REVIEW ARTICLE

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VACCINE FOR MALARIA – HOW FAR?

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This is a review of the progress made so far in the effort to produce a malaria vaccine. The problems that have made it impossible to get an effective vaccine for malaria are discussed. Also examined are the current efforts to produce the vaccine and the prospects for an effective vaccine in the future.

Key words: Vaccine, malaria, review.

INTRODUCTION

Vaccines are one of the most cost-effective and logistically feasible means of disease control, and have remarkable success in the control of many infectious diseases [1]. Examples include the eradication of smallpox and the near-eradication of polio [2]. It is surprising that a licensed potent vaccine for malaria has not arrived. The difficulties on the way of production of this vaccine have been identified for a long time but every advance towards the resolution of this problem, like chasing a mirage, has resulted in illusion.

Yet, the increasing resistance of the *Plasmodium* species to chemotherapeutic agents and the increasing resistance of the vectors, *Anopheles* mosquitoes, to insecticides [3] pinpoint the critical need for an effective vaccine against this infection. Although protective immunity is conferred by naturally acquired infection and by irradiated sporozoite immunization, no subunit malaria vaccine candidate so far has proven sufficiently efficacious for commercial development [4].

Hope for a potent malaria vaccine was raised when an international body of genome scientists and funding agencies was formed in 1996 [3]. This hope was on course with the publication of the genomic sequence of *Plasmodium falciparum* in 2002 [5] and simultaneous publication of the *Plasmodium falciparum* proteome representing stage-specific sporozoites, merozoites, trophozoites and gametocytes [6, 7].

Also completed is the gene expression profile of the parasite during the different stages of its life cycle [8]. The hope is that this knowledge will be exploited to identify and prioritize antigens and epitopes that may be targets of anti-malarial protective immunity. However, identification of the most effective epitopes is difficult because the genome of *Plasmodium falciparum* is large.

PROBLEMS HINDERING THE PRODUCTION OF AN EFFECTIVE MALARIA VACCINE

Despite intense research efforts over close to half a century now, which has resulted in clinical trials of many candidate vaccines, few humans have been protected from malaria through vaccination [4]. Unlike in the development of vaccines against bacteria and viruses, developing a vaccine against the malarial parasite is complicated

by the complexity of the parasite and that of the host's response [3].

The identification of antigens that will stimulate the most effective immunity against *Plasmodium* is problematic because of: (a) the multistage parasitic life cycle, (b) a large genome encoding more than 5,300 proteins, and (c) distinct proteins expressed at different stages of the parasite. Added to these problems is the poor understanding of what constitutes the protective immune mechanisms that target the different parasite stages; and sequence polymorphism of identified target epitopes.

It is well established that immunization of humans with radiation-attenuated *Plasmodium* sporozoites confers sterile protective immunity [9-12]. Irradiated sporozoites could invade hepatocytes and undergo limited development but could not mature into blood-stage parasites [13] thus eliminating clinical symptoms of the disease and transmission of the parasite. However, immunization with heat-killed, formalin-inactivated or lysed sporozoites was not effective [3], making sporozoite-based vaccine production difficult. These observations seem to emphasize the requirement for live sporozoites targeting the liver. Even then the targets of cellular immunity, induced by irradiated sporozoites are largely unknown, and correlates of protection after immunization are not clearly elucidated.

A dilemma is suggested by the possibility that the protective immunity induced by irradiated sporozoite immunization is due to the summation of many immune responses of low magnitude against multiple targets which result from low density of epitopes [3]. Thus, responses against characterized hepatic-stage antigens recognized by sporozoite-induced cellular immune responses are not as potent as those induced by subunit vaccination [14].

Repeated natural infections confer immunity against severe infection but this immunity is normally species and strain specific; and it is dependent on continuous boosting and is usually short-lived. A potent vaccine based on this type of natural immunity will then be required to generate "super-natural" immunity [15].

CURRENT APPROACHES TO MALARIA VACCINE DEVELOPMENT

Presently, most candidate malaria vaccines are designed to protect against pre-erythrocytic and/or erythrocytic stage antigens. Transmission blocking vaccines are formulated to protect the entire community by inducing protective antibodies directed at sexual stage antigens [3]. These vaccines have been developed with potency against a single or a few key antigens, such as *Plasmodium falciparum* circumsporozoite protein (CSP) and merozoite surface protein-1 (MSP1), by immunizing with synthetic peptides or recombinant proteins in an adjuvant [15]. Results of trials with these subunit vaccines [16] leave us with the question of whether all the antigens are the same and it is the vaccine delivery system that matters. It is also not clear yet whether key protective antigens have already been identified or not.

