Recurrent Exposure to *Histoplasma capsulatum* in Modern Air-Conditioned Buildings

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**Background** Between 1989 and 1996, an epidemic of histoplasmosis occurred on a medical school campus. There had been numerous construction projects on the campus that involved previously wooded land and were adjacent to a large bird sanctuary.

**Methods** We investigated the epidemic with active surveillance to detect cases, a histoplasmin skin-test survey, inspection of the air-filtration systems of the involved buildings, and cultures of soil samples. The investigation also included a simulation of entry into air-intakes of the buildings from spore sources by means of a wind-tunnel analysis of a model of the campus that used inert gas. After control procedures were instituted, sentinel population groups had follow-up with yearly serological tests.

**Results** From 1989 through 1996, there were 29 cases of histoplasmosis that occurred among school employees. All cases with a defined onset began during periods of ongoing campus construction. Positivity rates for histoplasmin skin testing were higher among on-campus personnel (47%) than among off-campus employee control subjects (28%) (P<.001); the rates were highest in employees who worked on the upper floors of 2 research buildings. The air-handling units on the roofs of these buildings were not designed to exclude *Histoplasma* spores. The wind-tunnel experiment indicated that spores aerosolized in the bird sanctuary were not taken into campus buildings.

**Conclusions** The major sources of employee exposure to *H. capsulatum* spores were the construction sites. Low-level, recurrent exposures occurred over several years inside modern research buildings. This phenomenon, which has not been previously described, may play a role in the epidemiology of spore-transmitted diseases in urban settings.

An epidemic of clinical histoplasmosis occurred on the campus of the University of Texas Southwestern Medical Center at Dallas from 1989 to 1996. Because faculty and employees were concerned that they were being infected on campus, a combined clinical, epidemiologic, and environmental investigation was initiated. The purpose of this report is to describe the epidemic, identify its underlying causes, and present the procedures that led to its control.

**METHODS AND MATERIALS**

A case of histoplasmosis was defined as a compatible clinical illness with laboratory documentation of infection, either in the form of a pathological report from tissue, positive culture results, or positive serologic test results [1–4]. Complement fixation (CF) and immunodiffusion tests for histoplasmosis were performed at the Mayo Medical Laboratories in Rochester, Minnesota. A case was considered to be confirmed serologically if the subject experienced a 4-fold rise in CF antibody titer against that of the yeast antigen, or if a serum specimen demonstrated both H and M bands by immunodiffusion. A case was defined as presumptive if the subject’s CF antibody test titer was ≥1:32. A case was considered to have a defined onset if the subject had acute illness with fever, cough, chest pain or dyspnea, and an abnormal finding of a chest radiogram or other radiological study, or an abnormal physical finding. If we could not precisely date the onset or exposures, the case was categorized as having an indeterminate onset.

In December 1994, we performed a skin-test survey using a single lot of mycelial antigen obtained from Parke-Davis Laboratories. The tests were applied according to a standardized protocol [5, 6]. After stratification of employees by age, duration of employment,
and office location, 640 persons were picked randomly by computerized number assignment from the 1314 university employees with offices in the following 5 campus buildings: K (housing 298 employees), L (housing 422 employees), G (housing 360 employees), V (housing 136 employees), and X (housing 109 employees). The K and L buildings were hypothesized to have the greatest increase in risk of infection because they had the largest number of clinical cases in December 1994, when the survey was designed; G was a nearby campus building hypothesized to have a moderately increased risk of infection because it did not house any employees with clinical cases; and the V and X buildings were remote from the main campus and hypothesized to have risks similar to that of the general population in Dallas [7]. Skin tests were administered and results were obtained from 404 of the 640 persons. Persons who did not undergo a skin test were more likely to be from the off-campus buildings (X and V), but, otherwise, there were no significant differences. The sizes of skin-test reactions and information obtained by the questionnaire were analyzed with SAS software version 8.0 (SAS Institute).

