Mixed Pneumocystis and Cryptococcus Cutaneous Infection Histologically Mimicking Xanthoma

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Abstract: Cutaneous *Pneumocystis jirovecii* infection is rare. It is thought that the disease emerges from a latent infection delivered via hematogenous and/or lymphatic dissemination from a primary lung infection in immunocompromised individuals. A 32-year-old human immunodeficiency virus–positive male was admitted for headache and vomiting. He was diagnosed with meningitis due to *Cryptococcus neoformans* and sputum tested positive for *Pneumocystis*. Six months later, he presented with a slightly crusted yellowish brown plaque and 2 similar but smaller papules with telangiectasia near the right angle of the mouth. Biopsy of the area featured histiocytes expanded by foamy cytoplasm as in a xanthoma except that the vacuoles were coarser. Special stains ultimately demonstrated the characteristic disks of *Pneumocystis* accompanied by a minor component of budding yeasts (*Cryptococcus*) in the same fields. This case illustrates the utility of adequate special stains in recognizing a mixed cutaneous infection, particularly in human immunodeficiency virus–positive patients, when microscopy presents an odd xanthoma-like lesion.

Key Words: pneumocystosis, cutaneous, xanthoma, extrapulmonary, HIV, *Pneumocystis jirovecii*

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INTRODUCTION

The Centers for Disease Control and Prevention has determined that the first opportunistic infection in human immunodeficiency virus (HIV)–infected individuals is typically *Pneumocystis jirovecii* pneumonia, formerly known as *Pneumocystis carinii* pneumonia.[1] *Pneumocystis jirovecii*, a fungus, is normally a commensal organism in man, not a zoonosis.[2] It is thought to spread through the air, making prevention of exposure difficult.

Rarely, *P. jirovecii* exhibits hematogenous spread beyond the lungs, typically in the setting of prophylaxis with aerosolized pentamidine but also with no prophylaxis at all.[3] A decade ago, a large review, during the ongoing HIV-1 and related *Pneumocystis carinii* pneumonia epidemic, identified at least 90 cases of extrapulmonary pneumocystosis.[4] Usually, examples of extrapulmonary *P. jirovecii* dissemination occurred without documented pulmonary infection and despite prophylaxis with high-dose dapsone. In addition, it may rarely present as primary otic pneumocystosis as the harbinger of underlying infection with HIV.[5] However, pathologic evidence suggests that extrapulmonary pneumocystosis occurs via the lungs in most cases.[6] It seems that extrapulmonary pneumocystosis is due to the reactivation of latent infection of hematogenous and/or lymphatic dissemination of organisms from the lung and rarely by primary infection.

Like *P. jirovecii*, cutaneous cryptococcosis occurs after hematogenous dissemination in immunocompromised individuals. In contrast to *P. jirovecii*, the cutaneous spread originates from cryptococcal meningitis.[7]

Here is presented an unusual case of cutaneous mixed pneumocystosis and cryptococcosis mimicking xanthoma histologically.

CASE REPORT

A 32-year-old HIV-positive male was admitted to the hospital for headache and vomiting. Initial blood smears revealed "fungal elements" represented by rare spore forms. A cerebrospinal fluid (CSF) specimen was india ink positive for *Cryptococcus*. A CSF culture grew *Cryptococcus neoformans*. Therapy with amphotericin B (0.7 mg/kg intravenously per day) plus flucytosine (100 mg/kg/day) for 2 weeks, followed by fluconazole (400 mg daily) for 8 weeks of consolidation therapy and 200 mg daily for maintenance therapy, was initiated. Cryptosporidium and Giardia antigen positivity were detected in a stool specimen. A repeat CSF specimen revealed cryptococcal-like organisms in an india ink preparation; however, cultures were negative, predicted by the nonviable appearance of the posttreatment organisms. Sputum for *Pneumocystis* was positive and a diffuse miliary nodular pattern was seen on x-ray (Fig. 1), which motivated the addition of trimethoprim/sulfamethoxazole at 2 tablets orally for every 6 hours for a 3-week course to his medication regimen. The patient dramatically improved and was discharged after a 3-week stay. Five months later, he returned to the Emergency Department with headache, fever to 101.3°F with chills, and some neck stiffness and began vomiting that afternoon. Lung fields were radiographically improved from 5 months earlier. A lumbar puncture had a white count of 60 cells per deciliter (normal = 0–5 cells/dL), 100% monocytes, glucose 21 mg/dL (normal = 50–80 mg/dL), and protein 109 mg/dL (normal = 15–60 mg/dL); no organisms were seen on Gram stain, and cultures were sterile. The patient spontaneously improved and was discharged. Soon thereafter, a 7-mm brownish plaque with slight crust and telangiectasia appeared on the right upper lip (Fig. 2A), followed by two smaller similar papules nearby below the right angle of the mouth (Fig. 2B). Clinically, these resembled basal cell carcinomas.

