Properties of IgG-Binding Proteins Expressed by Streptococcus pyogenes Isolates Are Predictive of Invasive Potential

Roberta Raeder and Michael D. P. Boyle

Department of Microbiology, Medical College of Ohio, Toledo

Recent clinical Streptococcus pyogenes isolates of the M1 serotype can be grouped according to the IgG-binding properties of their M proteins. One group expressed an IgG-binding M1 protein reactive with human IgG1, IgG2, IgG3, and IgG4 (type I10); the other expressed a protein with predominant reactivity with human IgG3 alone (type I1b). Both IgG-binding protein phenotypes were equally resistant to phagocytosis in human blood; however, when they were injected into a skin air sac on outbred CD1 mice, all mice injected with M1 isolates of the type I10 phenotype were dead within 70 h, while only 40% of those injected with M1 isolates of the type I1b phenotype died within the same period. Bacteria recovered from the spleens of animals that died after injection with type I1b phenotype isolates demonstrated a change in their IgG-binding profile and were indistinguishable, in vitro or in vivo, from isolates displaying the type I10 phenotype.

A resurgence of severe group A streptococcal disease beginning in the mid-1980s has precipitated a renewed interest in the pathogenic mechanisms of the causative organism. The predominant serotype associated with the recent outbreak of invasive streptococcal disease as well as the newly described toxic shock-like syndrome is M1 [1-4]. Detailed multifunctional enzyme analysis and genetic approaches by Musser et al. [5] have suggested that the M1 isolates are clonally related. Cleary et al. [6], using restriction fragment length polymorphism analysis of chromosomal DNA, were able to distinguish two patterns of group A isolates recovered from different sites. These differences suggested a unique form of the speA gene, which was present in all invasive or blood isolates. In addition, M1 isolates with this putative invasive genotype were internalized more efficiently by respiratory epithelial cells than were isolates lacking the genotype [7].

In a study of invasive group A streptococcal isolates collected as part of a Centers for Disease Control and Prevention (CDC) surveillance study of a recent outbreak of severe streptococcal disease [8], we identified two phenotypes of M1 isolates on the basis of their IgG-binding protein profiles [9, 10]. One group of isolates expressed a protein that was recognized specifically by an M1 serotyping antiserum and reacted with all four human IgG subclasses (type I10 IgG-binding profile), while in the second group, the antigenically related M1 protein reacted predominantly with human IgG3 (type I1b IgG-binding profile). These differences were qualitative and not quantitative and the activities could be shown to be distinct from another.

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Reprints or correspondence: Dr. Michael D. P. Boyle, Dept. of Microbiology, Medical College of Ohio, 3000 Arlington Ave., Toledo, OH 43699-0008.
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Methods

Bacteria. Ten Streptococcus pyogenes isolates of the M1 serotype were obtained from the CDC. These group A streptococcal isolates were collected as part of a surveillance study and extensively characterized at the CDC for a variety of phenotypic and genotypic characteristics [8]. Our laboratory has also analyzed these isolates for expression of IgG-binding proteins [9, 10]. The characteristics of each isolate are summarized in table 1. The isolates were maintained as glycerol stocks at −70°C and had not been passaged in the laboratory on more than three occasions.
Solubilization of IgG-binding proteins from group A streptococcal isolates. All isolates were grown overnight at 37°C as stationary cultures in Todd-Hewitt broth. IgG-binding proteins were solubilized by CNBr treatment (30 mg/mL in 0.2 N HCl) as described [19].

Plasma proteins. Human IgG myeloma proteins of each subclass were obtained from the World Health Organization/International Union of Immunologic Societies immunoglobulin committee.

Labeling of proteins. Proteins were radiolabeled with 125I (Amersham, Chicago) using a lactoperoxidase method [20]. A PD-10 desalting column (Pharmacia, Piscataway, NJ) was used to separate labeled proteins from free iodine. The specific activity of all radiolabeled proteins was ~0.3 mCi/mg.

PAGE and Western blotting techniques. Protein samples were denatured by boiling for 5 min in 0.5 M TRIS-HCl (pH 6.8) containing 2% (wt/vol) SDS, 5% (vol/vol) β-mercaptoethanol, 10% (vol/vol) glycerol, and 0.01% (wt/vol) bromphenol blue. Denatured proteins were electrophoresed in 10% polyacrylamide slab gels at 50 V for 16 h according to the method of Laemmli [21]. Prestained molecular weight standards (Bio-Rad, Richmond, CA) containing phosphorylase B (Mn ~106,000), bovine serum albumin (Mn ~66,000), ovalbumin (Mn ~45,000), carbonic anhydrase (Mn ~33,500), soybean trypsin inhibitor (Mn ~27,500), and lysozyme (Mn ~18,500) were included in each SDS-PAGE assay.

