

Fungal Diseases

For creatures your size I offer
a free choice of habitat,
so settle yourselves in the zone
that suits you best, in the pools
of my pores or the tropical
forests of arm-pit and crotch,
in the deserts of my fore-arms,
or the cool woods of my scalp

Build colonies: I will supply
adequate warmth and moisture,
the sebum and lipids you need,
on condition you never
do me annoy with your presence,
but behave as good guests should
not rioting into acne
or athlete's-foot or a boil.

from "A New Year Greeting" by W.H.
Auden.

Introduction

Most of the important contacts between human beings and the fungi occur outside medicine. Fungi give us beer, bread, antibiotics, mushroom omelets, mildew, and some devastating crop diseases; their ability to cause human disease is relatively small. Of approximately 100,000 known species of fungi, only a few hundred are human pathogens. Of these, only a handful are significant enough to be included in medical texts and introductory courses like this one.

On the other hand, while fungal virulence for human beings is uncommon, the fungi are not casual pathogens. In the spectrum of infectious diseases, they can cause some of the most devastating and stubborn infections we see. Most human beings have a strong natural immunity to the fungi, but when this immunity is breached the consequences can be dramatic and severe. As modern medicine becomes increasingly adept in prolonging the survival of some patients with naturally-occurring immunocompromise (diabetes, cancer, AIDS), and causing iatrogenic immunocompromise in others (antibiotics, cytotoxic and

immunomodulating drugs), fungal infections are becoming increasingly important.

Remember: in a sense, every fungal infection is "opportunistic." The fungi are taking advantage in some way, eluding the patient's ordinarily competent defenses against them. The exact ways they do this vary -- quantity of infecting organism, breach of skin or mucosal integrity, abnormality in leukocyte function can each play a key role. A familiarity with these details of pathogenesis is the most important lesson to learn from your reading and these lectures.

This syllabus consists of some tables to review aspects of fungal pathogenesis, followed by details of the fungal infections to be considered in the lectures.

Table 1: Glossary

<u>fungus</u>	(pl. fungi) A nonmotile eukaryotic protist characterized by the absence of chlorophyll and the presence of a rigid cell wall.
<u>yeast</u>	A fungus growing as a round or oval unicellular organism, reproducing by budding or fission.
<u>mold</u>	A fungus growing in hyphal form, reproducing by sexual or asexual spore formation.
<u>hypha</u>	(pl. hyphae) A branching, tubular filament.
<u>mycelium</u>	(pl. mycelia) A mass of hyphae.
<u>pseudohypha (pseudomycelium)</u>	A filament (or mass of filaments) composed of many elongated yeast cells adherent to each other end to end.
<u>dimorphic</u>	Having the ability to transform between yeast and mold stages in response to specific changes in environment.
<u>spore</u>	The sexual or asexual reproductive unit of a mycelium.
<u>mycosis</u>	(pl. mycoses) A human disease caused by a fungus.

Table 2: Important distinctions between fungi and bacteria

<u>size</u>	<u>fungi</u> diameter 2-10 microns volume (yeasts) 20-50	<u>bacteria</u> diameter 1 micron volume 1-5
<u>nuclear structure</u>	eukaryotic	prokaryotic
<u>cell membrane composition</u>	sterols present (ergosterol)	no sterols present
<u>cell wall composition</u>	chitin, glucans, mannans	peptidoglycans, lipopolysaccharides, lipoproteins
<u>respiration</u>	aerobes or facultative anarobes. No strict anaerobes	aerobic or anaerobic
<u>metabolism</u>	heterotrophic	heterotrophic or autotrophic
<u>preferred temperature</u>	usually 25-35 C	usually 37 C
<u>toxin production</u>	rarely; always irrelevant in human infection	occasionally; may be important in human infection
<u>laboratory stain of choice</u>	KOH preparation of smears; methenamine silver or PAS stains of tissue. Gram stain unreliable.	Gram stain
<u>important immunity</u>	cellular immunity most important; role of antibodies still under investigation	humoral and cellular immunity

Table 3: Clinical categories of fungal infections

1. Superficial/Cutaneous: infection of outer layer of skin by lipophilic or keratinolytic fungi **e.g. pityriasis, dermatophytosis**
2. Subcutaneous: infection of subcutaneous tissues from the traumatic implantation of the fungus into the skin **e.g. sporotrichosis**
3. Systemic:
 - A. "The true pathogenic fungi"-- can cause disease even in **immunocompetent** hosts: **histoplasmosis, coccidioidomycosis, blastomycosis**
 - B. "The opportunistic fungi"-- generally cause disease only in **immunocompromised** hosts: **cryptococcosis, candidiasis, aspergillosis, mucormycosis**

