Medical Mycology Case Reports 3 (2014) 14–16

Contents lists available at ScienceDirect

Medical Mycology Case Reports

journal homepage: www.elsevier.com/locate/mmcr

Cryptococcal infection in non-HIV immunosuppressed patients – Three case reports in a nephrology setting ☆

Pedro Ventura Aguiar [a,*], Virgínia Lopes [b], La Salete Martins [a], Josefina Santos [a], Manuela Almeida [a], Sofia Pedroso [a], Leonídio Dias [a], António Castro Henriques [a], Helena Ramos [b], António Cabrita [a]

[a] Nephrology Department – Oporto Hospital Centre, Largo Professor Abel Salazar, 4099-001 Porto, Portugal
[b] Microbiology Department – Oporto Hospital Centre, Largo Professor Abel Salazar, 4099-001 Porto, Portugal

ARTICLE INFO

Article history:
Received 26 September 2013
Received in revised form
12 November 2013
Accepted 14 November 2013

Keywords:
Cryptococcosis
Meningoencephalitis
Transplant
Lupus
Immunosuppression

ABSTRACT

Cryptococcal infection has been increasing among immunosuppressed population. We report three cases of *Cryptococcus neoformans* infection in immunosuppressed patients – two renal transplanted and one with lupus nephritis. Early infection (< 3months) was diagnosed in two – an allograft *Cryptococcus* infection and a central nervous system involvement. The third, a 10-year transplant vintage patient, presented with cryptococcal meningitis. Amphotericin B provided good clinical outcomes. We outline the importance of suspicion for cryptococcal infection in immunosuppressed patients.

1. Introduction

Infections are a major cause of hospitalization and death in immunocompromised patients, representing 20.9% [1] and 3.6% [2] of all death causes among renal transplant recipients and Systemic Lupus Erythematous (SLE) patients, respectively. Though most are due to bacterial infections, invasive fungal infection prevalence is increasing in these populations [1].

Cryptococcus neoformans is a yeast present in soil contaminated with pigeon and/or chicken droppings. The mainstay of host defense against this pathogen is innate and T-cell-mediated immunity [3], likely explaining its high incidence among immunosuppressed patients.

Approximately 20–60% of cryptococcosis in HIV-negative patients occurs in solid organ transplant recipients [4,5]. Kidney transplant recipients infection most often occurs after the first year, while in SLE patients it occurs at a median of 33months post-diagnosis. Mortality rates range from 73% [6] to 46.6% [7], respectively.

2. Case

2.1. Case 1

A 34 year-old male with chronic renal failure due to renal malformation, was admitted due to persistent headaches. Patient was diagnosed with bilateral renal atrophy at the age of 3. He was on hemodialysis since 1998 (day – 4783) and submitted to deceased-donor renal transplant in April 2001 (day – 3780). Immunosuppressive therapy consisted of cyclosporine A (CsA) and mycophenolate mofetil (MMF), with posterior conversion to sirolimus. High dose methylprednisolone (MP), plasmapheresis, monthly high-dose immunoglobulin infusions, and rituximab were administered due to acute humoral rejection in April 2010 (day – 548).

On November 2011 (day – 0) he was presented to the emergency department with a 5 day frontal headache, nausea and vomiting, photo- and phonophobia. No other symptoms were reported, namely fever. Physical examination was unremarkable. Hematologic and biochemical analysis demonstrated normal white blood cells count, no anemia nor thrombocytopenia, and a normal C-Reactive Protein (C-RP) level. Urinary sediment was bland. Chest X-ray, abdominal and graft ultrasounds were normal. Thoracic CT revealed condensation on the upper lobe of the left lung. Cerebral MRI presented asymmetric cortical

* Corresponding author. Tel.: +351 222077500; fax: +351 223325541.
E-mail address: pedro.ventura.aguiar@gmail.com (P. Ventura Aguiar).

2211-7539/$ - see front matter © 2013 International Society for Human and Animal Mycology. International Society for Human and Animal Mycology. Published by Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.mmcr.2013.11.003

lesions, with hyperintensity on T2-weighted and hipointensity on T1-weighted images.

Cerebral spinal fluid (CSF) analysis revealed 138 white blood cells (137 lymphocytes), glucose 0.69 g/L, and proteins 0.69 g/L. CSF was *India-ink and culture positive* for *C. neoformans*, and polymerase chain reaction (PCR)-positive for cytomegalovirus (CMV).