The proteins were electrophoretically transferred to nitrocellulose (Bio-Rad) by a modification of the method described by Towbin et al. [22]. Briefly, SDS slab gels were presoaked for 30 min in 25 mM TRIS, 192 mM glycine, and 20% methanol (pH 8.3), assembled into the high-field-intensity transblot system (Bio-Rad), and electrophoresed in that buffer at 70 V for 3 h. Nitrocellulose blots were washed four times for 15 min each at ambient temperature with 250 mL of 0.05 M Veronal buffer (VBS), pH 7.4, containing 0.15 M NaCl, 0.25% gelatin, and 0.25% Tween 20 (VBS-gel-Tween) to saturate remaining protein binding sites on the nitrocellulose. Blocked membranes were incubated for 3 h at ambient temperature in 25 mL of VBS-gel-Tween containing 3 × 10^5 cpm/mL of the appropriate 125I-labeled probe. The nitrocellulose membranes were then washed four times (for 15 min each time) with 250 mL of VBS containing 0.01 M EDTA (pH 7.2), 1 M NaCl, 0.25% gelatin, and 0.25% Tween 20 on a rocking platform at ambient temperature. The membranes were dried and autoradiographed using Kodak XAR-5 film and Kodak X-omatic intensifying screens at ~70°C for 1–3 days.

Mice. Outbred CD1 mice were obtained from Charles River (Portage, MI).

Air sac procedure. The air sac procedure was a modification of the procedure described in [23]. Briefly, an air and liquid tight connective tissue pouch was generated on the back of a mouse by slow subdermal injection of 0.9 mL of air via a 27-gauge needle on a 1.0-mL syringe. The syringe containing the air also contained 0.1 mL of a suspension of group A streptococci containing 10^10 cfu, which had been determined in previous experiments to be the dose required to kill ~50% of the animals via this route of infection [15]. Mice were provided with food and water ad libitum. Each experiment was continued for a period of 96–144 h after infection, and death was used as the end point. All surviving animals were euthanized by cervical dislocation at the termination of the experiment.

The spleen was removed from any animal that died of infection and from all animals euthanized at the termination of the experiment. A spleen cell suspension was generated by homogenizing the spleen in 1 mL of sterile 10 mM PBS, pH 7.4. An aliquot was cultured to determine whether bacteria were present and the purity of the culture, and the remainder of the spleen cell suspension was mixed with an equal volume of sterile glycerol and stored at −70°C for later analysis. No viable group A streptococci were recovered from the spleen of any animal that was alive when the experiment was terminated.

Analysis of IgG-binding protein expression by group A streptococci recovered from infected animals. Bacteria were recovered from infected spleens by plating an aliquot of the glycerol-stocked spleen cell suspension on blood agar plates. All of the group A colonies from the plate were used to inoculate Todd-Hewitt broth.
and expanded by growth as stationary cultures overnight at 37°C. IgG-binding proteins expressed by different group A streptococci were extracted from a standardized suspension of bacteria by treatment with CNBr using conditions described [19].

Opsonophagocytosis assay. Bactericidal assays were done as described by Schnitzler et al. [24]. Each test organism was grown overnight in Todd-Hewitt broth. One hundred microliters of the culture was added to 5 mL of prewarmed Todd-Hewitt broth and incubated at 37°C for exactly 90 min. Each culture was then diluted 1:10 in sterile PBS, and 100 μL was used to inoculate tubes containing 300 μL of human plasma or 300 μL of lightly heparinized whole blood with or without 100 μL of a 1:10 dilution of normal rabbit serum or rabbit anti-M1 antiserum. Control tubes containing 300 μL of Todd-Hewitt broth were included in each assay to determine the inoculum at time 0 and growth after 3 h of incubation. Tubes were incubated at 37°C with end-over-end rotation for 3 h. After the incubation, 3 mL of molten 0.7% Todd-Hewitt agar was added to each tube, and the contents were overlaid on Todd-Hewitt agar plates. The number of surviving bacteria was determined by counting colony-forming units after overnight incubation at 37°C. Values are averages of duplicate samples ± the range.

