New observations on the epidemiology of sporotrichosis and *Sporothrix schenckii* complex

1. Sources of infection and occupational risks

Key words: Sporotrichosis, occupational, epidemiology, sources of infection, zoonotic transmission.

Palabras clave: Esporotricosis, ocupacional, epidemiología, fuentes de infección, transmisión zoonótica.

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Abstract

Sporotrichosis is a chronic fungal disease usually limited to cutaneous and subcutaneous tissues. The most common way of the infection is the skin trauma by a prick from a thorn or a piece from a variety of barks, trees and bushes. Occupations that predispose persons to infections includes: farmer-agriculturist, gardener, florist, planters of trees, forestry workers, horticulturist, workers of the greenhouses, orchids growers, outdoor labor, masonry, veterinarians and laboratory workers (microbiologist), and people involved in activities and exposure to contaminated soils, vegetations, wood, *sphagnum* moss, wheat, straw, prairie hay, corn husks and strubble, barks, thorny plants and shrubs, reeds, rose bushes, cat´s skin ulcers, rodent bites and punctures of insects. A critical review of published epidemics and the mechanisms of zoonotic cat´s transmitted infection, and the recent advances were also described.

Resumen

La esporotricosis es una enfermedad micótica crónica, generalmente está limitada en los tejidos cutáneos y subcutáneos. La forma más común de la infección es el trauma de la piel, ocasionado por el pinchazo de una espina o de un fragmento de gran variedad de cortezas de árboles y arbustos. Las personas predispuestas a las infecciones incluyen: campesinos, jardineros, floristas, plantadores de árboles, trabajadores de la silvicultura, horticultores, trabajadores de los invernaderos, cultivadores de orquídeas, sujetos con trabajo al aire libre, albañiles, veterinarios y técnicos de laboratorio (microbiólogo), así como la gente implicada en actividades con exposición a suelos contaminados, vegetales, madera, musgo de *Sphagnum*, paja del trigo, heno de la pradera, hojas de maíz y puntas de rastrojo, cortezas, plantas y arbustos espinosos, cañas, rosales con espinas, y personas expuestas a úlceras en la piel de los gatos, mordeduras de roedores y picaduras de insectos. Este artículo presenta una revisión crítica de las epidemias publicadas, los mecanismos de transmisión zoonótica de la infección felina y los avances recientes.

Definition

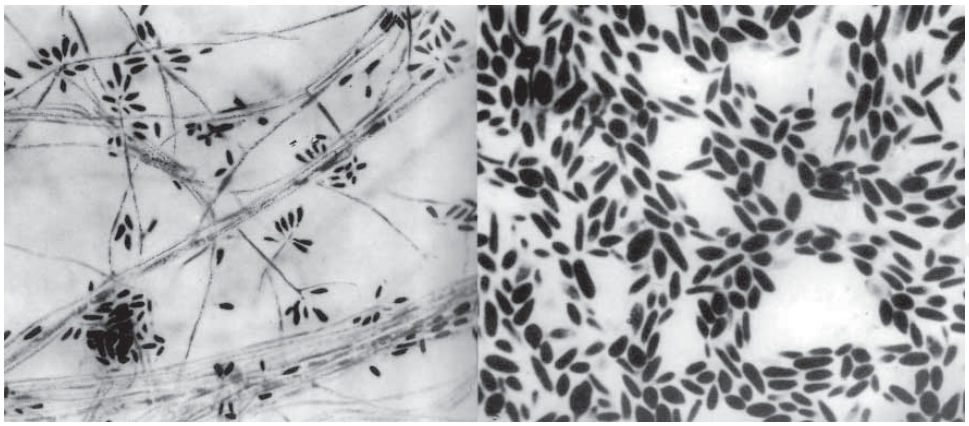
Sporotrichosis is a chronic infection usually limited to cutaneous and subcutaneous tissues, although it rarely become disseminated. The skin lesion develop where *Sporothrix schenckii* is introduced to dermal sites of injury, after scratches from thorns or splinters, cuts from sedge barbs, or handling of reeds, sphagnum moss, dry moldy hay, or grasses; also by bruising against infected timber or tree barks.[1,2] These modes of transmission emphasize the association of the fungus with decaying vegetation and soils. Rarely, Sporotrichosis infection can be acquired by inhalation or by deposition into the conjunctiva sac of the fungal spores. Secondary spread to articular surfaces, bones and muscle is infrequent, and the infection may also occasionally involve the lungs, eyes, central nervous system, and genitourinary tract.[3,4]

