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General review

COVID-19-associated mixed mold infection: A case report of aspergillosis and mucormycosis and a literature review

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ABSTRACT

COVID-19-associated mold infections have been increasingly reported, and the main entity is COVID-19-associated aspergillosis (CAPA). Similarly, COVID-19-associated mucormycosis has been reported in hematology, and its prevalence is high and has been increasing in the diabetic population in India during the third COVID-19 pandemic wave. Simultaneous infection with Mucorales and *Aspergillus* is rare and even rarer during COVID-19. Here, we report the case of a previously immunocompetent patient with severe SARS-CoV-2 infection complicated with probable CAPA and mucormycosis co-infection. Specific diagnostic tools for mucormycosis are lacking, and this case highlights the advantages of analyzing blood and respiratory samples using the quantitative polymerase chain reaction to detect these fungi. We further reviewed the literature on mixed *Aspergillus*/Mucorales invasive fungal diseases to provide an overview of patients presenting with both fungi and to identify characteristics of this rare infection.

Introduction

There have been few studies on mixed mold diseases, and they are rarely reported in immunocompromised patients with neutropenia, malignant hemopathy [1,2], solid organ transplantation [3], or poorly controlled diabetes mellitus [4,5]. Viral infections, especially severe influenza and COVID-19, which cause acute respiratory distress syndrome (ARDS), increase the susceptibility to mold infection in previously immunocompetent patients [6,7]. COVID-19-associated pulmonary aspergillosis (CAPA) and COVID-19-associated mucormycosis (CAM) are associated with a higher mortality rate in this patient population [8,9]. CAPA and CAM prevalence rates vary widely between studies, which may be explained by different awareness and diagnostic strategies [8]. Our center screened every mechanically ventilated COVID-19 patient who clinically worsened despite adequate standard of care for CAPA. This case is a patient with *Rhizopus*/*Aspergillus* coinfection in a previously immunocompetent COVID-19 patient and a discussion about mixed mold infections.

CASE

A 74-year-old retired man with a history of hypertension that was being treated with calcium channel blockers presented in the emergency ward on October 21, 2020 with a cough and a fever that lasted 4 days (Figure 1). He had evocative chest computed tomography (CT) scan findings, and the COVID-19 diagnosis was confirmed using real-time reverse-transcriptase polymerase chain reaction (PCR) on a nasopharyngeal swab (RealStar® SARS-CoV-2 Kit, Altona Diagnostics). The patient had a history of gastric lymphoma that was treated surgically in 1975, and he has subsequently been in remission.

Upon admission in the pneumology ward, the patient received ceftriaxone, azithromycin, and preventive anticoagulation by enoxaparin sodium (0.8 mL × two per 24 h subcutaneously). On October 23rd, 2020, the patient received dexamethasone 6 mg/day because of a persistent SpO_2 of 85% on room air and a fever (38.6 °C). After multiple desaturation episodes despite increased oxygen support (up to 6 L/min), he received a bolus of 120 mg methylprednisolone and was

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Table 1
Summary of mycological results.

	BAL Day 19	Serum Day 19	BAL Day 24	Plasma Day 24
DE	Negative	ND	*Aspergillus*- and Mucorales-type mycelium	ND
Culture	*Aspergillus welwischiae* *Candida parapsilosis*	ND	*Rhizopus delemar* *Aspergillus welwischiae*	ND
GM Ag	GM Ag > 4.63	GM Ag > 4.63	GM Ag > 5.12	ND
BDG	ND	30.7 pg/mL	ND	ND
PCR *Mucor / Rhizopus*	negative	positive* (Cq 30.87)	positive (Cq 25.65)	positive (Cq 24.62)
PCR *A. fumigatus*	negative	negative	negative	negative

BAL: Bronchoalveolar lavage ; BDG: Beta D glucan; Cq: Quantification cycle ; DE: Direct examination; GM Ag: Galactomannan Antigen; ND : Not done; PCR: Polymerase chain reaction; *PCR added retrospectively.