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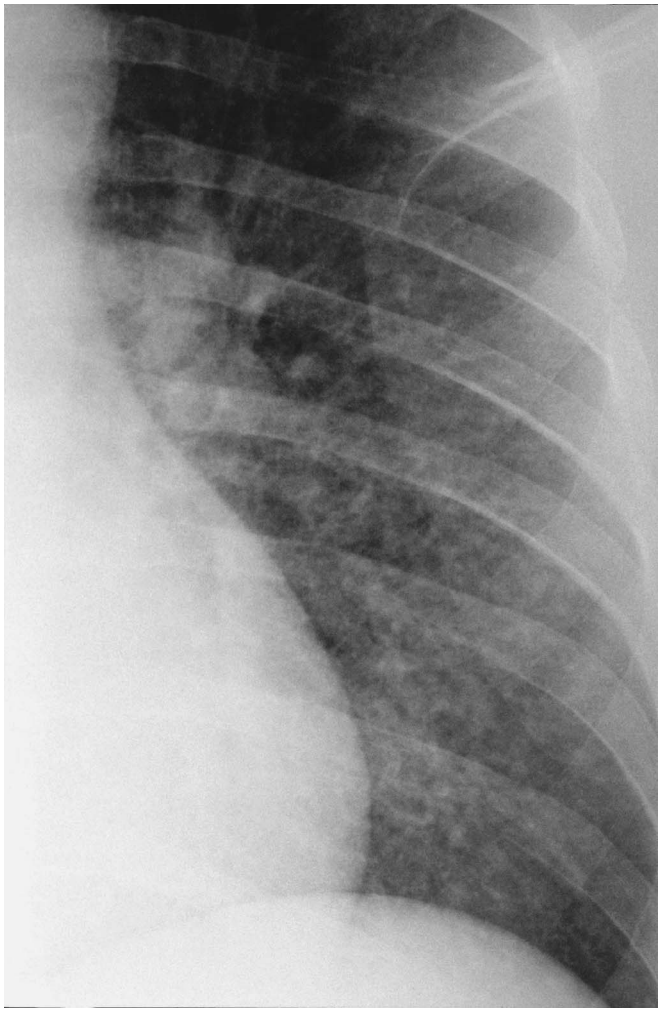

FIGURE 1. Radiograph of the patient's lung 6 weeks before appearance of 3 facial lesions.