Mycology

Sporothrix schenckii is isolated and identified by most clinical laboratories,[5] but recent research of microscopic morphology, physiologic and molecular properties have found this fungus is actually made up of a complex of several different phylo genetic species, and tend to cluster in some geographic regions.[6-10] At least four different clades, in addition to *S. schenckii* sensu stricto have now been identified, these include *S. brasiliensis* and *S. globosa*, and both have caused human infections; *S. mexicana* and *S. albicans* which have not been associated with infections in humans. *S. schenckii* has been isolated from soil, humus, the stems of beech trees, vegetable debris, moist wood and refrigerated meat. It has also isolated from many types of plants, such as: horsetails, rose bushes, cacti, salt meadow hay, residual packing straw, carnations, wood splinters, and most commonly sphagnum moss and mulch-hay.[5-10]

S. schenckii exhibits thermal-nutritional dimorphism: It grows on most artificial culture media at room temperature, as a characteristic moist, flat, creamy colony that becomes dry, wrinkled and feathery, and cream to tan, brown, or blacks with age.[2,5]

Microscopic examination of the filamentous-mycelia form reveals thin (less than 3 um diameter) hyaline, septate and branching hyphae, with right angled conidiophores producing at their apices, small, hyaline or dark, oval, globose or pyriform conidia, arranged in clusters resemblance of a flowerette *(figure 1)*. The yeast colony of the organism is creamy and grayish white, soft and bacterial in appearance. Microscopic examination reveals small ovoid-to-cigar-shaped cells, and has single or multiple buds.[5-10] When conidial suspension prepared



A) Mycelial **B) Levaduriforme**

Figure 1. A: Mycelial phase oval conidia borne sympodially on a lateral conidiophore and solitary conidia on the hyphal axis. **B:** Typical oval-to-cigar-shaped yeast cells, from a case of skin infection. (1,200 X).

from mycelia-phase is injected intra peritoneal into male mice, prominent orchitis is produced within 10 days to two weeks.[5] Kwon-Chung showed isolates with higher thermo tolerance at 37 °C, multiply well in the internal organs and grow faster in the testes. The isolates with lower thermo tolerance 35 °C, multiply slower in the testes, but not in the internal organs.[11] *S. schenckii* cannot grow from temperatures above 38.5 °C.[12] The effects of temperature on the manifestation of lesions in mice and rats infected with *Sporothrix schenckii* was noted by J. MacKinnon: The infected animals housed from 2 to 5 °C had military lesions in the internal organs and muscles of the hind legs. However, lesion was produced only in the muscles of the legs when the rodent was housed from 13 to 17 °C. At 31 °C the rats did not develop any lesions.[13]

Sources of infection

Classically, the most common method of infection has been trauma to the skin by a prick from a thorn or a piece of any one of a variety of bushes, trees, or hay.[14] This was confirmed by the fact that *S. schenckii* has been cultured from plant material: rotten palm-tree trunks, dry grass, sandy soil covered by mosses and protected from direct sunlight.[15]

FM Gastineau et al found *sphagnum* moss to be a common source of contamination for six cases of sporotrichosis[16] *(figure 2)*. In an outbreak of fourteen cases at a state nursery in Vermont, described by D´Alessio et al, the fungus was isolated from samples of moss in contact with all patients. They were able to identify *S. schenckii* in samples collected at the harvesting sites from the bog. In another clinical case of Sporotrichosis, the organism was isolated from a barn owned by the patient as well as from roses stored in moss in the barn.[17] Mc Donough pointed out that *S. schenckii* has never been cultured from fresh moss obtained directly from a bog.

