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Different Classes of Proteoglycans Contribute to the Attachment of
*Borrelia burgdorferi* to Cultured Endothelial and Brain Cells

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The Lyme disease spirochete, *Borrelia burgdorferi*, infects multiple tissues, such as the heart, joint, skin, and nervous system and has been shown to recognize heparan sulfate and dermatan sulfate proteoglycans. In this study, we examined the contribution of different classes of proteoglycans to the attachment of the infectious *B. burgdorferi* strain N40 to several immortalized cell lines and primary cultured cells, including endothelial cells and brain cells. Bacterial attachment was inhibited by exogenous proteoglycans or by treatment of host cells with inhibitors of proteoglycan synthesis or sulfation, indicating that proteoglycans play a critical role in bacterial binding to diverse cell types. Binding to primary bovine capillary endothelial cells or a human endothelial cell line was also inhibited by digestion with heparinase or heparitinase but not with chondroitinase ABC. In contrast, binding to glial cell-enriched brain cell cultures or to a neopallial cell line was inhibited by all three lysases. Binding of strain N40 to immobilized heparin could be completely inhibited by dermatan sulfate, and conversely, binding to dermatan sulfate could be completely blocked by heparin. As measured by 50% inhibitory dose, heparin was a better inhibitor of binding than dermatan sulfate, regardless of whether the substrate was heparin or dermatan sulfate. These results are consistent with the hypotheses that the species of proteoglycans recognized by *B. burgdorferi* vary with cell type and that bacterial recognition of different proteoglycans is mediated by the same bacterial molecule(s).

Lyme disease is a chronic, multisystemic infection caused by the tick-borne spirochete *Borrelia burgdorferi* (40). The spirochete initially establishes an infection at the site of the tick bite and then migrates through the skin, resulting in the characteristic expanding rash, erythema migrans. As the infection progresses, the bacterium can spread via the bloodstream to multiple sites, such as the joints, heart, skin, or nervous system. During this phase of the infection, Lyme disease patients may experience arthralgia, carditis, secondary erythema migrans lesions, or neurologic manifestations such as meningitis, cranial neuritis, or radiculoneuritis (40). Chronic infection may be established in at least some of these tissues, as reflected by the late manifestations of Lyme disease, which include arthritis and a variety of neurological syndromes, such as encephalopathy or polyradiculoneuropathy (15, 25, 30). The spirochete has been detected at a variety of these sites in Lyme disease patients and infected laboratory animals (2, 34, 36). Thus, although the pathophysiological mechanisms that result in the manifestations of Lyme disease are not known in detail, infection of the affected tissues is likely to be the critical trigger.

Most bacterial pathogens are able to attach to host cells in the target tissue, a step that is thought to contribute to the establishment of an infectious niche at that site (3, 4). During blood-borne dissemination from the site of tick inoculation, *B. burgdorferi* must cross the endothelial cell barrier. Reflecting this property, the spirochete has the ability to bind to and cross confluent endothelial cell monolayers in vitro (7, 8, 32, 42). Furthermore, the ability of *B. burgdorferi* to infect multiple tissues may result in part from its ability to bind to many different cell types, including neuroglia (15, 16), epithelial cells (44), fibroblasts (20, 28), lymphocytes (9), and platelets (5, 14).

*B. burgdorferi* binds to several classes of host cell molecules expressed on the cell surface or in the extracellular matrix. For example, the platelet-specific integrin αvβ3 and two widely expressed integrins, α5β1 and αvβ1, mediate bacterial attachment to human cells (5, 6), while galactocerebroside promotes attachment to Schwann cells (17). In addition, *B. burgdorferi*, like many other microbial pathogens (39), binds to proteoglycans (19, 23, 29). Proteoglycans consist of core proteins covalently linked to long, linear, negatively charged disaccharide repeats, termed glycosaminoglycans (27). They are widely expressed and involved in diverse biological phenomena, such as cell adhesion and migration, tumor metastasis, cell signaling, and hemostasis. Depending on the composition of the disaccharide repeat and the overall extent of sulfation, glycosaminoglycans can be classified into several different species, including heparin, heparan sulfate (formerly called heparitin sulfate), chondroitin-4-sulfate (chondroitin sulfate A), chondroitin-6-sulfate (chondroitin sulfate C), dermatan sulfate (chondroitin sulfate B), and keratan sulfate.