transferred to the intensive care unit (ICU) on October 26, 2020 (Day 1). The patient presented with polypnea (respiratory rate, 34 breaths/minute) with signs of respiratory distress such as abdominal breathing and SpO_2 91% under 15 L/min of oxygen. His-white blood cell count was 10,000 cells per mm^3 (normal range, 4000 to 10,000) with 90% neutrophils (9040, normal range 1700 to 7000 cells per mm^3) and 3.8% lymphocytes (400, normal range 1500 to 4500 cells per mm^3), and his C-reactive protein level was 194 mg/dL (normal value <5 mg/dL), procalcitonin was 1.49 μg/L (normal value <0.05 μg/L), fibrinogen was 7.77 g/L (normal range 2 to 4 g/L), and D-dimer was 2830 ng/mL (normal value <500 ng/mL). He had metabolic alkalosis with profound hypoxemia at 55 mmHg and lactatemia at 1.4 mmol/L. The simplified acute physiology score II (SAPSII) and Sepsis-related Organ Failure Assessment (SOFA) at 24 h were 32 and 4, respectively. Ceftriaxone and azithromycin were discontinued on Day 2 after negative microbiological results from respiratory samples.

On Day 6, the patient's condition deteriorated due to acute heart failure. Mechanical ventilation was initiated. Dexamethasone was increased to 10 mg twice daily and piperacillin/tazobactam was empirically prescribed. The patient subsequently presented with multiple organ failure.

Bronchoalveolar lavage (BAL) was performed on Day 7 and identified 5×10^3 colony forming units (CFU)/mL of *Pseudomonas aeruginosa* that was resistant to piperacillin/tazobactam, and a chest CT angiography showed a bilateral segmental pulmonary embolism with D-dimer >3000 ng/mL. Ceftazidim and intravenous unfractionated heparin were introduced on Day10.

On Day 15, a second BAL identified ventilation-associated pneumonia with 10^3 CFU/mL *P. aeruginosa*, 10^4 CFU/mL *Stenotrophomonas maltophila*, and herpes simplex virus (HSV)−1 reactivation at 8 log copies/mL. He was administered cefepime, amikacin, and sulfamethoxazole−trimethoprim (800 mg twice daily). Acyclovir was introduced on Day 16 before HSV-1 and cytomegalovirus viremia (4.37 log copies/mL and 2.2 log IU/mL).

The evolution was initially favorable, but a new degradation with hemodynamic and acute renal failure on Day 19 required initiation of renal replacement therapy. A new CT-scan highlighted a right apical excavation with peripheral condensation, which was associated with overall stability in the areas with a ground glass appearance and an increase in bilateral subpleural condensation in the postero-inferior segments (Figure 2). BAL performed on Day 19 was positive for *Aspergillus* section *Nigri* and was associated with a positive BAL galactomannan index >4.63 (Platelia *Aspergillus* enzyme immunoassay, Biorad). The concomitant serum galactomannan index was also >4.63 (Table 1). Voriconazole was introduced on Day 19.

A third BAL was performed on Day 24 due to symptom persistence despite antifungal treatment. BAL results were positive for *Aspergillus* and Mucorales type mycelium upon direct examination, *Aspergillus* section *Nigri* and *Rhizopus* sp. grew in culture (Figure 2), and *Mucor/Rhizopus* qPCR results on BAL and serum samples were positive [9] (Cq value, 25.6 and 24.6, respectively). *Mucor/Rhizopus* qPCR performed retrospectively on the Day 19 sample was positive and showed a 2 log increase of the circulating fungal load in only 5 days under voriconazole therapy (Cq from 30.8 to 24.6). *Aspergillus fumigatus*-specific PCR [10] was negative on all samples. Voriconazole was switched to liposomal amphotericin B on Day 25, but the patient died on Day 26.