Hay has been found to be a point source of infection for epidemics in children. In one instance, nine children were infected while playing in prairie hay[18] *(figure 3)*. Powell and Hodges reported two additional cases related to prairie hay.[19] Another case was found, at the Missouri State Chest Hospital in Mount Vernon, in a five-year-old boy who had previously played with prairie hay. An additional six cases related to handling of hay were described by Sanders: bricks stored in contaminated hay had been handled in bucket-brigade fashion by several college students. During an alcohol-induced melee the bricks landed on bare skin, resulting in Sporotrichosis in several of these students.[20]

In Australia, from January to October 1995, 16 clinical cases of Sporotrichosis were diagnosed at the Toowoomba´s Skin Clinic, in the Darling-Dows Region of Queensland.[21] This area usually had one or two cases per annum. Exposure histories pointed to a local store selling hay originated from a farm at Tara, the moldy hay was used as garden mulch. All affected patients were in directed contact with the blackened and foul smelling, stored-hay. Nine of 16 patients were males; the youngest was aged 11 and the oldest 67 years of age. Lymph cutaneous sporotrichosis was seen in 50% of patients, the rest had fixed forms. Three separate families reported more than one member involved, and a dog that had slept on the hay also developed ulcerated nodules on the paws, but cleared spontaneously. Histological examination performed in 12 of 16 cases confirmed the diagnosis; all patients had a positive culture for *S. schenckii*.[21]

In Western Australia, only eight cases of Sporotrichosis were reported from1997 to 1999, as compared to 41 mycological confirmed human cases from 2000 to 2003 at the Path Laboratory Medical Centre.[22] A careful review found: 22 cases were from the Busselton-Margaret River (BMR) small farming subtropical area, South of Perth, where no cases had previously been recorded. Epidemiologic research from July 2003 to July 2004, discovered 11 patients who had been in contact with moldy hay, used for domestic gardening and

commercial farming. Two of the affected Children had played together in hay, and one patient developed sporotrichosis after a camping trip. Fifty hay samples were collected around the BMR region for mycological culture. Half of the case-related properties tested were positive for *S. schenckii* as was a sample from a local commercial-hay supplier. Hay bales were reported as visually moldy.[22] The DNA-macro restriction digestion patterns for the recent Western-Australia (WA) clinical isolates and four hay-isolates were indistinguishable. The pulsed-field gel electrophoresis method used, showed that the dominant-WA strain of *S. schenckii* was present in hay and has caused endemic Sporotrichosis for at least 15 years, and it was quite different from clinical strains isolated in Queensland and other parts of Australia.[23]

Kedes et al.[24] reported sporotrichosis in a rose gardener, who often subdue the day´s problems by drinking wine while he tended rose bushes. He fell asleep while working and he subsequently came down with sporotrichosis, described as the first to be afflicted with "the syndrome of the alcoholic rose gardener". Alcohol has not yet proved to be a cause of sporotrichosis; however, it may produce underlying conditions which predisposes to the extra cutaneous infection. A review of sporotrichosis-cases seen at the Mayo Clinic in the twenty years period 1953-73, confirmed the most common sources of infection: rose bush 12 *(figure 4)*, corn husks 2, splinter 2, insect bite 2, Japanese shrub 1, raspberry bush 1, evergreen 1, sphagnum moss 1, cat bite 1, unknown vegetation 1, and unknown etiology 7.[14]

Sphagnum moss epidemic sporotrichosis

In the spring of 1988, the largest documented US-outbreak of cutaneous sporotrichosis occurred, with 84 culture-confirmed cases among persons from 15 States, who were exposed to Wisconsin-grown Sphagnum-moss used in packing evergreen tree seedlings. In New York State, 13 cases occurred among 76 forestry workers who had handled seedlings and moss, the attack rate = 17%. The risk of infection increased as working time exposure to moss increased and, the attack rates were: <10 hrs 33%. Environmental samples of moss from Wisconsin supplier were negatives, but *Sporothrix schenckii* was cultured from multiple samples obtained from one of six Pennsylvania tree nurseries, identified as the source for 79 (94%) of cases-associated with handling the 1-to-3 year old stored moss.[25]

Previous studies from the University of Wisconsin demonstrated: sphagnum moss in the presence of water was an excellent substrate for the growth of *S. schenckii*.[26] Other reported risk factors include minor trauma and lack of protective globes use.[25] Gastineau et al reviewed 206 cases of sporotrichosis reported in the United States 1898-1940: 46 (22%) were in farmers and 30 (15 %) in nursery workers, professional gardeners, or florist.[16] After 1941 we identified 11 epidemics reported in the US literature, Table 1, eight were associated with exposure to sphagnum moss and, three were related to hay exposure. Also thorny-yard-plants possessing rigid needles such as roses, bayberry, the Japanese barberry *Berberis thunbergui (figure 5)*, hawthorns, acacia and shrubbery with its exceedingly sharp, stiff and slender thorns inflicts injury on the slightest carelessness in handling, therefore, they are definitely a risk factor for contracting sporotrichosis.[14] Children after playing in yards and gardeners may develop facial lesions.[27] In the laboratory, S. *schenckii* population increased when inoculates on dead but not on living moss, supporting the contention that at some point after harvesting the moss becomes contaminated with the fungus, which then colonizes and reproduces on the dead plant under warn-moist conditions.[28] Mice-virulent *S. schenckii* has never been isolated from the bogs of Wisconsin.[29-31]

