Cutaneous *Purpureocillium lilacinum* and *Fusarium* coinfection in a heart transplant recipient

Leonard Farrugia [a], Veronica Baston [b], Laura Burfield [c], Lucy Melly [d], Andrew M. Borman [e,f], Abhijit M. Bal [a,*]

[a] *Department of Microbiology, Queen Elizabeth University Hospital, Glasgow, UK*
[b] *Department of Cardiology, Golden Jubilee National Hospital, Clydebank, UK*
[c] *Department of Dermatology, Inverclyde Royal Hospital, Greenock, UK*
[d] *Department of Pathology, Queen Elizabeth University Hospital, Glasgow, UK*
[e] *UKHSA Mycology Reference Laboratory, Southmead Hospital, Bristol, UK*
[f] *Medical Research Council Centre for Medical Mycology, University of Exeter, UK*

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ABSTRACT

Purpureocillium lilacinum and *Fusarium* species are increasingly recognized as significant opportunistic fungal pathogens. We report a rare case of co-infection in a 63-year old heart transplant recipient presenting with nodular skin lesions, treated successfully with voriconazole. We highlight the importance of being vigilant about co-infection with moulds as it impacts on the selection of appropriate antifungal agents. 2012 Elsevier Ltd. All rights reserved.

1. Introduction

Invasive fungal infections (IFIs) are well-recognized cause of morbidity and mortality in solid organ transplant recipients. Heart transplant recipients constitute an important subgroup in this cohort with an estimated 4 % risk of IFI [1]. Overall mortality at 3 months has been broadly estimated to be in the 20–40 % range, but is likely to be poor in heart transplant recipients [2]. While *Candida* and *Aspergillus* remain the most common causes of IFI in this cohort [3], infections with rare moulds are increasingly reported, with a global guideline recently published to help inform treatment options [4].

Both *Purpureocillium lilacinum* and *Fusarium* are saprobic filamentous fungi with ubiquitous environmental distribution, and commonly found in soil. *Fusarium* is the most clinically prevalent rare mould genus causing disseminated infection in immunocompromised individuals but infections with *Purpureocillium* spp are also of interest, with over 250 cases reported from 28 countries [4].

While cases of mixed infection with *Purpureocillium* spp and other fungi have been reported [5], we report a rare case of coinfection of *Purpureocillium* and *Fusarium* spp that was treated curatively with a prolonged course of voriconazole.

2. Case

A 63-year-old male cardiac transplant patient presented to the dermatology clinic (day 0) with multiple erythematous nodular skin lesions which developed over a period of 4 weeks (Fig. 1A–C). Initially assumed to be abscesses, these did not respond to incision & drainage or oral flucloxacillin. The patient was systemically well and apyrexial.

His past medical history included orthotopic cardiac transplant nine months previously for hypertrophic cardiomyopathy. He had subsequently been treated as presumed transplant rejection with methylprednisolone 1g daily for 3 days, followed by long-term prednisolone. The past history was also significant for mild renal impairment with a baseline eGFR of 40mL/min, diverticular disease with previous colovesical fistula repair, as well as recurrent urinary tract infections. His drug history included tacrolimus, mycophenolate mofetil, long-term prednisolone and a number of other cardiac medications.

Initial differential diagnosis included opportunistic infection as well as lymphoma, and a decision was therefore made to biopsy these lesions

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* Corresponding author.
  E-mail address: abhijit.bal3@ggc.scot.nhs.uk (A.M. Bal).

for histopathology and culture. Blood cultures were not taken as the patient had no signs of systemic sepsis. Following 5 days of incubation, *Purpureocillium lilacinum* was isolated from a skin swab of the buttock lesion taken on day +14 (Fig. 2), and from skin biopsy on day +21. The mould was identified on the basis of its morphological appearance. Both cultures were submitted to the United Kingdom Health Security Agency National Mycology Reference Laboratory (MRL). At the MRL, *P. lilacinum* and a *Fusarium* species were identified in the skin swab while the accession culture from the skin biopsy detected *P. lilacinum*, *Fusarium* species, as well as *Aspergillus flavus* by macroscopic and microscopic morphological examination with lactofuchsin from cultures grown on Sabouraud's Dextrose agar supplemented with chloramphenicol (SABC, Oxoid). For the *P. lilacinum* isolate, species-level identification was confirmed by matrix-assisted laser desorption time of flight mass spectrometry (MALDI-ToF MS) using established methods on the Bruker Biotyper Sirius, using Compass software (V4.1) with filamentous fungus database (V3) supplemented with an in-house mass spectral profile database developed and curated by the MRL (Mean LogScore [MLS] of the *P. lilacinum* = 2.30). The identification of the *Fusarium* was phenotypic and accurate to genus level, as inability to purify it from the more rapidly-growing *P. lilacinum* prevented species-level identification via MALDI-ToF MS. Histopathological analysis revealed necrotising granulomas (Fig. 3) with fungal spores and hyphae (Fig. 4) diagnostic of deep fungal infection.