We retrospectively studied the two strains that were identified. *Aspergillus* section *Nigri* was identified as *Aspergillus welwitschiae* by sequencing using calmodulin as a target with CL1/CL2a primers [11], and *Rhizopus* sp. was identified as *Rhizopus delemar* by sequencing with an internal transcribed spacer (ITS) as a target. *A. welwischiae* and *R. delemar* underwent antifungal susceptibility testing using the EUCAST method and showed minimal inhibitory concentrations (MICs), respectively, for posaconazole (0.5 mg/L and 0.5 mg/L), itraconazole (2 mg/L and 4 mg/L), voriconazole (1 mg/L and ≥8 mg/L), isavuconazole (2 mg/L and 2 mg/L), amphotericin B (4 mg/L and 0.125 mg/L), micafungin (≤0.008 mg/L and ≥4 mg/L), caspofungin (0.5 mg/L and ≥4 mg/L).

In the ICU that cared for the patient, 153 patients were admitted for severe COVID-19 between March 2020 and February 2021, of whom 84 were mechanically ventilated. Among these latter patients, five (6.0%) developed CAPA and one (1%) developed a mixed mold infection, highlighting the rarity of this triple association. No other mucormycosis cases were reported.

LITERATURE REVIEW

References for this review were identified by searching the PubMed database using a combination of title keywords that referred to mixed mold infection (mucormycosis AND (aspergillosis OR aspergillus), mixed fungal infection, mixed mold infection). Thirty five cases reporting *Aspergillus* and Mucorales were analyzed (Table 2). These cases were described mostly in males (n = 21; 60%) with a median age of 51 (IQR, 34−65) years. These mixed infections were reported in patients with hematological malignancy (n = 12; 34%), diabetes (n = 10; 29%), trauma (n = 3; 9%), solid cancer (n = 1; 3%), solid organ transplantation (n = 1), Castleman disease (n = 1), and while receiving high-dose corticosteroids (n = 4). Most infections involved the lungs (n = 21; 60%) and CT scan showed cavitations (n = 13; 62%). Sinuses, brain, and skin were involved in 11 (31%), 4 (11%), and 2 (6%) cases, respectively. When identified to the *Aspergillus* species level (n = 22; 69%), *A. fumigatus* was isolated in 15 (69%) patients followed by *A. flavus* (n = 5; 23%), and *A. niger* (n = 3; 14%). In two cases, multiple *Aspergillus* species were isolated. Mucorales were rarely identified to the species. The most frequently identified genera were *Mucor* (n = 11; 31%) and *Rhizopus* (n = 8; 23%) followed by *Lichteimia* (n = 4; 11%), *Cunninghamella* (n = 3; 9%) and *Rhizomucor* (n = 2; 9.1%). In fifteen cases, Mucorales order members were only identified upon histopathological biopsy examination. In twelve (34%) cases, aspergillosis was diagnosed before mucormycosis and voriconazole was prescribed, and the patient was subsequently switched to amphotericin B. Thirteen of 35 (37%) patients died. Among these thirteen patients, most had European Organization for Research and Treatment of Cancer (EORTC) host factors (hematological malignancy, n = 5; high dose corticosteroids, n = 3; cancer under chemotherapy,

Table 2
Published cases of mixed mucormycosis and aspergillosis infection.