Figure 2. Left: Living plants *sphagnum* moss collected in August from a bog at Wisconsin, USA**. Right:** Moss for horticulture and forestry consist of dried plants, with very high water-holding capacity, hence the various applications in floral arrangements.

Figure 3. Prairie-hay has been found to be a point source of epidemic sporotrichosis; children were infected while playing in Australia, Brazil and USA.

Figure 4. Gardeners and florist handling rose bushes, may suffer cutaneous sporotrichosis in Mexico, USA and Queensland, Australia.

Epidemic zoonotic sporotrichosis

In the 12 year period 1987-1988 only 13 human cases of sporotrichosis were recorded, at the Infectious Dermatology Service of the Research Center Evandro Chagas Hospital in Rio de Janeiro, Brazil. Epidemic sporotrichosis is an emerging zoonosis.[32] From 1998 to 2003, 497 humans and 1,056 cats culture-positive were recorded. A total of 421 (67.4%) human patients had a history of a cat´s scratch or bite, or reported contacts with infected cats, studied at the E. Chagas Clinical-Research-Institute (IPEC).[33] Most clinical cases of sporotrichosis diagnosed and treated at IPEC, came from outlying neighborhoods of Greater Metropolitan Rio de Janeiro, an area of very low socioeconomic conditions. The patients had an age range from 5 to 89 years (median 39), one hundred twenty-two (68%) were women. Housewives (30%) and students (18%), were most attacked groups, however, 5% were veterinarians with occupational sporotrichosis.[33]

A broad spectrum of clinical sings and symptoms was recorded in 347 cats with sporotrichosis: a) subclinical infection, b) solitary cutaneous lesion, c) fatal systemic disease. Lymph cutaneous form was observed in 19.3% of cats, mucosal involvement of the upper-respiratory and digestive tract was confirmed in 34.9%, and multiple cutaneous ulcers 39.5%.[34,35] *(figure 6)*. Two mycological studies of hundreds of cats were done to determine the sources of the zoonotic transmission: *S. schenckii* was recovered from the skin swabs, aspirates or biopsies 96-100%, nasal swabs specimens 66-70%, oral specimens 41-49%, and nails fragments pool 39%.[34] Also, the fungus was cultured from the oral and nasal cavities of 10 (9.9%) from 101 apparently healthy cats, that lived with sporotrichosis-infected felines.[36]

Most frequently observed cutaneous lesions were: crusted smooth nodules, gummas, multiple draining and purulent ulcers, usually teeming with highly infectious yeast-cells *(figure 7)*. Most

Figure 5. In 1926, Harry R. Foerster, described ten cases of occupational sporotrichosis, in employees of a tree nursery, they acquired the infection by inoculation with the slender, sharp, and stiff thorns of the barberry shrub *Barberies thunbergui*.

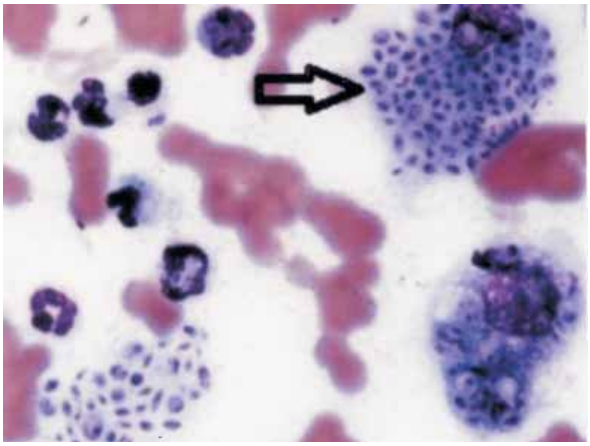


Figure 7. Infected cats, usually showed macrophages with large numbers of intra-and extracellular yeast-cells (arrow), present in skin ulcers. Transmission of infection can result even in the absence of human skin injury. (Giemsa stain 1,200 X).

