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Cryptococcus neoformans, *Candida albicans*, and Other Fungi Bind Specifically to the Glycosphingolipid Lactosylceramide (Galβ1-4Glcβ1-1Cer), a Possible Adhesion Receptor for Yeasts

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The role of glycosphingolipids as adhesion receptors for yeasts was examined. *Cryptococcus neoformans*, *Candida albicans*, and *Saccharomyces cerevisiae*, as well as *Histoplasma capsulatum* and *Sporotrichum schenckii* (in their yeast phases), bound specifically to lactosylceramide (Galβ1-4Glcβ1-1Cer), as measured by overlaying glycosphingolipid chromatograms with ^{125}I-labeled organisms. An unsubstituted galactosyl residue was required for binding, because the yeasts did not bind to glucosylceramide (Glcβ1-1Cer) derived from lactosylceramide by treatment with β-galactosidase or to other neutral or acidic glycosphingolipids tested that contained internal lactosyl residues. Interestingly, the yeasts preferentially bound to the upper band of the lactosylceramide doublet in human lung and bovine erythrocytes, suggesting that the ceramide structure also affects binding. Active metabolism of the yeasts was required for binding to lactosylceramide, as binding was maximal in buffer containing glucose and was almost completely abolished in nutrient-deficient medium. *C. neoformans* also bound to human glioma brain cells grown in monolayers, and this binding was inhibited by liposomes containing lactosylceramide but not by liposomes containing glucosylceramide. Lactosylceramide is a major glycosphingolipid in these cells and the only one to which the yeasts bound. As lactosylceramide is widely distributed in epithelial tissues, this glycosphingolipid may be the receptor for yeast colonization and disseminated disease in humans.

Yeasts are unicellular fungi that are widely distributed in nature and that can colonize humans and animals, often without harm. For example, *Candida albicans* and other yeasts are commensal colonizers of skin and mucosal tissues, and *Crytpococcus neoformans* may colonize the lung asymptomatically (9, 19). The disease caused by these opportunistic yeasts is influenced by the host response, and severe disseminated fungal syndromes can occur in immunocompromised patients (1). In patients with the acquired immunodeficiency syndrome, even yeasts with relatively low virulences, such as *Saccharomyces cerevisiae*, can cause severe disease (21).

Adhesion to host tissues is a critical step in colonization and subsequent infection by yeasts (2, 4, 13, 18, 20). Prior work to define yeast adhesion receptors by inhibition of binding by various sugars has been inconclusive (2, 4, 13, 18). Because animal cell surface glycosphingolipids (7) are adhesion receptors for many bacteria (8, 10–12, 14, 24, 25, 27) and viruses (16, 17), we investigated their possible role in yeast adhesion. In the present report we show that *C. neoformans* and *S. cerevisiae*, as well as *C. albicans*, *Histoplasma capsulatum*, and *Sporotrichum schenckii* in their yeast phases, bind specifically to lactosylceramide (see Table 1 for structures) and that this glycosphingolipid is probably responsible for the adhesion of yeasts to host tissues.

MATERIALS AND METHODS

Glycosphingolipids. Galactosylceramide, glucosylceramide, trihexosylceramide, globoside, and sulfatide were purchased from Supelco (Bellefonte, Pa). Bovine brain lactosylceramide was obtained from Sigma Chemical Co. (St. Louis, Mo.). Bovine erythrocyte lactosylceramide was obtained from M. Kyogashima (National Institutes of Health). Semisynthetic lactosylceramide (DL-dihydrolactocerebroside), which contains a short-chain synthetic fatty acid (*N*-palmitoyl), was obtained from Calbiochem-Behring (La Jolla, Calif.). Gangliotetraosylceramide (asialo-GM1), gangliotriaosylceramide (asialo-GM2), and paragloboside were prepared as described previously (11, 12). Lipids from normal lung and human polymorphonuclear leukocytes were extracted and separated into neutral and acidic fractions by anion-exchange chromatography on DEAE-Sepharose (Pharmacia, Inc., Piscataway, N.J.) in the bicarbonate form, as described previously (11). Human glioma brain cells were extracted as described above, phase partitioned (6), and subjected to silicic acid chromatography (Biosil; Bio-Rad Laboratories, Richmond, Calif.) (29). Glucosylceramide was prepared from bovine erythrocyte lactosylceramide by treatment with bovine testis β-galactosidase as described previously (11). Polar contaminants and detergent were removed by Sephadex G-25 and DEAE-Sepharose column chromatography as described previously (11, 15). Liposomes containing glycosphingolipids were prepared as follows. One milligram of lactosylceramide or glucosylceramide was added to cholesterol and phosphatidylcholine in a ratio of 3:2:4, by weight. The lipids were mixed in $CHCl_3$-CH_3OH (1:1), dried under nitrogen, and sonicated for 5 min in 0.01 M sodium phosphate (pH 7.2) containing 0.15 M sodium chloride (phosphate-buffered saline [PBS]). The liposomes were centrifuged at 4°C and 10,000 × *g* for 10 min, and pellets were washed twice with PBS and suspended to 1 mg of glycosphingolipid per ml of PBS.