Results

Effect of IgG-binding protein phenotype on invasive potential of group A isolates in a mouse model of skin infection. Animal studies from our laboratory have suggested that IgG-binding protein expression may be important in enabling group A streptococci to establish invasive skin infections [15, 25]. Consequently, the invasive potential of 5 representative type IIo-expressing and 5 representative type IIb-expressing group A isolates of the M1 serotype (table 1) were compared in a mouse model of skin infection. Groups of 5 outbred CD1 mice were injected with each isolate at a single dose, 10^10 cfu, into a skin air sac as described in Methods. These experiments were done in a blind fashion, in which the organisms were coded before injection.

Two distinct patterns of infectivity were observed. For 5 of the isolates, the majority of mice died between 24 and 48 h after infection, and 24 of 25 mice were dead within 72 h (figure 1A). Infection with the other 5 isolates demonstrated a second profile of infectivity, in which only 1 of 25 mice died within the first 24 h and only 9 of 25 mice died during the 96-h duration of the experiment (figure 1B). When the code was broken, all 5 isolates that demonstrated the rapid onset of lethal infection belonged to the group expressing type IIo IgG-binding proteins, while mice in the groups that showed the slow progression of infection had been infected with group A isolates expressing type IIb IgG-binding proteins. When all 25 animals infected with either a type IIo- or a type IIb-expressing isolate were grouped, the difference in infectivity of these isolates was statistically significant at the P < .005 level using Student’s t test (figure 1C).

In the course of the skin infection experiment, it was noted that animals in some groups demonstrated severe tissue damage at and around the injection site within 24 h after infection. The majority of these animals survived the infection, and the lesions were almost completely resolved when the experiment was terminated on day 5. All of these animals were found to be in groups that had received M1 isolates expressing type IIb IgG-binding proteins, and all of these isolates secreted a casein-hydrolyzing enzyme in culture supernatants (table 1).

The organisms recovered from the spleens of representative mice that died after inoculation with either type IIo- or type IIb-expressing isolates were expanded in vitro and their IgG-binding protein expression monitored after extraction with CNBr. The extracts were separated on SDS-polyacrylamide gels under reducing conditions and probed for antigenic and functional reactivity. Extracts of bacteria from an aliquot of a culture of each inoculating organism were included for comparison. Extracts of organisms recovered from the spleens of animals that died after infection with a type IIb organism demonstrated a different profile of IgG-binding protein reactivity than that of the infecting bacteria (figure 2). The recovered group A streptococci now demonstrated an IgG-binding profile characteristic of a type IIo-expressing organism. By contrast, the organisms recovered from the spleens of animals receiving a lethal infection with a type IIo organism demonstrated no qualitative difference in the IgG-binding profile, although enhanced expression of type IIo IgG-binding proteins was observed (figure 2).

Previous studies from our laboratory have demonstrated that organisms that were recovered from the spleens of mice lethally infected with a group A isolate and that expressed higher levels of IgG-binding proteins were more virulent when injected into naive animals [15]. Consequently, in the next series of experiments, organisms recovered from the spleens of 2 mice that died of lethal infection with a representative type IIb-expressing isolate (isolate 1881) were injected into the skin air sac of naive mice and compared for their virulence with the original isolate. The IgG-binding profile of the inoculating isolate was predictive of the virulence of the organism (figure 3). Bacteria recovered from the spleen of lethally infected mice and demonstrating the functional type IIo profile were more virulent than the type IIb-expressing parent when injected into naive mice and were equally as virulent as the 5 M1 isolates that expressed a type IIo binding protein (e.g., figures 1A, 3A). In contrast to the type IIb-expressing parental isolate, the spleen-recovered type IIo phenotype isolates no longer produced a casein-hydrolyzing protease in culture supernatants and no longer caused lesions when injected into the skin of mice (data not shown).

The conversion of a type IIb phenotype to a type IIo-expressing organism after selective pressures associated with mouse skin infection was a consistent finding for bacteria recovered from the spleen of any mouse that died after skin infection with any type IIb-expressing isolate (n = 5). Since the type IIb-expressing organisms used to infect the mice were not derived from a single colony, it was possible that host
Figure 1. Analysis of virulence of type IIo (A) and type IIb (B) *S. pyogenes* serotype M1 isolates when injected into mouse skin air sac; each symbol is individual group. C, Summary of animals dying of lethal infection with either type IIo or type IIb M1 strain. Difference between groups was significant by Student's *t* test (*P* < .005).