Figure 6. In Rio de Janeiro, Brazil over 2,200 human cases of epidemic sporotrichosis, were reported housewives were most affected, after a cat´s bite or scratch. Cats developed skin ulcers on the head and thoracic limbs.

lesions were located on the head (56.8%), especially the nasal septum (28%), ears (21.6%), and thoracic limbs.[37] In contrast to other animal species, the presence of large numbers of intra and extracellular fungal elements and the absence of organized granulomas, lack of asteroid bodies' formation with a massive plethora of *Sporothrix* infection yeast is a characteristic of feline sporotrichosis.[38] The isolation of the fungus from different clinical specimens obtained from cats during both the preclinical and clinical phase of sporotrichosis provides support and strong evidence of the zoonotic potential and, reinforces the necessity of individual's protection specially those handling cats in endemic areas.[36] At the end of December 2009 over 2,200 human cases were diagnosed, at IPEC, plus 3,244 cats and 120 dogs with sporotrichosis accumulated; however, canine cutaneous disease has no relevance in the epidemiologic chain transmissions of the epizootic sporotrichosis.[39,40]

Highest incidence of cat's sporotrichosis was observed in the males with a mean reproductive age of 2 to 3 years. Given the cats natural habits of sharpening claws on the trunks, digging holes and covering wastes with sand or earth, healthy animals frequently carried the fungus in their claws (29.1%).[36] Free-roaming and housed, mongrel cats were usually infected through males fights (71%), contacts with other *Sporothrix* infected cats (22.3%), and by iatrogenic transmissions (5.6%).[37] A complex of many biologic-epidemiologic factors may help to explain the highest feline attack-rate and the unprecedented development of the ongoing Brazilian epizootic.[31]

Endemic zoonotic sporotrichosis

Extensive epidemics involving wide geographic areas or a large number of cases, are related to a common source of contamination in the environment.[29] Different animal species can be attacked by *S. schenckii* usually as isolated cases, and can be rarely transmitted to man by accident. K. Meyer described an epizootic outbreak in the United States affecting horse; however, just two from 400 human cases were of equine origin, because horses were installed on land recently deforested by colonization.[41]

Most clinical cases of Sporotrichosis in Uruguay were associated with armadillo (mulita-*Dasypus septemcinctus*) hunting: human infections were linked to animal scratches and skin injures from

Table I. U.S. Epidemics of sporotrichosis involving *sphagnum* moss 1941-1994.

No. of cases	Location	Occupation	Moss source*	Author	Year
6	Indiana	Florist	Not reported	Gastineau et al.	1941
8	Florida	Nursery worker	Wisconsin	Crevasse-Ellner	1960
5	Michigan	Nursery worker	Wisconsin	Hayes	1960
14	Vermont	Nursery worker	Wisconsin	D´Alessio et al.	1965
7	Oregon	Botanist	Not reported	CDC	1969
17	Mississippi	Forestry worker	Wisconsin	Powell et al.	1978
3	Minnesota	Nursery worker	Wisconsin	Grotte-Younger	1981
4	Wisconsin	Garden-shop	Wisconsin	CDC	1982
84	New York, Ill	Forestry worker	Wisconsin	Coles FB et al.	1988
9	Florida	Topiaries producer	Wisconsin	Hajjeh et al.	1994

**Sporothrix schenckii* was cultured from moss samples at the epidemic sites, but not from the bogs in Wisconsin.

tools used to dislodge the armadillo from burrows. In these cases the dirty tools and the armadillo´s claws act primarily as mechanical vectors of fungal spores from inside the burrows; armadillos are in permanent contact with soil, digging holes and galleries. *S. schenckii* has been also recovered from armadillos nest in South America.[42,43] Rarely the fungus infection was reported in three Florida´s armadillos in captivity, in such cases superficial wounds and abrasions are commonly related to poor sanitation, overcrowding or self trauma from borrowing on in appropriate surfaces. Contamination of armor wounds with subsequent hematogenous spread to organs-systems, appears to mode of infection in *D. novemcintus*, facilitated by animal captures stress, shipment to a zoo and confinement which may predispose to systemic dissemination of the fungus.[44] Singer and Muncie presented six patients who contracted the infection within a limited area of Long Island from grass mulch used on bulb farms. In Guadalajara City, Mexico Barba Rubio attached importance to rat's bites.[1-3]