A meningoencephalitis to *C. neoformans* concomitant with CMV infection was assumed. On day 2 treatment with liposomal amphotericin B associated to fluconazole was initiated, along with valganciclovir and anti-CMV immunoglobulins. Lumbar puncture repeated on day 15 revealed a CSF negative for both agents. Transition to fluconazole (400 mg per day, oral) for 12 months and valganciclovir (450 mg every other day) for 6 months was performed. Sustained clinical response was achieved.

2.2. Case 2

A 56 year-old caucasian male was admitted due to fever of unknown origin. He had end stage renal disease due to a rapidly progressive IgA nephropathy treated with cyclophosphamide for 6 months (cumulative dose 21.6 g) plus prednisone. Was on hemodialysis since April 2005 (day – 2355). He also had a prior history of Polycythemia Vera diagnosed in 2001 (day – 3566), arterial hypertension, rickettsiosis diagnosed and treated on May 2010 (day – 441), and pulmonary tuberculosis infection, with clinical response to treatment.

Patient was submitted to deceased-donor renal transplant on 1st of September 2011 (day – 0). Induction immunosuppression was performed with basiliximab, tacrolimus, MMF, and prednisolone. Post-surgery was complicated with infected retroperitoneal hematoma (treated with meropenem and vancomycin), surgical wound infection by *Pseudomonas aeruginosa* (treated with ciprofloxacin plus ceftazidime), and *Candida albicans* oropharyngitis (resolved with fluconazole).

Allograft presented delayed graft function and later, acute vascular rejection (Banff IIA) for which ten doses of Anti-thymocyte globulin (ATG) were administered (day – 35).

Due to persistent fever and sepsis with negative blood cultures, and failure to recover renal function, patient was submitted to transplantectomy 3 months later (day – 88). *Cultured fluid collected from the abdominal drain placed in the area of the removed allograft was positive for C. neoformans*. Allograft histology also revealed the presence cryptococcus. Normal opening pressure was found on lumbar puncture, with negative India-ink CSF.

On day 89 patient was started on liposomal amphotericin B (1.0 mg/kg per day IV) for four weeks, with complete resolution of symptoms. He progressively recovered and was discharged under fluconazole (200 mg per day, oral).

2.3. Case 3

An 18 year-old male with unremarkable history until July 2011 (day – 173), was diagnosed with SLE due to anemia, thrombocytopenia, malar rash, and positive anti-nuclear (ANA) and double-strand DNA antibodies (dsDNA). He was prescribed oral prednisone, but failed to comply with. On December 2011 (day – 67) presented to the emergency department with nephritic syndrome. Kidney biopsy revealed a class IV lupus nephritis. He was prescribed high dose methylprednisolone (total 1.5 g) followed by oral prednisone (1 mg/kg), along with twice monthly administrations of intravenous (IV) 500 mg of cyclophosphamide (total 3 doses). Patient was discharged on the 08th of January 2012 (day – 42).

Six weeks later patient presented to the emergency department (day – 0) due to fever (39 °C) and diarrhea. No other symptoms were reported. Physical examination was unremarkable. Analysis revealed normal leukocyte count, C-RP 70 mg/dL. Chest X-ray revealed no suspicious images. Empirical prescription with ciprofloxacin to an infectious diarrhea was performed and was admitted to the ward. Pneumonia was diagnosed on day 2 due to hypoxemic respiratory failure and a lower right lung condensation on the X-ray. Antibiotherapy spectrum was broadened to meropenem.

C. neoformans was isolated on blood cultures retrieved at admission (day – 6). Lumbar puncture presented an opening pressure of 26 cm/H_2O. Though clear and colorless, CSF fluid was India-ink and *culture positive for C. neoformans*. Immunosuppression was reduced and iposomal amphotericin B (1 mg/kg per day, IV) plus flucytosine (100 mg/Kg per day, IV) was prescribed. CSF fluid collected on day 28 was sterile and India-ink negative. Patient presented good clinical outcome and was discharged on day 41 prescribed with fluconazole (200 mg per day, oral) for 12 months.

3. Discussion

Fungal infections are a rare but frequently fatal disease in immunosuppressed patients. Herein we report three cases of different clinical settings and presentations of invasive cryptococcal infections in non-HIV immunocompromised patients.

