Diagnosing pulmonary aspergillosis is much easier than it used to be: a new diagnostic landscape

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SUMMARY

Significant innovations in the past decade have resulted in more sensitive and faster diagnosis of allergic, chronic and invasive pulmonary aspergillosis, as well as *Aspergillus* bronchitis and *Aspergillus* nodules. This new diagnostic landscape has revealed that the incidence and prevalence of aspergillosis is substantially higher than previously understood, and is summarised in this review. Oral and intravenous antifungal treatment offers good clinical response rates for affected patients. Nevertheless, missed diagnoses mean that patients are over-treated with antibacterial agents, corticosteroids and anti-TB drugs, resulting in continuing illness and often death. The clinical introduction of several high performing diagnostic tests is helping to redefine patient management. It is well-known that *Aspergillus* antigen can be detected in 70–95% of bronchoscopy samples in patients with invasive and chronic aspergillosis in less than 1 hour. *Aspergillus* immunoglobulin G (IgG) (precipitins) is >90% sensitive and >85% specific for chronic and allergic aspergillosis. High-volume respiratory fungal culture and *Aspergillus* polymerase chain reaction have 3–5-fold higher sensitivity than routine bacterial culture. *Aspergillus* IgE (or skin prick testing) diagnoses *Aspergillus* sensitisation in asthma, cystic fibrosis, chronic obstructive pulmonary disease and post-TB, and correlates well with poorer lung function and/or exacerbations. Clinicians and laboratorians across the world need to mainstream these excellent new tools to improve clinical outcomes by delivering results in a more timely and accurate fashion.

KEY WORDS: *Aspergillus*; antibody; immunodeficiency; HIV/AIDS; leukaemia

ASPERGILLUS FUMIGATUS IS RESPONSIBLE for remarkably diverse disease manifestations, ranging from infection in healthy and immunocompromised people to several allergic and locally invasive conditions. This diversity of different disease patterns is greater than those caused by any other microorganism, and reflects a complex fungal/host interaction, including invasive and chronic forms of pulmonary and sinus disease, dissemination to every organ, post-operative aspergillosis, trauma/burn and corneal local invasion, onychomycosis, external otitis and allergic disease. Every breath drawn includes *Aspergillus* spores, and in general, innate immune defences are able to eradicate these slowly. *Aspergillus* spores are found in normal lungs; 70% of normal lungs yielded positive culture results when sampled after death from other causes.[1] The clinical and imaging manifestations of all forms of aspergillosis are subtle in their early stages and mimic other conditions. Only in the more advanced cases are the clinical features more apparent, but even then, are rarely distinctive. All forms of pulmonary and bronchial aspergillosis are 'stealth' conditions.

The most common comorbid conditions for invasive, chronic and allergic aspergillosis are shown in Table 1.[2–35] Also documented are approximate frequencies (incidence or prevalence) in these different clinical backgrounds.

NEW DIAGNOSTIC TOOLS AND DATA ON ASPERGILLOSIS

Given the subtle nature of most forms of aspergillosis, and its clinical and radiological overlap with other pulmonary conditions, mycological tests are mandatory to confirm or rule out the diagnosis. The turnaround times (TATs) quoted below are in-laboratory ones, and do not include transport to the laboratory or delays in delivering the result, which are often considerable for send away tests.

Fungal culture

Culture testing of respiratory samples should be

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Table 1   Manifestations of pulmonary aspergillosis and their approximate incidence and prevalence in different patient groups