On day +29, the patient was commenced on oral voriconazole 400 mg twice daily for two doses followed by 200 mg twice daily and underwent CT Thorax, which was normal. Mycophenolate was stopped and therapeutic drug monitoring (TDM) was carried out for both voriconazole and tacrolimus. The dose of tacrolimus was reduced in view of its predictable drug-drug interaction with voriconazole, and the steroid dose was tapered down to 5 mg daily over several months. Antifungal susceptibility testing was performed on the *P. lilacinum* isolate using CLSI broth microdilution methodology [6], with minimum inhibitory concentrations (MICs) interpreted using clinical breakpoints for *Aspergillus fumigatus* in the absence of specific breakpoints for *P. lilacinum*. Testing confirmed likely susceptibility to voriconazole but raised MICs to amphotericin B and itraconazole (voriconazole = 0.5mg/L, itraconazole = 2mg/L, anidulafungin = 0.03mg/L, amphotericin B = >16mg/L). No MICs were obtained for the *Fusarium* isolate as repeated attempts to purify this organism were unsuccessful. Drug monitoring of voriconazole revealed therapeutic drug levels, with trough serum voriconazole levels of 3.85, 3.65, 3,50, 1.40 and 5.22 mg/l at days 28, 35, 83, 100 and 160, respectively.

In view of the unusual finding of co-infection with *Fusarium* in a patient who was systemically well, a repeat biopsy was performed. The

Fig. 2. Floccose,lilac-brown colonies of *P.lilacinum* on sabouraud's Dextrose agar. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

repeat histology showed filamentous fungal elements on direct microscopic examination and both *P. lilacinum* (MALDI-ToF MLS = 2.44) and a *Fusarium* species were again isolated on prolonged incubation but yet again, the *Fusarium* could not be obtained in pure culture to enable the laboratory to undertake species identification and susceptibility testing. Repeated attempts to purify the isolates for susceptibility testing failed. *Aspergillus flavus* was not isolated on the repeat biopsy and was deemed to most likely have been a slope contaminant, growing over the top of the other two moulds.

The patient's skin nodules resolved within the first two months after starting voriconazole. An ophthalmology review excluded oculomycosis. He completed 6 months of treatment with voriconazole but developed side-effects of alopecia, phototoxicity and renal dysfunction, due to which voriconazole was discontinued with cautious monitoring. The patient currently remains in good health and under regular review, with no relapse noted several months without antifungal therapy.
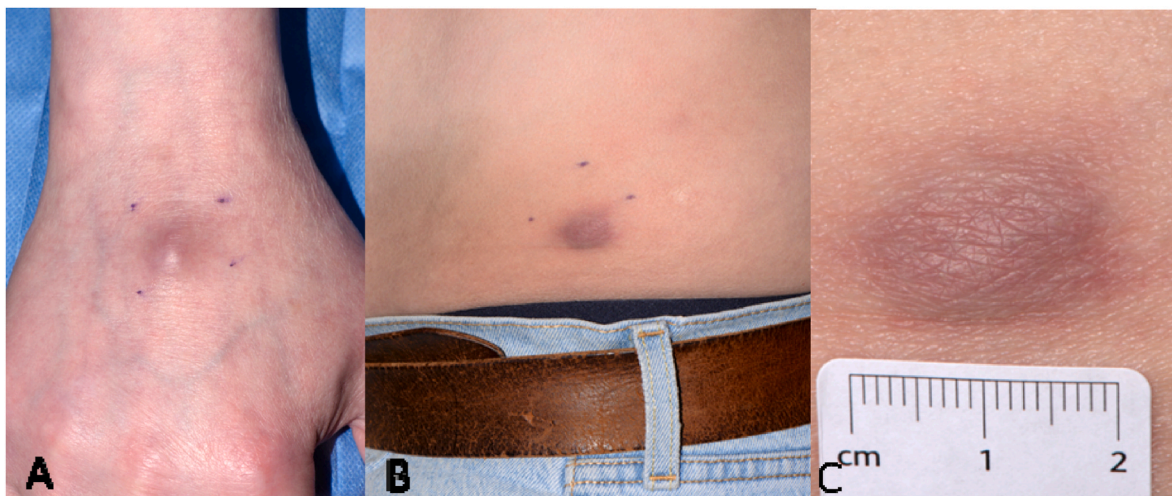

