Status of Vaccine Research and Development of Vaccines for *Streptococcus pyogenes*
Prepared for WHO PD-VAC

I. About the Disease and Pathogen

Basic information on pathogen, including transmission, estimated global disease burden for those at risk, for morbidity and for mortality, including uncertainties/data gaps, geographical distribution, economic burden if available, age groups affected and target groups for vaccination.
Existing preventive, diagnostic and treatment measures and their limitations.

Streptococcus pyogenes or Group A Streptococcus (GAS) causes a massive disease burden that has been underestimated by global health authorities. A 2005 study estimated that there are > 500,000 deaths annually due to the bacteria, mostly occurring in low and middle income countries [1].
The agent is a Gram-positive bacteria with the human as its unique reservoir and an array of virulence factors allowing for a very broad spectrum of clinical expression. The nasopharyngeal mucosa and the skin, the two principal sites of asymptomatic colonization of GAS represent the primary reservoirs responsible for the maintenance and transmission of GAS to a new host. The ability of GAS to colonize and persist in skin tissue permits transmission through person-to-person contact. GAS can also overcome innate and acquired immune mechanisms present in saliva to remain viable for long periods, enabling transmission from infected persons or asymptomatic carriers via respiratory droplets. In addition, numerous outbreaks caused by food-borne GAS have also been reported [2].

GAS pharyngitis and impetigo are responsible for the greatest absolute number of GAS infections each year. GAS pharyngitis affects approximately 8%–15% of school-aged children per year, whereas GAS impetigo is a very common infection in children, especially in tropical developing countries with a prevalence of >10%, and even over 50% in some settings.

GAS bacteria are able to penetrate normal tissue barriers leading to invasive infection at local (e.g. retropharyngeal abscess or necrotizing fasciitis) as well as distant sites (e.g. septic arthritis). It produces an array of superantigens that can result in streptococcal toxic shock syndrome, which carries a high case fatality rate (>50%). In addition, invasive GAS disease is a frequent cause of sepsis in children and adults and has a high-case fatality rate leading to at least 150,000 deaths annually worldwide, although this figure is almost certainly an underestimate because of sparse data from many developing countries. GAS can also cause invasive infection in infants (neonatal sepsis) and the mother (puerperal sepsis); indeed, in the UK, GAS is the leading single cause of maternal death.

The immune response to GAS can be disordered and lead to post-streptococcal sequelae, including acute rheumatic fever (ARF), which in turn leads to chronic rheumatic heart disease (RHD), as well as post-streptococcal glomerulonephritis (PSGN). ARF/RHD is an uncommon disease today in most resource-rich countries including the United States, but it remains the major cause of acquired heart disease in children, adolescents and young adults in the developing world, responsible for at least 350,000 premature deaths per year. Available data on the prevalence of RHD suggest that there are over 30 million people affected by RHD worldwide. PSGN is thought to contribute to the high rates of end-stage renal failure in many GAS endemic regions.

Although data are sparse on the economic burden of GAS disease, a recent study in Fiji found that the cost of RHD over a 5 year period was ~US$48 million, or about one third of one percent of total GDP, representing a significant economic burden for the country. An economic evaluation of interventions for ARF/RHD found that a vaccine against GAS would be the most cost-effective intervention for ARF/RHD in ARF-endemic regions at a cost of between US$137–458 per DALY averted, assuming 80% efficacy and 65-95% coverage (compared with 22-33 thousand dollars for treatment of sore throat). Even wealthy

countries are affected - a study of GAS pharyngitis in the US suggested that GAS pharyngitis costs at least US$500 million per annum. Few data are available for impetigo, invasive disease, scarlet fever and PSGN.