To estimate the total number of cases of infection that might have occurred on the campus, we grouped the floors of all campus buildings into strata at different distances from the putative source of spores and with different numbers of clinical cases. The attributable risk for each stratum was estimated by subtracting the baseline proportion of positive skin test results for patients in the control building X (0.15) from the proportion of positive skin test results for patients in the surveyed building floor in the stratum, multiplying the remainder by the number of employees who worked on that building floor, and adding the attributable number of cases from all building floors on the campus. For example, floors 8 and 9 of building Y were located next to the fifth floor of building L, and persons who worked on those floors had clinical cases. The rate of skin-test indurations \( \geq 12 \text{ mm in diameter} \) among persons on the fifth floor of building L was 0.55, and subtracting the rate in control building X (0.15) yielded an attributable risk of 0.40. Multiplying this by the 192 persons working on those floors yielded an estimated 77 cases of infection. The risk associated with on-campus outdoor recreation was not included.

The air-handling units in the K and L buildings were inspected in August 1994, and a formal air-filtration study [7] was performed in September 1996. It involved examining the air-conditioning–filtration equipment, measuring particle counts upstream and downstream of filter housings, and measuring particle counts in buildings K, L, and Y.

Soil samples from the bird sanctuary (obtained in 1975, 1981, and 1984) and from the former blackbird roost (obtained in 1994) were cultured for *Histoplasma capsulatum* by inoculating plates with antibiotic-treated extracts [8]. In 1996, an additional 50 samples were cultured after antibiotic treatment with direct intraperitoneal inoculation into mice.

A scale model of the campus was constructed and placed in a wind tunnel [9]. Fourteen air intake vents with suction were constructed on the models of the buildings: 7 air intakes were located on building L; 2 air intakes were located on each of buildings K and Y; and 1 air intake was located on each of buildings E, G, and J. Seven spore sites were constructed at the bird sanctuary and various construction sites. Aerosols of an inert, measurable gas (ethane) were created at the spore sources, and the gas intake at each vent was quantitated.

To monitor the effects of the control measures, yearly CF testing was performed for the groundskeepers and for a subset of persons working in building L (in 1995 and 1996) who were found to have negative skin-test results during the skin-test survey conducted in 1994. The Infectious Diseases Division maintained active surveillance of clinical cases from 1994 through 1996 and from 2001 through 2002, during construction of the student union building.

## RESULTS

The University of Texas Southwestern Medical Center campus is located on a former grassy plain with groves of trees and is next to a protected bird sanctuary that is the home of \( \geq 1000 \) birds, mainly great egrets and cattle egrets. From 1985 through 1992, blackbirds roosted in the trees surrounding garages I and II and in and around the bird sanctuary (figure 1). In 1992, a connecting bridge was constructed from the south campus to the north campus (campus connector phase 1). An overhead walkway connecting garage I and building L over Inner Campus Drive (campus connector phase 2) was built in 1993–1994, and in 1996 a new sidewalk was placed along the Inner Campus Drive sidewalk, just west of buildings K, L, and Y.

From 1989 through 1996, there were 27 documented clinical cases of histoplasmosis that occurred among employees of University of Texas Southwestern Medical Center. Two additional cases were encountered during the skin-test survey and are listed as possible cases, because CF antibody titers determined after skin testing would not have been valid. One patient with a possible case had a skin-test induration that measured 31 mm in diameter, dyspnea, miliary nodules revealed by CT scanning of the chest, and a clinical response to ketoconazole. The other patient with a possible case had a skin-test induration that measured 20 mm in diameter and superior vena cava syndrome. Pathologic analysis showed fibrosing mediastinitis but no yeast forms [10].

Fifteen cases had acute onset, with fever usually being the feature that denoted the time the case began (table 1 and figure 2). In addition, a visiting scientist was on campus for a short time in 1993–1994; later, in his home country, he received a diagnosis of histoplasmosis based on examination of a coin.
lesion. Thirteen cases did not have a defined onset or a discrete, limited period of exposure. No patient had acute diffuse pulmonary histoplasmosis, as has been reported to occur after massive inoculation [11]. Although persons with positive skin-test results in the survey often recalled unexplained episodes of fever, no epidemic of acute febrile illness was recognized.

The first case occurred in 1989; additional cases occurred in 1991, 1992, and 1993 (figure 2). The peak of the epidemic occurred in 1994, when 8 cases with defined onset occurred. After the skin-test survey in December 1994, a moratorium on construction was put into place. No cases occurred in 1995. The moratorium was lifted in early 1996 for construction of the Inner Campus Drive sidewalk. Although major efforts were made to wet the soil, this activity took place in the midst of a regional drought. After 3 cases occurred in 1996, the construction moratorium (with the exception of minor projects) was again imposed.