Entity	Comorbid condition	Frequency in that condition	Comment	Reference
Community-acquired *Aspergillus* pneumonia	Influenza, COPD, diabetes, mild immunosuppression	Uncommon	Difficult to document as diagnostic approach unclear	2
Chronic	TB and NTM infection	~10%	Co-infection and post-TB most common	3, 4
	COPD	<1%		5
	Sarcoidosis	~4%		6, 7
	Lung cancer surgery	3.5%	Cumulative incidence after 10 years	8
	Asthma, ABPA	<5%	Uncertain	9
	Pneumothorax	?	No follow-up study published; probably rare and linked to bullae	10
ABPA*	Adult asthma	0.7–5%	More common in the Indian subcontinent	11, 12
	Childhood asthma	?	Described in several case series in India	13, 14
	Cystic fibrosis	10–25%	Peak risk period is late teenage years or early adulthood	15
Severe asthma with fungal sensitisation*	Severe asthma in adults	30–70%	Prevalence depends on which fungi are tested for and the definition of severe asthma	16
Aspergillus sensitisation *Aspergillus* colonisation	COPD exacerbations	Doubles hospitalisations, increases mortality	Airborne fungal sensitisation and home exposure in South East Asia strongly linked to hospital admission and worse COPD	17, 18
Aspergillus bronchitis	Bronchiectasis and cystic fibrosis	30% in cystic fibrosis	Data are evolving and incomplete	19
Extrinsic allergic alveolitis	Occupational exposure	Industry-specific	Malt worker's lung, tobacco worker's lung; occupational asthma may be induced by fungal exposures	20
Invasive	Leukaemia and lymphoma, multiple myeloma, myelodysplasia	2–15%	Primarily linked to neutropenia, corticosteroids and ibrutinib; reduced by prophylaxis	21–23
	Transplantation	0.5–26%	Airways disease most common in lung transplantation; linked to GvHD in allogeneic HSCT	21
	ICU patients	1–12%	Most common in influenza (8–24%), COVID-19 (~20%) and medical patients	24–28
	Lung cancer	2.6%	Many probably sub-acute invasive	29
	Autoimmune disorders such as SLE	<4%	Especially linked with corticosteroid and other immunocompromising treatments	30,31
	COPD admissions to hospital	1.3–3.9%	Some cases linked to oral corticosteroid use	5
	Liver failure	5–14%	Partially linked to corticosteroids	21, 32
	Severe fever with thrombocytopenia syndrome	20%	Tick-borne infection in Northeast Asia	33
	HIV/AIDS	~4%	Advanced disease, corticosteroids and neutropenia	34,35
	Chronic granulomatous disease and other primary immune deficiencies†	~40% lifetime risk	Lifelong prophylaxis reduces risk	21

* Also known collectively as fungal asthma, a catch-all term for several entities linking fungal allergy/sensitisation and airway infection with worse asthma.
† Other primary immunodeficiency disorders with moderate or high risk of invasive aspergillosis include caspase recruitment domain family member 9 deficiency, hyper-immunoglobulin E syndrome, MonoMAC syndrome due to defects in GATA2 and other even rarer disorders.
COPD = chronic obstructive pulmonary disease; NTM = non-tuberculous mycobacteria; ABPA = allergic bronchopulmonary aspergillosis; GvHD = graft versus host disease; HSCT = haematopoetic stem cell transplant; ICU = intensive care unit; SLE = systemic lupus erythematous

performed on fungal (not bacterial) media, as the yield is 30% higher for *Aspergillus* (Table 2).[36] As overall culture positivity rates in all forms of aspergillosis are under 30% with standard fungal culture techniques,[37,38] these cannot be used to exclude infection. High-volume culture techniques boost culture-positive rates by at least three-fold.[39] Bronchoalveolar lavage fluid (BAL) is inferior to

Table 2 Diagnostic sensitivity of rapid tests for aspergillosis (all figures are rounded for ease of comprehension)

