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Review Article

Preventing rheumatic fever: M-protein based vaccine

*Rajendra Tandon**

Consultant Cardiologist, Sitaram Bhartia Institute of Science & Research, B-16, Mehrauli Institutional Area, New Delhi 110 016, India

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ABSTRACT

Group A beta hemolytic streptococcus (GAS), the organism which initiates rheumatic fever (RF) continues to be sensitive to penicillin. However, penicillin cannot prevent RF if the preceding sore throat is asymptomatic in more than 70 percent children. Prevention of rheumatic fever (RF) may be possible only with the use of a vaccine. Efforts to design a vaccine based on emm gene identification of GAS, M-protein going on for more than 40 years, is unlikely to succeed. M-protein is strain specific. Infection with one strain does not provide immunity from infection with another strain. Based on the emm gene identification, of 250 or more identified strains of GAS, the distribution is heterogenous and keeps changing. The M-protein gene sequence of the organism tends to mutate. A vaccine prepared from available strains may not be effective against a strain following mutation.

Lethal toxic shock syndrome due to GAS infection has been described with organisms without identifiable or functional M-protein. M-protein has been excluded as the antigen responsible for acute glomerulonephritis (GN). Therefore M-protein plays no role in one suppurative (toxic shock syndrome) and one non-suppurative (acute GN) manifestation due to GAS infection. Lastly there is no direct evidence to indicate that M-protein is involved in inducing RF. The role of M-protein and the GAS component resulting in the suppurative manifestations of GAS infections like pyoderma, septic arthritis or necrotizing fasciitis etc is unknown. For a vaccine to be effective, an epitope of the streptococcus which is stable and uniformly present in all strains, needs to be identified and tested for its safety and efficacy. The vaccine if and when available is expected to prevent GAS infection. Preventing GAS infection will prevent all the suppurative as well as non-suppurative manifestations including RF.

It is generally accepted that rheumatic fever (RF) follows group A beta hemolytic streptococcal (GAS) infection. Since GAS infection spreads through droplets, overcrowding causes an increased transmission from person to person. Undernutrition or malnutrition can increase the susceptibility to infection. Poor socio-economic status results in an inability to obtain optimal medical care. Hence a higher prevalence in developing countries is predominantly related to low socio-economic status. The resurgence of RF in the Utah area in USA occurred in middle class families with a healthy lifestyle without overcrowding, a suburban population with facilities for good medical care indicates that improvement in socio-

* Tel.: +91 011 42111111; fax: +91 011 26533027.
E-mail address: dr_rajtandon@yahoo.com.
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economic status alone cannot control RF.[1] Virulence of RF is related to its capacity to cause more or less permanent cardiac damage. In Utah epidemic, carditis based on clinical combined with echocardiogram findings, occurred in almost 90 percent patients.[1] Hence prevention of rheumatic heart disease requires preventing RF. The purpose of this communication is to indicate that it is unlikely that a vaccine based on M-protein will succeed in preventing RF. Prevention of RF can be considered under two approaches – primary prevention and secondary prevention.

1. Secondary prevention

Secondary prevention consists in giving injections of intramuscular benzathine penicillin every two to three weeks depending on age and muscle mass to patients who have suffered from acute RF to prevent recurrences. Children weighing more than 20 kg can be given 1,200,000 units of benzathine penicillin every three weeks whereas younger children weighing less than 20 kg can be given 600,000 units every two weeks, since they do not have enough muscle mass for the higher dose. Secondary prophylaxis is ethically mandatory. If given properly, it reduces overall heart damage from subsequent attacks and in some patients disappearance of the auscultatory findings of valvar damage. However, secondary prophylaxis cannot reduce the burden of disease. Disappearance of murmurs does not mean that the heart has become normal. Recurrences bring out the former damage and increase the cardiac damage further.

2. Primary prevention

Primary prophylaxis consists in identifying that the patient has a sore throat, that it is GAS infection and giving penicillin to eradicate the GAS infection. Primary prophylaxis has too many loopholes and is almost impossible to practice even for individual patients. Resurgence of RF in USA indicates that GAS sore throat was asymptomatic in 22–71 percent patients; 18 percent with sore throat asked for medical treatment; one third to one half of acute RF occurred in asymptomatic patients and a 10 day oral penicillin course failed to protect the patients from acute RF in 15–48 percent cases.[1,2] Thus primary prevention is impossible if patients (parents) do not seek medical help, do not complete the treatment course and if the prescribed oral penicillin fails to prevent RF. As such, a 10 day intramuscular penicillin course is mandatory if we want to prevent RF. Intramuscular penicillin is painful, and is known to give rise to anaphylactic reactions, even causing death (very rarely). Many physicians refuse to give injectable penicillin and in some parts of India, injectable penicillin is officially banned.