Growth and labeling of fungi. The strains used in this study are listed in Table 2. All fungi were grown in yeast nitrogen base medium (Difco Laboratories, Detroit, Mich.) with 1%

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glucose (J. T. Baker Chemical Co., Phillipsburg, N.J.) without shaking at 26 or 37°C with the exception of the dimorphic fungi *H. capsulatum* and *S. schenckii*, which were grown only at 37°C as required for production of the yeast phase (19). After 2 to 7 days of growth, yeasts were harvested, washed twice by centrifugation (1,000 × *g* for 10 min) in PBS, and suspended to 2×10^8 cells per ml of PBS. Yeasts were radioiodinated as described previously for bacteria (11), with minor modifications. Briefly, 0.5 ml of the fungal suspension was reacted with 1 mCi of $Na^{125}I$ at 4°C in a tube (10 by 75 mm) coated with 100 μg of Iodogen (Pierce Chemical Co., Rockford, Ill.). After 5 min, the suspension was transferred to a tube containing 0.5 ml of Hanks balanced salt solution (HBSS) containing 1% bovine serum albumin (BSA; pH 7.4), washed by centrifugation (1,000 × *g*, 10 s), and suspended to 10^6 to 10^7 cpm/ml of HBSS-BSA. Expression of the *C. albicans* hyphal phenotype was obtained after incubating the yeasts in RPMI 1640 medium (GIBCO Laboratories, Grand Island, N.Y.) for 2 h at 37°C in 5% CO_2 (19). For some experiments, yeasts were fixed with 0.5% glutaraldehyde in PBS at 0°C for 10 min and then washed with PBS.

Chromatogram overlay assay for binding of fungi to glycosphingolipids. The chromatogram overlay assay was performed as described previously for bacteria (8, 11). Briefly, glycosphingolipids were chromatographed on aluminum-backed silica gel high-performance thin-layer plates (E. Merck AG, Darmstadt, Federal Republic of Germany) developed with chloroform–methanol–0.25% aqueous KCl (5:4:1). The plates were coated with polyisobutylmethacrylate (0.1% in hexane), air dried, soaked for 1 h in Tris-BSA buffer (0.05 M Tris hydrochloride [pH 7.8] containing 0.15 M sodium chloride and 1% BSA), and overlaid for 3 h at room temperature with ^{125}I-labeled fungi (2×10^6 cpm/ml of HBSS-BSA). The plates were gently washed to remove unbound organisms, dried, and exposed for 24 h to XAR-5 X-ray film (Eastman Kodak Co., Rochester, N.Y.).

Solid-phase assay for binding of fungi to glycosphingolipids. The solid-phase binding assay was performed as described previously for bacteria (11). Briefly, serial dilutions of purified glycosphingolipids in methanol (25 μl) containing cholesterol and phosphatidylcholine (0.1 μg each) were added to polyvinylchloride microdilution wells (Falcon 3912-111; Becton Dickinson and Co., Paramus, N.J.) and dried by evaporation. The wells were blocked with Tris-BSA for 1 h, rinsed with HBSS-BSA twice, and incubated with 25 μl of ^{125}I-labeled yeasts (10^7 cpm/ml of HBSS) for 3 h at 25°C. After the wells were washed five times with PBS, the polyvinylchloride wells were cut with scissors and placed in counting tubes. Binding was quantified in a gamma spectrometer.

