Candidal Endocarditis Presenting with Bilateral Lower Limb Ischemia

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ABSTRACT

The incidence of fungal endocarditis is increasing. While the pathogenic mechanisms are not fully understood, infection is associated with underlying heart disease and is most often attributable to *Candida* species. Candidal endocarditis complications include heart damage, inflammation, and emboli with resulting ischemia and tissue death. Candidal endocarditis is difficult to diagnose as blood cultures are often negative. Treatment includes surgical intervention and antifungal therapy. This case study describes a 41-year-old female complaining of acute onset of pain with numbness and tingling in both lower extremities. Prior history was significant for mycotic valve aneurysm and replacement secondary to culture-negative endocarditis. Evidence of limb-threatening ischemia led to a bilateral thrombectomy. During the thrombectomy white debris, later identified as *Candida albicans*, was encountered. A transesophogeal echocardiogram revealed a pedunculated mass which was determined to be the source of infection. The patient was placed on micafungin and voriconazole and discharged with a diagnosis of *C. albicans* fungal infection with descending aorta fungal mass. This case study illustrates an unusual presentation of candidal endocarditis with discussion of disease epidemiology, pathogenesis, diagnosis, and treatment.

ABBREVIATIONS: Spp. - species, KOH - Potassium Hydroxide, PNA FISH - peptide nucleic acid fluorescence in situ hybridization, DVT - Deep Vein Thrombosis, WBC - White Blood Cell

INDEX TERMS: *Candida albicans*, Candidiasis, Endocarditis

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Case Study

A 41-year-old female presented to a local hospital complaining of acute onset of pain with numbness and tingling of the lower extremities and evidence of limb-threatening ischemia. Upon review of previous history, it was noted the patient had a repair of a mycotic aneurysm with concomitant aortic valve replacement secondary to endocarditis seventeen months prior to arrival. A second episode of endocarditis occurred post repair, however, no organism was recovered and the patient was treated empirically with combination antibacterial therapy consisting of vancomycin and ceftriaxone.

The patient had no recent history of fever, chills, sweating, respiratory infections, gastroenteritis, or urinary tract infection. However, the patient did have a history of a hypercoagulable state secondary to presence of Factor V Leiden and was on long-term anticoagulation therapy consisting of warfarin. Patient history also revealed a central nervous system bleed secondary to warfarin, deep vein thrombosis (DVT), and previous intravenous drug use. The patient denied any recent substance abuse.

On admission, laboratory results indicated an elevated white blood cell (WBC) count with neutrophilia and anemia. Polychromasia, anisocytosis, ovalocytes, and hypersegmented neutrophils were observed. Chemistry studies revealed a decreased serum iron, an increased ferritin, normal vitamin B12 and haptoglobin values,

and an increased lactate dehydrogenase. Coagulation results were abnormal. Laboratory results are summarized in Table 1.

Table 1. Laboratory Results on Admission

Test	Patient Result	Reference Range
WBC Count	$19.0x10^9$/L	$3.2\text{-}11x10^9$/L
Neutrophils	78%	50-70%
Lymphocytes	4%	20-50%
RBC Count	$3.42x10^{12}$/L	$3.6\text{-}4.9x10^{12}$/L
Reticulocyte	2.05%	0.2-1.6%
Hemoglobin	9.3g/dL	11.4-14.1g/dL
Hematocrit	28.5%	33.4-42%
PT	19.9 seconds	9.2-12.1 seconds
PTT	42.0 seconds	20-32 seconds
INR	1.80	2.0-3.0
Blood Culture	No Growth at 5 days	No Growth at 5 days
Ferritin	177.7µg/dL	3-105µg/dL
Serum Iron	10µg/dL	28-170µg/dL
LDH	355 U/L	98-192 U/L

WBC = White Blood Cell, RBC = Red Blood Cell, PT = Prothrombin Time, PTT = Activated Partial Thromboplastin Time, INR = International Naturalized Ratio, LDH = Lactate dehydrogenase

Due to the limb-threatening ischemia, a bilateral femoral-iliac and aorto-iliac thrombectomy was conducted to correct the occluded distal aortae. Bilateral white debris was encountered and sent for culture. Upon potassium hydroxide (KOH) preparation of both the left and right iliac clot, many fungal elements were observed. No acid fast bacilli or anaerobic bacteria were isolated. Three sets of blood cultures revealed negative results except for growth of coagulation negative Staphylococcus spp. which was determined to be a contaminant.

A tissue culture resulted in moderate growth of suspected *Candida albicans*, confirmed with microscopic morphology and biochemical tests. This led to a diagnosis of *C. albicans* fungal infection of the descending aorta.

