

(Notes on Anti-TB agents are included in the TB syllabus)

## **Antifungal Agents**

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### General background:

1. Fungi are eukaryotes, with more cellular similarities to human cells than to bacterial cells.
2. Until recently, fungal infections were far less common and less important than bacterial infections.
3. The in vitro testing of fungal drug sensitivities is difficult and highly technique-dependent. And, it appears to have inconsistent relevance to the ability of antifungal drugs to actually treat disease.

So...

1. Antibacterial agents don't treat fungal infections.
2. The development of antifungal compounds toxic enough to be efficacious against fungi, without being too toxic for humans, has been a) intellectually difficult and b) not (until recently) worthwhile for the pharmaceutical industry. In other words, it has been very slow. The gold standard of antifungal treatment remains a compound developed in 1956 which is one of the most toxic medicines used in clinical medicine.

### **Classes of antifungal agents to be discussed in lecture:**

1. **Polyenes**
2. **Azoles**
3. **Echinocandins**
4. **Miscellaneous others**

#### **1. Polyenes**

History: The first antifungal antibiotics. Produced by *Streptomyces* species ("higher bacteria")

Structure: Polyenes: multiple conjugated double bonds

Macrolides: large ring structures with a lactone linkage

Properties:

Lipophilic

Amphipathic (both hydrophilic and hydrophobic)

Amphoterics (both acid and base)

Mechanisms:

A. action: The rodlike macrolide ring inserts into the fungal cell membrane parallel to the acyl chains of the phospholipid. Apparently bind to the sterol moiety of the membrane, although the exact mechanism is unclear. Several molecules of the drug form a cylindrical channel in the membrane that allows cations, then macromolecules to leak out. When macromolecules leak, cell dies.

B. Toxicity: Destabilizes human cell membranes. Triggers release of cytokines from human mononuclear cells.

C. Resistance: Present in some species (most importantly, dermatophytes) but rarely inducible. Resistant strains have diminished membrane ergosterol with less membrane affinity for the polyene

Individual polyenes

A. **Nystatin**

History: isolated in the N.Y. State Health Labs from S. nodosus in 1950 by two biochemists (Elizabeth Hazen, Rachel Brown)

Pharmacology: Topical administration only

Toxicity: minimal (occasional hypersensitivity)

Use: skin and mucosal candidal infections. No effect on dermatophytes.

B. **Amphotericin B**

- History: isolated from S. noursei from soil of the Orinoco Valley, Venezuela, in 1950s.
- Pharmacology: Not orally absorbed. Sometimes given as a slurry for oral or esophageal infection. Most commonly administered intravenously as a colloidal dispersion in deoxycholic acid, a bile salt. Protein bound in serum and tissue. Tissue stores persist for weeks to months, slowly released. Urine and CSF concentrations low.
- Toxicity: immediate: dramatic fever, chills, nausea, vomiting, diarrhea during infusion
- cumulative: after days-weeks of therapy, characteristic K- and Mg-wasting renal failure, anemia.
- Use: Still the gold standard for systemic fungal disease caused by both yeasts (cryptococcus, candida) and molds (dimorphs). Not active against dermatophytes. Only slowly being supplanted by less toxic alternatives. Toxicity usually mandates confirmation of diagnosis before drug is used.
- Modifications:
- Amphotericin encased in liposomes or highly lipid-associated has less nephrotoxicity with similar efficacy. (Mechanism unclear, may stem from reduced free AmB in solution, on which human cell toxicity seems to depend). These preparations permit higher cumulative doses with less cumulative toxicity but greater drug costs (as much as 30-fold more \$\$\$).
- Preparations:
- Liposomal Amphotericin ("AmBisome")
  - Amphotericin B Lipid Complex ("Abelcet")
  - Amphotericin B colloidal dispersion with cholesteryl sulfate ("Amphotec")
- Generally used in persons with:
- documented systemic fungal infection

- predicted need for long course of amphotericin
- documented or likely nephrotoxicity

## 2. Azoles

### History:

Synthetic antifungal agents developed over last 25 years. Early ones too toxic for any but topical use. Later ones have opened a new age in antifungal treatment because they allow ORAL treatment of systemic infections.

