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COCCIDIOIDOMYCOSIS AND HISTOPLASMOSIS A REVIEW OF THEIR EPIDEMIOLOGY AND GEOGRAPHICAL DISTRIBUTION

by

L. AJELLO*)

Coccidioidomycosis and histoplasmosis are two of the most important human mycoses. In their endemic areas, millions of infections have occurred and thousands continue to occur among susceptibles. Hundreds of active cases that require hospitalization develop each year. The deaths attributed to these diseases, in the United States alone, have averaged over 120 yearly (1). However, because the mycoses are not reportable diseases, data on the morbidity and mortality attributable to *Coccidioides immitis* and *Histoplasma capsulatum* undoubtedly are understated. The number of cases reported must represent a mere fraction of those that actually occur in the general population.

Because of their public health importance, a great many studies have been carried out on coccidioidomycosis and histosplamosis. For these proceedings it has been my task to review developments, during the past five years, regarding their epidemiology and geographical distribution.

Coccidioidomycosis

This disease, first described from Argentina by ALEJANDRO POSADAS (2) in 1892, is caused by a true soil-inhabiting fungus. MADDY (3) directly observed the growth of colonies of *C. immitis* in desert soil at a site near the outskirts of Phoenix, Arizona. Through the use of fluorescent antibody procedures, the infectious arthrospores of this fungus have been detected in aqueous suspensions of Arizona soils (4). Since 1932, when STEWART & MEYER (5) first isolated *C. immitis* from California soil, this fungus has been isolated from soil on many occasions. The most recent records are of desert areas in Arizona (3), California (6-8) and Mexico (9).

Soil surveys are useful in determining whether or not *C. immitis* is present in a given area. This is especially true where population den-

*) Present address: Center for Disease Control, Atlanta, Georgia 30333.
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sities are low or skin test surveys cannot be conducted. SWATEK & OMIECZYNSKI (10) recently evaluated four techniques for the direct and indirect isolation of *C. immitis* from soil. They found that a double-pour plate technique, originally described in 1965 by OMIECZYNSKI, SWATEK & BECKER (11) and modified for soil studies by OMIECZYNSKI & SWATEK in 1967 (12) was superior to and safer than any of the other methods tested. This technique, because of its simplicity and low cost, should prove useful in a wide variety of ecological and epidemiological studies.

As reviewed by SWATEK (13) in Teheran, ecological, epidemiological and microbiological studies have revealed that *C. immitis* is a highly specialized fungus. Rather than being haphazardly distributed in nature, it has definite ecological requirements. *C. immitis* is a fungus attuned to survive and flourish in arid regions. Studies by the EGEBERGS, ELCONIN & MADDY (reviewed in 1, 14) have revealed that during prolonged periods of high temperature and low soil moisture, *C. immitis* survives below the hostile surface at depths as great as 20 cm. After the rainy season and in periods of less intense heat, the surface is re-invaded and the fungus sporulates heavily. With the return of adverse conditions, the winds disseminate the infectious spores before the heat and dryness kill the surface growth.

The areas of the world where *C. immitis* has been proven to be endemic are in the semi-arid regions of the United States, Mexico, Guatemala, Honduras, Colombia, Venezuela, Paraguay and Argentina (1).

During the Teheran Congresses reviews on the coccidioidomycosis areas in Mexico and Central America and South America were presented by MAYORGA & ESPINOZA (15), CAMPINS (16) & EDWARDS (17).

In Mexico there are three endemic zones - Northern, Pacific and Littoral. These zones encompass the following states: Baja California, Chihuahua, Coahuila, Colima, Durango, Guanajuato, Guerrero, Jalisco, Michoacan, Nayarit, Nuevo Leon, Sinaloa, Sonora, and Tamaulipas. Surprisingly coccidioidin reactor rates of 5-30% occur in three tropical areas in the states of Colima, Michoacan (10-30%) and Guerrero (5-10%). These areas warrant detailed ecological and epidemiological studies to determine the factors that permit *C. immitis* to exist in a tropical climate. (Valleys of Tecoman-Colima; Apatzingan-Michoacan and Valley of Arcelia-Guerrero).