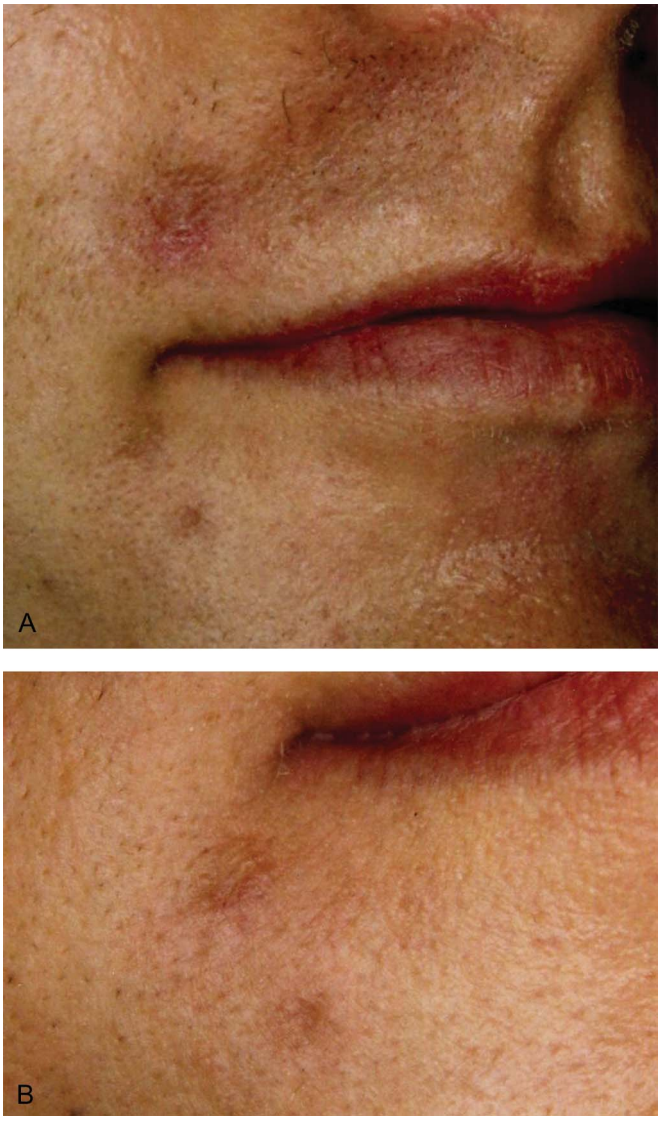

FIGURE 2. Facial lesions consisted of a brownish plaque with slight crust and telangiectasia above the lip (A) and 2 small papular lesions at the corner of the mouth (B).

A shave biopsy was performed of the right upper lip lesion. The 4 × 4 × 1-mm shave biopsy specimen was fixed in 10% buffered formalin and routinely processed. Periodic acid–Schiff stain with and without diastase, Gomori methenamine silver (GMS), Giemsa, and mucicarmine special stains were performed using positive and negative controls.

In the hematoxylin and eosin slide, under medium power the lesion resembled a xanthoma (Fig. 3), with sheets of abundant foamy histiocytes distending the dermis and elevating the overlying epidermis. On higher magnification, the vacuoles were coarser than in histiocytes of a xanthoma (Fig. 3, inset). By lowering the condenser and thus increasing the refractive index, unstained translucent discoid structures approximately the size of red blood cells were visualized in the vacuoles. These motivated special stains, becoming more visible with Giemsa and particularly with GMS (Fig. 4) but not with Periodic acid-Schiff with distase. With GMS, the majority organisms were a uniform red blood cell size. Their discoid shape and staining characteristics fit well with *P. jirovecii*. However, some budding and variability of the size and shapes were puzzling. A standard 1-hour mucicarmine stain performed in search of the mucinous capsule of *Cryptococcus* did not show it. A 48-hour mucicarmine stain (Fig. 5) revealed larger ovoid structures (arrowhead) demonstrating a few budding yeast (Fig. 5, inset) consistent with capsule-deficient *C. neoformans*, in addition to the numerous small discoid structures (Fig. 5, arrow) consistent with *P. jirovecii*, indicating a mixed infection. Both Fite and Ziehl-Neelsen stains for acid-fast bacilli showed no mycobacteria. Giemsa was negative for sporozoites.

Fresh tissue was not available for detection of *P. jirovecii* by direct immunofluorescence. Immunohistochemistry for *P. jirovecii* was attempted on paraffin-embedded tissue but failed due to exhaustion of the block by special stains. Although it is ideal to have either direct immunofluorescence or immunohistochemistry confirmation for diagnosis, the present case serves to underline the reality that when these tools are not available, dermatopathologists can and should apply their critical thinking to clinicopathologic–radiologic correlation to solve unusual puzzles.

Due to his recent improvement with negative sputum and CSF cultures and the fact that no cultures of the cutaneous lesions were performed, no intravenous therapy was initiated. Because pathologic findings were consistent with a mixed cutaneous infection,

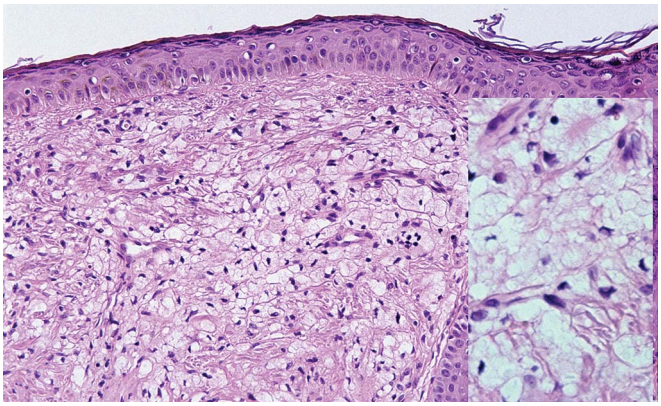

FIGURE 3. Abundant interstitial infiltrate of histiocytes with abundant foamy cytoplasm and scattered lymphocytes slightly elevates the overlying epidermis. At higher magnification (inset), the foamy cytoplasm is coarser than in typical xanthoma cells (hematoxylin and eosin, original magnification ×20 and ×40).

fluconazole was increased to 400 mg daily for 8 weeks and a 3-week course of trimethoprim–sulfamethoxazole was repeated.