Test	Turnaround time (hours)	Acute invasive (%)	Sub-acute invasive (%)	Chronic cavitary (%)	*Aspergillus* nodule (%)	Allergic (%)	*Aspergillus* bronchitis (%)
Culture on respiratory samples	48–96	<30*	<30*	<30	<10	<30	~80[†]
Microscopy on respiratory samples	<6	<10	<10	<10	<5	<25[‡]	? 10–25
Antigen on BAL	2–48[§]	70–95[†]	70–90[†]	~75[†]	ND	ND	ND
Antigen on serum	2–48[§]	20–80[†¶]	? 20–30	10–65	ND	ND	ND
IgG antibody	48–96	<25	?30–60	80–90[†]	65	80–90[†]	60–80[†]
IgE antibody	48–96	ND	ND	60–70	ND	90–100[†]	ND
Beta-D-glucan on serum	2–48[#]	50–75	ND	~20–75	ND	<5	
Aspergillus PCR on respiratory samples	24–72	75–90[†]	75–90[†]	~60	ND	~60	~75[†]
Aspergillus PCR on serum/blood	24–72	60-80[†¶]	ND	<20	ND	ND	ND

* Yield may be higher in COPD patients with invasive aspergillosis– not carefully studied.
[†] Best tests to request in these clinical circumstances.
[‡] Sputum plugs from ABPA usually show fungal hyphae, eosinophils and Charcot-Leyden crystals.
[§] Lateral flow antigen tests are quick. Single-sample *Aspergillus* ELISA is <2 hours, other ELISA assays are batched.
[¶] Highest sensitivity in profoundly neutropenic patients without mould-active prophylaxis.
[#] Single-sample assays are quicker, otherwise batch tested.
BAL = broncholaveolar lavage; ND = no data; Ig = immunoglobulin; PCR = polymerase chain reaction; COPD = chronic obstructive pulmonary disease; ABPA = allergic bronchopulmonary aspergillosis; ELISA = enzyme-linked immunosorbent assay.

bronchial aspirate and sputum obtained pre- or post-bronchoscopy,[40] indicating that most *Aspergillus* resides in the larger airways, not the alveoli, unlike cytomegalovirus and *Pneumocystis jirovecii*. The more samples submitted, the higher the yield, which may go up to ~60%.[41] Culture rates in chronic obstructive pulmonary disease (COPD) patients may be higher, as airway macrophages and epithelial cells are poor at destroying ingested spores in these patients.[42] There is no relationship between colony counts and severity of disease:[43] a single colony can reflect rapidly invasive aspergillosis,[44] whereas high colony counts may signal tracheobronchitis rather than parenchymal disease. Blood cultures are almost always negative for *Aspergillus*.[45] Positive cultures can be tested for azole resistance. Culture TAT is usually 48–96 h.

Microscopy

The most rapid methodology for direct microscopy is the fluorescent brightener and microscope; slides can be scanned in under 3 min by an experienced technician.[46] Alternatives include potassium hydroxide, Gram staining and silver staining (after cytocentrifugation), all of which are probably slightly less sensitive.[45] Most samples are microscopy-negative; a positive is highly correlated with disease, often tracheobronchitis, but also invasive and chronic aspergillosis (Table 2). The TAT should be under 6 h.

Aspergillus *antigen*

Commercially available for over 25 years, the *Aspergillus* galactomannan (GM) antigen enzyme-linked immunosorbent assay (ELISA) from BioRad (Marnes-la-Coquette, France) is substantially more sensitive than culture or microscopy on BAL. Prior to antifungal therapy, sensitivity and specificity of GM ELISA is 80–90% for all forms of invasive aspergil-

losis[25,28,38,47] and ~75% for chronic pulmonary aspergillosis (CPA)[48] (Table 2). Blind tracheal aspiration in intensive care unit (ICU) patients has not been well studied. As sputum yields much higher GM values than serum or BAL,[49] an alternative cut-off to define positivity is required. Serum samples need heating before analysis. The TAT is 24–48 h.

Recently, there have been three developments in this field, which could lead to more rapid results.[50,51] The most recent is a single sample, 1-h semi-automated ELISA launched by Vircell (Granada, Spain). Performance appears to be similar to the long-established ELISA product sold by BioRad and more recently by Dynamiker (Tianjin, China) and Era Biology (Tianjin, China). A new lateral-flow GM assay yields a result in <1 h on BAL (Immy, Norman, OK, USA). The performance appears similar to the GM ELISA assay and can be read visually or with a reader. Another lateral-flow device detects a different *Aspergillus* antigen in <1 h and has a sensitivity apparently slightly less than GM, possibly because the current comparator uses GM as a gold standard (OLM Diagnostics, Newcastle, UK).