Although GAS remains susceptible to penicillin, there is no evidence to suggest the burden of GAS diseases is decreasing, and antibiotic treatment of pharyngitis is an extremely resource intensive method for controlling ARF at a population level. Furthermore, many cases (possibly over 60%) of ARF occur without a history of symptomatic pharyngitis and it has been hypothesized that GAS skin infection may also contribute ARF. If GAS skin infections are involved, this further limits the potential benefits of penicillin treatment of pharyngitis for ARF prevention. For those diagnosed with ARF, secondary penicillin prophylaxis is effective in preventing recurrent ARF episodes and consequent worsening of RHD when efficiently delivered, but it requires monthly penicillin injections over many years. Although serious GAS diseases appear to be waning in some middle-income countries, probably because of improved living conditions and access to health services, GAS diseases continue to exact a toll in terms of mortality, morbidity and economic costs, even in wealthy countries [3].

Better data, particularly on the contribution of RHD to premature mortality in developing countries and on rates of invasive disease, including in the newborn and the new mother, are needed in most developing countries. However, current data on disease burden make a convincing case for the need for an effective vaccine that could offer a practical strategy for disease control and prevention, especially for ARF and RHD.

II. Overview of Current Efforts

A. Biological feasibility for vaccine development

Evidence that vaccine development is biologically feasible including from development of naturally acquired immunity, from vaccine development for related pathogens, from animal models or in vitro data

Although there are no currently licensed GAS vaccines yet, the biological feasibility for GAS vaccine development is supported by the following observations:

- The natural history of GAS infection: Pre-school and school-aged children experience repeated episodes of GAS pharyngitis and skin infection until they reach early adulthood when these infections become far less common. This pattern of infection suggests generation of immunity that requires repeated presentation of conserved antigens before a threshold level of protective immunity is achieved, *or* that immunity is type-specific.
- Available serologic data from natural history studies: The latter paradigm is supported by elegant longitudinal studies that have observed that infection with a single strain of GAS leads to generation of strain specific (M-protein) antibodies that lead to a long period (up to 30 years) of protection against the homologous strains but not against other strains.
- Animal data from pre-clinical vaccine studies: Pre-clinical (murine) studies of GAS vaccine candidates (predominantly M protein vaccines) have demonstrated protection against challenge infections.
- Evidence of protection from challenge in subjects immunized with purified M proteins: Subjects vaccinated with purified M proteins from GAS were protected against challenge with virulent homologous strain of GAS [4, 5, 6]. These GAS pharyngeal challenge studies, involving a total of 178 healthy adult volunteers in 3 separate studies were successfully used to demonstrate efficacy of prototype M protein vaccines in the 1970s. Importantly, GAS challenge was safe, with all participants responding to penicillin therapy without complications or sequelae developing.

<u>Serotype/strain coverage</u>

A potential barrier to a type-specific M protein-based vaccine is that there are > 200 *emm* types of GAS (the N-terminal part of the M protein has a variable amino acid sequence resulting in antigenic diversity and is the basis for this widely used nucleotide based *emm*-typing scheme). If type-specific antibody protection is the major mechanism by which immunity is generated against GAS then this clearly raises the issue of potential coverage for type-specific vaccines. This was highlighted in an article published in 2009 that identified that the distribution of *emm*-types was quite different in developing compared to developed settings. The study observed that there was a higher diversity of strains in lower to middle income settings versus high income settings, and indicated that the theoretical coverage of the 26-valent (strain specific) vaccine would be favorable in developed settings (>72%) while it would be much lower in settings were serious GAS disease is more common (e.g. Africa 39% and Pacific 24%)[7]. However, recent data from epidemiologic, genomic and *in vitro* studies suggest that there may be immunologic "cross-protection" between *elm*-types of GAS that may overcome this issue [8, 9, 10]

B. General approaches to vaccine development for this disease for low and middle income country markets

What are the scientific approaches and indications and target/age/geographic groups being pursued? What public health needs will these vaccines meet if successfully developed? Where there are several different possible indications/target groups, how much consensus is there as to prioritization between these for vaccine development in LMIC.