Structure: Imidazoles have 2N in 5-membered ring  
Triazoles have 3N in 5-membered ring

### Mechanisms:

#### A. Action:

Inhibit cytochrome P450 enzymes by binding to a heme moiety, and interfering with certain mixed oxidase functions. Specifically: stop alpha-demethylation of lanosterol, blocking the formation of ergosterol and leading to an accumulation of alpha-14-methyl esters in the cell. Both of these effects slow/stop fungal growth. May have other direct effects on membrane integrity. Generally considered to be fungistatic, although in high doses they may be fungicidal.

#### B. Toxicity:

Inhibit cholesterol-dependent steroid hormone synthesis (cortisol, testosterone). May lead to aldosterone-like effects from accumulation of metabolites. May also affect the metabolism of other cytochrome p 450-dependent drugs. Liver toxicity and skin rash also seen.

#### C. Resistance:

Rare, but increasing with increasing use (inducible). Appears to result either from alteration in p450 enzymes or from change in fungal membrane lipids

resulting in decreased fungal cell permeability to the azole.

### Individual azoles

#### **A. Clotrimazole, Miconazole, Terconazole**

History: Older azoles

Pharmacology: Topical. (parenteral miconazole used briefly 1970s but extremely toxic).

Toxicity: minimal

Use: Dermatophytes and mucosal candidal infections. (inexpensive and effective)

#### **B. Ketoconazole**

History: Released 1983 The first oral agent with good activity against some systemic fungal disease

Pharmacology: Insoluble in water save at acid pH. No parenteral form available. Requires gastric acidity for oral absorption. Circulating drug bound to plasma proteins/ RBC membranes. Extensive metabolism in liver. No CSF or urine penetration. Most excreted in feces.

Toxicity: Dose-related adrenal hormone suppression: low serum testosterone (libido, gynecomastia); low serum cortisol response to ACTH. Rare adrenal insufficiency. Hepatitis.

Use: Mucosal candida. Sporotrichosis. Some success with Cocci, Histo. May be the most effective azole against pityriasis and the dermatophytes in vitro.

**C. Fluconazole**

History: Released 1990.

Pharmacology: Soluble in water at neutral pH. Rapid, excellent oral absorption, excellent CSF penetration. IV form available. Minimal hepatic metabolism; most excreted unchanged in the urine.

Toxicity: Relatively little (occasional GI distress, hepatitis, rash).

Use: Cryptococcosis (high CSF levels), cocci, disseminated and mucosal candidiasis.

**D. Itraconazole**

History: released 1992.

Pharmacology: Almost insoluble in water. Oral absorption enhanced by food/acid; no iv form available. Low serum levels (all protein-bound), higher tissue levels, very high adipose tissue and keratinized tissue levels. Highly metabolized in the liver. Some renal excretion of active metabolite.

Toxicity: similar to fluconazole

Use: Potential still under investigation. Histoplasmosis, sporotrichosis, onychomycosis.

**E. Voriconazole**

History: released 2002

Pharmacology: A synthetic derivative of fluconazole, with a broader spectrum of activity. Addition of a methyl group to fluconazole's propyl backbone increases affinity of the drug for the 14-alpha sterol demethylase of *Aspergillus fumigatus* tenfold. Also active

against fluconazole-resistant non-albicans *Candida*. Good oral absorption, shorter half-life than fluconazole, 50-60% protein-bound. Primary route of elimination hepatic.

**Toxicity:**

Unique visual disturbances in 10% (bright lights, flickering, blurry vision, ?mechanism). Otherwise similar to fluconazole.