In Central America only two endemic areas have been uncovered. They are located in parts of the Motagua River Valley of Guatemala and the Comayagua Valley of Honduras. Coccidioidin sensitivity in the inhabitants of the endemic zone of the Motagua Valley (Gualán, Rio Hondo, Teculutan, Zacapa) was found to be 42.4% (898 positive among 2,120 tested). In the non-endemic sections of the Motagua Valley (Puerto Barrios, Bananera) and other areas of the republic the sensitivity levels ranged from 1.7 to 3.8% (115 of 4,614 individuals tested).

Ten human cases (Guatemala -8 and Honduras -2), and nine animal cases (7 dogs, 2 cattle - Guatemala) have been described. A dog, shipped from Nicaragua, was diagnosed as having histoplasmosis shortly after arrival in Norway.

In South America endemic areas are known to exist with certainty in only four countries: Argentina, Colombia, Paraguay and Venezuela.

In Argentina the endemic area, based on coccidioidin skin test surveys and the diagnosis of 27 cases, ranges from the 27th to the 40th south parallels. NEGRONI (18) divides this area into three zones with each one having specific climatological and botanical characteristics.

The Paraguayan endemic area is located in the open plains of the Gran Chaco. Here the climate is hot with annual rainfall as high as 1600 mm in one area and less than 475 mm in another. Few cases of coccidioidomycosis have been diagnosed in Paraguay, but coccidioidin skin test surveys have revealed that among the Indians that live in the western part of the Paraguayan Chaco, the sensitivity level is 43.9%.

Detailed information on the small Colombian endemic area has become available only recently. Cases of coccidioidomycosis and coccidioidin skin test reactors have originated or have been located in the Departments of Guajira, Magdalena and Cesar. All are in the northwestern part of Colombia adjacent to the Venezuelan border. This region is semi-arid. In the departments of Guajira and Magdalena rainfall barely exceeds 500 mm per year and generally falls during a 3-month period. In the department of Cesar, rainfall is more abundant - 1,000 to 2,000 mm-yearly and the vegetation is of the very dry tropical forest types (19). To date only two autochthonous cases of coccidioidomycosis have been diagnosed in Colombia.

Venezuela has the highest incidence and prevalence of coccidioidomycosis in South America. CAMPINS (16) noted that since 1949, 35 cases have been confirmed. Coccidioidin sensitivity levels are as high as 46%. The endemic areas are located in the states of Falcon, Lara and Zulia. Cacti of various genera and species dominate the vegetation of these states and the climate is desertic.

Reports of the occurrence of coccidioidomycosis in Bolivia, Ecuador and Peru have not been confirmed.

Histoplasmosis

In contrast to *C. immitis,* areas suitable for the survival and proliferation of *Histoplasma capsulatum* occur throughout the world. In Teheran reports by RANDHAWA (20), SOTGIU et al (21), CHIN (22) and EDWARDS (17) summarized current knowledge concerning histoplasmosis in Asia, Europe, the United States, and the world.

RANDHAWA'S critical review of Asian studies on histoplasmosis revealed how limited our knowledge of the prevalence and incidence of histoplasmosis in that continent is at the present time. Of 30 human cases reported, only 13 were confirmed by culture. These originated in: Malaya -4, Indonesia -3, Thailand -2, India -1, Japan -1, Singapore -1, and Vietnam -1. Malaya has the distinction of being the only Asian country where *H. capsulatum* has been isolated from soil (23).

A limited number of histoplasmin skin test surveys indicate that low levels prevail in most of the areas tested. Levels of 10% or more were observed in Burma (24), Indonesia (25), Pakistan (26), and Thailand (27).

Solid evidence for the existence of *H. capsulatum* in Europe is provided by the isolation of this fungus from soil in Italy and Rumania. The Italian positive soils came from a village in the region of Emilia-Romagna (28) and the Rumanian specimens were collected in the Topolnita bat cave in the district of Oltenia (29). That *H. capsulatum* is endemic in Europe receives further support by the diagnosis of animal and human cases of histoplasmosis that are considered autochthonous in Albania, Austria, Hungary, Italy, Portugal, Rumania, Switzerland, Turkey and the Union of Soviet Socialist Republics.