Figure 2. SDS-PAGE analysis of functional and antigenic reactivity of organisms recovered from spleen of mice lethally infected in skin with either type IIo or a type IIb *S. pyogenes* serotype M1 isolate. CNBr-extracted polypeptides from inoculating strains are included on each gel for comparison. Parallel blots probed with human IgG2 and IgG4 gave results identical to those indicated for human IgG1 (data not shown). Lane 1, CNBr extracts of strain 906 (type IIo) or strain 1881 (type IIb) mouse inoculum. Lanes 2 and 3, organisms isolated from spleens of 2 mice receiving lethal infection with either strain 906 (type IIo) or strain 1881 (type IIb). Molecular weights (K) are indicated at left.
pressure, present at the site of infection, selected for and permitted expansion of a subpopulation of organisms with a type II0 IgG-binding protein phenotype. Consequently, these experiments were repeated using 10 individual colonies selected at random from a representative type IIb isolate (1881).

An aliquot from a culture of an individual colony was either extracted with CNBr and analyzed for IgG-binding reactivity or injected at a concentration of 10^10 cfu into an air sac on outbred mice (5 mice/group). While there was some quantitative variation in the amount of antigenic and functional protein recovered in CNBr extracts of bacteria from cultures derived from individual colonies, the polypeptides solubilized in each case bound only human IgG3. The results of the skin infection studies using type IIb phenotype bacteria derived from a single colony demonstrated a more variable time course than observed in the initial experiment. Of the 50 mice injected with single colonies (10 groups of 5 animals each), 3 animals, all from different groups, died within the first 24 h, and 39 of the 50 animals had died when the experiment was terminated 96 h after infection. There was no evidence for a significant difference in lethality among the 10 groups.

Selected organisms recovered from the spleens of mice that had died from infection with bacteria derived from a single type IIb colony were extracted and found to display a consistent profile of type IIo reactivity (data not shown). Taken together, these results demonstrate that the type IIb phenotype can be converted to a type IIo phenotype by biologic pressures present in the infected host.

**Effects of IgG-binding phenotype on opsonophagocytosis in human blood.** Since the M protein is primarily associated with the ability of group A streptococci to resist opsonophagocytosis, the next series of experiments was designed to determine if the observed phenotypic differences associated with the protein recognized by the anti-M1 serotyping antiserum altered their ability to be phagocytosed in human blood. All 10 isolates, regardless of their IgG-binding phenotype, were resistant to phagocytosis in human blood.

Similar studies were done with a representative type IIo isolate (906) and a type IIb isolate (1881) and their variants recovered from the spleens of lethally infected mice. The change to the type IIo IgG-binding phenotype associated with the 1881 isolate recovered from the spleen was not associated with any change in the ability to survive phagocytosis in human blood (table 2). Addition of a rabbit anti-M1 typing antiserum, but not normal rabbit serum, led to effective phagocytosis of both the parent and the spleen-recovered variant of either isolate 906 or 1881 (table 2). Thus, despite the quantitative difference in extractable antigenic M1 protein and the difference in

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**Figure 3.** Comparison of virulence of 2 spleen-recovered *S. pyogenes* type IIb M1 isolates (□, ▲) with that of original infecting isolate, 1881 (○), when injected via skin air sac into naive animals at 10^8 cfu (A), 10^7 cfu (B), or 10^6 cfu (C).
functional binding profiles previously observed for these isolates (figures 2, 3), both expressed sufficient antiphagocytic activity to survive in human blood. All isolates studied also expressed sufficient M1 protein to facilitate efficient opsonization in the presence of specific antibody. Thus, neither quantitative nor qualitative differences in IgG-binding protein expression significantly alter the behavior of any of the group A isolates in phagocytic assays using human blood.

### Discussion

Recent analysis of group A streptococcal IgG-binding proteins has provided evidence for an association between expression of these proteins and skin invasive potential in a mouse model or resistance to phagocytosis in human blood [15, 25]. In addition, epidemiologic studies have suggested a potential role for these functional proteins, particularly in skin infections [14]. The recognition that many of these IgG-binding proteins are coded for by genes within the M protein gene family, which are present in a coordinately regulated virulence locus in group A streptococci [26–32], suggests that these gene products may play some critical role in the interaction between the bacteria and the human host.