Occupations

Foerster[45] described Sporotrichosis as an occupational dermatosis; most cases occurred among manual workers such as farmers, and rural corn cultivators, greenhouse and nursery worker, floral work, masonry and construction workers, rose-growers, horticulturists, orchid growers, coffee-garden workers, outdoor labor (brick-makers), tree planters and forestry workers, and people involved in activities and exposure to contaminated soil, and vegetation such as sphagnum moss, salt marsh hay, prairie hay, or thorny plants. The most common form of trauma to the skin involves punctures from sharp thorns, splinters, cuts from sedge barbs, or handling of reeds, grasses and corn stalks. Other types of trauma reported include: bites from rodents, cats, dogs, horses, badger, iguana; pecks from hens and other birds and insect stings. This was later demonstrated by Gastineau[46] in a study of 146 clinical cases which gave the percentages associated with most common occupations as follows: farmers, 32%; laborers, 17%; housewives, 12%. Gonzalez-Ochoa[47] reported the occupational incidence as follows: school-children and students, 26%; peasants, 24%; and housewives, 24%. The ratio male to female was 1:1. Men acquired the disease on the legs from thorns and sedges or on the fingers and wrist from gathering wood or grass; the women on the fingers from cultivating decorative house plants an making baskets; and the children on the face from scratches of thorny branches, exposure to hay and various plants during play, 60% of Mexican cases were found in persons less than 30 years of age. Sub acute fixed lesions were recorded most often in the young, whereas chronic disease was found with greater frequency in older patients. He also has shown that a group of grass handlers-either gathering it or using it as a packing material those who had been on the job for more than ten years had no clinical disease, but 100% of them had a positive sporotrichin skin-test. When disease does occur in this population, it is the fixed cutaneous type, and many cases appear to heal spontaneously.[48] Repeated exposure to small amounts of spores in the hyper endemic areas may confer protective immunity.[47,48]

Population-based surveillance and case control study were conducted in Abancay, Highlands of Peru (elevation 2 750 m) to estimate the burden of lymph cutaneous (LC) sporotrichosis and to determine risk factors, in a poorest and rural District with a population of 73, 003 persons, and three local laboratories with capability of isolating fungi from clinical specimens.[49] Clinical records from hospitals for the years 1997-1998 and prospective surveillance was conducted 1999. The mean annual incidence was 98 cases per 100 000 persons. A total 201 culture confirmed cases of LC-sporotrichosis were identified, 66% of which occurred in children aged < 15 years (mean calculated incidence 156/; against 52 in adults x 100 000), therefore, children had an incidence three times higher than adults,

and were more likely to have LC-lesions on face and neck. Identified risk factors included: playing in crop fields; having a dirty floor in the house, owning a cat. It was evident the higher incidence of sporotrichosis in adults whose occupations require outdoor exposure: work with hay; work in brick making and the relevance of conditions associated with poverty and lower socioeconomic status, such as home is walls made of adobe and ceilings made of unfinished wood.[49-51]

Discussion

In human medicine, general practice physicians and dermatologists consider Sporotrichosis to be an environmental and occupational acquired disease,[52] which occurred primarily after handling sphagnum moss, hay-mulching,[53] thorny plants (domestic gardening) such as rose, bushes, cactus,[54] corn, stalk[55] and the horticultural workers in a Disney World topiary.[56] The largest epidemic reported was in Witwatersrand, South Africa, involving 14 gold workers between 1941-1944, nearly 3 000 miners were infected cases in a space of two years, the source was traced to the growth of *S. schenckii* on the humid timbers of the mine.[57] Findley found that the fungus over grown on untreated mine poles at a temperature of 26 to 27 °C and a relative humidity of 92 to 100%.[58] Sporotrichosis has been rarely reported from geographic areas with dry and deserted climate, such as Central and North Chile,[1,2,5] although micro niches favoring growth of *S. schenckii* may exist even in such arid regions.[5] Attention may be called to a sporotrichin skin test survey conducted by Chakravarty et al in the rural and urban inhabitants of Delhí and Mathura district of Uttar-Pradesh, India: sporotrichin (+) hypersensitivity occurred in 22.4% (201 out of 897 individuals) of urban-population, against 37.2% (101 out of 273) of the rural areas.[59] In a more recent skin survey by Ghosh et al. conducted in endemic areas of Himachal Pradesh, the prevalence of cutaneous hypersensitivity to sporotrichin was significantly higher in the villages with several clinical cases, range 22.9% to 40%; against the controls without reports of human cases, range 6.5% to 7.5%.[60] In Central Rio Grande do Sul, Brazil Lopes JO et at compared two different periods 1957-1987 and 1988-1997, they found that in the last ten years the number of infected women and children was reduced in parallel with a decrease in horticultural farming activities. Authors attributed such drastic changes to ongoing urbanization plus changes in occupation and lifestyles recorded in the recent industrialized and richer State.[61]