The patient's history of previous DVT increased the risk of an additional thrombotic event, potentially complicated by the Factor V Leiden mutation which subjected the patient to a hypercoagulable state. Iron studies performed during hospitalization revealed an increased ferritin level and decreased serum iron level indicative of anemia of chronic disease, attributable to the fungal infection. The intravascular hemolysis was demonstrated in this case by the elevated lactate dehydrogenase, decreased red blood cell count, and decreased hemoglobin and hematocrit.

Given the previous history of a mycotic aneurysm with concomitant aortic valve replacement secondary to endocarditis, a transesophogeal echocardiogram of the heart valve was ordered. The procedure revealed a mass in the descending aorta and it was determined to be the source of the infection.

Previous antibacterial therapy, empiric vancomycin and ceftriaxone, was ineffective against the fungal organism and may have facilitated the pathogenesis. Laboratory results demonstrated a steadily decreasing WBC count from $19.0x10^9$/L at admission to $12.2x10^9$/L. Warfarin dosage was appropriately adjusted to therapeutic range with an International Normalized Ratio of 2.39. The patient was placed on combination antifungal therapy consisting of micafungin and voriconazole and discharged. No further follow-up information was available.

Disease Overview

The discovery of human disease caused by *Candida spp.* was first described by Gruby in 1842. The number of cases of pathogenic fungi causing disease has since risen with *Candida spp.* now accounting for approximately 8-15% of nosocomial blood stream infections.[1] Among this genus, *Candida albicans* ranks as the main cause of yeast infections in the world and is currently the fourth most common cause of blood-borne infection in the United States.[2]

Although the prevalence of nosocomial candidemia is high, the occurrence of fungal endocarditis is rare (approximately 1.3 – 6% of all cases).[3,4] This rare but serious infection appears to mainly affect younger patients with an average age of 44.5 years.[5] Common etiologic agents of fungal endocarditis are *C. albicans* (30.3%) and *Aspergillus fumigatus* (28%), of which, *C. albicans* has a better prognosis.[4,5] Fungal endocarditis carries a morbidity and mortality rate of 50% which is partially due to issues such as systemic embolization and the difficulty in isolating the fungi with routine blood cultures.[5] The difficulty in isolation is attributed to the intermittent presence of fungi in the bloodstream.[6]

Candida albicans possesses many virulence factors which contribute to its pathogenicity. Among these factors are

phenotypic switching and the pleomorphic nature of the organism. It is estimated that one colony in $10x10^4$ colonies exhibits phenotypic switching, such as a change in morphology, cell surface proteins, and metabolism.[7] Among the factors regulating the morphogenesis between yeast and hyphal forms are temperature and pH. The formation of hyphae is favored at higher temperatures (37ºC) and neutral pH, while the yeast form is favored at lower temperatures (<30ºC) and acidic pH (<6.0).[8] Darkowska-Kuleta et al.[7] have hypothesized that the yeast form occurs during dissemination in the environment and the hyphal form occurs during tissue damage and invasion.

The survival of *Candida* may be enhanced through the formation of biofilms. Biofilms are communities of microbial cells, attached to each other and/or a substratum. Both yeast and hyphal forms of *C. albicans* are found in biofilms located on artificial devices. A polysaccharide matrix associated with the biofilm plays an important part in drug resistance. *C. albicans* cells within this matrix express several other genes which influence pathogenicity, adhesion, carbohydrate synthesis, regulation of efflux pumps, and quorum sensing.[7]

Contributing to the virulence of *C. albicans* infections is the ability of the organism to uptake iron from host sources. Several iron acquisition systems exist: reduction of extracellular ferric chelates; production of siderophores with high iron binding affinity; and heme iron uptake. The majority of circulating iron is bound to hemoglobin in erythrocytes. *C. albicans* hemolyzes the cell in order to obtain heme iron. This hemolytic activity relies on the intracellular enzyme heme oxygenase in order to release iron from porphyrin chelate.[9]

Although they are normal biota of various locations, including skin, oral mucosa, and digestive tract, *Candida spp.* are commonly associated with yeast infections. Upon altered immune status in the host, this organism is capable of causing a range of diseases from superficial skin infections to disseminated disease.[2] Predisposing factors which can lead to candidemia include underlying valvular disease, malignancy, indwelling vascular catheters, use of antimicrobial agents, heroin addiction, immunosuppression and surgery.[10,11] These patients may develop thrush, an oral mucosa infection of *C. albicans*, which may disseminate and cause more serious systemic disease.[2] Many other disseminated infections may occur, frequently due to neutropenia.[9] In addition, the loss of the capability of neutrophils to phagocytize and kill pathogens can lead to extensive invasive disease.

