in clinical trials

From: Advances in malaria vaccine development: report from the 2017 malaria vaccine symposium

Parasite stage	Vaccine classification	Current status
Pre-erythrocytic stage		
PfSPZ vaccine	Whole organism (radiation attenuation)	Phase II
GAP vaccines	Whole organism (genetic attenuation)	Phase I
RTS,S	Subunit	Phase IV
CVac	Whole organism (chemical attenuation)	Phase I
Blood stage		
Chemically attenuated parasites		
PlasProtect	Whole organism	Phase I*
AMA1-RON2	Subunit	Preclinical
PfRH5	Subunit	Phase I
Mosquito stage (TBVs)		
Pfs25	Subunit	Phase I
Pfs230	Subunit	Phase I
Pfs47	Subunit	Preclinical

*PlasProtect was at the Preclinical stage during the 2017 Malaria Vaccine Symposium but has now advanced to early stage human trials.

Pre-erythrocytic, blood-stage and transmission-blocking vaccines (TBVs) are being evaluated in clinical trials (denoted as Phases I to IV) or are being tested in rodent or non-human primate models (preclinical status).

Recent Research Developments (not an exhaustive list)

A team at Imperial College London in collaboration with a team from the University of Maryland has identified a protein (HAP2) which is involved in the *Plasmodium* fertilisation process between male and female parasites within the mosquito. By blocking a small easily targetable part of the HAP2

protein fertilisation is disrupted and the malarial parasites cannot reproduce efficiently. Disruption of the fertilisation process prevents the parasites from travelling to the salivary glands of the mosquito, and thus reduces the further transmission of parasites to humans.

With the aid of the particle accelerator at the Australian Synchrotron, the team from the Walter and Eliza Hall Institute (WEHI) recently announced that they had determined the structure of a key protein used by *Plasmodium vivax* to infect the youngest of the red blood cells. By generating antibodies to the protein the WEHI have shown that they can block the parasite's ability to hook onto and subsequently infect red blood cells. The recent findings provide a basis for moving forward to the potential development of vaccine strategies for *Plasmodium vivax* in particular.

The Global Context:

The World Health Organisation (WHO) has established a Strategic Advisory Group of Experts (SAGE) on Immunization to provide guidance on the work of WHO. SAGE is the principal advisory group to WHO for vaccines and immunization. It is charged with advising WHO on overall global policies and strategies, ranging from vaccines and technology, research and development, to delivery of immunization and its linkages with other health interventions. SAGE produced a "Malaria Vaccine Technology Roadmap (2006) and published an update in 2013. Further updates will occur every five years. At its most recent meeting in October, 2017, SAGE reported the following in respect of the development of vaccines for malaria:

"*The complexity of the malaria parasite makes development of a malaria vaccine a very difficult task. Recent progress has been made with the completion of a Phase 3 trial of the RTS,S/AS01 candidate vaccine and review by the European Medicines Agency and WHO. There is currently no commercially available malaria vaccine. Over 20 other vaccine constructs are currently being evaluated in clinical trials or are in advanced preclinical development.*"

WHO published the *World Malaria Report 2017* in November drawing on data from 91 countries and areas with ongoing malaria transmission. The 2017 report shows that after an unprecedented period of success in global malaria control, progress has stalled. In 2016, there were an estimated 216 million cases of malaria, an increase of about 5 million cases over 2015. Deaths reached 445 000, a similar number to the previous year.

Recent Announcement from Bill Gates

Rotarians and Rotaractors are hopefully aware of the recent announcement from Bill Gates (https://www.theaustralian.com.au/news/health-science/bill-gates-asks-for-australias-help-in-ridding-asia-of-malaria/news-story/474ade9f0e0f8c84d4033b6b5953476b0) in which he called on Australia to become the regional champion of a drive to eradicate malaria from Asia within 10 years. The Australian government has announced that Foreign Minister Julie Bishop has joined a key global body, the End Malaria Council, co-chaired by Mr Gates, and will continue to invest strongly in health security, including malaria control and elimination, with a focus on increasing impact through partnerships. Mr Gates sees a role for Australia "to be a regional champion of getting rid of malaria" and believes that if Australia leads a concerted regional drive against malaria, the disease could be eradicated from Southeast Asia as early as 2025.

Implications for RAM (Rotarians Against Malaria):

Given the limited financial resources of RAM we cannot presume to be able to fund the development of any candidate vaccine strategy through to clinical development. However, within our means, I believe it is appropriate for us to provide resources to local researchers to undertake

basic research, and where appropriate, provide funds to kick-start pre-clinical and Phase-I testing. A reasonable approach for RAM to adopt in the Australian context would be to maintain a watching brief over local scientific developments at WEHI, the Institute for Glycomics at Griffith University and other research institutes, without attempting to "pick a winner" in terms of likely long term success.

There are varying reports about the effectiveness of the current RTS,S vaccine which has been administered in a pilot implementation project in three African countries. The RTS,S vaccine requires 4 shots which presents logistical difficulties for widespread deployment and follow-up. In another report, protection from the RTS,S vaccine was found to fade after 7 years, and the vaccine achieved an effectiveness level of just 4% in children older than 7 in the phase-II clinical trial in Kenya.

Given doubts about the effectiveness of the RTS,S vaccine in its current form, and given that all other candidate vaccines listed in Table 1 are mostly in pre-clinical or phase 1 status, it is not possible to predict which vaccine development strategy will ultimately result in the most effective vaccine. Further, as noted during the 2017 Malaria Vaccine Symposium, successful candidates could be combined to achieve greater activity than achievable by individual vaccine candidates alone.

The development cost to advance RTS,S to stage IV trials has been huge (many hundreds of million USD$s) and as a result potential vaccine development funding agents such as drug companies, the Global Fund, UNITAID and the Gates Foundation are now reluctant to fund early stage trials until a vaccine candidate has demonstrated strong consistent positive results.

As noted earlier, the candidate PlasProtect vaccine has completed Preclinical trials and is now in early Stage human trials. The current fund-raising is aimed at completing phase-I trials on a larger number of naïve human subjects (volunteers).

In the context of the recent challenge by Bill Gates for malaria to be eliminated from Southeast Asia as early as 2025, and the current state of vaccine development, malaria elimination in Southeast Asia by 2025 will have to be achieved without the benefit of a vaccine, using all the basic RAM strategies that have proved effective in reducing malaria in the countries upon which the RAM effort is focused (Solomon Islands, Papua New Guinea, Timor Leste). Professor Dennis Shanks, recently retired from the Army Malaria Institute in the Department of Defence, believes that it is unlikely we will have vaccines available to make a significant contribution to malaria elimination outside Africa. On the basis of the reading undertaken to prepare this short position paper I agree with the view held by Professor Shanks.

Dr Bruce Anderson
National Scientific Committee Coordinator
January 2018.
*for scientific keynote presentations from the 2017 RAM National Conference, including Professor Shanks' talk, please refer to http://ram.rawcs.com.au/gallery/media-resources/