Fungal adhesion to cultured human glioma cells. Human glioma brain cells (ATCC HTB 138) were grown in Dulbecco modified Eagle medium supplemented with 10% fetal bovine serum at 37°C in 5% CO_2–95% air. Cells were grown until they formed confluent monolayers, usually 7 days, on round cover slips (diameter, 13 mm) placed in 24-well tissue culture plates. For some experiments, cells were fixed in 0.5% glutaraldehyde at 4°C for 1 h. The wells were washed with serum-free medium and then incubated with 1 ml of ^{125}I-labeled fungi (2×10^6 cpm/ml of HBSS). After incubation for the indicated periods, the cover slips were washed five times with PBS and the bound radioactivity was measured. For inhibition tests, the fungi were preincubated for 1 h at 25°C with liposomes or free sugars.

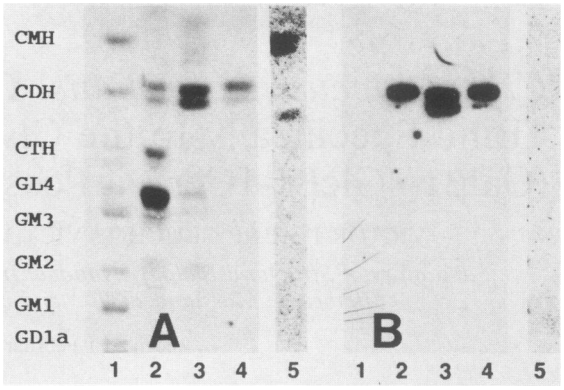

FIG. 1. Binding of *C. neoformans* to glycosphingolipids separated on thin-layer chromatograms. Glycosphingolipids were extracted and chromatographed in duplicate high-performance thin-layer plates as described in the text. Glycosphingolipids on the chromatograms were visualized with orcinol reagent (A) or overlaid with ^{125}I-labeled cryptococci followed by autoradiography (B). Lanes 1, 2 μg of galactosylceramide (CMH); 1 μg of chemically synthesized lactosylceramide (CDH); 1 μg of trihexosylceramide doublet (CTH); 1 μg of globoside (GL4); and 2 μg each of the gangliosides GM3, GM2, GM1, and GD1a; lanes 2, neutral glycosphingolipids from 100 mg (wet weight) of human lung tissue; lanes 3, neutral glycosphingolipids from 2×10^6 polymorphonuclear cells; lanes 4, 1 μg of purified bovine erythrocyte lactosylceramide (doublet); lanes 5, 2 μg of glucosylceramide derived from lactosylceramide by treatment with β-galactosidase. Similar binding patterns were also observed with the other fungi listed in Table 2 (data not shown).

RESULTS

Binding of *C. neoformans* and other fungi to glycosphingolipids on chromatograms. The binding of ^{125}I-labeled *C. neoformans* to glycosphingolipids from normal human lung and human polymorphonuclear leukocytes was examined by the chromatogram overlay assay. As shown by a thin-layer chromatogram of glycosphingolipids detected by orcinol reagent (Fig. 1A) compared with an identical chromatogram overlaid with radiolabeled yeasts and subjected to autoradiography (Fig. 1B), the organisms bound to pure bovine erythrocyte lactosylceramide (Fig. 1B, lane 4) and to glycosphingolipids from human lung (Fig. 1B, lane 2) and human polymorphonuclear cells (Fig. 1B, lane 3) with the same mobilities. That these glycosphingolipids from human tissues were indeed lactosylceramide was shown by their specific immunostaining (15) with monoclonal antibody A_5T_7, which is directed against lactosylceramide (26) (data not shown). Interestingly, no binding was detected to semisynthetic lactosylceramide (DL-dihydrolactocerebroside) (Fig. 1B, lane 1), which contains a short-chain *N*-palmitoyl fatty acid. Also, while binding was better to the upper band than it was to the lower band of the lactosylceramide doublet obtained from human lung (Fig. 1B, lane 2) and bovine erythrocytes (Fig. 2B, lane 4), binding was the same for both bands of lactosylceramide from human polymorphonuclear cells (Fig. 1B, lane 3). The cryptococci did not bind to glucosylceramide derived from lactosylceramide by treatment with β-galactosidase (Fig. 1B, lane 5) or to the other neutral or acidic glycosphingolipids tested that contained internal lactosyl residues (Fig. 1B, lane 1, and Table 1). Lactosylceramide from bovine erythrocytes separated chromatographically into approximately equal amounts of upper and lower bands (Fig. 2A, lane 4). The ceramide moiety in