Although few patients remembered aerosol exposures that might have contained *Histoplasma* spores, 1 groundskeeper became ill in 1994 shortly after working in the bird sanctuary. Two patients who worked on the second and fifth floors of building K experienced onset of infection in 1994 after exposure to dust caused by the construction of a steam line between buildings S and K. All patients with cases in 1996 were exposed to dust raised by construction of the Inner Campus Drive sidewalk.

A prior study of US Navy recruits found the rate of skin-test positivity in metropolitan Texas to be 29.6% [5]. In our skin-test survey, persons working in campus buildings X and V had a similar rate of positivity (28%; positivity was defined as a skin-test induration measuring ≥5 mm in diameter). In contrast, persons working in campus buildings G, K, and L had a significantly higher rate of positivity than those in buildings X and V (47% of persons in buildings G, K, and L versus 28% of persons in buildings X and V; *P* < .0001) (table 2). This risk was also present when a positive test result was defined as a skin-test induration measuring ≥12 mm in diameter (37% of persons in buildings G, K, and L versus 15% of persons in

![Figure 1](https://academic.oup.com/cid/article-abstract/41/2/170/529686)
Table 1. Clinical presentation of cases of histoplasmosis on a medical school campus, 1989–1996.

<table>
<thead>
<tr>
<th>Clinical characteristic, by onset category</th>
<th>No. of cases</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acute onset</td>
<td></td>
</tr>
<tr>
<td>Fever and pulmonary infiltrate</td>
<td>9</td>
</tr>
<tr>
<td>Tracheoesophageal or mediastinal lymphadenopathy with fever</td>
<td>6</td>
</tr>
<tr>
<td>Uncertain time of onset or duration</td>
<td></td>
</tr>
<tr>
<td>Pulmonary coin lesion</td>
<td>9</td>
</tr>
<tr>
<td>Retinal histoplasmosis</td>
<td>1</td>
</tr>
<tr>
<td>Tracheoesophageal adenopathy and odynophagia without fever</td>
<td>1</td>
</tr>
<tr>
<td>Disseminated histoplasmosis</td>
<td>1</td>
</tr>
<tr>
<td>Pleural effusion</td>
<td>1</td>
</tr>
<tr>
<td>Superior vena cava syndrome</td>
<td>1</td>
</tr>
</tbody>
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buildings X and V; \( P < .0001 \). A skin-test induration measuring \( \geq 12 \) mm in diameter correlated best with cases with recent exposure on campus. The rate of skin-test positivity was highest among employees who worked in building L, was lower among employees who worked in buildings K and G, and was lowest in employees who worked in the off-campus control buildings V and X (\( P < .0001 \); \( \chi^2 \) test for trend). The rate increased by \( \sim 17\% \) (OR, 1.2; 95% CI, 1.0–1.3) with each higher floor in the building where the employees’ offices or laboratories were located (building L, \( P = .003 \); building K, \( P = .0003 \); building G, \( P = .01 \)) (table 2). The percentage of persons with a positive skin test result increased significantly with increased time worked on the fifth floor of buildings K and L (\( P < .0001 \); \( \chi^2 \) test for trend) (figure 3). The diameter of induration increased significantly with each increase of floor in buildings L, K, and G (\( P = .01 \)) but not with increased floors in the control buildings V and X (\( P = .35 \)) (figure 4).

For the entire campus, we estimated that 600–700 cases of \textit{Histoplasma} infection may have been acquired on the campus in the years before the survey. An inspection of the air-filtration units in buildings K and L in 1994 concluded that the standard filters were not capable of excluding \textit{Histoplasma} spores. In addition, defects in the equipment allowed unfiltered air to enter the air-conditioning system.

Cultures of soil samples performed in 1975, 1981, and 1994 yielded \textit{H. capsulatum} in 3 (12%) of 26 cultures. Of 50 soil samples obtained in 1996, isolates were obtained from 6%. Soil samples that yielded \textit{H. capsulatum} were obtained only in or near the bird sanctuary.