Reference	Sex	Age	Underlying condition	Localization	Imaging	Aspergillus sp.	Mucorales sp.	Concomittant isolation	Classification*	Treatment	Outcome
Bellanger (2021) [15]	M	55	COVID-19; Neutropenia; Hematological malignancy	Lung	Not contributive	*Aspergillus fumigatus*	*Rhizopus microsporus*	YES	Probable°	AMB	Died
Johnson (2021) [16]	M	79	COVID-19; Diabetes	Lung	Cavitation	*Aspergillus fumigatus*	*Rhizopus oryzae*	Aspergillus first	Probable°	VCZ->AMB	NA
Bretagne (2021) [37]	M	73	COVID-19; Diabetes	Lung	Not contributive	*Aspergillus fumigatus*	*Rhizopus microsporus*	YES	Putative✧	AMB	Alive
Buil (2021) [38]	M	Late 50 s	COVID-19	Lung	Cavities, reversed halo-sign	*Aspergillus fumigatus*	*Lichtheimia ramosa*	Aspergillus first	Probable°	VCZ→AMB+PCZ	Died
Buil (2021) [38]	M	Late 60 s	COVID-19, Hematological malignancy, Diabetes, Obesity	Lung	Progression of pulmonary lesions,dissemination to the kidneys	*Aspergillus fumigatus*	*Rhizopus. microsporus*	Aspergillus first	Proven	VCZ+MCF→ICZ →AMB	Died
Present case	M	74	COVID-19; High dose corticosteroids	Lung	Cavitation	*Aspergillus niger*	*Rhizopus delemar*	Aspergillus first	Probable°	VCZ->AMB	Died
Bergantim (2013) [39]	F	58	Hematological malignancy	Lung	Consolidation; Cavitation	*Aspergillus* sp.	*Mucor* sp.	YES	Proven*	AMB	Alive
Chermetz (2016) [40]	F	17	Cerebral glioma; Chemotherapy	Brain; Sinus	Sinus opacification; Brain lesion	*Aspergillus* flavus	*Rhizomucor* sp.	YES	Proven*	AMB	Died
Davoudi (2014) [2]	F	24	Hematological malignancy; Neutropenia	Lung	Opacities; Cavitation	Uncultured	*Rhizomucor* sp.	YES	Proven*	AMB+VCZ+CAS	Alive
Hu (2021) [41]	F	51	Hematological malignancy	Lung, gastrointestinal	Massive high-density shadow in the right superior lobe, free abdominal gas under the diaphragm, and peritoneal fluid	*Aspergillus flavus*	*Cunninghamella bertholletia*	YES	Proven*	VCZ→ D-AMB →AMB→ Posaconazole	Alive
Johnson (1993) [42]	F	38	Hematological malignancy; HSCT	Skin	NA	*Aspergillus niger*	*Rhizopus oryzae*	YES	Probable*	AMB	Died
Kishel (2008) [43]	F	49	Hematological malignancy; Neutropenia	Sinus	*NA*	*Aspergillus* sp.	*Mucor* sp.	YES	Probable*	AMB+CAS	Died
Lai (2021) [44]	M	70	COVID-19, High dose corticosteroids	Lung	Infiltrations	*Aspergillus terreus*	*Cunninghamella bertholletiae*	Aspergillus first	Probable°	VCZ+ ADF → AMB	Died
Leelawattanachai (2019) [45]	M	44	Trauma	Lung	Cavitation	*Aspergillus fumigatus; Aspergillus flavus*	*Lichteimia corymbifera; Rhizopus microsporus*	YES	Putative✧	AMB->PCZ	Alive
Lin (2019) [46]	M	52	Diabetes	Lung	Cavitation	Uncultured	Uncultured	Aspergillus first	Proven*	VCZ->AMB	Alive
Madan (2021) [47]	F	11	Beta thalassemia	Sinus, eye	maxillary sinusitis,erosion of orbital bone,extension into right orbit	*Aspergillus* sp.	*Mucor* sp.	YES	Proven*	AMB→VCZ	Alive
Mahadevaiah (2013) [5]	F	27	Diabetes	Lung	Consolidation; Cavitation	*Aspergillus fumigatus*	*Mucor* sp.	YES	Proven*	AMB+VCZ	Died
Maiorono (2005) [48]	M	66	Castleman disease	Sinus	NA	Uncultured	Uncultured	YES	Proven*	AMB	Alive
Mantero (2019) [17]	F	55	High dose corticosteroids; Dermatomyositis	Brain	Abcess	*Aspergillus* sp.	*Mucor* sp.	Aspergillus first	Proven*	VCZ->AMB	Died
McLintock (2005) [1]	M	19	Hematological malignancy; Neutropenia	Lung	Cavitation	*Aspergillus fumigatus*	*Lichteimia corymbifera*	YES	Proven*	AMB+ITZ	NA
Moorthy (2021) [49]	M	45	COVID-19	Sinus, eye	NA	Uncultured	Uncultured	NM	Proven*	AMB	Alive
Obradovic-Tomasev (2014) [50]	M	28	Trauma	Skin	NA	*Aspergillus* sp.	Uncultured	Aspergillus first	Proven*	AMB+VCZ	Alive
Point (2017) [51]	M	61	Diabetes	Lung; Sinus	Consolidation; Sinus opacification	*Aspergillus fumigatus*	Uncultured	Aspergillus first	Proven*	VCZ->AMB	Alive
Pouvaret (2019) [52]	F	52	Hematological malignancy; Ibrutinib	Brain; Kidney	Abcess	*Aspergillus fumigatus*	*Lichteimia* sp.	Aspergillus first	Proven*	VCZ->AMB	Alive
Radowsky (2011) [53]	M	22	Trauma	Lung	NA	*Aspergillus niger; Aspergillus terreus; Aspergillus flavus*	*Cunninghamella* sp.	YES	Proven*	AMB	Died
Ravindra (2021) [54]	M	65	Alcoholic	Lung	Ground-glass opacity, consolidation (reverse halo sign), cavitating consolidation	Uncultured	Uncultured	YES	Proven*	AMB	Alive
Ravindra (2021) [54]	M	70	None	Lung	Intra-cavitary mass, crescent of air, thick-walled cavity	*Aspergillus* sp.	*Mucor* sp.	YES	Proven*	AMB	Alive
Safai Nodeh (2019) [55]	F	34	Hematological malignancy; Neutropenia	Lung; Sinus	Sinus opacification; Bone lysis	*Aspergillus* sp.	Uncultured	Aspergillus first	Proven*	VCZ->AMB	Alive
Saltini (2021) [56]	M	72	COVID-19, Hematological malignancy, Diabetes, High dose corticosteroids	Lung, sinus	Pseudo-nodular cavitary, consolidations, inflammatory thickness of the left maxillary sinus	*Aspergillus fumigatus*	*Mucor circinelloides*	Aspergillus first	Probable*	VCZ+ ADF → AMB +CPF	Died
Singh (2021) [57]	F	50	Diabetes	Sinus	NA	*Aspergillus fumigatus*	*Mucor* sp.	YES	Proven*	AMB→ VCZ	Alive