TABLE 1. Structures of glycosphingolipids tested for binding of yeasts

Glycosphingolipid (abbreviation)	Structure
Galactosylceramide (CMH)	Galβ1-1Cer
Glucosylceramide	Glcβ1-1Cer
Lactosylceramide (CDH)	Galβ1-4Glcβ1-1Cer
Trihexosylceramide (CTH)	Galα1-4Galβ1-4Glcβ1-1Cer
Paragloboside	Galβ1-4GlcNAcβ1-3Galβ1-4Glcβ1-1Cer
Globoside (GL4)	GalNAcβ1-3Galα1-4Galβ1-4Glcβ1-1Cer
Gangliotriaosylceramide (asialo GM2)	GalNAcβ1-4Galβ1-4Glcβ1-1Cer
Gangliotetraosylceramide (asialo-GM1)	Galβ1-3GalNAcβ1-4Galβ1-4Glcβ1-1Cer
GM3	NeuAcα2-3Galβ1-4Glcβ1-1Cer
GM2	GalNAcβ1-4[NeuAcα2-3]Galβ1-4Glcβ1-1Cer
GM1	Galβ1-3GalNAcβ1-4[NeuAcα2-3]Galβ1-4Glcβ1-1Cer
GD1a	NeuAcα2-3Galβ1-3GalNAcβ1-4[NeuAcα2-3]Galβ1-4Glcβ1-1Cer

the upper band contains predominantly $C_{24:0}$, $C_{24:1}$, and $C_{22:0}$ fatty acids; the ceramide moiety in the lower band contains predominantly $C_{16:0}$ and $C_{18:0}$ fatty acids (28). As was the case with lactosylceramide from human lung and bovine erythrocytes (Fig. 1B, lanes 2 and 4, respectively), *C. neoformans* bound better to the upper band than it did to the lower band (Fig. 2B). Less than 50 ng of upper band lactosylceramide was detected (Fig. 2B).

The fungi that bound to lactosylceramide in the chromatogram overlay assay are listed in Table 2, and all fungi produced binding patterns similar to that shown in Fig. 1B for *C. neoformans*. Binding was detected with the yeast phase of *C. albicans*, but not with the pseudohypha phase. Also, the dimorphic fungi *H. capsulatum* and *S. schenckii* in their yeast phases bound to lactosylceramide, as did the yeast *S. cerevisiae* (data not shown).

Binding of *C. neoformans* to lactosylceramide immobilized in microdilution wells. The binding of *C. neoformans* to lactosylceramide by a solid-phase assay was measured (Fig. 3A). Binding was concentration dependent, and half-maximal binding required about 0.2 μg of lactosylceramide per well. In terms of specific activity, approximately 10% of the radiolabeled yeast cells bound to lactosylceramide in this solid-phase assay. The other yeasts listed in Table 2 also produced similar half-maximal binding values (data not shown). No binding was detected to asialo-GM1 or asialo-GM2, which contain internal lactosyl residues. The effect of viability and growth on binding was also examined (Fig. 3B). Fungi suspended in Tris-BSA buffer alone did not bind, but binding was restored by the addition of glucose to the buffer. The absence of Ca^{2+} and Mg^{2+} in the incubation medium or the addition of the chelating agents ethylene glycol-bis(β-aminoethyl ether)-*N*,*N*,*N*′,*N*′-tetraacetic acid (EGTA) and EDTA had no effect. Glutaraldehyde-fixed yeasts did not bind to lactosylceramide (data not shown).