The wind-tunnel experiments that modeled the prevailing wind characteristics and eddy currents indicated that sources of spores in or around the bird sanctuary did not create aerosols that were measurable in the air intakes of the buildings. A source of spores (spore source 5) located north of building L, representing dust clouds in the former blackbird roost that were caused by construction of campus connector phases 1 and 2, produced the highest concentration of simulated spores at 8 of the 14 building air intakes and was considered to be the major contributor to concentrations in campus building (figure 1).
negative skin-test results in 1994 (who accounted for 25 person-in persons working on the fifth floor of building L who had positive for *Histoplasma* antibodies. In 2002, only 3 (15%) of 20 tested groundskeepers tested positive for antibodies. In 1995, 12 seroconversions per 100 person-years of exposure). The other two seroconversions per 100 person-years of exposure during September 1995–June 2002, only 2 showed a 4-fold rise in antibody titer (a value that corresponds to 2.2 increase in risk of a positive skin-test induration with each higher floor. The OR for floor (1.2) indicates the difference from off-campus control buildings V and X, .

Control measures included discouraging employees from entering the bird sanctuary, minimizing campus construction, wetting the soil exposed by excavation during the daytime and covering it with tarpaulins at night, and retrofitting buildings K, L, and Y with filters that excluded 95% of particles ≥2 microns in diameter. These filters were placed after the occurrence of cases in 1996. All buildings on campus now have comparable filters. A regular inspection system was put into place to repair the air filters. When a new student union building was built in 2000–2002, a 6-foot high, 80% fabric, chain-link fence was constructed around the excavation site. It prohibited nonconstruction workers from entering the site and prevented the bird sanctuary, minimizing campus construction, wetting the soil exposed by excavation during the daytime and covering it with tarpaulins at night, and retrofitting buildings K, L, and Y with filters that excluded 95% of particles ≥2 microns in diameter. These filters were placed after the occurrence of cases in 1996. All buildings on campus now have comparable filters. A regular inspection system was put into place to repair the air filters. When a new student union building was built in 2000–2002, a 6-foot high, 80% fabric, chain-link fence was constructed around the excavation site. It prohibited nonconstruction workers from entering the site and was especially designed to take advantage of the prevailing wind directions to create areas of low pressure that trapped dust at the base of the fence [9]. After wetting the soil, the top 9 inches of topsoil were removed from the entire construction site and transported to a landfill before construction began.

Of 22 groundskeepers observed for 91 person-years of exposure during September 1995–June 2002, only 2 showed a 4-fold rise in antibody titer (a value that corresponds to 2.2 increase in risk of a positive skin-test induration with each higher floor. One of the 2 groundskeepers with a negative histoplasmin skin-test induration failed to react to 3 control antigens. No seroconversions occurred in 1994, another in March 1994, and the third in April 1994. In particular, 3 members of a single laboratory became ill: one in 1989, another in March 1994, and the third in April 1994. They parked in different garages and walked different paths to work, but all of them worked on the fifth floor of building L. Moreover, skin test positivity rates were higher among subjects who had worked longer on the fifth floor of buildings R and L (figure 3). The size of skin-test indurations also increased.

### DISCUSSION

We documented the occurrence of histoplasmosis on a medical school campus for >5 years. All subjects who had cases with a defined time of onset worked in campus buildings or as groundskeepers. The skin-test survey revealed a significantly higher positivity rate among persons working on campus than among persons working in the 2 off-campus control buildings (buildings X and Y). Sizes of skin-test indurations were also significantly larger in campus personnel, >600 of whom may have become infected during the outbreak.

Although both infection and clinical cases resulted from multiple outdoor exposure events, a critically important conclusion of the investigation is that most transmission of *Histoplasma* spores occurred inside large research buildings. This is best illustrated by cases on the fifth floor of building L, where 5 of the 29 total cases and 4 of 15 cases with defined onset occurred. In particular, 3 members of a single laboratory became ill: one in 1989, another in March 1994, and the third in April 1994. They parked in different garages and walked different paths to work, but all of them worked on the fifth floor of building L. Moreover, skin test positivity rates were higher among subjects who had worked longer on the fifth floor of buildings R and L (figure 3). The size of skin-test indurations also increased.
significantly with each higher floor in on-campus buildings but did not increase by floor in control buildings (figure 4). This was particularly true for buildings K and L and may reflect the proximity of the upper floors to the air-intake systems located on the roofs of these buildings.