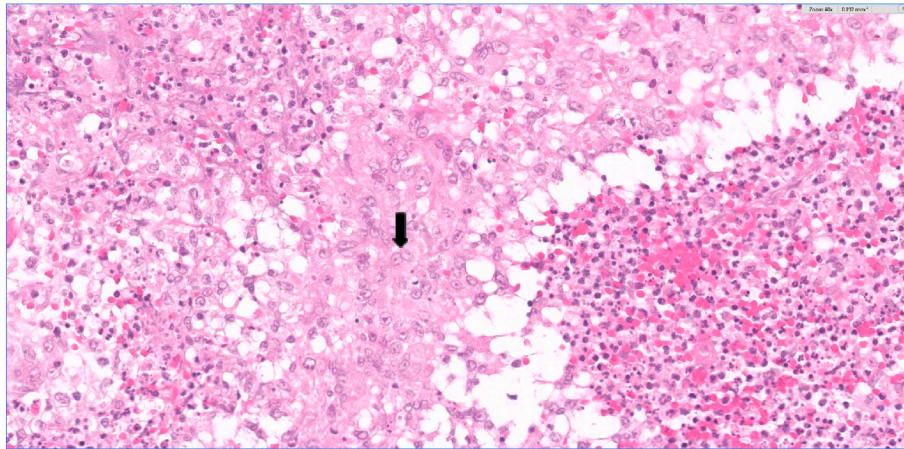**Fig. 1.** Nodular skin lesions over right hand (A) & left hip (B&C).

Fig. 3. Necrotising granulomas within the subcutis. (Hematoxylin & eosin stain,x40).
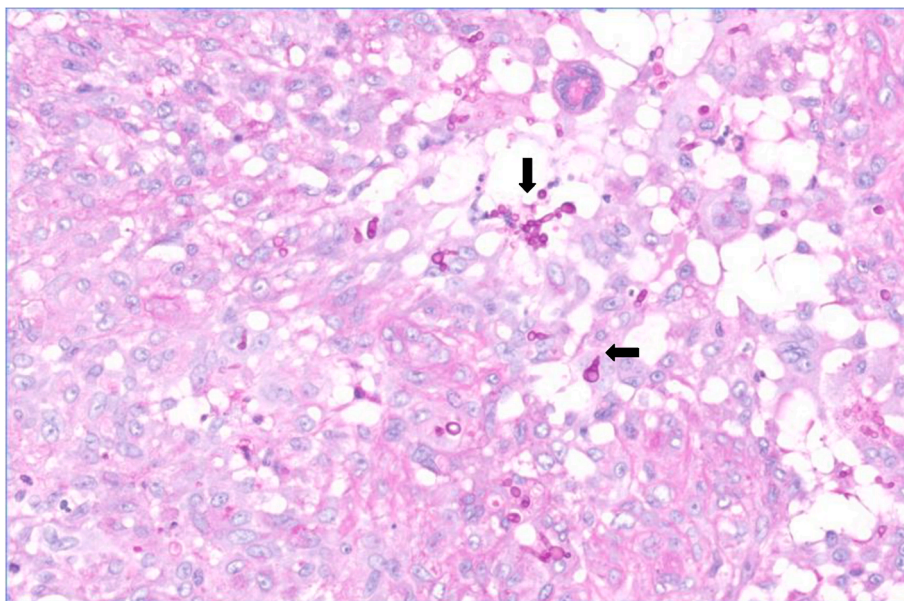

Fig. 4. Fungal spores within the granulomatous inflammation. (Periodic acid-Schiff,x40).

3. Discussion

Purpureocillium and *Fusarium* share a number of interesting similarities as well as important differences. They are both hyaline hyphomycetes with widespread environmental distribution. Both are important causes of fungal keratitis with a tropism for ocular structures, but can also cause disseminated bloodstream infections in immunocompromised individuals. Route of entry is through direct inoculation of the skin or inhalation of airborne microconidia. Despite being isolated from skin lesions and having normal lungs on CT imaging, we postulate inhalation with subsequent immune evasion and multifocal dissemination to skin and subcutaneous structures as a possible route of acquisition in our case. However, we are not aware of any history of exposure to moulds so direct inoculation cannot be ruled out.

Skin lesions are a distinctive feature of both infections, and a hallmark feature of systemic fusariosis. Lesions are generally accompanied by fever and systemic inflammatory response, which were notably absent in our case. Data from an international web-based registry for rare and emerging IFI show that skin is the most prevalent site of isolation of *Purpureocillium,* with 36.6 % of isolates originating from dermal samples and approximately 10 % of patients showing nodular lesions. There was

a male preponderance (61.4 %) and the median age of diagnosis was 53 years. The investigators list cases of mixed infection with *Alternaria* and *Fusarium*, both also isolated from skin samples, with all other coinfections being pulmonary. In the registry data, a patient with mixed infection with *Fusarium*, to our knowledge the only other case reported in the literature, passed away despite antifungal therapy, highlighting the potentially life-threatening nature of such infections. While a majority of patients had an immunosuppressive condition, only 3 % of patients in the registry had undergone cardiac transplant [5].