Ecological studies have revealed that *H. capsulatum* is a soil inhabiting fungus but its occurrence there is not haphazard (1). As reviewed by CHIN (22) and MAHVI (30), *H. capsulatum* has a predilection for avian and chiropteran habitats. Soil enriched with bat and bird guanos favors the growth of this fungus and gives it competitive advantages over other soil fungi. Through epidemiological studies acute outbreaks of histoplasmosis have been traced to point sources. These sites frequently have proven to be bat caves, chicken coops, and the roosts of such gregarious birds as grackles and starlings.

The importance of bird roosts as sources of *H. capsulatum* was emphasized by CHIN. Soil from 9 of 32 blackbird roosts in the states of Arkansas and Missouri (28%) yielded *H. capsulatum*. In Mason City, Iowa, two separate outbreaks of histoplasmosis associated with the clearing of a starling (*Sturnus vulgaris*) roost resulted in 115 acute of histoplasmosis with two fatalites. Histoplasmin skin test surveys revealed that 1/3 to 1/2 of the population of that city became infected.

At Mason City, Iowa, studies were carried out by TOSH et al. (31) to decontaminate soil containing *H. capsulatum*. These investigators found that 3% formalin effectively eliminated *H. capsulatum* from the surface of the soil. A total of 138 gallons of 3% formalin was applied per 100 square feet of the study area in three separate applications.

EDWARDS (17) pointed out that localized outbreaks of this type "account for only a negligible fraction of the vast number of human infections." She emphasized that more epidemiological studies are required to determine where the vast number of individuals "who become infected but do not develop clinical disease get their infections."

Clinical, ecological and epidemiological studies carried out by SUTLIFF & AJELLO (32) provided evidence for exogenous reinfection in ten cases of acute pulmonary histoplasmosis. All of these patients had preexisting calcified nodules in their lungs or hilum. *H. capsulatum* was recovered from the environments of six of these patients. In contrast *H. capsulatum* was not recovered from the environment of any of the 21 cases of chronic histoplasmosis with pre-existing calcified nodules. It was concluded that chronic pulmonary histoplasmosis in these cases was not the result of exogenous reinfection but rather provided evidence for endogenous reinfection.

In two papers SYMMERS (33-34) pointed out that clinicians outside the endemic area should be prepared to diagnose cases of coccidioidomycosis and histoplasmosis. He described cases diagnosed in Great Britain in travellers who had acquired their infections during short or prolonged stays in the naturally infected regions of the world. A few patients apparently had become infected following contact with contaminated fomites exported from endemic areas. Several infections had been acquired through the handling of cultures of *C. immitis* and *H. capsulatum* in the laboratory.

A case of coccidioidomycosis in an Australian who had never been in the Americas opened up the intriguing possibility that a hitherto unknown coccidioidomycosis endemic area awaits discovery in Australia. But SYMMERS concluded that in all likelihood, the patient had come in contact with contaminated material that had originated in the New World.

ASGARI & OURANY (35) reported on their search for evidence of coccidioidomycosis and histoplasmosis in Iran. Nine reactors to coccidioidin were found among 3,468 individuals tested (0.3%). The investigators did not regard these findings to be significant and concluded that *C. immitis* was not present in Iran.

In contrast 129 of 4,562 subjects reacted to histoplasmin skin tests (3%). The geographic distribution of these reactors showed that the highest percentage of reactors dwelled in the Central Plateau (32%).

All attempts to isolate *H. capsulatum* from clinical materials and soil ended in failure. These negative findings coupled with the absence of clinical or laboratory proven cases of histoplasmosis led to the conclusion that other soil fungi were responsible for the histoplasmin reactions.

Some of the most significant advances in the study of coccidioidomycosis and histoplasmosis have been made in the development of immunological procedures for their diagnosis and eventual control.