Although *H. capsulatum* was recovered only from the bird sanctuary, soil samples were never taken from the former blackbird roost until late 1994 and 1996, after the construction projects north of building L had been completed and the topsoil had been replaced. The wind-tunnel experiments showed that sources of spores in the bird sanctuary did not create aerosols that could be measured in any building. The source of spores north of building L, at the site of the former blackbird roost, was considered the major contributor to indoor concentrations in on-campus buildings.

The air-handling systems for buildings K and L were not designed to exclude *Histoplasma* spores. The clinical characteristics and the temporal occurrence of the cases are consistent with the conclusion that some persons on the fifth floors of buildings K and L were exposed to low-level infection on multiple occasions [12, 13]. The evidence suggests that exposure on the fifth floors of buildings K and L was intermittent and recurrent and occurred from at least 1989 through 1994. Such recurrent exposure inside modern buildings has not previously been described.

Two other epidemics have involved air-treatment facilities. In each, exposure occurred only once over a short period [14, 15]. In 1970, an outbreak of histoplasmosis occurred in 384 (40%) of the students in a junior high school (Willis Junior High School) in Ohio [14]. On Earth Day, a courtyard was raked and swept, and the entire school building was contaminated with air containing *Histoplasma* spores. The epidemic was explosive but the acute illness, the so-called “Willis flu,” was short-lived and influenza-like. In 1975, bird droppings swept from the roof of a courthouse in Arkansas were distributed through the building by window air-conditioners [15]. Again, the resulting epidemic was explosive, with 52% of 84 employees inside the courthouse developing histoplasmosis in the 2 months after exposure.

What was the role of the bird sanctuary in the outbreak we describe? Although 1 case occurred in a groundskeeper, and
the rate of skin test positivity was high among persons with exposure to the sanctuary (e.g., groundkeepers) and increased levels of outdoor activity (e.g., groundkeepers, joggers, and basketball players) (table 2), the bird sanctuary’s role in the epidemic was relatively minor. Most cases occurred in employees who had no direct contact with the sanctuary and began during times when campus construction was underway. New cases did not occur after campus construction ceased. The similarity of skin test positivity rates among subjects in building V (located directly downwind from the sanctuary) and subjects in building X (located in the opposite direction) also attests to the limited role of the sanctuary in generating infective aerosols. Leaving the sanctuary undisturbed appears to provide adequate protection [16]. Destroying it could potentially create dangerous aerosols, and environmental concerns limited the other options [17, 18].

There is an assumption that one is safe from airborne infectious agents when working inside a modern building with contemporary air-handling systems. In this outbreak, we found evidence that employees were repeatedly exposed to Histoplasma spores while working in 2 modern research buildings. The sporadic nature of the resulting disease and the fact that this phenomenon had not been described previously delayed recognition of the problem. Remediation required installation of new air filters and prevention of the formation of spore aerosols whenever movement of earth was necessary. For the construction of new office buildings, professional organizations (such as the American Society of Heating, Refrigerating, and Air-Conditioning [19]) currently recommend the installation of filters that exclude only 25%–35% of particles 1–3 μm in size. The epidemiology of modern urban histoplasmosis is in the process of being elucidated [20]. Recurrent transmission of histoplasmosis in large buildings next to construction and excavation sites may be an important part of this unfolding story.

The vulnerability of people in large office buildings to exposure to spore-transmitted diseases, such as histoplasmosis, probably includes other infections that can be spread by aerosol that might be used in bioterrorism attacks, such as anthrax. The realization that infective spores can penetrate inefficient air-filtration systems and infect workers inside office buildings should be considered in plans for protection against bioterrorism.

Acknowledgments

We are indebted to Ted Pass (Moorhead State University, Moorhead, Kentucky) for collecting and culturing the soil samples obtained on-campus in 1996. We would also like to thank the following: Engineered Solutions (Fort Worth, Texas), who performed the study on the air-filters for the campus buildings; Cermak, Peterka, Petersen (Fort Collins, Colorado), who performed the wind-tunnel experiment; Donald E. Carlson and Esther J. Robinson of Environmental Health and Safety and William B. Neaves of the University of Texas Southwestern Medical Center; and fellows in infectious diseases and medical students for their assistance in performing the histoplasmin skin-test survey.

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176 • CID 2005:41 (15 July) • Luby et al.