Table 4: The pathogenic vs. the opportunistic fungi

	<u>pathogens</u>	<u>opportunists</u>
habitat	generally confined to "endemic" areas	omnipresent
morphology	dimorphic	no true dimorphs
route of infection	usually pulmonary	varies
host infected	often "normal"	usually immunocompromised
host response	pyogenic or granulomatous	necrosis to pyogenic to granulomatous, depending on degree of host impairment
immunity	resolution imparts strong specific immunity	no specific resistance to reinfection
prognosis	most resolve spontaneously	depends on degree of host impairment

Table 5: Host Defenses against Fungi

<u>Fungus</u>	<u>Host defenses of primary importance</u>
Dermatophytes	Intact skin barrier
Sporothrix	Intact skin barrier
Histoplasma	Lymphocyte function
Blastomyces	Lymphocyte function
Coccidioides	Lymphocyte function
Cryptococcus	Lymphocyte function; possibly humoral immunity
Candida	cutaneous infection: Intact skin barrier mucocutaneous infection: Lymphocyte function disseminated infection: Neutrophil function; intact skin and mucosal barriers
Aspergillus	invasive infection: Neutrophil function
Mucorales	invasive infection: Neutrophil function

Superficial Fungal Infections

<u>disease</u>	Dermatophytosis
<u>organisms</u>	Dermatophytes (members of the genera Trichophyton, Microsporum, Epidermophyton)
<u>mycology</u>	Related species of mold all possessing <u>keratinases</u> .
<u>epidemiology</u>	omnipresent;
<u>pathogenesis</u>	Spores on shed skin or hairs adhere to stratum corneum, germinate, invade. Individual susceptibility varies. Organisms confined to stratum corneum, with surrounding inflammation penetrating deeper layers of skin
<u>clinical</u>	tinea corporis (ringworm) tinea pedis (athlete's foot) tinea cruris (jock itch) tinea capitis (scalp and hair) onychomycosis (nails)
<u>diagnosis</u>	characteristic appearance (KOH preps or Wood's light exam); smear and culture of specimen
<u>treatment</u>	topical (azoles) systemic (griseofulvin, azoles, allylamines)

<u>disease</u>	Pityriasis versicolor
<u>organism</u>	<u>Malassezia furfur</u> .
<u>mycology</u>	A <u>lipophilic yeast</u> , part of the normal skin flora.
<u>epidemiology</u>	omnipresent; disease more prevalent in tropics
<u>pathogenesis</u>	Yeasts proliferate in settings of lipids and sweat. Rare in kids, appears in adolescence.
<u>clinical</u>	pityriasis = tinea versicolor. Seborrheic dermatitis (?) Infusion of lipid-containing nutritional solutions is rarely associated with <u>M. furfur</u> bacteremia.
<u>diagnosis</u>	characteristic appearance; UV light fluorescence
<u>treatment</u>	topical (selenium sulfide or azoles) catheter removal for fungemia

The Subcutaneous Fungal Infections

<u>disease</u>	Sporotrichosis
<u>organism</u>	<u>Sporothrix schenckii</u>
<u>mycology</u>	dimorphic: grows as mycelium in culture; as yeast with variably sized and shaped cells in host.
<u>epidemiology</u>	worldwide distribution in soil, especially Mexico and South America.
<u>pathogenesis</u>	Thorny plants (roses) or splinters inoculate fungus into subcutaneous tissues. Infection spreads slowly along draining lymphatics. Tissue reaction mixed pyogenic and granulomatous. Fungi often difficult to identify in lesions.
<u>clinical</u>	Small hard nodule at primary inoculation site discolors, ulcerates. Multiple similar nodules along lymphatics, which themselves become hard and cordlike. Rare bone, joint involvement; rarer dissemination. Likelihood of severe disease appears increased with poor nutrition or repeated exposure.
<u>diagnosis</u>	Culture of tissue or drainage (fungi rarely seen on smear or section). Skin test determines exposure but not disease.
<u>treatment</u>	Ketoconazole, itraconazole Amphotericin B