(*continued on next page*)

Table 2 (*Continued*)

Reference	Sex	Age	Underlying condition	Localization	Imaging	Aspergillus sp.	Mucorales sp.	Concomitant isolation	Classification*	Treatment	Outcome
Singh (2021) [57]	F	60	Diabetes	Rhino-ocular	NA	*Aspergillus fumigatus*	*Mucor* sp.	YES	Proven*	AMB→ ITRA	Alive
Singh (2021) [57]	M	35	Diabetes	Sino-nasal	NA	*Aspergillus fumigatus*	*Mucor* sp.	YES	Proven*	AMB→ VCZ	Alive
Weng (2012) [58]	M	10	Hematological malignancy; HSCT	Lung;	Disseminated	Cavitation	*Aspergillus fumigatus*	*Rhizopus* sp.	Mucorales first	Proven*	AMB
Alive											
Zayet (2021) [4]	F	69	Diabetes	Sinus; Eyes; Brain	Sinus opacification; Bone lysis	*Aspergillus flavus*	Uncultured	Mucorales first	Proven*	AMB	Alive
Zhan (2008) [3]	M	41	SOT (kidney and liver)	Liver	NA	*Aspergillus* sp.	*Mucor* sp.	YES	Proven*	NA	Died

ADF: Anidulafungin ; AMB: liposomal amphotericin B; CPF : caspofungin EORTC/MSG: HSCT: hematopoeitic stem cell transplant, ICZ: isavuconazole ; MCF: micafungin; NA: not assessed ; NM : not mentionned; PCZ: posaconazole; SOT: solid organ transplant; VCZ: voriconazole. Classification used was *ECMM/ISHAM for COVID-19 patients [7], *EORTC/MSGERC (European Organization for Research and Treatment of Cancer and the Mycoses Study Group Education and Research Consortium), ⟡Blot et al. [28]for patients in intensive care unit with EORTC/MSGERC host factor and proven infection; ->: switched to.

n = 1; solid organ transplantation, n = 1; diabetes, n = 1; COVID-19 only, n = 1; Trauma, n = 1). Nine mixed infections were reported in COVID-19 patients including our patient, and among these three patients, five had underlying risk factors (hematological malignancy and diabetes) and two received high-dose corticosteroids for COVID-19. Seven patients had lung infections. Overall, no specificities emerged from these cases compared to other patients with mixed *Aspergillus*/Mucorales infection.