ARF occurs predominantly in school-aged children, although in some settings earlier skin infection may prime the immune system. Invasive disease occurs in all ages, with an increased incidence in infants. Impetigo is most common in pre-school children, with PSGN also following this age distribution because most cases of PSGN in endemic countries are due to skin infection. Therefore, while consensus in the field has not been reached on a target age group for vaccination, an infant schedule will likely be the most appropriate schedule for most endemic settings, possibly with a school entry booster dose. In non-endemic settings, a school entry schedule that coincides with the schedule for final TDap, IPV, and MMR doses may be appropriate. In areas where GAS is an important cause of maternal and neonatal sepsis, maternal immunization may also be considered.

A successful vaccine could address a huge unmet public health demand, and a vaccine that can prevent ARF (and thus RHD) as well as invasive GAS disease has the potential to save over 500,000 premature deaths per year. In addition, prevention of GAS pharyngitis and impetigo would have an enormous impact on reductions in morbidity through improved quality of life as well as a major economic impact through reduced health care expenditure on these exceedingly common infectious disease problems of childhood.

III. Technical and Regulatory Assessment

Highlight perceived positive/negative aspects in clinical/regulatory pathways e.g. well established product development and regulatory pathway to licensure, accepted immune correlates and/or functional assays, accepted surrogate efficacy endpoints, existence of well accepted animal or challenge models, agreed trial designs and endpoints. Possibilities to develop case for correlates/surrogates should be included.

Despite considerable progress having been made with a number of vaccine candidates, there remain a number of significant barriers to vaccine development. GAS vaccines, despite a long history of development, remain in their infancy [11]. There is no clear pathway agreed by multi-disciplinary consensus for a pathway for vaccine licensure, although global efforts are beginning to come together through a rudimentary roadmap for vaccine development [12]. GAS vaccines are now considered "impeded vaccines". The major issues include, but are not limited to: safety concerns, an incomplete understanding of immune protection in humans, inadequate epidemiological data and minimal development of combination antigen vaccines.

Concerns regarding vaccine safety are based upon a theoretical risk of autoimmune reactions in vaccinees leading to the development of ARF. One small study of a crude M protein vaccine suggested that there may be an increased risk of ARF in vaccine recipients; however, there are a number of concerns about the design of this trial that make it difficult to interpret, and autoimmune reactions have not been observed in the other human GAS vaccine trials involving thousands of study subjects. Understanding of human GAS immunity remains incomplete. More information is needed regarding immune protection against GAS skin infection, the role of T-cell immunity and the relative contributions of non-M type-specific antigens (common antigens) in inducing protective immunity. Better epidemiologic data are also required, for assessing burden of disease to strengthen the case for GAS vaccine development and for assessing vaccine coverage more systematically with high quality, standardized molecular typing studies in more countries, particularly in Africa and Asia.

Combination vaccines may be a viable approach to overcoming "gaps" in *emm* type coverage achieved with multivalent vaccines alone and to potentially broaden the immune response. However, to date there has been minimal progress in combining antigens in a single vaccine, and such a move would need to overcome proprietary interests and intellectual property rights. It is unclear exactly why there has been an apparent reluctance of large pharmaceutical companies to invest in clinical development of GAS vaccines. The obstacles listed above, together with the perception of a questionable market for a vaccine in affluent countries, likely combine to create the impression of adverse commercial risk.

A potential strategy to improve understanding of GAS immunology and also to create a pathway for relatively rapid testing of new GAS vaccine candidates is through the development of human GAS (pharyngeal) challenge studies. Previous studies (in the 1970's) in over 170 volunteers have shown that this approach is feasible, and proposals are under consideration for funding for a revival of this approach.

IV. Status of vaccine R&D activities

Summarize status of vaccine design, pre-clinical and clinical trial activity, including platforms, vectors, and adjuvants. Note academic, government, biotech and industry entities engaged. Summarize antigenic targets (if subunit approaches). Section on major advances in last 3-5 years, including key opportunities highlighted by recent science developments in the area.