In addition to the horticultural-farming gardening sources of Sporotrichosis, increasing attention is being focused on the role of domestic cats in the epidemic transmission of *S. schenckii* to humans.[62] The primary importance of cats as an important source of zoonotic sporotrichosis was highlighted by the development of the disease in a veterinarian in Stillwater, Okla. One week after being scratched on the left thumb by a cat with ulcerated cutaneous abscesses, the veterinarian also developed a swollen red and painful lesion on the dorsum of the scratched thumb.[62] However, trauma is not being required for the natural transmission of *S. schenckii* from sick cat to humans.

In a clinical report,[63] a veterinarian acquired sporotrichosis from a cat without clear evidence of trauma; after molecular analyses of DNA restriction fragment-length profiles, the isolates of *S. schenckii* identified in samples obtained from the cat and veterinarian were identical. From this finding, it was suggested that exposure to the large plethora of infectious yeasts present in the cat´s skin ulcers, can result in transmission of infection, even in the absence of skin penetrating injury, therefore, *Sporothrix schenckii* has been listed as an emerging zoonotic disease. Six principal factors have contributed to the unprecedented emergence of the zoonotic fungal disease: the increasing transportation of human and animals between geographic locations; increased contacts between animals and humans; drastic changes in the environment and husbandry

practices; larger populations of stray cats and immune compromised humans; increased scientific awareness of the zoonotic origin of fungal diseases; and the recent identification of new fungal virulent strains and species not previously known.[7,9]

Owners should be advised of the zoonotic potential of cutaneous sporotrichosis and the need to take precautions when handling their affected pets. After handling dogs or cats with sporotrichosis, hands and arms should be washed with soap, or antiseptic solution of known antifungal activity such as povidone iodine or chlorhexidine solutions. Primary care physicians, dermatologist and veterinarians may mistake lesions of Sporotrichosis with leishmaniasis, tuberculosis, and many causes of wounds. Misdiagnosis and difficulty of treating fungal diseases should emphasize the importance of early diagnosis, and proper clinical evaluation and good use of laboratory resources, when presented with patients having cutaneous lesions.[1,2,64] Vegas[65] attributed failure to recognize the fixed and minimal forms of sporotrichosis. Early confusion with bacteriological infections has occurred frequently, delaying diagnosis while the patient was treated with antibiotics.[21] There are more than 15 cases of laboratory acquired sporotrichosis.

The general belief that *S. schenckii* can grow on all plants and woods need to be questioned,[66] the fungus has some predilection for untreated eucalyptus and wattle-wood, this puts rural builders and farmers at risk, especially where lumber is stacked and comes in contact with contaminated potting and garden moist soil.[67,68] To which compost mosses, or mulching-hay is added.[53]

Mexico "country of corn, flowers and roses", sporotrichosis is endemic in the raining subtropical Central-Plateau and the Western central farming area of the Bajio, also in the humid coastal subtropical bands of the Pacific Ocean and Gulf of Mexico. There are hyper endemic urban centers: Mexico City and Guadalajara, Jalisco, plus the States of Guanajuato, Puebla, Morelos and Veracruz,[69] those places have lush gardeners, with amazing variety of roses, indigenous cactus, exotic plants and avenues planted with *Eucaliptus* sp. and conifers. In springtime there is abundance of rose growers and tropical orchidariums; in the rainy season from June to September corn stalks and straws are produced and collected everywhere. Occupational and domestic handling of potting soil and plants, are "new chances" to acquire sporotrichosis infections all the year round, with a slight increase from November to February (dry season).[70,71]