Aspergillus *IgG and IgM antibody*

Detectable *Aspergillus* immunoglobulin (Ig) G is the cornerstone of diagnosis of CPA[52] and is usually positive in allergic bronchopulmonary aspergillosis (ABPA) and *Aspergillus* bronchitis in asthma, cystic fibrosis and bronchiectasis patients. The value of IgM detection alone has not been fully evaluated, but it is less sensitive than IgG in CPA.[53] Studies in CPA show a sensitivity of *Aspergillus* IgG from ~70% to >90%, depending on the assay and cut-off used.[52] Multiple studies from many countries have evaluated the optimum cut-offs. Compared with blood donors, the specificity of *Aspergillus* IgG assays is ~98%, but this falls to ~85% if patients with respiratory disease

are used, probably because of some true-positives for other forms of aspergillosis among 'controls'. The new lateral flow assay for both *Aspergillus* IgG and IgM is the best performing assay,[54] but is not quantitative, unlike the other assays. Counter-immunoelectrophoresis (CIE) to detect precipitins is slower and less sensitive than ELISA and lateral flow testing.[55] False-negative tests in CPA are linked to subtle immune defects such as mannose-binding lectin deficiency, low natural killer or plasma cell populations and poor pneumococcal antibody responses.[56] Lateral flow assay TAT is <1 hour, 24–72 h for ELISA and automated assays as these are usually batched, and about 7 days for CIE.

Aspergillus *polymerase chain reaction*

The challenges to providing reliable, uncontaminated, sensitive polymerase chain reaction (PCR) to detect *Aspergillus* spp. commercially were first overcome in 2011 with the launch of the MycAssay kit (Myconostica, Camberley, UK). Since then, several companies have brought *Aspergillus* PCR tests to the market with slight variations in design.[46,57] With respiratory samples, as with culture, *Aspergillus* PCR cannot distinguish colonisation from infection, and consequently will always require clinical interpretation, supported by other diagnostic tests and imaging appearances. PCR is more sensitive than culture, depending on the DNA extraction, PCR and culture methods used. Different samples can yield different answers because fungal hyphae and DNA are not evenly distributed in respiratory samples. BAL is less sensitive than bronchial and tracheal samples, probably because of dilution.[40] Although the actual laboratory processes for DNA extraction and PCR only take a few hours, in practice, it takes 24–48 h to yield a result after the receipt of a sample in Cat 3 laboratory. The performance of PCR assays developed in-house may or may not adequate, including generic PCR when using targets such as internal transcribed spacer or 18S ribosomal RNA targets, with or without subsequent sequencing.[56]

PCR on blood or serum samples is really only useful in profoundly immunocompromised patients.[58] As blood tubes containing *Aspergillus* DNA may occasionally yield a false-positive result,[59] confirmation on a second sample is helpful. However, as *Aspergillus* DNAaemia may not be continuous, clinical correlation is required with all available data.

Respiratory sample acquisition

Expectorated or induced sputum can be submitted for microscopy, culture and PCR. Physiotherapy-assisted cough can yield good specimens,[60] as does sputum induction. Bronchoscopy is useful for inspecting the airways if tracheobronchitis is considered, and BAL testing for *Aspergillus* antigen the most sensitive test.

But the bronchial wash specimen is superior to BAL for culture and PCR, probably because more *Aspergillus* is present in the airways than in the alveoli.[40] Bronchial biopsy is useful for suspicious lesions, especially in tracheobronchitis,[61] and aspirated mucus should be submitted for microscopy or cytology, culture and histopathology.

MANIFESTATIONS OF ASPERGILLOSIS (INCLUDING RADIOLOGY)

Brief summaries of each major entity are provided below. The severity of clinical features varies substantially. Early in the disease, there are no symptoms and imaging findings are minor, or easily mistaken for other conditions. Physical examination of the chest may be abnormal, but any signs heard are rarely specific to any form of aspergillosis, except for a monophonic wheeze, which is a rare but characteristic sign of invasive *Aspergillus* tracheobronchitis.