Candida spp. may be isolated from many clinical specimens such as skin, oral mucosa, and the digestive tract. A 10% solution of KOH is useful for observing fungal elements in skin, hair, nails, and tissue. The KOH preparation of a specimen suspected of containing *C. albicans* should be examined for small, oval to round, thin-walled budding yeasts and pseudohyphae. A modification to this procedure which uses a fluorescent dye, such as calcofluor white, can be used to observe fungal structures. Histological stains like periodic acid-Schiff and Gomori methenamine-silver nitrate can also be used to visualize fungal elements.

In addition to preliminary results obtained from wet preparations and stains, a diagnosis of *C. albicans* requires identification of the isolate from a clinical specimen. Although the recovery of yeasts from conventional blood culture systems is difficult due to the slower growth of the fungi compared to bacteria, recovery of pathogenic fungi can be enhanced with the lysis-centrifugation method. This method produces a concentrated sample of blood for direct inoculation onto solid media as opposed to conventional incubation in broth media. Increasing the incubation period to 4 weeks may also increase recovery.[2]

After incubation at 37ºC on Sabouraud Dextrose Agar *Candida spp.* produce creamy white colonies with a smooth and waxy appearing surface. Tiny projections, commonly referred to as feet, appear at the base of the colony. Although media containing chromogenic substrates, such as CHROMagar™, can assist in the identification of *C. albicans*, differentiation of *C. albicans* from other *Candida spp.* is accomplished using germ tube production, growth on Cornmeal agar, or carbohydrate assimilation.

When incubated in serum or plasma at 37ºC for 3 hours, both *C. albicans* and *Candida dubliniensis* produce germ tubes. A germ tube represents the initial stage of hypha formation in the yeast cell. Cornmeal agar induces spore formation and is important in

determining yeast morphology. While both yeasts produce thick-walled vesicles called chlamydospores, they can be differentiated by growth of *C. albicans* at 42°C and the lack of growth of *C. dubliniensis* at this temperature. The carbohydrate assimilation tests are valuable for determining aerobic sugar utilization of sugars as a carbon source. Results of carbohydrate assimilation by *Candida spp.* are shown in Table 2. Assimilation patterns may be established by manual, commercial, or automated methods. Examples of commercially available systems include API *Candida* (bioMerieux) and the MicroScan WalkAway→ System (Dade Behring).

Table 2. Differentiating Characteristics of Candida spp.*

Biochemical Characteristics										*Candida*
Morphology				Assimilation						Species
PH	GT	CS	42°C	G	S	L	T	R	C	
+	+	+	+	+	+	-	+	-	-	*C. albicans*
+	-	-	+	+	+	-	+	-	+	*C. tropicalis*
-	-	-	+	+	-	-	+	-	-	*C. glabrata*
+	-	-	-	+	+	-	+	-	-	*C. parapsilosis*
+	-	-	+	+	-	-	-	-	-	*C. krusei*
+	+	+	-	+	+	-	+	-	-	*C. dubliniensis*

* modified from references 1 and 15.
PH = Pseudohyphae, GT = Germ Tube, CS = Chlamydospore, G = Glucose, S = Sucrose, L = Lactose, T = Trehalose, R = Raffinose, C = Cellobiose

In addition, definitive identification may be achieved using molecular methods. Fluorescence in situ hybridization assays employing peptide nucleic acid probes (PNA FISH) to target species-specific RNA in yeast have been developed. The PNA-FISH *C. albicans* assay (AdvanDx, Inc.) can identify *C. albicans* from a positive blood culture in as little as three hours.[12]

Given the wide range of infections, from non-life-threatening mucocutaneous infections to systemic illness, a broad range of antifungal strategies is required to successfully treat *Candida spp.* infections. The azole antifungal agents, such as miconazole and fluconazole, are the therapeutic agent of choice for mucocutaneous candidiasis while amphotericin B, the azole antifungal agents, and echinocandin antifungal agents have been the standard treatments for invasive candidiasis.[13] Current management of endocarditis caused by *Candida spp.* calls for a combination of amphotericin B and flucytosine followed by prolonged suppression with fluconazole. It should be noted, long-term suppressive therapy can result in the formation of biofilms and development of multidrug resistance which complicates treatment.[14]

It has been demonstrated that within biofilms, *C. albicans* cell membranes have a reduced ergosterol content. This may explain the poor activities of antifungal agents, such as amphotericin B, that target ergosterol. In contrast the echinocandins, such as capsofungin, anidulafungin, and micafungin, target the cell wall and may be more active than amphotericin B against *C. albicans* biofilms. Inhibition of DNA synthesis with flucytosine has also been shown to be effective against biofilms.[14]

CONCLUSION

This case illustrates an unusual presentation of candidal endocarditis: pain and numbness of the lower extremities. Surgical intervention to correct occulsion of the distal aortae revealed white debris, later identified as *C. albicans*. Vegetation in the descending aorta was determined to be the source of the infection. The patient was treated with antifungal therapy and released.

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