The "Pathogenic" Fungi

<u>disease</u>	Histoplasmosis
<u>organism</u>	<u>Histoplasma capsulatum</u>
<u>mycology</u>	dimorphic fungus: grows as mycelium in culture, as small budding yeasts in host
<u>epidemiology</u>	worldwide distribution in soils with high nitrogen content -- guano of birds, poultry, bats. Caves. Starling habitats. Endemic in Ohio-Mississippi Valley, Central and South America, Puerto Rico.
<u>pathogenesis</u>	Spores inhaled from soil, transform to yeast in lungs, phagocytosed by macrophages. Intracellular multiplication, dissemination, granulomatous host response as in TB. May resolve, resolve with scarring, remain active in lungs, or, especially in settings of immunocompromise, disseminate. Moves through reticuloendothelial organs (liver, spleen, lymph nodes) Like TB, may reactivate years after initial exposure (AIDS).
<u>clinical</u>	<ol style="list-style-type: none">1. self-limited disease: asymptomatic (95%) or flu-like syndrome. Noninfectious complications of healing may include mediastinal fibrosis, pericarditis.2. chronic pulmonary disease mimics TB; most common in settings of prior lung damage.3. disseminated disease (0.01%): fever, wasting, variable hepatosplenomegaly, ulcerations in mouth, GI tract, adrenal insufficiency, pancytopenia.
<u>diagnosis</u>	Skin test confirms exposure, but not infection. It may artificially boost subsequent serologic testing. Antibody titers are often unreliable, especially in immunocompromise. Antigen detection in serum and urine is sensitive but not commercially available.
<u>diagnosis</u>	pulmonary: exposure + compatible illness + compatible x-ray + antibody titer +/- sputum culture. disseminated disease: organisms seen intracellularly in blood/bone marrow/ liver/urine/tissue and/or grown in culture.
<u>treatment</u>	mild pulmonary: none severe pulmonary or disseminated: Amphotericin B

The "Pathogenic" Fungi

<u>disease</u>	Coccidioidomycosis ("Cocci")
<u>organism</u>	<u>Coccidioides immitis</u> (<u>immitis</u> = not mild (Lat.))
<u>mycology</u>	dimorphic: grows as mycelium in culture; as spherules producing endospores in host.
<u>epidemiology</u>	Endemic in arid, hot areas of North, Central, South America ("the lower Sonoran life zone.") Creosote bushes and rodent burrows. Soil organisms aerosolize late summer-fall. Note outbreak of cocci in Southern California immediately following LA earthquake of fall 1994.
<u>pathogenesis</u>	Inhaled spores swell into spherules in lung, burst releasing hundreds of endospores. Spherules evoke mononuclear response, endospores neutrophilic. Pulmonary pathology: consolidation, caseation, necrosis, cavity and nodule formation. Hematogenous dissemination of endospores to meninges, skin, bone, liver, spleen with lesions ranging from pyogenic abscesses to granulomas. Reactivation years after exposure may occur.
<u>clinical</u>	60% asymptomatic or mild pulmonary infection. 40% significant pulmonary infection: fever, cough, malaise, sputum production, erythema multiforme ("Valley Fever") 5% progressive pulmonary disease, residual pulmonary nodule or cavity. 0.5% disseminated disease (more common in men, pregnancy, dark-skinned races, immunocompromise). skin: fungating masses or ulcers; bone: osteolytic and/or blastic lesions; meninges (25%): chronic meningitis.
<u>diagnosis</u>	coccidioidin skin test positive 1-3 weeks after infection but not reliable: anergy common in disseminated disease. Complement fixation antibody may be + in disseminated disease, but not helpful in immunocompromised hosts. Best: smear and culture of pus, tissue.
<u>treatment</u>	(for severe primary disease, for primary disease in persons at risk of dissemination, for disseminated disease) Amphotericin B; fluconazole; itraconazole.

The "Pathogenic" Fungi

<u>disease</u>	Blastomycosis
<u>organism</u>	<u>Blastomyces dermatitidis</u>
<u>mycology</u>	dimorphic fungus: grows as mycelium in culture, as large yeasts with refractile cell walls and broad-based buds in host.
<u>epidemiology</u>	primarily in humid, wooded areas of North America. Prefers decaying organic matter and wood to soil. Outbreaks associated with log cabin building, peanut harvesting, camping, beaver dam viewing.
<u>pathogenesis</u>	Conidia inhaled, convert to yeast form in lungs. Evoke neutrophilic and granulomatous response. Dissemination via infected macrophages to skin, bone, urinary tract. In skin, pseudoepitheliomatous hyperplasia can simulate squamous cell cancer.
<u>clinical</u>	Frequency of asymptomatic infection unknown. Pulmonary infection mimics histo, TB. Disseminated disease: Skin involvement most common: ulcerating, heaped-up pigmented nodules face/hand/legs/mucosa +/- sinus tracts. 25-50% bone disease: well-demarcated osteolysis long bones/skull. Urogenital disease 5-20%: epididymitis/prostatitis/scrotal ulcers. Disease of liver, spleen, GI tract rare.
<u>diagnosis</u>	No adequate skin test, antibody or antigen assays exist. Best option is smear or culture of affected site.
<u>treatment</u>	Amphotericin B ketoconazole itraconazole