Community-acquired Aspergillus *pneumonia*

This uncommon disorder is linked to massive airborne exposures and sometimes, influenza or a recent course of corticosteroids.[2] The exposure history includes bark chippings, waste disposal, extensive cleaning of damp properties, composting and gardening. Patients have occasionally had influenza. Fever, anorexia, cough and breathlessness are typical, with no improvement with antibacterial agents. Patients usually present acutely with 'pneumonia' (usually consolidation which cavitates) or a bilateral miliary or nodular pattern which is very unusual. Undetected, severe disease is rapidly fatal; sub-acute community-acquired *Aspergillus* pneumonia may resolve or develop into CPA.

Invasive pulmonary aspergillosis

Early clinical features are mostly absent, but patients may have fever, dry cough, chest discomfort and malaise.[21,22] Occasional patients produce sputum or have haemoptysis. Other patients present with non-pulmonary features such as stroke or rhinosinusitis. Patients with sub-acute invasive pulmonary aspergillosis have clinical and radiological features for 4–12 weeks, and are almost always symptomatic with the above features plus weight loss, anorexia and fatigue.[52,62] Inflammatory markers may or may not be raised.

Chest X-ray (CXR) is falsely negative in at least 10% of patients; computed tomography (CT) is a much better investigation. Contrast has often not been used to avoid the possibility of nephrotoxicity, but recent work shows that contrast is valuable because vessel occlusion can be seen.[63] This radiological sign is useful in both neutropenic and non-neutropenic patients, but will not be discriminatory
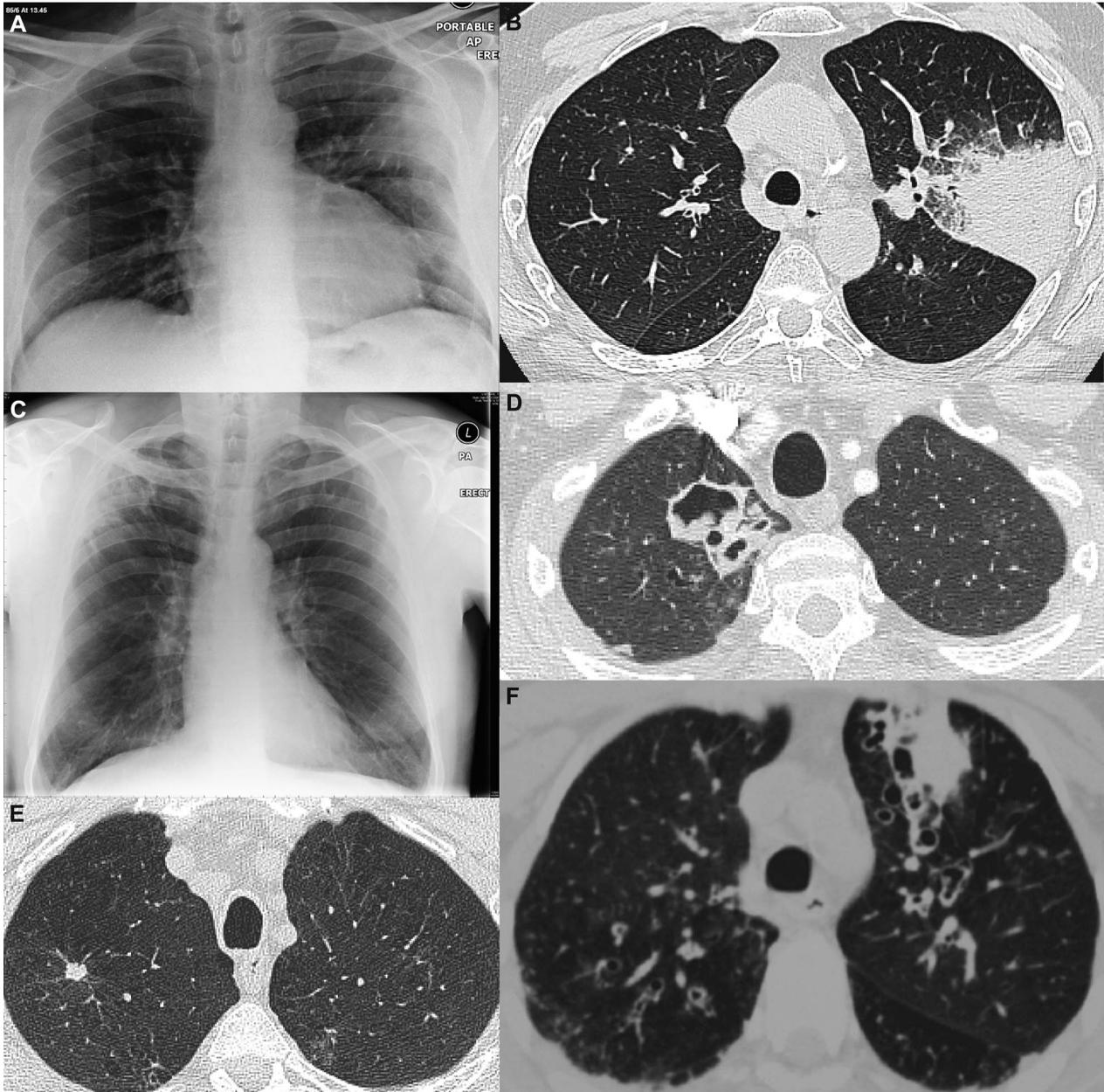
Figure 1   Radiological examples of different patterns of aspergillosis. **A)** Chest X-ray of a patient with left upper lobe consolidation typical of pneumonia, but actually invasive aspergillosis. **B)** CT scan of the thorax in the same patient as **A)**, only 2 days apart showing dense consolidation of the posterior segment of the left upper lobe, sharply demarcated by the fissure, a sharp edge against the pleura and anterior ground glass appearance, typical