In a series of studies of the distribution of IgG-binding proteins on invasive group A clinical isolates, collected as part of a CDC surveillance study [8], we have demonstrated two distinct phenotypes of IgG-binding reactivity associated with M1 isolates [9, 10]. The IgG-binding proteins extracted from these isolates differed in quantity but were antigenically related to each other and could be identified specifically by an anti-M1 serotyping antiserum. One form of IgG-binding M1-related protein bound all four human IgG subclasses (type IIo), while the second form of the antigenically related protein bound preferentially to human IgG3 (type IIb). In a previous study, these distinctions were shown to be qualitative and not merely quantitative [10].

In this study, a detailed comparison of the behavior of the two IgG-binding M1 phenotypes, in two defined biologic systems, was done. Although there were differences in the quantity of antiphagocytic M protein that could be extracted from the 2 groups of M1 isolates [10], both type IIo and type IIb pheno-

### Table 2. Sensitivity of type IIo or type IIb S. pyogenes M1 isolates to opsonophagocytosis in human blood.

<table>
<thead>
<tr>
<th>Strain</th>
<th>M1 group</th>
<th>Inoculum</th>
<th>Human plasma</th>
<th>Human blood</th>
<th>Normal rabbit serum</th>
<th>Rabbit anti-M1 serum</th>
</tr>
</thead>
<tbody>
<tr>
<td>906</td>
<td>IIo</td>
<td>61.5 ± 6.5</td>
<td>&gt;500</td>
<td>&gt;500</td>
<td>&gt;500</td>
<td>21.5 ± 10.5</td>
</tr>
<tr>
<td>906SR</td>
<td>IIo</td>
<td>65.5 ± 7.5</td>
<td>&gt;500</td>
<td>&gt;500</td>
<td>&gt;500</td>
<td>2 ± 0</td>
</tr>
<tr>
<td>1881</td>
<td>IIb</td>
<td>55 ± 0.5</td>
<td>&gt;500</td>
<td>&gt;500</td>
<td>&gt;500</td>
<td>3 ± 1</td>
</tr>
<tr>
<td>1881SR</td>
<td>IIo</td>
<td>41.5 ± 0.5</td>
<td>&gt;500</td>
<td>&gt;500</td>
<td>&gt;500</td>
<td>51.5 ± 4.5</td>
</tr>
</tbody>
</table>

NOTE. SR, spleen-recovered variant. Values are mean cfu ± range for duplicate samples.

Types were found to be resistant to phagocytosis in human blood and were efficiently opsonophagocytosed in the presence of rabbit antiserum to M serotype 1 but not in the presence of normal rabbit serum (table 2). These results indicated that there was no significant difference in the antiphagocytic properties of the M1 proteins from these isolates, despite differences in their IgG-binding profiles.

The major reproducible difference between the type IIo and type IIb phenotypes of these M1 isolates was their behavior in a mouse model of skin infection. Isolates expressing type IIo IgG-binding proteins were found to lead to a more rapid invasive infection than was observed after infection with organisms expressing the type IIb phenotype (P < .005). These distinct differences among isolates were observed only when mice were injected via a skin air sac. In similar studies in which the isolates were injected intraperitoneally, there was no association between IgG-binding protein type and virulence (data not shown). These findings are consistent with earlier mouse studies using group A isolate 64/14 that demonstrated marked differences in IgG-binding protein expression when subjected to host pressure in the skin versus the peritoneum [15].

Of particular interest in these studies was that organisms recovered from the spleens of all animals that died of lethal infection after injection with a type IIb isolate demonstrated the type IIo IgG-binding phenotype. These findings raised concerns that the inoculum might contain populations of group A isolates and that selection for a subpopulation of a more virulent phenotype might be occurring. Studies using individual colonies of 1 group A isolate expressing the type IIb IgG-binding phenotype indicated that the conversion of the type IIb to the type IIo phenotype was a consistent finding, suggesting that biologic pressures could affect the properties of the expressed M1 gene product. In addition, organisms recovered from the spleens of animals lethally infected with a type IIb isolate and displaying the type IIo phenotype were more virulent when inoculated via the skin air sac into naive animals.