The "Opportunistic" Fungi

<u>disease</u>	Cryptococcosis
<u>fungus</u>	<u>Cryptococcus neoformans</u>
<u>mycology</u>	A yeast without a clinically relevant mold phase. Characterized by unique thick polysaccharide capsule.
<u>epidemiology</u>	The pigeon and its excreta, worldwide. Some species colonize eucalyptus trees instead.
<u>pathogenesis</u>	Aerosolized yeasts inhaled. Pulmonary disease (nodular, miliary) may occur. Hematogenous dissemination to central nervous system, where symptomatic human disease most common. Result: edematous, mucoid, cloudy meninges, usually with a poor inflammatory response. Intracerebral masses of yeasts may form (cryptococcomas).
<u>clinical</u>	Acute or chronic meningitis, with headache, fever, elevated intracerebral pressure. Often associated with AIDS or other immunocompromise, although cases in apparently normal hosts do occur. Cell-mediated immunity of paramount importance. Antibody is formed against capsule and may play a role in host defense.
<u>diagnosis</u>	India ink preparation, culture of cerebrospinal fluid (or other body site). Polysaccharide capsule allows for easy cryptococcal antigen assay in serum or cerebrospinal fluid -- sensitive and specific when titers are high.
<u>treatment</u>	Amphotericin B ± 5-flucytosine fluconazole

The "Opportunistic" Fungi

<u>disease</u>	Candidiasis
<u>organism</u>	<u>Candida albicans</u> ; other candida species (less frequent)
<u>mycology</u>	Yeast cells, pseudohyphae and hyphae in both culture and host. Organism stains Gram +.
<u>epidemiology</u>	Normal inhabitant of alimentary tract and mucosa (mouth, vagina, anus). <u>Not</u> on normal skin, although damage, moisture or any biochemical change leads to rapid colonization. Also predisposing to colonization: infancy, debilitation, pregnancy, antibiotics.
<u>pathogenesis</u>	

Primary host defenses consist of **intact skin and mucosa with normal pH and flora** (oral, gi tract, vaginal). Antibiotics, pregnancy, skin maceration leaves even healthy individuals susceptible to superficial Candida infection.

Dissemination occurs via defect in mucosa, or formation of biofilm on catheter, other foreign body. Once organism penetrates through the dermis into the blood, PML are the first line of defense. Can phagocytize and kill blastospores and damage pseudohyphae. Monocytes and eosinophils also ingest and kill organism. Phagocytes without myeloperoxidase or ability to generate hperoxide or superoxide anion cannot kill effectively. Serum or plasma alone even with antibodies and complement cannot kill candida -- cells are necessary. However, complement is also necessary for optimal opsonization, and IgG opsonizes. Patients with disseminated candidiasis may have a significant antibody response to the infection (although not reliably enough for antibody to be a diagnostic tool).

The role of lymphocytes in anticandidal immunity is immensely complex. The simplified version: they appear to protect primarily against proliferation on the skin or mucosa. Hence, HIV infection with its abnormal lymphocyte function predisposes primarily to oral/vaginal candidiasis. NK cells with anti-candidal activity have been identified. Lymphocyte cytokines are essential to recruit phagocytes in disseminated disease.

Virulence factors:

Are there hypervirulent species of Candida? NO. Many studies DNA sequencing species that colonize and infect find they are identical. In other words, people are infected by their own previously nonpathogenic flora, not someone else's monster strain.

But, overall, some qualities enable pathogenicity:

a. **environmental tolerance**. Candida can survive acid pH (stomach), in bloodstream, on mucosal surfaces.

b. secreted **hydrolases**. Produce extracellular proteases, phospholipases and other enzymes that can destroy connective tissue and kill host cells.

c. **adherence**. May stick to host cells as well as dentures, catheters and prosthetic devices.

d. Ability to grow in **hyphal** form. Induced by presence of serum and long associated with invasive disease in tissue sections. Specifics still under investigation. Hyphal form appears to protect yeast after phagocytosis, allowing escape from phagocytic cell and progression of infection. In mouse model, a strain of mutant Candida deficient in two genes enabling hyphae formation was avirulent. Still, unclear if directly related to filamentation or other virulence factors.