Because of their ubiquitous expression on the cell surface and in the extracellular matrix, proteoglycans could mediate spirochetal attachment to diverse tissues. *B. burgdorferi* attachment to monkey kidney (Vero) cells is mediated by heparan sulfate (29). The spirochete binds to heparan sulfate and dermatan sulfate on human epithelial (HeLa) cells (23) and to decorin, a dermatan sulfate/chondroitin sulfate proteoglycan.
associated with collagen fibrils (19). It is not clear whether binding to different proteoglycans is mediated by the same bacterial molecule(s) or by multiple molecules, each of which recognizes a distinct subset of proteoglycans. The present study was designed to investigate whether (i) the attachment of *B. burgdorferi* to diverse cell types is mediated by glycosaminoglycans, (ii) the host cell determinants of these interactions exhibit cell type specificity, and (iii) binding to different classes of glycosaminoglycans is likely to be mediated by the same bacterial molecule(s).

**MATERIALS AND METHODS**

**Bacteria and mammalian cells.** *B. burgdorferi* N40, clone D10/E9, is an infectious *B. burgdorferi* (serum stricto) isolate (5). These strains were cultured in MKP base medium (MKP-S) supplemented with human serum as described previously (5, 37). Briefly, 100 ml of 10× CMRL medium, 3 g of neopeptone, 6 g of HEPES, 0.7 g of sodium citrate, 3 g of glucose, 0.8 g of sodium pyruvate, 0.8 g of N-acetylglucosamine, and 2 g of sodium bicarbonate were added to 900 ml of distilled H2O, and the pH was adjusted to 7.6. Then 200 ml of autodewater 7% gelatin, 35 ml of 35% bovine serum albumin (BSA) filtered through a 0.45-μm-pore-size filter, and 70 ml of heat-inactivated human serum were added prior to filtration through a 0.22-μm-pore-size filter. Radiolabeled *B. burgdorferi* was prepared by growth on modified MKP medium supplemented with 100 Ci of [35S]methionine per ml, washed, and stored as aliquots at −80°C as previously described (5).

Vero cells were cultured in RPMI 1640 supplemented with 10% NuSerum (Collaborative Research). 293 human embryonic kidney cells were cultured in a 1:1 mix of Dulbecco modified Eagle medium (DMEM; low glucose; Gibco-BRL, Bethesda, Md.) and Ham’s F12 medium (Gibco-BRL) supplemented with 10% fetal bovine serum (FBS). Primary bovine endothelial cells, provided by Judah Folkman, Catherine Butterfield, and Marsha Moses (13), were grown on gelatin-coated plastic in DMEM (low glucose; Gibco-BRL) supplemented with 10% newborn calf serum and 3 ml of basic fibroblast growth factor (Gibco-BRL) per ml. EA-HY926 is a human endothelial cell line that expresses a wide range of differentiated endothelial cell markers, such as von Willebrand factor antigen, Weibel-Palade bodies, and factor VIII-related antigen with morphological distributions similar to those of primary endothelial cells (11, 12). These cells, provided by Cora-Jean Edgell, were cultured in DMEM (high glucose; Gibco-BRL) supplemented with 1% hypoxanthine-aminopterin-thymidine (Gibco-BRL) and 10% FBS. Primary cultures of telencephalic cells, consisting primarily of astrocytes, with approximately 5% oligodendrocytes and 10% microglia, were prepared from 1- to 2-day-old Sprague-Dawley rats. This cell preparation responds to *B. burgdorferi* by the production of nitric oxide and interleukin-6 and was prepared as described previously (43). The CATHa cell line, provided by Dona Chikaraishi, was derived from a tumor induced in mice by a transgene provided by Cora-Jean Edgell, were cultured in DMEM (high glucose; Gibco-BRL) supplemented with 10% NuSerum (Collaborative Research). 293 human embryonic kidney cells were cultured in a 1:1 mix of Dulbecco modified Eagle medium (DMEM; low glucose; Gibco-BRL, Bethesda, Md.) and Ham’s F12 medium (Gibco-BRL) supplemented with 10% newborn calf serum and 3 ml of basic fibroblast growth factor (Gibco-BRL) per ml. EA-HY926 is a human endothelial cell line that expresses a wide range of differentiated endothelial cell markers, such as von Willebrand factor antigen, Weibel-Palade bodies, and factor VIII-related antigen with morphological distributions similar to those of primary endothelial cells (11, 12). These cells, provided by Cora-Jean Edgell, were cultured in DMEM (high glucose; Gibco-BRL) supplemented with 1% hypoxanthine-aminopterin-thymidine (Gibco-BRL) and 10% FBS. Primary cultures of telencephalic cells, consisting primarily of astrocytes, with approximately 5% oligodendrocytes and 10% microglia, were prepared from 1- to 2-day-old Sprague-Dawley rats. This cell preparation responds to *B. burgdorferi* by the production of nitric oxide and interleukin-6 and was prepared as described previously (43). The CATHa cell line, provided by Dona Chikaraishi, was derived from a tumor induced in mice by a transgene provided by Cora-Jean Edgell, were cultured in DMEM (high glucose; Gibco-BRL) supplemented with 10% NuSerum (Collaborative Research). 293 human embryonic kidney cells were cultured in a 1:1 mix of Dulbecco modified Eagle medium (DMEM; low glucose; Gibco-BRL, Bethesda, Md.) and Ham’s F12 medium (Gibco-BRL) supplemented with 10% newborn calf serum and 3 ml of basic fibroblast growth factor (Gibco-BRL) per ml. EA-HY926 is a human endothelial cell line that expresses a wide range of differentiated endothelial cell markers, such as von Willebrand factor antigen, Weibel-Palade bodies, and factor VIII-related antigen with morphological distributions similar to those of primary endothelial cells (11, 12). These cells, provided by Cora-Jean Edgell, were cultured in DMEM (high glucose; Gibco-BRL) supplemented with 1% hypoxanthine-aminopterin-thymidine (Gibco-BRL) and 10% FBS. Primary cultures of telencephalic cells, consisting primarily of astrocytes, with approximately 5% oligodendrocytes and 10% microglia, were prepared from 1- to 2-day-old Sprague-Dawley rats.