The specificity of binding of *C. neoformans* to lactosylceramide was further defined by inhibition experiments with liposomes containing glycosphingolipids (Fig. 3C). Liposomes containing lactosylceramide, but not glucosylceramide, inhibited binding. About 6 μg of lactosylceramide per ml inhibited binding by 50%, whereas glucosylceramide at 100 μg/ml had no effect. Glucose, galactose, and lactose at 100 μg/ml did not inhibit binding (data not shown).

Adhesion of *C. neoformans* to human brain cells. Because the brain is a common site of human cryptococcal infections, the adhesion of cryptococci to human glioma cells grown in monolayers was tested. ^{125}I-labeled *C. neoformans* adhered readily to brain glioma cell monolayers, and adhesion was both time and temperature dependent (Fig. 4). Adhesion was almost complete at 2 h and was markedly reduced at 4°C as compared with that at 37°C. Concentration-dependent binding to glioma cells was also observed with *C. albicans* and *S. cerevisiae* (data not shown).

As was the case with the binding of *C. neoformans* to immobilized lactosylceramide (Fig. 3C), binding of yeasts to glioma cells was strongly inhibited (>80%) by liposomes containing lactosylceramide, whereas an equal amount of liposomes containing glucosylceramide had no effect (<4%). Glucose, galactose, and lactose at 100 μg/ml did not affect

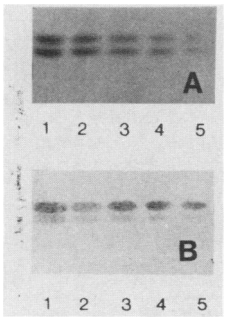

FIG. 2. Binding of *C. neoformans* to purified lactosylceramide. Various amounts of bovine erythrocyte lactosylceramide on duplicate thin-layer chromatograms were visualized with orcinol reagent (A) or overlaid with ^{125}I-labeled organisms followed by autoradiography (B), as described in the text. Lactosylceramide was used at 0.5 μg (lanes 1), 0.35 μg (lanes 2), 0.25 μg (lanes 3), 0.15 μg (lanes 4), and 0.05 μg (lanes 5).

TABLE 2. Fungi that bind specifically to lactosylceramide[a]

Fungus	Strain	Comment
Cryptococcus neoformans	ATCC 34877	Type culture
Cryptococcus neoformans	NIH 68A	Clinical isolate
Cryptococcus neoformans	IMT 43	Clinical isolate
Cryptococcus neoformans	HFH 32	Clinical isolate
Candida albicans[b]	ATCC 18804	Type culture
Candida albicans[b]	HFH 44	Clinical isolate
Candida albicans[b]	HFH 45	Clinical isolate
Histoplasma capsulatum[b]	CDC B923	Clinical isolate
Histoplasma capsulatum[b]	IMT 55	Clinical isolate
Sporotrichum schenckii[b]	CDC B4668	Clinical isolate
Sporotrichum schenckii[b]	IMT 58	Clinical isolate
Saccharomyces cerevisiae	ATCC 18824	Type culture
Saccharomyces cerevisiae	HFH 21	Clinical isolate

[a] Determined by the chromatogram overlay assay with 1 μg of purified bovine erythrocyte lactosylceramide, as described in the text.
[b] Tested in the yeast phase, as described in the text.