By the end of the treatment regimen, the lesions resolved, and at a visit 2 years after the biopsy, his viral load was significantly lower. No new lesions had occurred, and the patient was working and fully integrated into daily life.

DISCUSSION

Cutaneous involvement by *Pneumocystis* organisms is rare. The first reported case of skin involvement by *P. jirovecii* in a patient with AIDS was a 42-year-old man with a 9-month history of progressive hearing loss and bilateral external auditory canal masses.[7] Delayed treatment in another HIV-infected patient resulted in *Pneumocystis* extending from skin of the external auditory canal into the middle cranial fossa.[8] Otic foaming exudates can be the first sign of ear infection,[9] and early treatment typically results in resolution.[5]

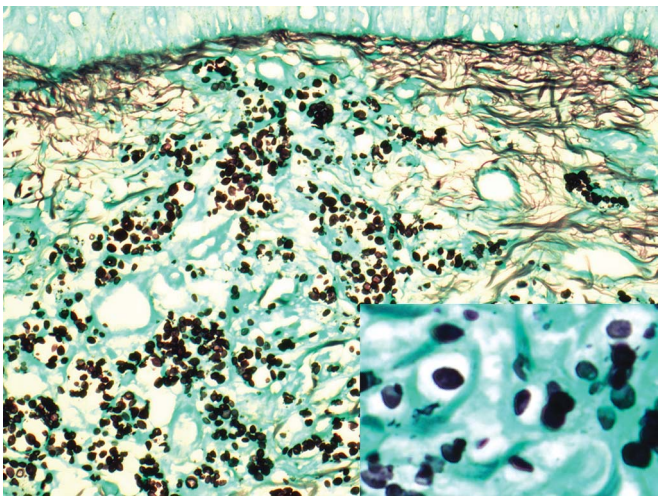

FIGURE 4. Gomori methenamine silver staining reveals abundant, small, discoid staining organisms within the foamy cytoplasm of engorged macrophages. The majority of the organisms (*Pneumocystis*) are of uniform size at approximately the size of erythrocytes, and occasional larger ovoid organisms (*Cryptococcus*) are admixed (inset) (Gomori methenamine silver, original magnification ×20 and ×40).

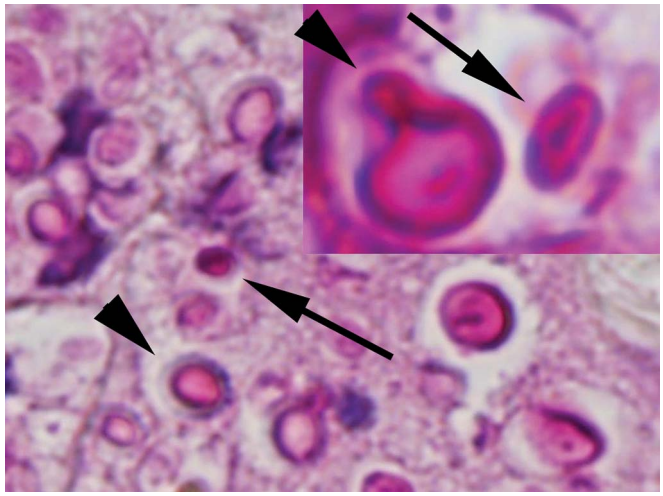

FIGURE 5. On mucicarmine special staining, numerous oval structures are seen within the abundant frothy cytoplasm. The majority of structures are the size of erythrocytes (arrows: *Pneumocystis*) with admixed larger ovoid structures (arrowheads: *Cryptococcus*) exhibiting occasional budding (inset). A thin mucicarmine-positive capsule is brought out around the larger organisms (48-hour mucicarmine stain, original magnification ×20 and ×40).

At sites other than the ear canal, *P. jirovecii* infection's clinical presentation varies from ill-defined papules to multiple discrete nodules[2,3] and may mimic granulation tissue[5] or Kaposi sarcoma.[10] Mixed skin infection with both *P. jirovecii* and *C. neoformans* is extremely rare[11] and may present as in this case as small plaques or papules.