GAS vaccines can be broadly divided into M protein–based and non–M protein–based vaccines. The GAS has a broad armamentarium of virulence factors, but it is the M protein that is the major virulence determinant of the organism. The M protein is a coiled-coil protein consisting of 3 domains: an A-repeat/N-terminal domain, which is highly variable and is used for epidemiologic molecular typing (*emm* typing); a B-repeat domain (antibodies against this region are not opsonic and some are cross-reactive with human tissues) and a conserved C-repeat domain. The 2 vaccines that have entered or are nearing clinical investigation are the N-terminal M protein-based multivalent vaccines (26-valent and 30-valent vaccines) and conserved M protein vaccines (the J8 vaccine and the StreptInCor vaccine) [13]. There are a variety of other vaccine candidates that are at various stages of discovery and development, some of them identified using reverse genomics (Table 1).

<u>26-Valent and 30-Valent M protein Vaccines</u>

These vaccines consist of fused recombinant peptides from the N-terminal region of M proteins from multiple different *emm* types of GAS. The original prototype multivalent vaccine was a hexavalent vaccine that was evaluated in a phase I trial and later expanded to a 26-valent vaccine and most recently a 30-valent vaccine. The 26-valent vaccine underwent a phase I/II clinical trial in human adult volunteers and was shown to be safe and immunogenic [14]. Functional opsonic antibodies were induced against all *emm* types of GAS in the vaccine. The 26-valent vaccine was reformulated into a 30-valent vaccine to increase "coverage" of circulating *emm* types in the United States, Canada and Europe as well as

developing countries. Epidemiologic surveys suggest that the 26-valent vaccine would provide good coverage of circulating strains of GAS in industrialized countries (over 72%) but poor coverage in many developing countries (as low as 24% in the Pacific region). In preclinical studies, the 30-valent vaccine has been shown to induce functional opsonic antibodies against all *emm* types of GAS represented in the vaccine. An intriguing finding of the studies of the 30-valent vaccine is that antibodies produced by the vaccine were shown to cross-opsonize a proportion of non-vaccine *emm* types of GAS, implying that cross-protection may mitigate, to a greater or lesser extent, the limited coverage of the 30-valent vaccine in many tropical developing settings where GAS disease is endemic. A phase I clinical evaluation of the 30-valent vaccine in adult volunteers is anticipated in 2014.

Conserved M Protein Vaccines

These vaccines contain antigens from the conserved C-repeat portion of the M protein. The StreptInCor vaccine incorporates selected T and B-cell epitopes from the C-repeat region, whereas the J8 and J14 vaccines contain single minimal B cell epitopes from this same region. Extensive studies in mice, particularly of the J8 vaccine candidate, have shown that these antigens produce opsonic antibodies that protect against challenge. These vaccines have the clear advantage of being comprised of single antigens. Limited data available for the J8 peptide indicate that its structure is highly conserved among multiple *emm* types of GAS and across regions. The J8 vaccine has recently entered a phase 1 trial in adult volunteers.

Other Vaccines

Cell wall and secreted virulence factors, such as streptococcal C5a peptidase, GAS carbohydrate and streptococcal fibronectin-binding proteins, among others, have been the subject of vaccine research for up to 20 years with some encouraging results, particularly for C5a peptidase, but none of these candidates has entered clinical trials. More recently, a number of promising, apparently conserved, vaccine candidates have been identified using reverse genomics [15]. In a large study, a number of vaccine candidate antigens delivered by both subcutaneous and intranasal routes were tested in murine intravenous and intranasal challenge models. Evidence of protection was demonstrated in at least 1 of the models for 9 of the antigens. However, their role in protection in human disease remains unknown, and none of these candidates has entered clinical trials.