of invasive aspergillosis. **C)** Right upper lobe cavitary lesion of chronic pulmonary aspergillosis. **D)** Thick walled cavity containing fungal material (a very small aspergilloma) adjacent to enlarged and thickened airways and one very small cavity posteriorly. Highly characteristic appearance of chronic pulmonary aspergillosis, although no pleural thickening it does not abut the pleura. **E)** *Aspergillus* nodule in an emphysematous lung, one of two in this patient, diagnosed using biopsy. **F)** Severe bronchiectasis of long-standing ABPA with focal areas of distal consolidation or airway obstruction in the inferior lingular segment during an exacerbation. CT = computed tomography; ABPA = allergic bronchopulmonary aspergillosis.

in Covid-19 patients, who often have intrapulmonary infarction.[28] Bilateral findings are a strong clue to the diagnosis of invasive aspergillosis. Nodules, some with cavitation or with surrounding ground glass (halo sign), are most common, but areas of non-specific consolidation are also common (Figure 1A and B). Pleural effusions are rare, but useful if present, as they can be sampled for *Aspergillus* antigen.

Chronic pulmonary aspergillosis

Patients with *Aspergillus* nodules and simple aspergilloma usually have no specific symptoms for aspergillosis.[52,64] Early symptoms of cavitary CPA may be minor and inconsistent with radiological appearances, which often appear more severe. Symptoms of cavitary CPA may be systemic (weight loss, marked fatigue, rarely fever and/or night sweats) or

Figure 2 An example of left-sided fibrosing CPA (destroyed lung) from long-standing CPA. CPA = chronic pulmonary aspergillosis.

pulmonary (cough, haemoptysis, chest discomfort and/or breathlessness), or any combination of these. Fatigue is often profound. Symptoms typically fluctuate minimally while gradually progressing over the course of weeks to months. Multiple treatments are usually attempted with little respite. Concomitant bacterial infection is common, with slight improvement with antibiotics noted. Patients with fibrosing CPA are breathless on exertion, and usually systemically unwell.[64] Inflammatory markers may or may not be raised.