Epidemiology research and Ecology occupy the principal places in the traditional study of sporotrichosis.[72] Knowledge of the sources of the fungus is important in establishing the mode of infection and has implications in public health and prevention. In clinical sporotrichosis, traumatic skin implantation is the primary step of infection, the hyphae and conidia of the etiological agent are melanized by a pigment known as dihydroxy naphthalene (DHN)-melanin. The presence of DHN in the fungal cell wall is thought to confer a selective advantage on the organisms, allowing them to survive desiccation and ultraviolet irradiation, DHN is also considered as a virulence factor.[73]

Sporothrix schenckii has been isolated from humid straw, prairie hay, desiccated mushrooms, potting soil, armadillo burrows, and from stored sphagnum moss, but not from any moss directly from bogs in Wisconsin. Most likely the moss after harvesting comes into contact with bare soil if left outdoors several days, before packing. We know of no evidence to support the hypothesis that *S. schenckii* was present in the bogs. In fact, sporotrichosis is not a risk factor for workers harvesting moss. Additionally, in a serological study of the 1988 epidemic, there was no evidence of diagnostic agglutinating, precipitating or complement-fixing antibodies in the bog workers, including one worker with more than 30 years of occupational exposure.[73] The fungus remains relatively quiescent as long as the moss is dry, but when it is wetted before gardening uses, conditions become conductive for microbial growth on the dead and humid substrate, which

makes it more likely that persons handling moss will become infected.[17] My own observations in the laboratory "in relatively undisturbed potting soils a small number of microbial species assume dominance, as a result of particular, favorable conditions, including: microclimate, chemical characteristics of the substrate, and associated micro floral and micro faunal species".

DNA-typing method have proven useful in elucidating the probable source of sporotrichosis outbreaks, and also provides researches with the ability to investigate subtle differences between clinical isolates and to solve difficult epidemiological problems, separating species of fungal infection, as well as environmental strains differentiation.[74] In 1988 the largest North American epidemic of sporotrichosis occurred in 15 States. A total 84 cases were reported, with the majority of patients having contact with moss from a single nursery. Approximately 100 clinical and environmental strains were collected and studied in the laboratory. Standard mycological investigation of morphological and physiological similarities, as well as their virulence (mouse model), suggested six distinct groups: DNA isolated from the organism was subjected to restriction endo nuclease digestion, and the resulting fragments were separated by gel-electrophoresis. The DNA banding patterns (RFLP), was used to compare relatedness of isolates. Dixon and colleagues after a careful evaluation of results suggested the group I was the only source of the epidemic, because it was indistinguishable from clinical isolates. In contrast, a variety of DNA-types were associated with isolates in group II to VI. All clinical isolates from environment group I with ability to grow at 37^0 C produced fatal infection in mice.[75]

Human sporotrichosis is very rare in the Arid lands Israel, where only two cases have been reported. However, in 1976 a pigmented strain of *S. schenckii* was cultured from unopened nodules of the left hand and forearm of an 80-year man, after a trauma when working with pieces of wood. Experimental intra testicular mice inoculation of the clinical isolated fungus was followed by orchitis and abscess formation. The source of infection was found to be soil adhering to wood´s fragments. From such soil samples two strains of a black fungus, morphological identical to *S. schenckii*; were isolated through the mouse-virulence test procedure, also from soil samples collected in the vicinity patients residents, since this type of infection is so very rare, it may be overlooked for months or years, it is of much scientific interest to report and to publish all new cases recorded.[76]

High quality medical education and good clinical records;[64] awareness and perseverance of histological diagnoses: it is uncommon for yeast and asteroid bodies to be seen on the initial section, but if multiple serial sections are cut and stained with H&E and PAS, chances of specific diagnosis are very high (90-100%), at least 20 sections should be examined when sporotrichosis is suspected.[77] Although it has been more 110 years since Hektoen and Perkins so thoroughly and elegantly described sporotrichosis disease and its causative agent, there is some technical difficulty in distinguishing member of the complex-group *S. schenckii*, from its closely related, non-sporotrichosis causing relatives: characterization of species includes macroscopic and microscopic morphology, conversion to yeast phase, exo antigens reactions, virulence test in mice and DNA-RFLP and hybridization.[73,74] Epidemiologic investigations.[78,79] Sporotrichin test[79] and use of the more sensitive methods with purified cell-wall antigens.[80] The proper and timely application of the available resources surely will help to uncover the hidden sources of this fungal infections.[14,15,73]

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