**Use:**

Under investigation. Superior to Amphotericin B in one trial for invasive aspergillosis.

**Comparison of newer azole drugs**

	Ketoconazole Oral	Fluconazole Oral/IV	Itraconazole Oral/IV	Voriconazole Oral/IV
Formulations available				
Steady-state plasma conc (mg/L)	4-6	5-10	1-2	2-5
Oral availability	requires acid	>90%	55%	90%
Elimination $\frac{1}{2}$ life	8h	30h	24h	6h
Protein binding	99%	12%	99.8%	50-70%
Volume of dist'n		.7-1.0	10-11	2
Primary route of elimination	liver	kidneys	liver	liver
CSF penetration	little	good	little	?considerable

### 3. Echinocandins

History: semisynthetic drugs (first was isolated from *Aspergillus* species in 1974 and then structurally modified)

Mechanisms of

- a. Action: Inhibit fungal cell wall synthesis by disrupting glucan formation. Noncompetitive irreversible inhibitors of 1,3 beta glucan synthase. Considered fungicidal.
- b. Toxicity: Target enzyme not present in human cells, implying little direct human toxicity.
- c. Resistance: Attempts to induce in-vitro resistance by exposing isolates to drug have not succeeded. Some isolates (i.e. *C. krusei*) have relative inherent resistance.

First available agent: **Caspofungin** ("Cancidas") (approved January 2001)

Pharmacology: Intravenous only; infused slowly over 1 hour once daily. Half-life 10 hours. 95% protein bound. Elimination probably hepatic.

Use: Fungicidal against wide range of pathogens, including *Histoplasma*, *Candida*, *Aspergillus*. Approved for treatment of invasive aspergillosis failing other therapy.

Toxicity: Infusion may cause histamine release but otherwise less renal and hematologic toxicity than Amphotericin B.

### 4. Miscellaneous agents

#### A. Griseofulvin.

Venerable oral antifungal active only against the dermatophytes. Isolated from *penicillium griseofulvum* in 1939. Inhibits fungal mitosis by disrupting microtubules composing the mitotic spindle. Still used, relatively nontoxic, long courses of tx required; will most likely be supplanted by the azoles and the allylamines (below).



**B. 5-Fluorocytosine**

Fungal cells deaminate this compound to 5-FU, which is a potent antimetabolite (used for human chemotherapy). It is incorporated into RNA itself, or further metabolized to 5-fluorodeoxyuridylic acid, which is a potent inhibitor of thymidylate synthetase. Rarely used alone as an antifungal agent since resistance emerges quickly, but is often used for an additive or synergistic effect against severe candidal, cryptococcal, sometimes aspergillus infections.

Note: mammalian cells don't deaminate 5-FC but the bacteria in the human gut do. Hence one of its major toxicities, a characteristic necrotizing enteritis confined to the colon where the bacteria are.

**C. Allylamines/ Thiocarbamates**

Structured around a naphthalene ring, they inhibit squalene epoxidase, an essential enzyme in the synthesis of ergosterol. Active only against dermatophytes, but fungicidal against them. Available in both oral and topical over-the-counter forms.

**Pharmacologic treatment of common fungal infections**

Infection	Drug alternatives
<b>Superficial</b>	
Candida	Topical polyene (nystatin)
Skin dermatophyte	Topical azole; oral for severe Combine with topical steroids for symptomatic relief.
Nail dermatophyte	Itraconazole, terbinafine
<b>Subcutaneous</b>	
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Sporotrichosis	Itraconazole, amphotericin B
<b>Systemic</b>	
Histo	Amphotericin B, itraconazole
Cocci	Amphotericin B, itraconazole, fluconazole
Blasto	Amphotericin B, itraconazole, fluconazole
Cryptococcosis	Amphotericin B, fluconazole
Candidiasis	Amphotericin B, azoles, caspofungin
Aspergillosis	Amphotericin B, voriconazole, caspofungin