Discussion

Here, we report the case of a patient with severe SARS-CoV-2 infection with the probable complications of CAPA and CAM.

One important aspect of our case is the co-infection with both *A. welwischiae* and *R. delemar*. *Aspergillus* and *Rhizopus* are two fungal genera with an angioinvasive ability. This association has been rarely described in the immunocompromised subject with an often fatal outcome in pulmonary and brain localizations (Table 2). More cases are described in case series of mucormycosis with up to 44.4% of *Aspergillus* co-infection [12]. Furthermore, the VITAL trial that assessed the efficacy of isavuconazole for treatment of mucormycosis also reported six cases among 37 patients, suggesting that this co-infection may be underdiagnosed [13,14]. The triple association COVID-19/*Aspergillus*/Mucorales has been reported in severely immunocompromised patient after stem cell transplantation and in patients with diabetes mellitus [15,16]. In one other case, a mixed mold infection was reported in a patient receiving high-dose corticosteroids for dermatomyositis [17].

There have been several publications on the subject of COVID-19 and mold co-infection since the beginning of the first COVID-19 pandemic wave. The main co-infection that was studied was aspergillosis [7,18,19], and these studies highlighted several potential risk factors including intubation and mechanical ventilation, high-dose corticosteroids [20,21], azithromycin for ≥3 days [22], tocilizumab [23,24], and immunological storm including high inflammatory cytokine levels [18]. CAM was reported less frequently especially in the first two COVID-19 pandemic waves, but it appears to be an increasing problem in India [25]. Although mucormycosis and mixed *Aspergillus*-Mucorales infection have been described less frequently, risk factors are expected to be similar [25,26]. Most cases of CAM are reported in patients with underlying conditions who are at a high risk of contracting these invasive infections (i.e. EORTC and the Mycoses Study Group Education and Research Consortium [EORTC/MSGERC] host factors or diabetes mellitus), and is it unclear whether COVID-19 infection is associated with a higher risk in this population or if there is a publication bias toward mucormycosis in patients with concomitant COVID-19 infection. Our patient was immunocompetent before contracting COVID-19. The risk factors for co-infection with mold disease were azithromycin administration for 7 days, high-dose corticosteroids, and the use of mechanical ventilation. This combination of factors probably led to the development of mixed invasive fungal disease.

Similar to any environmental mold that is isolated in respiratory specimens, there is always a discussion about colonization and infection. Because most COVID-19 patients have no classical host factors for invasive mold disease, other classifications besides the EORTC/MSGERC classification [27] were developed and used to identify invasive fungal diseases. These classifications are AspICU [28], an influenza-associated pulmonary aspergillosis classification developed by Verweij et al. [29], and most recently, a classification that was proposed by ECMM/ISHAM [7]. Both aspergillosis and mucormycosis are consistent with putative or probable CAPA or CAM because both BAL direct examination and culture results were positive. Additionally, a CT-scan showed patterns that were compatible with invasive fungal disease. Furthermore, a highly positive galactomannan index in both blood and BAL and a positive Mucorales qPCR result in the blood