Aspergillus nodules may be single or multiple (Figure 1E).[64] They are increasingly being recognised during screening for lung cancer. They are most often found on a background of COPD, asthma that has required courses of oral corticosteroids or rheumatoid arthritis. They usually show low to moderate fluorodeoxyglucose uptake on positron emission tomography.

Simple aspergilloma is always unilateral and non-progressive and sometimes found incidentally. Some patients present with haemoptysis, wheeze or cough. A cavitary lesion with a fungal ball showing an air crescent on CXR or CT is typical. Fungal ball mobility is no longer a criterion for diagnosis.

In cavitary CPA, >40% of patients have bilateral abnormalities,[65] and the upper lobes are involved in

>85% of cases. The cardinal features of cavitary CPA are one or more cavities, which expand or coalesce over time, usually have thick walls, pleural thickening and pericavitary infiltrates (Figure 1C and D).[51] On CT scan, the inside of the cavity shows irregularity of the inner cavity wall (which is fungal growth) or a fungal ball or non-descript contents, occasionally a fluid level.[66] The presence of a fungal ball or aspergilloma is a late feature of CPA.[65] Fat ingress adjacent to pleural thickening is common, as is associated bronchiectasis. Effective therapy reduces the thickness of cavity walls, peri-cavitary infiltrates and pleural thickening. Cavitary CPA can progress to fibrosing CPA (Figure 2),[67] involving two or more lobes, usually with some persistent cavitation and often an aspergilloma.

Allergic bronchopulmonary aspergillosis

In patients with asthma, ABPA presents in several ways,[41,68] the most common presentation being poorly controlled asthma and cough. Some patients present with typical symptoms of bronchiectasis with productive cough and green sputum. A remarkable presentation is the production of brown plugs, described variously as 'slugs' or 'peas' or 'jelly babies'. Occasional patients present with 'pneumonia', having an infiltrate on their CXR, but without being as systemically ill as the CXR might suggest. Profound fatigue, faint rash or worse eczema, or a surprise finding at bronchoscopy of mucous plugging are occasional presentations. Positive sputum cultures for bacteria are common and can be misleading.

In cystic fibrosis, an exacerbation that does not remit with systemic antibiotics, sometimes with a new infiltrate is the most common presentation of ABPA,[15,69] but often the diagnosis is made by screening using total IgE or *Aspergillus* IgE. Forced expiratory volume in 1 sec (FEV_1) is usually substantially reduced in an ABPA exacerbation.

Occasional patients present with ABPA who do not have asthma or cystic fibrosis. Some are heterozygous for a cystic fibrosis transmembrane receptor (CFTR) gene abnormality or have a rare and mild CFTR defect and late-onset cystic fibrosis.[70] Some patients with a very similar syndrome to ABPA are allergic to other fungi, an entity called allergic bronchopulmonary mycosis.[71]

Patients with ABPA may have normal CXR, but more often have bronchiectasis or an infiltrate.[72] Bronchiectasis was previously thought to be more central, but may be seen anywhere, and becomes more prominent in an area previously affected by mucous plugging. A highly characteristic feature of ABPA is hyper-attenuated mucus, best seen on CT bone windows.[71,72] Patients may manifest ABPA together with a form of CPA with pleural thickening and peripheral fibrosis at presentation or years later.[9]

Aspergillus *sensitisation*

Aspergillus sensitisation is defined by *Aspergillus* IgE, detectable in serum or on positive skin prick testing.[5,17,73] Sensitisation to *Aspergillus fumigatus* is usually (but not always) accompanied by sensitisation to multiple other allergens, some airborne fungi. *A. fumigatus* has about 60 IgE-binding proteins, some of which cross-react with other organisms, including homologous human proteins.[74,75]

The most common groups to manifest *Aspergillus* sensitisation are patients with ABPA (100% by definition), severe asthma (30–70%), COPD (35–55%), cured TB (∼30%), bronchiectasis (∼30%), cystic fibrosis (∼40%) and CPA (∼70%).[5,15,17,20,68,69,71,73] In most cases, *Aspergillus* sensitisation is linked to worse lung function and/or more exacerbations. There is no radiological correlate for *Aspergillus* sensitisation, and it is not directly linked to airway or lung infection with *Aspergillus* or *Aspergillus* IgG positivity.