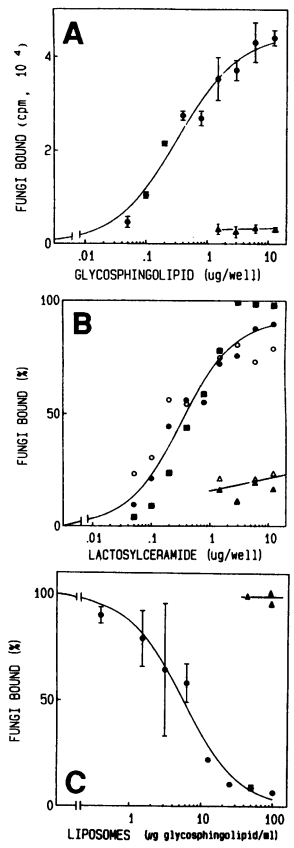

FIG. 3. Binding of *C. neoformans* to glycosphingolipids in a solid-phase assay. (A) Binding of ^{125}I-labeled *C. neoformans* (3 h at 25°C) to serial dilutions of purified glycosphingolipids immobilized in microdilution wells as described in the text. Symbols: ●, lactosylceramide; ▲, asialo-GM1 or asialo-GM2. (B) Binding of ^{125}I-labeled *C. neoformans* (percent binding relative to maximum bound) to serial dilutions of lactosylceramide in Tris-BSA buffer (▲), Tris-BSA buffer supplemented with 5 mM Ca^{2+} and Mg^{2+} (△), Tris-BSA buffer supplemented with 2 mM glucose (○), HBSS-BSA (■), and HBSS-BSA supplemented with EGTA-EDTA (●). (C) Inhibition of binding by liposomes containing glycosphingolipids. Binding of labeled *C. neoformans* to 1 μg of lactosylceramide was determined after incubation of the organisms for 1 h at 25°C with the indicated concentrations of liposomes containing lactosylceramide (●) or glucosylceramide (▲). Data represent means ± standard deviations of quadruplicate samples.

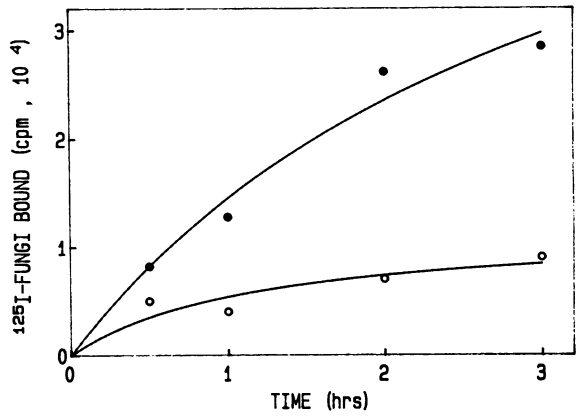

FIG. 4. Adhesion of *C. neoformans* to cultured human glioma cells. Brain glioma cells grown as monolayers on cover slips were incubated with ^{125}I-labeled organisms for various times at 37°C (●) or 4°C (○) and then washed free of unbound cryptococci. Data are expressed as bound radioactivity per cover slip as described in the text. Results represent means of quadruplicate samples.

binding (data not shown). *C. neoformans* bound as well to glutaraldehyde-fixed glioma cell monolayers as it did to unfixed monolayers (98% of control).

Lactosylceramide is a major glycosphingolipid in the glioma cell line, as shown in Fig. 5A. Labeled *C. neoformans* bound only to the glycosphingolipid in this target tissue that migrated with authentic lactosylceramide (Fig. 5B).

DISCUSSION

C. neoformans, *C. albicans*, and the other yeasts bound specifically to lactosylceramide, as measured by an overlay assay (Fig. 1 and 5 and Table 2) and a solid-phase assay (Fig. 3A). The terminal galactosyl residue of lactosylceramide is required for binding, as *C. neoformans* did not bind to glucosylceramide derived from bovine erythrocyte lactosylceramide by treatment with β-galactosidase (Fig. 1, lane 5). Substitution of the lactosyl residue with other sugars blocked binding, as the yeasts did not bind to asialo-GM1 or asialo-GM2, which are receptors for many bacterial lung pathogens (11), or to other glycosphingolipids with internal lactose sequences that were tested (Fig. 1, 3A, and 5 and Table 1). Thus, the yeasts require an unsubstituted lactosyl residue for binding, in contrast to bacteria such as *Neisseria gonorrhoeae*, *Propionibacterium granulosum*, and *Bordetella pertussis* (24, 25, 27), which bind to lactosylceramide but also bind to glycosphingolipids with internal lactosyl sequences.