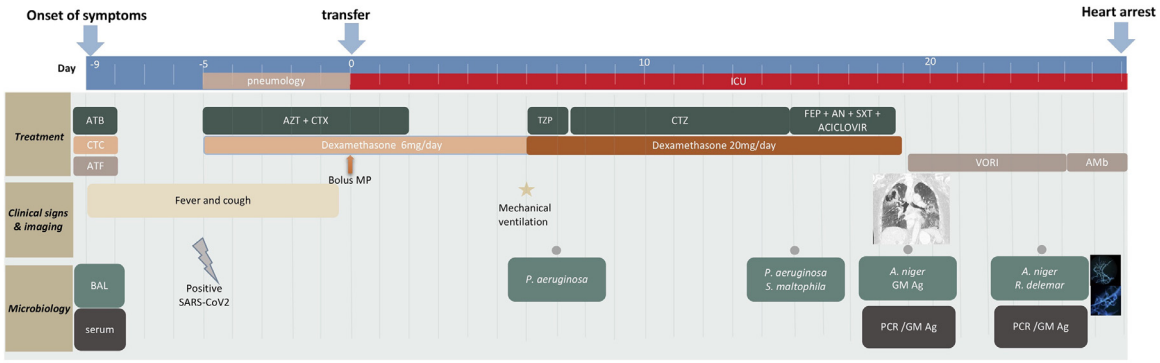

Fig. 1. Time course of the patient with COVID-19-associated pulmonary aspergillosis and COVID-19-associated mucormycosis.
AMb, liposomal amphotericin b; AN, amikacin; ATB, antibiotic; ATF, antifungal; AZT, azithromycin; BAL, bronchoalveolar lavage; CTC, corticosteroids; CTX, cefotaxime; DXM, dexamethasone; FEP, cefepime; GM Ag, galactomannan antigen; ICU, intensive care unit; MP, methylprednisolone; PCR, polymerase chain reaction; SXT, sulfamethoxazole–trimethoprim; VORI, voriconazole.

strongly suggested an invasive disease. Therefore, the patient could have been classified as having a CAPA and CAM infection. However, careful investigation of our patient's corticosteroids doses indicated that he met the EORTC criteria [27] because he received a prednisone equivalent dose >0.3 mg/kg/day for 3 weeks (0.08 mg/kg/day of dexamethasone for 8 Days [0.6 mg/kg/day prednisone equivalent], 0.3 mg/kg/day of dexamethasone for 20 Days [1.9 mg/kg/day prednisone equivalent], and a methylprednisolone bolus of 120 mg [150 mg prednisone equivalent]) [27,30]. Therefore, a careful evaluation of the cumulative dose is required before concluding that the patient has no known EORTC/MSGERC risk factor for invasive mold infection.

Aspergillus section *Nigri* is also worth discussing because it is rarely responsible for infection compared to *Aspergillus fumigatus* due to its physiological characteristics such as its large conidia, which makes it more difficult to reach alveoli, and its optimal germination temperature, which is approximately 30 °C [31]. Invasive aspergillosis due to *A.* section *Nigri* is mostly reported in severely immunocompromised patients [32]. One other case report describes a fatal CAPA infection due to *A.* section *Nigri* with a high galactomannan index at Day 10 after ICU admission [33]. The delay between hospitalization in the ICU and CAPA infection of 19 days was longer than the median time that was published in CAPA cohorts of approximately 6 days [29]. This could be explained because reaching a critical dose of corticosteroids was required for invasive disease to develop in a previously immunocompetent patient.

Molecular biology tools to diagnose invasive fungal infections (IFIs) have only recently been developed. *Aspergillus* qPCR have been included in international guidelines for invasive aspergillosis since 2020 [27], and they are included when making a CAPA diagnosis [7]. However, detection of circulating Mucorales DNA (cmDNA) is not recommended by default to diagnose mucormycosis [34] despite data showing a high sensitivity and its ability to predict a diagnosis and quantify the fungal burden [9]. The important angioinvasive ability of Mucorales makes it possible to detect the fungus in blood samples. Regular screening of at-risk patients such as severely burned patients suggests that cmDNA detection allows an earlier diagnosis of invasive mucormycosis in this population and earlier treatment initiation [35]. Unlike galactomannan detection for the diagnosis of aspergillosis, there are no tools that target the Mucorales antigens, which emphasizes the need to include cmDNA detection in the mucormycosis diagnosis standards. This could be considered to be a screening tool for COVID-19 patients who are clinically worsening despite an appropriate standard of care and who have additional risk factors such as uncontrolled diabetes or high-dose corticosteroids.