C. neoformans is an encapsulated yeast, that has been isolated in pigeons' and chickens' droppings. In humans it can colonize the upper airway system. Nevertheless, no animal-to-human nor person-to-person respiratory transmission has been documented.

Pappas PG et al., from the Transplant-Associated Infection Surveillance Network (TRANSNET), reported a cryptococcal infection incidence of 3.7% in renal transplant recipients, with a 12 month death rate of 73% [7]. Among 8672 kidney recipients, the median time to diagnosis was 575 days, with the lower quartile just over the 6 month period, and with only 22 patients presenting after the 3rd year post-transplant.

The first case reported here presents a patient with 10 year transplant vintage, whose immunosuppression was previously converted from cyclosporine to sirolimus.

Both calcineurin inhibitors (cyclosporine and tacrolimus), the mainstay of immunosuppression in solid organ transplant, have been shown to suppress the growth of the *C. neoformans in vitro* [8]. It has been proposed that cyclosporine's lower concentration in CSF, when compared to tacrolimus, explains the higher incidence of central nervous system (CNS) cryptococcal infection in patients under CsA [8].

Though there was no time-relation to the development of symptoms, two events might have positively influenced the risk to fungal infection. First the cyclosporine-to-sirolimus conversion, and additionally a probable state of over-immunosuppression due to the previously administered methylprednisolone and rituximab.

Primary infection is the main form of cryptococcus transmission, but donor-derived and unrecognized pre-transplant latent disease have also been described [9,10]. A donor-derived transmission is suggested by a more localized form of disease found in these patients, often restricted to the transplanted organ or drainage fluid [11].

The second case reports a cryptococcal infection less than 3 months post-transplant, with cryptococcus isolated solely on the removed allograft and post-transplantectomy drainage fluid.

Immune response to cryptococcal infection is mainly mediated through innate (macrophages and natural killer cells) and adaptive immune system [3]. It has been proposed that the decreased production of pro-inflammatory cytokines, and consequent reduction on the development of T helper cells (Th-1), increases the risk for early post-transplant cryptococcal infection [9].

To the author's comprehension, there was likely a conjunction of factors that lead to infection in the second patient. A latent undiagnosed cryptococcal infection in the donor may have been delivered to an uremic patient, with innate immune depression due to corticosteroid therapy, and a restrained T-cell response due to cyclosporine and MMF therapy.

Autoimmune diseases, like transplant recipients, require high doses of immunosuppression to control disease symptoms and reduce organ damage. KDIGO [12] and ACR/EULAR [13] guidelines recommend high doses of corticosteroids and either intravenous cyclophosphamide or oral MMF for the treatment of lupus nephritis classes III and IV.

Even though, only a few hundred cases of fungal invasive infections in SLE patients can be found in literature. In the largest report ($n=15$) from Chen et al. [7], 11 were infected with cryptococcus. The median time from SLE diagnosis to disease was 33months, and mortality rate 46.6% [7].

The early presentation of infection post-immunosuppression is more suggestive of a reactivation of latent infection. Even though the patient with lupus reported here lived in a suburban area, with low social-economic and hygiene status, he denied contact with pigeons.

In the presented cases culture was performed using yeast VITEK2® card, standard culture medium currently at our center. This medium is not able to distinguish between *C. neoformans* and *Cryptococcus gattii* [14]. The later frequently presents as outbreaks, but has mainly been described in North America. Moreover, it affects equally immunocompromised and immunocompetent patients. Even though it has been previously described in Portugal a single case of *C. gattii* infection [15], to the authors' comprehension the clinical presentation and response to therapy makes it likely that the presented cases correspond to *C. neoformans* infection.

Guidelines for cryptococcus treatment recommend a combined therapy of flucytosine plus amphotericin [16]. Though the combination was only administered to one patient, a favorable outcome was achieved in the 3 cases.

A mainstay of the treatment relies on the management of immunosuppression, which must be individualized to each patient. Caution must be taken when reducing immunosuppression, since a rapid reduction can result in the development of organ rejection and/or immune reconstitution inflammatory syndrome (IRIS) – an inflammatory tissue response due to improvement in cellular immunity, presenting as lymphadenitis, cellulites, aseptic meningitis, pulmonary nodes, or allograft loss in kidney transplant recipients. The prevalence of IRIS in solid organ transplant is about 4.8% [17].