Once selected in the mouse, the type IIo phenotype remained stable after laboratory culture and did not require the continued presence of the biologic environment that was initially required to select for this mouse skin invasive phenotype. Previous studies have demonstrated that the change in IgG-binding profile
could not be attributed to expression of a related IgG-binding protein, protein H, which has been identified previously in certain M1 isolates [10–13]. The functional and antigenic properties of protein H are distinct from those of either antigenically related IgG-binding M1 protein [10], and none of the isolates studied has been found to express protein H before or after mouse passage.

Cleary et al. [6] defined two genotypes of M1 group A streptococci on the basis of restriction fragment length polymorphism analysis and the presence of the speA gene. These organisms were grouped into invasive or noninvasive on the basis of their original site of isolation. Invasive organisms were those recovered from cultures of normally sterile body fluids or deep wounds, and noninvasive isolates were isolated from pharyngeal or uncomplicated skin infection sites. Subsequent studies by this group demonstrated that organisms with the invasive genotype were more efficient in their ability to invade respiratory epithelial cells than were noninvasive isolates [7]. Eight of these isolates, 4 invasive and 4 noninvasive, were tested in our mouse model. Only 1 of these isolates, classified as invasive, expressed proteins in CNBr extracts with significant type IIb IgG-binding reactivity and demonstrated 100% lethality in the mouse skin infection model. The other 7 isolates demonstrated low levels of CNBr-extractable protein and were either avirulent (2 isolates) or weakly virulent (20%–40% lethality) in the mouse skin infection model. In all instances, however, isolates recovered from the spleens of lethally infected mice demonstrated significant levels of extractable IgG-binding proteins in CNBr extracts and all demonstrated type IIb binding characteristics. In addition, it was noted that 5 of the 8 isolates produced casein-hydrolyzing proteinase in culture supernatants and also caused lesions at the skin injection site. All of the isolates analyzed, regardless of their quantitative or qualitative expression of IgG-binding proteins, were equally resistant to phagocytosis in human blood while being efficiently opsonophagocytosed in the presence of specific antibodies (data not shown).

All of the isolates we analyzed in this study were derived from blood cultures and would therefore be defined as invasive according to the criteria described by Cleary et al. [6]. On the basis of our studies in the mouse model of skin infection, we conclude that invasive isolates could be further subgrouped on the basis of invasive potential as well as on their pattern of IgG-binding protein expression. It is also of interest that many of the isolates studied by Cleary et al. had been in laboratory collections for long periods of time and demonstrated very low levels of IgG-binding protein expression. In our studies, we have found that the expression of IgG-binding protein decreases markedly over time. This observation is not new [18] but is of concern in attempting to identify virulence factors of group A streptococci using isolates that have been manipulated in the laboratory. The studies presented herein demonstrate that the expression of the M1 protein, while being variable in both quantity and human IgG subclass–binding properties, does predict the invasive behavior of the organisms when injected into the skin of mice.

At present, the molecular change responsible for the different IgG-binding profiles of the M1 protein has not been identified. Polymerase chain reaction amplification of the chromosomal DNA from each of the 10 M1 isolates studied, using primers that amplify class 1 M protein genes selectively [31], resulted in a single product of a similar size and with identical restriction fragment length polymorphism profiles. These results suggested that the phenotypic difference between the M1 proteins was not due to a major difference at the gene level. Subtle variation in the nucleotide sequence of emm genes from M1 isolates has been reported [33]. It is not known whether these sequence changes would alter the IgG-binding properties of the expressed protein or if the mutation frequency is sufficient to account for the conversion of a type IIb form to a type IIo IgG-binding form observed after skin infection in mice. Alternatively, the difference in IgG-binding protein phenotype of antigenically related M1 proteins may be due to some unique combination of transcriptional, translational, or posttranslational processing.

Our studies demonstrated that fresh clinical group A streptococcal isolates of the M1 serotype have two distinct patterns of IgG-binding reactivity that are associated with proteins recognized specifically by an M1 serotyping antiserum. The difference, while having no marked effect on the ability of the organism to resist opsonophagocytosis in human blood, is predictive of the invasive potential of the organism when measured in a mouse skin infection model. These phenotypic variations in group A streptococcal IgG-binding proteins may be related to other variations noted among group A M1 serotype isolates [5, 6, 34] and may help to explain the different patterns of disease observed after infection with group A isolates of the same serotype.

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References


34. Norgren M, Norby A, Holm S. Genetic diversity of TIMI group A streptococcal IgG-Binding Proteins and Invasiveness 895