**Quantitation of bacterial attachment to mammalian cells.** One to two days prior to each assay, the mammalian cells that were to be tested were lifted and plated in Nunc 96-well break-apart microtiter plates coated with *Yersinia pseudotuberculosis* invasin protein, which promotes cell attachment by binding a subset of glycosaminoglycans (19). It is not clear whether binding to different proteoglycans is mediated by the same bacterial molecule(s) or by multiple molecules, each of which recognizes a distinct subset of proteoglycans. The present study was designed to investigate whether (i) the attachment of *B. burgdorferi* to diverse cell types is mediated by glycosaminoglycans, (ii) the host cell determinants of these interactions exhibit cell type specificity, and (iii) binding to different classes of glycosaminoglycans is likely to be mediated by the same bacterial molecule(s).

**RESULTS**

**Bacterial attachment to Vero and 293 cells is mediated by different species of glycosaminoglycans.** To investigate proteoglycan-mediated attachment of *B. burgdorferi* to host cells, Vero cells and 293 cells were infected with the infectious *B. burgdorferi* strain N40, clone D10/E9. Patterns of inhibition of N40 binding to both cell lines by different proteoglycans were remarkably similar: heparin, heparan sulfate, and dermatan sulfate showed better inhibitory activity than chondroitin-4-sulfate or chondroitin-6-sulfate (Fig. 1A). As previously shown (29), Vero cell binding was inhibited by digestion of the monolayer with heparinase, which cleaves heparin-related glycosaminoglycans, and with heparitinase, which...
cleaves heparan sulfate proteoglycans (Fig. 1B). Chondroitinase ABC, which cleaves chondroitin-4-sulfate, dermatan sulfate, and chondroitin-6-sulfate, had no effect on binding to this cell line. In contrast, parallel digestions with heparinase or heparitinase had no significant effect on N40 binding to 293 cells, while chondroitinase ABC digestion inhibited attachment by more than 60% (Fig. 1B). These results indicate that glycosaminoglycans play an important role in recognition of both cell lines by *B. burgdorferi*, but that the specific class of proteoglycans that plays the major role in bacterial attachment varies with host cell.