Table 1: Development Status of Current Vaccine Candidates (POC = Proof of concept trial)

Candidate Name/Identifier	**Preclinical**	**Phase I**	**Phase II**	**POC**	**Phase III**
M protein: 6-valent N-terminal	X				
M protein: 26-valent N-terminal	X	X	X		
M protein: 30-valent N-terminal	X				
M protein: minimal epitope J8	X	X			
M protein: minimal epitope J14/p145	X				
M protein: Whole C-repeat conserved region	X				
M protein: C-repeat epitope (StreptInCor)	X				
M protein: C-repeat epitopes	X				
Four conserved antigens (COMBO)	X				
GAS carbohydrate	X				
GAS carbohydrate defective for GlcNAc side-chain	X				
GAS C5a peptidase	X				
Fibronectin-binding protein	X				
Streptococcal protective antigen	X				
Serum opacity factor	X				

Streptococcal pyrogenic exotoxin B	X				
Streptococcal pyrogenic exotoxin C	X				
Streptococcal pili (T antigen)	X				
Serine protease (SpyCEP)	X				
Serine esterase (Sse)	X				
GAS 40	X				
Nine common antigens	X				
G-related α2-macroglobulin binding protein	X				
Metal transporter of streptococcus (MtsA)	X				
Superoxide dismutase	X				
Lipoproteins	X				

References

1. Carapetis JR, Steer AC, Mulholland EK, Weber M. The global burden of group A streptococcal diseases. Lancet Infect Dis. 2005 Nov; 5(11): 685-94
2. Walker M et al. Disease Manifestations and Pathogenic Mechanisms of Group A *Streptococcus*. Clinical Microbiology Reviews 2014; 27(2): 264-301.
3. Steer AC, Dale JB, Carapetis JR. Progress towards a global group A streptococcal vaccine. Pediatric Infect Dis J. 2013 Feb; 32(2): 180-2
4. Fox EN, Waldman RH, Wittner MK, Mauceri AA, Dorfman A. Protective Study with a group A streptococcal M protein vaccine. Infectivity challenge of human volunteers. J Clin Invest. Aug 1973; 52(8): 1885-92
5. Polly SM, Waldman RH, High P, Wittner MK, Dorfman A. Protective studies with a group A streptococcal M protein vaccine. II. Challenge of volunteers after local immunization in the upper respiratory tract. J Infect Dis.1975; 131(3): 217-224.
6. D'Alessandri R, Plotkin G, Kluge RM, et al. Protective studies with group A streptococcal M protein vaccine. III. Challenge of volunteers after systemic or intranasal immunization with Type 3 or Type 12 group A Streptococcus. J Infect Dis. 1978;138(6):712-718.
7. Steer AC et al. Global emm type distribution of group A streptococci: systematic review and implications for vaccine development. Lancet Infectious Diseases 2009;9(10):611-616
8. Sanderson-Smith M et al. A systematic and functional classification of Streptococcus pyogenes that serves as a new tool for molecular typing and vaccine development. J Infect Dis. 2014 (epub ahead of print)
9. Dale, JB et al. Potential coverage of a multivalent M protein-based group A streptococcal vaccine. Vaccine 2013; 31: 1576-81
10. Engel, ME et al. Group A streptococcal emm type prevalence among symptomatic children in Cape Town and potential vaccine coverage. Pediatr Infect Dis J. 2014;33(2):208-10
11. Steer AC et al. Group A streptococcal vaccines: facts versus fantasy. Current Opinion in Infectious Diseases 2009;22(6):544-52
12. Dale JB et al. Group A streptococcal vaccines: Paving a path for accelerated development. Vaccine. 2013;31S:B216-B222
13. Batzloff MR et al. Protection against group A streptococcus by immunization with J8-diptheria toxoid: contribution of J8-and diptheria toxoid-specific antibodies to protection. J Infect Dis 2003;187:1598-608
14. McNeil SA, et al. Safety and immunogenicity of 26-valent group a streptococcus vaccine in healthy adult volunteers. Clin Infect Dis 2005;41(8):1114-22
15. Fritzer A et al. Novel conserved group A streptococcal proteins identified by the antigenome technology as vaccine candidates for a non-M protein-based vaccine. Infect Immun 2010;78(9):4051-67.