Precisely, why the macrophages/histiocytes develop abundant vacuolated cytoplasms in the setting of engulfed *Pneumocystis* organism is unknown but likely relates to the loss of the ability to mount an effective immune response. In any event, the abundant foamy cytoplasmic expansion around the discoid organisms, resembling the "foamy alveolar casts"[12] familiar in bronchial lavage specimens, mimics xanthoma histologically. By electron microscopy, this "honeycombed material" has been described as mainly composed of dilated trophozoite-type organisms, containing scattered thick-walled cysts.[13] Contributing to it are the membranous materials beyond the cyst wall of *Pneumocystis*. An outer membrane in the cyst wall of *P. jirovecii* can be detected by a combination of techniques, including transmission electron microscopy, freeze-fracture electron microscopy, and membrane labeling with fluorescent lipid analogues.[14] This sugar-rich second membrane may have an important role in osmoregulation and nutrient utilization and may mediate *P. jirovecii*–host interaction, playing an important protective role in resisting host defenses and antimicrobial drugs by creating a permeability barrier around the cyst.[15]

Ultrastructurally, the foamy alveolar cast familiar in bronchial lavage specimens is composed of intimately

associated cysts and trophozoites and contains little or no fibrin,[12] unlike "exudates." By electron microscopy, *P. jirovecii* conglomerates are held together by slender membranotubular extensions growing from their concave and convex surfaces. Voids, formed by the juxtaposition of the uneven contours of the organisms and their intertwined membranotubular extensions, contribute significantly to the foamy alveolar casts. Taken together, the foamy alveolar cast material housing the organisms consists of partly digested organisms, especially denatured proteinaceous, polysaccharide, and lipid residues of the plasma membrane after excystation of intracystic bodies described by Yoshikawa et al.[16]

Infection in an immunocompromised individual, as in the present case, poses additional challenges in distinguishing *C. neoformans* from *P. jirovecii*. The host's weak or absent immune response to the organism obviates the need for *C. neoformans* to produce its protective capsule,[17] making it look very similar to the trophozite stage of *P. jirovecii*. However, subtle morphologic differences between these 2 organisms still allow differentiation, namely, mode of reproduction, shape, and size. *Cryptococcus neoformans* is spherical, reproduces by budding (Fig. 5, inset), and varies widely from 5 to 20 μm in size, whereas *P. jirovecii* is discoid, reproduces by bursting and releasing up to 8 spores, and is smaller staying within the range of 1.5–5 μm in largest diameter. Size difference and budding indicate a mixed infection in the present case.

Three patients with HIV-1 infection developed "facial papular xanthomatosis" associated with hypergammaglobulinemia and an immunoglobulin A gammopathy.[18] Special stains were negative for bacteria, mycobacteria, and spirochetes. No stain for fungal agents was included. Phagocytosed nuclear debris and hyalinization with areas of hyaline necrosis of collagen fibers were in their specimens. When these clinical and histologic features are absent as in this present case, it may be wise to exclude infection by *P. jirovecii*, including a mixed infection with *C. neoformans*.

Clinically, cutaneous *Cryptococcus* infection may present as molluscum contagiosum,[19] basal cell carcinoma,[20,21] keloid,[22] and histologically as a "granulomatous process."[21] Cutaneous cryptococcosis is typically secondary to dissemination after cryptococcal meningitis in immunocompromised individuals.[7] Hicks et al[23] reported perioral "umbilicated nodules " of *Cryptococcus* in a 27-year-old HIV-seropositive man with a history of *P. jirovecii* pneumonia and *Cryptococcus*-positive CSF. The polysaccharide capsules of *Cryptococcus* are typically identified by conventional 1-hour mucicarmine staining; however, mucicarmine stain is an empirical stain, and when capsules are partly digested by engulfing macrophages, as in the present case, 24- to 48-hour incubation in the mucicarmine stain may be required to stain capsule remnants (R. Neafie, oral communication, December 2010).[24]

The variable clinical presentation and the predominantly macrophage-based histologic findings of these cutaneous fungal infections relate to decreasing T-cell function, antigen response, and shifting cytokine expression and a propensity for autoimmune reactions occurring in the setting of HIV infection.[12] Cutaneous mixed fungal infections have been previously reported in HIV-infected individuals including histoplasmosis and cryptococcosis and, as in the present case, *Pneumocystis* and *Cryptococcus*.[11] This latter mixed infection clinically presented with multiple necrotic papules and nodules on the face associated with smaller flesh colored papules and, unlike the present case, histologically did not mimic xanthoma.[11]

This is only the second case of mixed cutaneous *Pneumocystis* and *Cryptococcus* in a patient with AIDS[11] and the first to emphasize the unusual xanthoma-like lesions that may arise from systemic dissemination of these organisms. It is advised to carry out adequate special staining to identify potential mixed infections in a xanthoma-like lesion in an immunocompromised patient.

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