**Binding of *B. burgdorferi* to heparan sulfate on endothelial cells.**

To determine which, if any, class of glycosaminoglycan might play a role in bacterial interactions with the endothelium, we investigated bacterial binding to both primary bovine capillary endothelial cells and to a human endothelial cell line. Binding of N40 to primary endothelial cells was almost completely inhibited by heparin or dextran sulfate, partially inhibited by dermatan sulfate, but only minimally inhibited by chondroitin-4-sulfate or chondroitin-6-sulfate (Fig. 2A, Soluble Inhibitor). Pretreatment of the cells with heparinase or heparitinase diminished N40 binding by 50%, whereas digestion with chondroitinase ABC had no effect (Fig. 2A, Lyase).

EA-Hy926 is a human endothelial cell line that expresses a wide range of differentiated endothelial cell markers (11, 12). N40 attachment to this cell line was inhibited by platelet factor 4, a chemokine that binds to glycosaminoglycans, and by heparin but not by chondroitin-6-sulfate (Fig. 2B, Soluble Inhibitor). Bacterial binding was blocked by pretreatment of EA-Hy926 cells with β-D-xyloside, which inhibits linkage of the heparin/heparan sulfate and dermatan/chondroitin sulfate chains to the protein core of proteoglycans (26). Pretreatment with a control sugar, α-D-galactoside, had no effect (Fig. 2B, Inhibitor of Synthesis). Sulfation is apparently required for attachment of *B. burgdorferi* to EA-Hy926 cells, because pre-treatment of EA-Hy926 cells with chlorate, an inhibitor of proteoglycan sulfation (1), reduced N40 attachment by almost 90% (Fig. 2B, Inhibitor of Sulfation). This effect was specific for chlorate: chloride treatment had no effect, and the addition of sulfate along with chloride partially reversed the inhibition of binding. Pretreatment of EA-Hy926 cells with heparinase or heparitinase reduced binding by 90%, while chondroitinase ABC treatment had little effect (Fig. 2B, Lyase). Taken together with the analysis of primary endothelial cells, these experiments provide evidence that proteoglycans are critical in the attachment of *B. burgdorferi* to endothelial cells and that heparin/heparan sulfate in particular play an important role in this recognition process. In contrast, there was no evidence that chondroitin or dermatan sulfate proteoglycans were required for efficient bacterial binding to endothelial cells.

**B. burgdorferi** binding to heparinase- and chondroitinase-sensitive proteoglycans expressed by cultured brain cells. To investigate the interaction of *B. burgdorferi* with a second potential target tissue, a glial cell-enriched population of primary mixed telencephalic brain cells was cultured from neonatal rats (43). Heparin, dextran sulfate, and platelet factor 4 reduced N40 binding, as did inhibition of sulfation by pretreatment with the sulfation inhibitor chlorate (Fig. 3A). Digestion of cultured telencephalic cells with heparinase or heparitinase diminished bacterial attachment by about 60% (Fig. 3A, Lyase), indicating that heparan sulfate proteoglycans expressed by telencephalic cells contribute to bacterial binding, as was the case for binding to endothelial cells. In contrast to the results with endothelial cells however, chondroitinase ABC digestion of telencephalic cells inhibited N40 attachment by about 45%. These results suggest that for this population of neural cells, chondroitinase-sensitive as well as heparinase-sensitive proteoglycans contribute to bacterial recognition.

To determine which species of proteoglycans contribute to *B. burgdorferi* attachment to neuronal cells, we assayed the effect of lyase digestion on bacterial binding to the catecholaminergic neuron-derived CATH.a cells (41). Heparinase, heparitinase, and chondroitinase ABC digestion of CATH.a cells each resulted in a significant inhibition of N40 attachment (Fig. 3B), indicating that a chondroitinase-sensitive component of CATH.a cells promotes attachment of *B. burgdorferi*. Lyase digestion of the pheochromocytoma cell line, PC12, gave similar results (data not shown). Thus, in this sampling of two neuronal cell lines and central nervous system-derived primary
neurons and glia, chondroitinase ABC-sensitive proteoglycans contributed to *B. burgdorferi* attachment.