The binding of *C. neoformans* to lactosylceramide is affected by the structure of the ceramide moiety, as was previously shown with antibodies (26) and bacteria (12, 25). These pathogenic fungi do not bind to semisynthetic lactosylceramide, which contains a short-chain fatty acid, and bind better to the upper band than to the lower band of the lactosylceramide doublet obtained from human lung and bovine erythrocytes (Fig. 1 and 2). The upper band of bovine erythrocyte lactosylceramide contains longer fatty acids than does the lower band (28). Thus, *C. neoformans* binds better to lactosylceramide with long fatty acids than it does

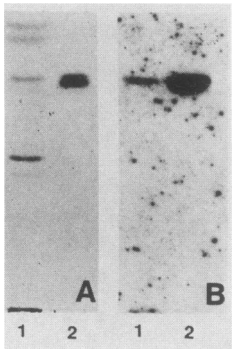

FIG. 5. Binding of *C. neoformans* to neutral glycosphingolipids from human glioma cells. Neutral glycosphingolipids from 100 mg (wet weight) of human glioma cells (lanes 1) and 1 μg of bovine brain lactosylceramide (lanes 2) were visualized with orcinol reagent (A) or overlaid with ^{125}I-labeled organisms followed by autoradiography (B).

to lactosylceramide with short fatty acids, as do some bacteria (25) and an antibody directed against lactosylceramide (26).

C. neoformans also bound strongly to brain cells (Fig. 4), a major site of infection in humans and animals (1, 19). Lactosylceramide is present in substantial amounts in brain cells and is the only glycosphingolipid to which the organism binds (Fig. 5). Furthermore, the binding of *C. neoformans* to cultured human brain cells and to purified lactosylceramide immobilized in microdilution wells was strongly inhibited by liposomes containing the receptor but not by liposomes containing glucosylceramide (Table 2 and Fig. 3C). Free lactose at the same concentration did not inhibit binding, probably because the binding of yeasts to liposomes containing lactosylceramide is multivalent and of high affinity, whereas the binding to lactose is monovalent and of low affinity, as is common with many other ligand-receptor interactions (5).

All of the yeasts that we studied require glucose but not divalent cations for optimal binding (Fig. 3B) and binding is temperature dependent (Fig. 4), suggesting that only metabolically active organisms adhere, as has been previously found for the adherence of bacteria to carbohydrate ligands (10, 11, 31). Of note, sugar alone as a carbon source promotes adhesion of *C. albicans* yeasts by increasing the expression of surface fibrillar adhesins (4). Glutaraldehyde-fixed yeasts do not bind, while the unfixed organisms bind well to glutaraldehyde-fixed brain cell monolayers. This resembles the binding of lymphocytes to high endothelial venules: lymphocytes bind to glutaraldehyde-fixed endothelium but glutaraldehyde-fixed lymphocytes no longer bind (23). The activity of the carbohydrate-binding protein on lymphocytes is destroyed by glutaraldehyde, whereas the carbohydrate receptor on the endothelium is unaffected (30).

The adhesins of various yeasts that recognize lactosylceramide are probably similar since all species examined, including the yeast phase (tissue invasive form) of the dimorphic fungi *H. capsulatum* and *S. schenckii*, bound to this glycolipid (Table 2). That *C. albicans* no longer bound to lactosylceramide after induction of the hyphal phase may reflect the inhibition in cell division and other important functions that occur during this phase (19, 22). Alternatively, phenotypic changes in cell wall components specific for this phase might be involved (22). Although yeasts vary in their pathogenic potentials, they may all share the ability to colonize the host. While *C. albicans* and *C. neoformans* are the species that most frequently cause disease in humans, other opportunistic yeasts can cause severe disease in immunocompromised patients (1, 9, 19, 21). Similarly, the pathogenicity of several cryptococci was found to be the same when it was tested in immunocompromised mice (3). Thus, factors other than the ability to colonize host tissues must account for the differences in pathogenicity among yeasts. Taken together, the results presented above suggest that lactosylceramide may be a host receptor for yeast adhesion. Since all yeasts examined in this study bound specifically to lactosylceramide, this property may be essential for other unknown fungal cell functions. Nevertheless, the ubiquity of this glycosphingolipid in mammalian tissues may favor the potential of these organisms to colonize host tissues such as mucosae and lungs and may account for the multiple organ involvement in patients with disseminated fungal disease.

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