3. Vaccine

Primary prophylaxis is possible if an anti-GAS vaccine becomes available. Although a number of components of GAS organism are being studied in order to make a vaccine, the most common and maximum effort has been directed toward an M-protein based vaccine. The specific role of streptococcal M-protein in the pathogenesis of RF is not clear. Lancefield isolated M-protein and designated it as the virulence factor of GAS.[3,4] M-protein has been extensively studied.[5] It has thermal stability, anti-phagocytic properties, capacity to initiate immunological response and has a structural similarity to tropomyosin. The M-protein, however, is strain specific and produces type specific antibody response. This results in each strain with its own specific M-protein failing to provide cross immunity from infection by another strain. As of today, according to the M-protein based classification, 250 strains of GAS organism are known.[6] M-protein has an alpha helical coiled coil structure, similar to myosin and tropomyosin.[7,8] Molecular mimicry between these proteins result in cross reactive antibodies. It is likely that because of similarities in the coiled coil structure of host proteins and streptococcal M-protein a rise in titer of a variety of auto-antibodies against these proteins is elicited in RF. Antibodies against a wide array of antigens of cardiac, nuclear and streptococcal origin are present in RF.[9,10] The presence of antimyosin antibodies is not indicative of cardiac muscle damage. Myosin specific antibodies are present in polymyositis, cocksackie B myocarditis and in patients following cardiac operations.[11,12] A multivalent vaccine based on hypervariable amino (N) terminal peptides of M-protein from 26 emm classified strains of GAS from Europe and USA has been utilized in clinical trials.[13] Subsequently a 30 valent M-protein peptide based vaccine has been evaluated.[14] It produced bactericidal antibodies against all the serotypes of GAS in the vaccine. In addition the vaccine produced antibodies against 24 of the 40 non vaccine GAS strains indicating a wider utility. However, with more than 250 identified strains of GAS it is unlikely to be effective globally not only because of the heterogeneity of GAS emm strain distribution but also because of the capacity to mutate.[15] The reasons why efforts for the M-protein based vaccine, going on for more than 40 years, are unlikely to succeed are outlined below.

- Emm gene typing for all the GAS strains isolated from children with sore throat in North India (Chandigarh) and South India (Vellore) was performed.[16] Of the 71 isolates of GAS in North India 14 emm types were found. Of the 227 isolates of GAS in South India 59 emm types were found. Comparing the isolates from North and South, only emm type 11 was common to both places. The GAS emm types varied from school to school and from one village to another both in North and South India. In North India two surveys were conducted three years apart. The GAS strains circulating in the community were entirely different in the two surveys (un-published data). Since immunity to GAS M-protein is strain specific, it is obvious that a polyvalent vaccine against most common isolates circulating in North India would be useless in South India or even in North India three years later.
- Studies of Kaplan and associates in a semi-closed community indicate that rapid changes occur in the serotypes causing infections and “broad non type specific immunity” does not occur.[17] Infection of GAS M1, strain may be followed by M6 strain within 4.3–27.7 weeks even in a semi-

closed community. Thus infection by M1 strain does not protect against infection by M6 even within a short period. Mutations occur in the emm gene sequence frequently. Six out of 106 M6 strains found in the study differed in the emm gene sequence from those in the data bank of the WHO streptococcal reference lab. The authors stated that "data also raise an important point regarding vaccine effectiveness for candidate group A streptococcal vaccine that are to be directed either towards conserved epitopes of the M-protein or extracellular products of the organism".[8] Mutation will result in the vaccine becoming ineffective against infection by the mutated organism.

- During the recent resurgence of RF in USA, heavily encapsulated M18 and M3 strains which produced mucoid colonies were found.[18] In the intermountain area the mucoid colonies of the strain did not result in a more severe pharyngitis. The change in colony character and the fact that RF resurgence occurred at the same time indicates that a change in the organism had occurred.[2] The change resulted in its developing a capsule, grow as mucoid colony and become virulent to result in RF. Krishna Kumar et al discussing the epidemiology of streptococcal infection state that "It is probable that the rheumatogenicity of certain serotypes of streptococci is due to the clonal emergence of a more virulent strain, by the acquisition of virulence genes … A waxing and waning of normal 'background' group A streptococcal infections occurs with one serotype becoming prevalent in a given population. A virulent clone of this specific serotype may 'emerge' resulting in an epidemic".[19]
- Bennett-Wood and associates have described toxic shock syndrome, secondary to GAS infection in two unrelated children from Australia.[20] One child died of toxic shock syndrome. GAS were isolated from the blood culture in both children. The two isolated GAS strains did not express M-protein. Electron microscopy could not identify M-protein fibrils on the surface of the organism which failed to resist phagocytosis suggesting the absence of "functional M-protein[20]". The inference of the study would be that the toxic shock syndrome – even lethal – could not be related to M-protein, since M-protein was not involved. The virulence must be due to some other component of the GAS organism and M-protein cannot be the only virulence factor of the GAS organism.
- GAS infection results in two non-suppurative clinical manifestations–acute RF and acute glomerulonephritis (GN). Logically similarities should be present in the pathogenesis of the two immunological manifestations. Studies in acute GN have excluded M-protein as the antigen responsible for the disease.[21]
- Rheumatic carditis was believed to result in myocarditis and myosin damage. M-protein designated as the virulence factor of GAS was chosen as being responsible for myosin damage because of its structural similarity to myosin. At present we know that RF does not cause myocarditis or myosin damage.[22] Hence, utility of M-protein which targets myosin to formulate anti-GAS vaccine does not seem to be justified. Despite all the studies, at present, there is no evidence in the literature to indicate that M-protein is the GAS antigen responsible for inducing RF.