**Dermatan sulfate inhibits bacterial binding to immobilized heparin, and heparin inhibits binding to immobilized dermatan sulfate.** The analysis of lyase-treated Vero and 293 cells (Fig. 1B) is consistent with the suggestion that heparin/heparan sulfate mediates *B. burgdorferi* attachment to Vero cells, while a chondroitinase ABC-sensitive glycosaminoglycan, e.g., dermatan sulfate, mediates attachment to 293 cells. The observed cell-specific difference in the species of glycosaminoglycans recognized by *B. burgdorferi* could be due to the expression of a single glycosaminoglycan-binding molecule that recognizes both heparan sulfate and dermatan sulfate. Alternatively, the spirochete could utilize two independent mechanisms, one recognizing heparin/heparan sulfate and the other recognizing dermatan sulfate. In the latter case, one might expect that Vero cell binding would be inhibited most efficiently by heparin, whereas dermatan sulfate would be the more effective inhibitor of attachment to 293 cells. Heparin, dermatan sulfate, and chondroitin-6-sulfate were titrated as inhibitors of strain N40 attachment to Vero or 293 cells. Both heparin and dermatan sulfate blocked binding to both cell lines, and regardless of cell line, heparin was the best inhibitor of N40 attachment, with a 50% inhibitory concentration (IC$_{50}$) 4- to 45-fold lower than that for dermatan sulfate (Table 1).

Because intact mammalian cells present a complex mixture of potential receptors for *B. burgdorferi*, we also determined whether bacterial binding to immobilized heparin could be inhibited by dermatean sulfate and, conversely, whether binding to immobilized dermatan sulfate could be inhibited by heparin. Each proteoglycan inhibited binding of *B. burgdorferi* N40 to the other (Fig. 4). Heparin was the more potent inhibitor regardless of which glycosaminoglycan was immobilized, with an IC$_{50}$ three- to sixfold lower than that for dermatan sulfate (Table 1). Chondroitin-6-sulfate showed some inhibitory activity but was the poorest inhibitor of the three tested. That heparin was the most potent inhibitor of *B. burgdorferi* N40 attachment to purified heparin, dermatan sulfate, Vero cells, or 293 cells is consistent with the hypothesis that a single glycosaminoglycan-binding pathway recognizes multiple species of glycosaminoglycans.

**DISCUSSION**

Given that endothelial damage is one of the hallmarks of Lyme disease (2, 10) and that neurologic manifestations are a prominent feature of this illness (30), it is likely that the interactions of *B. burgdorferi* with endothelial cells and cells in the nervous system play an important role in vivo. We analyzed bacterial attachment to primary endothelial cells, primary central nervous system-derived glial cells, and cell lines of endothelial or neural origin and found that binding of *B. burgdorferi* to all of these cells was mediated by proteoglycans. Binding to all cells tested was inhibited by the addition of heparin, heparan sulfate, or dermatan sulfate and by platelet factor 4, a chemokine that binds glycosaminoglycans. In addition, cell attachment was diminished by pretreatment of these cells with an inhibitor of proteoglycan synthesis (β-d-xyloside) or sulfation (sodium chlorate) or by digestion of cell surface glycosaminoglycans with lyases. Given the widespread expression of proteoglycans, it is likely that proteoglycan binding by the spirochete contributes to the recognition of other cell types as well.

Although glycosaminoglycans mediate attachment to many cell types, removal of specific classes of glycosaminoglycans with lyases indicated that the particular populations of glycosaminoglycans that contribute to spirochetal attachment vary with cell type. Heparin/heparan sulfate appeared to play the most critical role for spirochetal binding to primary endothelial

**TABLE 1. Comparison of heparin and dermatan sulfate as inhibitors of bacterial attachment to immobilized proteoglycans or mammalian cells**

<table>
<thead>
<tr>
<th>Inhibitor</th>
<th>Vero cells</th>
<th>293 cells</th>
<th>Heparin</th>
<th>Dermatan sulfate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heparin</td>
<td>8.0</td>
<td>1.3</td>
<td>0.2</td>
<td>0.5</td>
</tr>
<tr>
<td>Dermatan sulfate</td>
<td>32</td>
<td>60</td>
<td>0.6</td>
<td>3.2</td>
</tr>
<tr>
<td>Chondroitin-6-sulfate</td>
<td>&gt;500</td>
<td>&gt;500</td>
<td>&gt;500</td>
<td>18</td>
</tr>
</tbody>
</table>