Another approach to malaria vaccine development focuses on all of the currently known promising candidate antigens to give rise to a multivalent and multistage vaccine; produced mainly through the technology of DNA-based vaccines [17, 18]. First generation DNA vaccines were found suboptimal but

immune enhancement strategies, such as the use of adjuvants, show promise [19-21]. This approach to vaccine production faces limitations regarding the size of antigens that can be included in a given vaccine delivery system and the number that can be formulated for simultaneous administration without inducing antigenic competition.

Despite the challenges facing malaria vaccine production, there are lines of evidence showing that a vaccine is feasible. These include the age-related acquisition of immunity against severe clinical malaria in endemic regions [22, 23] and the ability of passively transferred antibodies from immune adults to protect against natural and challenge infections with *Plasmodium falciparum* [24-26]. Blood-stage vaccines against the parasite are aimed at preventing complications of the infection, such as cerebral malaria and anaemia.

Both *Plasmodium falciparum* and *Plasmodium vivax* can cause severe anaemia but only *Plasmodium falciparum* causes the many complications of cerebral malaria: hypoglycaemia, metabolic acidosis and respiratory distress [15]. Most effort has been devoted to the production of *Plasmodium falciparum* vaccines because it is responsible for majority of deaths from malarial infections.

Among the recombinant blood-stage antigens that have been proposed for development as candidate vaccines, the leading erythrocyte stage antigens, merozoite surface protein 1 (MSP1) and apical membrane antigen 1 (AMA-1) are expressed in all species of *Plasmodium* [15]. A phase 1 trial of a vaccine based on $MSP1_{19}$ fused to CD4 T cell epitopes from tetanus toxoid concluded that the vaccine was immunogenic but had a high rate of adverse reactions [27]. $MSP1_{42}$ formulated in ASO2 adjuvant has gone into phase 1 trials in the United States and Kenya [28-30]. Also, *Escherichia coli*-produced $MSP1_{42}$ and RTS, S combined with $MSP1_{42}$ has been phase 1 tried in the United States [31].

It is likely that many recombinant blood-stage vaccines will undergo safety and immunogenicity studies in malaria endemic regions soon. The short time of exposure of the merozoites to antibodies between release from one infected red blood cell and attachment and entry into another dictates that very high antibody levels are required to block entry [32]. Such levels of antibody may not be achieved with alum; therefore, a lot of effort is focused on the testing and development of new adjuvants for asexual blood-stage antigens.

The choice of adjuvants for use in man includes alum, MF 59, Montanide ISA 720, Montanide ISA 51. These are combined with immuno-stimulators, which include MPL, QS 21 and CpG. [15]. A number of human phase II trials have been carried out using SPf 66 [33, 34], a multi-component vaccine which includes a blood-stage antigen.

PROSPECT FOR A POTENT MALARIA VACCINE

Will an effective vaccine against malaria parasite eventually emerge? Advances in the fields of genomics, proteomics and molecular immunology offer new hope for the development of a malaria vaccine. However, algorithms that can effectively identify the targets of protective immune effectors for malaria from genomic data are just being developed [35]. Doolan *et al* [3] have indicated data showing that

protective immune responses induced by immunization with irradiated sporozoites are directed against multiple epitopes on multiple antigens, with variable potency. Thus, a multi-antigen vaccine that induces cell-mediated immune responses against antigens of the liver stage may be required to mimic irradiated sporozoite-induced protection. A vaccine that prevents blood stage infection and clinical disease is likely to emerge if the antigenic targets of irradiated sporozoite-induced immunity are identified and packaged in a vaccine formulation that is immunogenic and suitable for manufacture and administration.

Evidence that natural exposure to *Plasmodium falciparum* results in acquisition of anti-malarial immunity in humans include the observation of decrease in the incidence of infections, reduction in prevalence and density of parasitaemia and the lowering of morbidity and mortality associated with repeated infection [36-38]. The existence of naturally acquired immunity and the fact that erythrocytic stage immunity can be induced by natural exposure [36, 37] to repeated blood stage infection provide a strong rationale for the identification of the antigenic targets of naturally acquired immunity and the development of vaccines designed to include high levels of antibody responses against these antigens.

CONCLUSION

A malaria vaccine that is potent and protective against the infection is likely to emerge in the near future but disappointing experiences of the past counsels caution concerning a time-frame prediction. It is difficult to say now whether such a vaccine will target pre-erythrocytic stage parasite or blood stage infection or both.

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