Finally, there are no guidelines or standard practice for IFI management in COVID-19 patients, and clinical effectiveness of antifungal administration in these cases has not been demonstrated [7]. The treatment that is being promoted for patients with CAPA is intravenous voriconazole or isavuconazole, the latter of which covers the Mucorales species and *Aspergillus* species, and is showing promise in

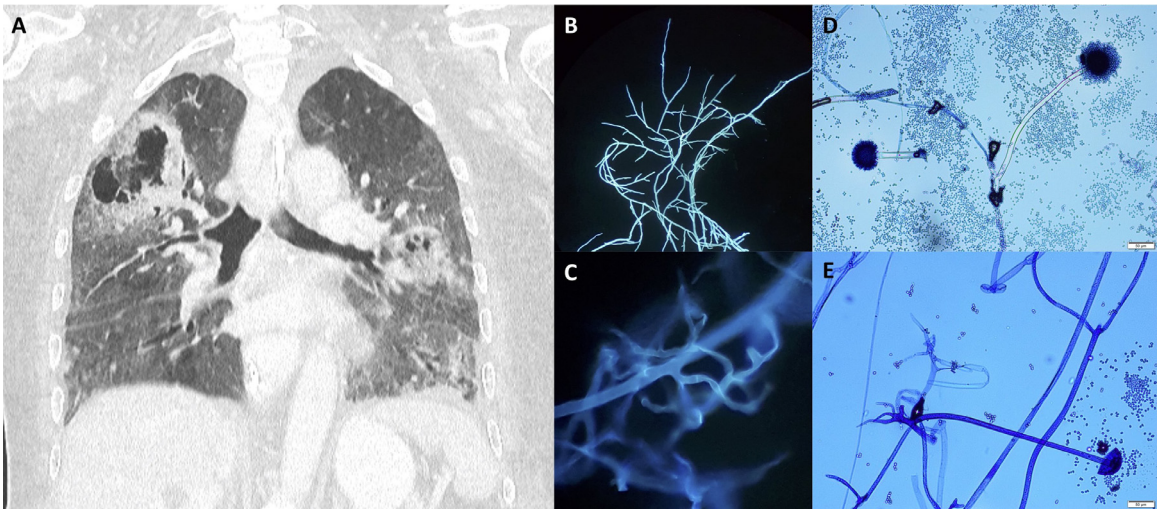

Fig. 2. (A) Chest CT on day 19 after symptom onset, which shows a right apical excavation with peripheral condensation. Direct examination of the bronchoalveolar lavage with calcofluor white showing (B) *Aspergillus*-type mycelium (original magnification × 200) and (C) Mucorales-type mycelium (original magnification × 400). Microscopic culture examination with cotton blue showing (D) *Aspergillus niger* and (E) *Rhizopus delemar* (original magnification × 200).

mixed mold infections [7,14]. In 34.2% of co-infections that were analyzed in this literature review, *Aspergillus* was identified and treated first with voriconazole suggesting that the mucormycosis may be a breakthrough IFI that could be avoided using isavuconazole as a first-line treatment. Our patient was treated with voriconazole, against which *Rhizopus delemar* has high a MIC, which was confirmed by our data [36]. He was later switched to liposomal amphotericin B after Mucorales-type mycelium was identified in the direct BAL examination. However, this antifungal modification may have occurred too late in the infection's course considering that the serum PCR was retrospectively found to be positive 6 days before the antifungals were changed.

CONCLUSION

The SARS-CoV-2 virus has highlighted the existence of multiple fungal superinfections in patients who were not previously immunocompromised and who did not have common risk factors for invasive mold disease. However, the cumulative steroid dose for concomitant COVID-19 infection should be considered to be a risk factor for fungal infection in these patients. These superinfections, particularly CAPA and mucormycosis and the association between the two infections, as shown in the case of our patient, require adapting the management of these patients by screening respiratory and serum samples using biomarkers.

Declaration of Competing Interest

The authors have nothing to declare

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