Prompt diagnosis of IFI with rare moulds continues to be a main barrier to improving outcomes. Both *Fusarium* and *Purpureocillium* may exhibit a phenomenon of adventitious sporulation, enabling the separation of conidia from invading hyphae and entrance into the bloodstream [7,8]. This facilitates culture from bloodstream, with rates of between 40 and 70 % positivity reported for *Fusarium*, [9,10] making blood cultures the primary diagnostic test for systemic fusariosis. In contrast, only 17.8 % of *Purpureocillium* isolates in the aforementioned study [5] were from blood cultures. The detection of adventitious sporulation may also aid presumptive identification during histological examination [8]. The inability to isolate *Fusarium* in pure culture despite repeat attempts prevented both species-specific identification as well as

formal susceptibility testing, but this was not felt to significantly impact clinical management given voriconazole is the treatment of choice for both infections.

Amphotericin B may be a useful adjunct in the initial treatment of fusariosis but its use as monotherapy is discouraged if alternatives are available [4]. In contrast, amphotericin B is not active against *Purpureocillium*, with elevated MICs *in vitro* as well as significantly higher mortality observed with its use *in vivo* [5]. In our case, the patient had already been commenced on oral voriconazole in the outpatient setting prior to identification of *Fusarium*, and voriconazole was therefore continued as monotherapy with TDM. Withholding mycophenolate to decrease net state of immunosuppression as well as preemptively decreasing the dose of tacrolimus were both key interventions to optimize host factors and limit risk of toxicity.

In summary, this case highlights the complexity of mixed invasive fungal infections in transplant recipients. Heightened clinical awareness, together with better diagnostics and drugs for the detection and treatment of rare moulds would be of great value in this neglected area of transplant infectious disease.

Conflict of interest

The authors have no conflicts of interest to disclose.

CRediT authorship contribution statement

Leonard Farrugia: Writing – review & editing, Writing – original draft, Visualization, Supervision, Conceptualization. **Veronica Baston:** Writing – review & editing. **Laura Burfield:** Writing – review & editing. **Lucy Melly:** Writing – review & editing, Visualization, Resources. **Andrew M. Borman:** Writing – review & editing, Investigation. **Abhijit M. Bal:** Writing – review & editing, Supervision.

References

[1] P.G. Pappas, B.D. Alexander, D.R. Andes, et al., Invasive fungal infections among organ transplant recipients: results of the Transplant-Associated Infection Surveillance Network (TRANSNET), Clin. Infect. Dis. 50 (8) (2010) 1101–1111.

[2] S. Shoham, K.A. Marr, Invasive fungal infections in solid organ transplant recipients, Future Microbiol. 7 (5) (2012 May) 639–655.

[3] A.S. Rabin, M.M. Givertz, G.S. Couper, M.M. Shea, D. Peixoto, D.S. Yokoe, et al., Risk factors for invasive fungal disease in heart transplant recipients, J. Heart Lung Transplant. 34 (2) (2015 Feb) 227–232.

[4] M. Hoenigl, J. Salmanton-García, T.J. Walsh, M. Nucci, C.F. Neoh, J.D. Jenks, et al., Global guideline for the diagnosis and management of rare mould infections: an initiative of the European confederation of medical Mycology in cooperation with the international society for human and animal Mycology and the American society for microbiology, Lancet Infect. Dis. (2021), https://doi.org/10.1016/S1473-3099 (20)30784-2.

[5] R. Sprute, J. Salmanton-García, E. Sal, X. Malaj, Z. Ráčil, C. Ruiz de Alegría Puig, et al., Invasive infections with Purpureocillium lilacinum: clinical characteristics and outcome of 101 cases from FungiScope® and the literature, J. Antimicrob. Chemother. 76 (6) (2021 May 12) 1593–1603.

[6] Clinical and Laboratory Standards Institute. Reference Method for Broth Dilution Antifungal Susceptibility Testing of Filamentous Fungi. Approved Standard CLSI Document M38-A2. third ed.

[7] W.A. Schell, New aspects of emerging fungal pathogens. A multifaceted challenge, Clin. Lab. Med. 15 (1995) 365–387.

[8] K. Liu, D.N. Howell, J.R. Perfect, W.A. Schell, Morphologic criteria for the preliminary identification of Fusarium, Paecilomyces, and Acremonium species by histopathology, Am. J. Clin. Pathol. 109 (1) (1998 Jan) 45–54.

[9] M. Nucci, K.A. Marr, M.J. Vehreschild, et al., Improvement in the outcome of invasive fusariosis in the last decade, Clin. Microbiol. Infect. 20 (2014) 580–585.

[10] M. Nucci, F. Carlesse, P. Cappellano, et al., Earlier diagnosis of invasive fusariosis with Aspergillus serum galactomannan testing, PLoS One 9 (2014) e87784.