3.1. *Comments*

GAS infection results in two non-suppurative manifestations, acute RF and acute GN as well as a number of suppurative manifestations like toxic shock syndrome, necrotizing fasciitis, pyoderma, septic arthritis and others. Preventing GAS infection should help prevent both types of manifestations.

The evaluation of findings related to M-protein indicate that:

- M-protein is strain specific. Infection from one strain does not provide immunity for infection from another strain even in a short period of four to six weeks. Since there are 250 strains, a polyvalent vaccine based on emm gene classification of M-proteins would be very difficult to achieve, whether conserved or non conserved areas of M-protein are utilized.[1,6]
- On the basis of emm gene classification, the distribution of GAS strains in the community is so variable and heterogenous that a polyvalent vaccine from one area will not be of value in another area (North and South India) or even in the same area sometime later.[16]
- Mutation in GAS emm gene occurs frequently enough that the mutated strain may be able to result in RF despite being a component (in the non mutated state) of a polyvalent vaccine because of altered gene structure.[17]
- Absence of expressed M-protein in a lethal GAS organism indicates that it cannot be the main or the only virulence factor of the GAS organism.[20]
- M-protein has been excluded as the causative antigen for acute GN, the other immunological manifestation of GAS infection.[21] At present there is no direct evidence that M-protein is the antigen causing RF.

The questions which need to be answered are:

(i) why is it necessary to start from RF and its relation with GAS M-protein in trying to make a vaccine ?
(ii) what is the role of M-protein in causing pyoderma or other suppurative diseases?
(iii) why are we not looking for the virulent epitope of GAS from infections causing toxic shock, pyoderma or septic arthritis for its suitability for anti-GAS vaccine ?

If GAS infection can be prevented by a vaccine designed from organisms causing arthritis, sepsis or pyoderma, it should be good enough to prevent all GAS related disease manifestations. M-protein is not involved in the pathogenesis of acute GN the other non-suppurative manifestation of GAS infection. GAS infection has resulted in death from toxic shock syndrome in the absence of M-protein being present, or expressed. Therefore acute GN a non-suppurative manifestation and toxics shock syndrome a suppurative manifestation may not be preventable by a vaccine based on M-protein. It is necessary to look for an epitope for vaccine which resulted in the death of a child from toxic shock syndrome due to GAS infection without identifiable or functional M-protein. The approach using M-protein has not provided the necessary break through in more than 40 years.

In order to make a vaccine, an attempt has to be made to identify a component with immuno-reactive properties which is uniformly present in all the GAS strains; does not alter or retains its specificity even after mutation. A second possibility

could be that all the emm based identified GAS strains be reclassified utilizing one or more features like – cell wall/membrane protein, glycoprotein, streptococcal polysaccharide etc. into groups with identical features, thus reducing the requirement of the numbers of strains for making a vaccine.

A polyvalent vaccine based on emm gene identification of more than 250 M-protein strains, which result in strain specific immunity; do not provide immunity against another strain even for a few weeks, have a heterogenous distribution in the community; keep mutating with time with the mutated strain may be having capacity to cause infection and disease in spite of being a component (in non mutated state) in the vaccine, is unlikely to succeed.

If a vaccine can prevent GAS infection it will prevent all the clinical manifestations of GAS infection. Preventing disability from RF or death from toxic shock syndrome from a vaccine designed to prevent pyoderma from GAS infection will be a tremendous achievement.

4. Conclusions

Primary prevention of RF is possible only with a vaccine. Injectable penicillin can prevent RF, however, it may not be possible to protect even an individual from RF utilizing penicillin if the antecedent sore throat is asymptomatic. Since emm typing has resulted in the identification of 250 strains, each providing specific individual immunity, utilizing M-protein as the basis of an anti-GAS vaccine is unlikely to succeed. At present there is no evidence to indicate that M-protein is responsible for RF. It has already been excluded as a cause of acute GN, the other non-suppurative manifestation of GAS infection. An attempt needs to be made to identify an epitope of GAS organism which is present, uniformly in all strains, is stable in spite of mutation and can be utilized for making a vaccine.

Conflicts of interest

The author has none to declare.

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