* a Bacterial attachment of radiolabeled *B. burgdorferi* N40 to mammalian cells or immobilized proteoglycans was determined in the presence of various concentrations of heparin, dermatan sulfate, or chondroitin-6-sulfate (see Materials and Methods). At the concentration of inhibitor indicated, bacterial attachment was 50% of the level of binding in the absence of inhibitor (Fig. 4).
cells, the EA-Hy926 endothelial cell line, and Vero cells. A chondroitinase ABC-sensitive glycosaminoglycan, presumably dermatan sulfate, mediated attachment to 293 cells. Binding to primary telencephalon cells and two neuronal cell lines appeared to be mediated by both heparin/heparan sulfate and dermatan sulfate. It was previously shown that binding of *B. burgdorferi* 297 to HeLa cells was inhibited by digestion with either heparinase or chondroitinase ABC (23). Of the glycosaminoglycans that are substrates for chondroitinase ABC (chondroitin-4-sulfate, chondroitin-6-sulfate, and dermatan sulfate), only dermatan sulfate is a potent inhibitor of bacterial attachment to mammalian cells. Thus, it is likely that the chondroitinase ABC-sensitive glycosaminoglycan that is critical for *B. burgdorferi* recognition is dermatan sulfate. Consistent with this hypothesis, chondroitinase AC, which does not cleave dermatan sulfate, had no effect on *B. burgdorferi* attachment to HeLa cells (23).

At the present time, we do not know whether the relative importance of a given species of glycosaminoglycan in bacterial binding to a particular cell type reflects its relative affinity for the bacterium, its abundance on the cell surface, or both. Heparan sulfate and chondroitin sulfate are expressed to a variable extent on virtually all cells, while dermatan sulfate is less common (27). Although dermatan sulfate did not appear to participate in bacterial binding to endothelial cells in this study, cultured bovine aortic endothelial cells have been shown to express this glycosaminoglycan (35). Dermatan sulfate appeared to promote bacterial attachment to cultured rat brain cells, even though it is apparently poorly expressed in rat brain (21, 33). It is difficult, however, to directly compare these previous studies to our results, because proteoglycan expression can vary considerably with culture conditions. For this reason, multiple representatives of both endothelial and neural cells were evaluated in this study.

The recognition of multiple classes of proteoglycans by *B. burgdorferi* could reflect the expression of several proteoglycan receptors, or the expression of a single receptor that recognizes different species of glycosaminoglycans. We found no evidence for independent mechanisms for binding dermatan sulfate and heparin by strain N40: heparin was a better inhibitor than dermatan sulfate, regardless of whether the substrate was heparin, dermatan sulfate, Vero cells, or 293 cells. Previous results demonstrating that 100 μg of heparan sulfate or dermatan sulfate per ml partially (46 or 59%, respectively) inhibits heparin binding by *B. burgdorferi* also suggest a promiscuous glycosaminoglycan-binding pathway that binds heparin with the highest affinity (23). It is not uncommon for glycosaminoglycan-binding receptors to recognize multiple species of glycosaminoglycans, because the polyanionic nature of these molecules is a critical determinant in these interactions (27).

Although our results are consistent with a single proteoglycan-binding mechanism, we cannot rule out the possibility that binding of one proteoglycan to the surface of the spirochete can inhibit attachment to other molecules nonspecifically, e.g., by steric hindrance, or by conferring a strong negative charge to the bacterial surface. Guo et al. showed that binding of *B. burgdorferi* B31 to decorin, a dermatan sulfate/chondroitin sulfate proteoglycan, was not inhibited by 10 μg of heparin per ml, whereas the same concentration of exogenous decorin blocked binding (19). This finding could indicate that the decorin binding and heparin binding are mediated by different bacterial molecules. Alternatively, the specificity of proteoglycan binding varies somewhat among strains of Lyme disease spirochete (35a), and strain B31 may express a proteoglycan-binding receptor that binds to decorin with much higher affinity than it does to heparin. Resolution of the question of one versus multiple proteoglycan-binding pathways awaits further characterization of the bacterial molecules that mediate the varied interactions that have been described to date.

While it is clear that proteoglycan recognition promotes bacterial attachment to a wide variety of cells, these results do not preclude the involvement of other host molecules, and the degree of residual binding upon inhibition of the proteoglycan pathway may reflect the activity of additional binding pathways. For example, integrins contribute to bacterial attachment to platelets (5) and other cell types (6), while galactocerebroside promotes binding to Schwann cells (17). Glycosaminoglycans often act in concert with other cell surface receptors to promote ligand binding (22, 38, 45), and attachment to proteoglycans by the Lyme disease spirochete could facilitate binding to other classes of molecules. The precise sequence of events that occur during bacterial attachment to host cells, as well as the way in which this interaction may promote colonization of specific tissues, will be the subject of future investigations.
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