PROCKNOW (36) reviewed the basic clinical characteristics of histoplasmosis which enable a physician to carry out an accurate differential diagnosis. He emphasized that the respiratory tract serves as the primary site of infection by *H. capsulatum*. The resultant infection may range from the insignificant to severe respiratory and influenza-like disease.

H. capsulatum is an intracellular parasite that multiplies within macrophages. Following their release the yeast-like cells are disseminated via the lymphatics and bloodstream. Demonstration of intra-cellular yeast-like organisms by special fungus stains and their cultural identification as *H. capsulatum* in clinical material serves as the only confirmed basis for the diagnosis of histoplasmosis.

Procedures

GORDON (37) discussed the development of specific fluorescent antibody conjugates for the detection of *C. immitis* and *H. capsulatum* cells in clinical material and soil and the use of fluorescent inhibition techniques for the rapid screening of sera for histoplasmosis. These procedures are not only rapid and sensitive but the results obtained are in good agreement with those obtained with the old conventional methods.

Through use of specific fluorescent antibody conjugates for the yeast-form of *H. capsulatum*, KAUFMAN & BLUMER (38) discovered five different antigenic variants or serotypes. One proved to be indistinguishable antigenically from *H. capsulatum* var *duboisii*. The value of the serotypes in antigens for diagnostic complement-fixation tests remains to be determined.

The antigens commonly used for the serological diagnosis of both coccidioidomycosis and histoplasmosis are crude substances of various types that are produced with isolates and conditions that vary from laboratory to laboratory. Obvious needs have developed for specific and standardized antigens and much attention is being given to their development and production. KOBAYASHI & PAPPAGIANIS (39) and LARSH (40) critically reviewed the properties of the various types of antigens currently in use for skin tests and diagnostic serological procedures. Due to the shortcomings of these antigens, new methods are being used to separate and purify antigenically active cellular components of *C. immitis* and *H. capsulatum*. Current work in immunological studies is expected to lead to the physiochemical characterization of these substances.

Impressed by the remarkable range of variation in cultures of *C. immitis*, HUPPERT (41) expressed the need to reinvestigate the geographic distribution of *C. immitis*. Is this fungus really limited to the semi-arid zones of the New World? Does it occur in the Soviet Union? He proposed that a search be carried out for coccidioidomycosis, not only in new areas of both the New and Old World but in places where the ecology differs from the semi-arid conditions that *C. immitis* seems to require. Suggested methods include soil and skin test surveys as well as the use of the newly developed particle agglutination (42) and the agar gel immunodiffusion tests (43).

The ultimate goal in the study of all diseases is their cure and prevention. Introduced in 1956 amphotericin B was the first truly effective therapeutic agent for coccidioidomycosis and histoplasmosis (44, 45, 46). With this antibiotic a large percentage of severe infections can be cured.

A number of investigators have sought to develop vaccines against the systemic mycoses (47). The greatest progress has been made in the development of anti-coccidioidomycosis vaccines. One group has investigated the use of a live attenuated vaccine (48, 49) and another a killed vaccine (50-53). In-vitro grown spherules of *C. immitis* were found by both groups to be the most effective immunogenic agent. As summarized by HUPPERT (14) the work by LEVINE et al. has shown "(1) that there was an increase in immunogenicity as endospores developed into mature spherules; (2) that the efficacy of the vaccine dose was dependent; (3) that the immunogen (s) were located in the cell wall of the spherule, and (4) that resistance to a challenge dose of arthrospores was revealed best when the challenge was administered by the respiratory route -----."

Critical studies (54) revealed that although a live attenuated and a formalin-killed vaccine were immunogenic, the attenuated strain of *C. immitis* once again became virulent, following a single mouse passage. Until this problem is satisfactorily solved, field trials with live vaccines cannot be carried out. However, preliminary tests with a killed vaccine in human volunteers have been encouraging (52, 55). The efficacy and practicality of such a vaccine, however, must await a well-controlled large scale study.

This review shows that during the past five years a great deal of new information has been obtained on the epidemiology and geographical distribution of *C. immitis* and *H. capsulatum.* But much remains to be discovered. These two fungi will continue to intrigue medical mycologists for many years to come.

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