In conclusion, the three cases presented highlight the vital importance for early recognition and microbiological diagnosis of cryptococcal infection among non-HIV immunocompromised patients. Clinicians' awareness in transplanted and auto-immune disease patients is recommended. A good communication and collaboration between the laboratory and the clinic is essential to achieve the best results.

Conflict of interest

The authors decline any conflict of interest.

Acknowledgments

Nothing to declare.

References

[1] ⟨http://www.usrds.org/default.aspx⟩.
[2] Bernatsky S, Boivin JF, Joseph L, Manzi S, Ginzler E, Gladman DD, et al. Mortality in systemic lupus erythematosus. Arthritis Rheum 2006;54(8):2550.
[3] Gupta RK, Khan ZU, Nampoory MR, Mikhail MM, Johny KV. Cutaneous cryptococcosis in a diabetic renal transplant recipient. J Med Microbiol 2004;53:445–9.
[4] Vilchez RA, Fung J, Kusne S. Cryptococcosis in organ transplant recipients: an overview. Am J Transplant 2002;2:575–80.
[5] Husain S, Wagner MM, Singh N. Cryptococcus neoformans infection in organ transplant recipient: variables influencing clinical characteristics and outcome. Emerg Infect Dis 2001;7:375–81.
[6] Pappas PG, Alexander BD, Andes DR, Hadley S, Kauffman CA, Freifeld A, et al. Invasive fungal infections among organ transplant recipients: results of the Transplant-Associated Infection Surveillance Network (TRANSNET). Clin Infect Dis 2010;50(8):1101–11.
[7] Chen HS, Tsai WP, Leu HS, Ho HH, Liou LB. Invasive fungal infection in systemic lupus erythematosus: an analysis of 15 cases and a literature review. Rheumatology 2007;46(3):539–44.
[8] Odom A, Del Poeta M, Perfect J, Heitman J. The immunosuppressant FK506 and its nonimmunosuppressive analog L-685,818 are toxic to Cryptococcus neoformans by inhibition of a common target protein. Antimicrob Agents Chemother 1997;41:156–61.
[9] Castro-Ferreira I, Carvalho C, Coentrão L, Gaião S, Bustorff M, Santos J, et al. Early-onset of disseminated cryptoccal infection in two renal transplant recipients. Clin Nephrol 2011;76:542–6.
[10] Singh N, Lortholary O, Alexander BD, Gupta KL, John GT, Pursell K, et al. Allograft loss in renal transplant recipients with cryptococcus neoformans associated immune reconstitution syndrome. Transplantation 2005;80:1131–3.
[11] Sun HY, Alexander BD, Lortholary O, Dromer F, Forrest G, Lyon GM, et al. Unrecognized pretransplant and donnor-derived cryptococcal disease in organ transplant recipients. Clin Infect Dis 2010;51:1062–9.
[12] KDIGO clinical practice guideline for glomerulonephritis. Kid Int Supp. vol. 2; 2012.
[13] Hahn BH, McMahon MA, Wilkinson A, Wallace WD, Daikh DI, Fitzgerald JD, et al. American College of Rheumatology guidelines for screening, treatment, and management of lupus nephritis. Arthritis Care Res 2012;64:797–808.
[14] Harris JR, Lockhart SR, Sondermeyer G, Vugia DJ, Crist MB, D'Angelo MT, et al. *Cryptococcus gattii* infections in multiple states outside the US pacific northwest. Emerg Infect Dis 2013;19(10):1621–7.
[15] Maduro AP, Mansinho K, Teles F, Silva I, Meyer W, Martins ML, et al. Insights on the genotype distribution among *Cryptococcus neoformans* and *C. gattii* Portuguese clinical isolates. Curr Microbiol.; 2013 Sep 28. [Epub ahead of print].
[16] Perfect JR, Dismukes WE, Dromer F, Goldman DL, Graybill JR, Hamill RJ, et al. Clinical practice guidelines for the management of cryptococcal disease: 2010 update by the infectious diseases society of America. Clin Infect Dis 2010;50:291–322.
[17] Sing N, Lortholary O, Alexander BD, Gupta KL, John GT, Pursell K, et al. An immune reconstitution syndrome-like illness associated with *Cryptococcus neoformans* infection in organ transplant recipients. Clin Infect Dis 2005;40:1756–61.