Recent synthesis: gene INT1 encodes surface protein Int1p. Enables the organism to grow esp in filamentous form, and seems linked to virulence. In vitro, superantigen-like effects (T cells activated; cascade of cytokines ---> sepsis syndrome) result when Int1p cleaved by heparin (common clinical anticoagulant). (JAMA 11/28/01 p. 2531).

clinical

Mucocutaneous: thrush (oral candidiasis); vaginitis, balanitis, onychomycosis; esophageal candidiasis; intertriginous candidiasis
Urinary tract: dysuria, fever
Disseminated: fever, chills, renal dysfunction, endophthalmitis, skin lesions.

diagnosis

1. pathognomonic appearance (thrush)
2. smear/culture of usually sterile site (distinction between colonization and infection can be tricky)
4. pathologic confirmation: organism in tissue

treatment

1. remove the breach of host defense
2. local (nystatin, clotrimazole); systemic (amphotericin B, fluconazole, voriconazole, caspofungin)

The "Opportunistic" Fungi

<u>disease</u>	Aspergillosis
<u>organism</u>	<u>Aspergillus fumigatus</u> , <u>A. flavus</u> , others (<u>aspergillum</u> = a perforated globe for sprinkling holy water (Lat)).
<u>mycology</u>	A mold without a yeast phase. Hyphae are septate and dichotomously branch at acute angles, like extended fingers. Fruiting heads seen in vitro.
<u>epidemiology</u>	Ubiquitous in the environment. Air, compost, soil, debris. Isolated from snow in the Antarctic, winds over the Sahara, an English apple orchard in the spring.... Also birds and chickens.
<u>pathogenesis</u>	Airborne spores are inhaled. May lead to: 1. hypersensitivity in airways (allergic bronchopulmonary aspergillosis). 2. massive local growth in preexisting abnormality in lungs (aspergilloma); 3. invasive disease in settings of neutropenia, steroid use: organism penetrates lung parenchyma and vasculature, causing dense pulmonary consolidation, necrosis, cavitation. May occur in sinuses as well. Hematogenous dissemination to skin, bone, heart.
<u>clinical</u>	allergic: wheezing, cough, eosinophilia aspergilloma: hemoptysis invasive pulmonary: fever, dyspnea, systemic toxicity Phagocytic defenses of paramount importance (neutrophils, monocytes, macrophages). Cell-mediated immunity less so.
<u>diagnosis</u>	Stain and culture of tissue biopsy specimen. (Positive culture of a secretion may represent contamination)
<u>treatment</u>	Allergic: corticosteroids aspergilloma: surgery invasive: Amphotericin B; itraconazole; voriconazole; caspofungin.

The "Opportunistic" Fungi

<u>disease</u>	Mucormycosis (AKA phycomycosis, zygomycosis)
<u>organisms</u>	species of the order Mucorales, esp. rhizopus.
<u>mycology</u>	Molds without a yeast phase. Broad, nonseptate hyphae, haphazardly branching at right angles. Thrive in warm, acid, sugary solutions.
<u>epidemiology</u>	Ubiquitous in decaying organic debris -- e.g. moldy bread.
<u>pathogenesis</u>	Spores are inhaled into the alveoli and the nasal turbinates. If unchecked, grow with blood vessel invasion. Erode through sinuses, nerves, bone. Thrombosis, infarction, tissue necrosis, surrounded by neutrophilic cellular infiltrate.
<u>clinical</u>	The most fulminant fungal infection known. Disease almost uniformly confined to patients with acidosis (diabetes, diarrhea, uremia, salicylate ingestion) or leukemia, occasionally other forms of immunocompromise. Neutrophils primary defense, so neutropenia major risk. Rare in AIDS but reported. Rhinocerebral: facial pain, headache, sinusitis, facial swelling, orbital cellulitis, cranial nerve signs. Pulmonary: cough, hemoptysis, rapidly progressive pulmonary insufficiency
<u>diagnosis</u>	Stain and culture of biopsy specimen
<u>treatment</u>	Correct hyperglycemia, acidosis, immunocompromise Amphotericin B