Severe asthma with fungal sensitisation

By definition (of which there are several), these patients have severe asthma, usually poorly controlled, with fungal sensitisation.[76] Their presentation is with moderate or severe asthma symptoms, exacerbations, including hospitalisation and ICU admission and associated low mood. Many are chronically unwell and effectively disabled by their asthma. It is uncommon for these patients to have bronchiectasis – bronchial wall thickening is more common on CT scanning. Some have eosinophilic asthma, but as corticosteroid usage is often high, this may be long in the past. They do not produce brown sputum plugs.

The radiology is not distinctive: these patients do not present with infiltrates, mucous obstruction or marked bronchiectasis as ABPA does.

Aspergillus *bronchitis*

Patients with *Aspergillus* bronchitis present in two ways, primarily with productive cough and breathlessness, and almost all grow bacteria in their sputum alongside *Aspergillus*.[19] First, they present with recurrent bronchitis or chest infections, which only partially clear with antibacterial therapy. Second, they present with thick tenacious sputum, violent cough and sometimes acute dyspnoea. Most, but not all, have bronchiectasis and are not immunocompromised. A similar, but often acute presentation in immunocompromised patients, is referred to as obstructing bronchial aspergillosis,[77] which is sometimes an immune reconstitution syndrome. The CXR usually shows only the features of bronchiectasis, but occasionally, opacification due to airway obstruction. If bronchoscopy is done, multiple airways may be found to be inflamed, some with contact bleeding, and there may be multiple mucus plugs.

Aspergillus *colonisation*

Airway *Aspergillus* colonisation is relatively common, especially in COPD patients.[78] This is distinguished from *Aspergillus* bronchitis by the lack of symptoms referable to the airways (i.e., bronchitis, sputum plugs), a single positive culture or PCR and lack of evidence suggesting invasive, chronic or allergic aspergillosis.[19] The one common exception is cystic fibrosis, in which long term colonisation is relatively common and can only reliably be distinguished from *Aspergillus* bronchitis or ABPA by the absence of a raised *Aspergillus* IgG or IgE.[69]

Occupational or exposure-related extrinsic allergic alveolitis

These patients present, as do others with extrinsic allergic alveolitis (EAA), with cough and breathlessness and sometimes, fatigue.[20] If there is no known occupational risk, the exposure history is sometimes difficult to elicit, and is occasionally occult, as for example, with contaminated air conditioning or air intake ducts in offices or homes, undetected dampness at home or work, breathing apparatus and other examples. Imaging is highly characteristic with CT findings of extensive ground glass opacities, which are usually bilateral and symmetrical, but sometimes patchy. If linked to *Aspergillus*, the *Aspergillus* IgG antibody is raised.[79]

Pulmonary aspergillosis in children

In children with cystic fibrosis, ABPA and *Aspergillus* bronchitis occur with increasing frequency in teenage years.[14,15] Other children without cystic fibrosis may develop invasive and allergic aspergillosis, including fungal asthma, but CPA has yet to be described.

DIAGNOSTIC APPROACH

Diagnostic approaches have been described below by clinical context to support physicians working in different settings. Outpatients and clinic patients are addressed first, then hospitalised patients and finally, bronchoscopy. Only major clinical groups are addressed. Much guidance is published for transplant patients, who are not covered here. However, there are some common threads: 1) culture is insensitive and multiple specimens and supporting antigen or antibody data are extremely helpful; 2) most diagnoses (but not all) depend on recognised risk factors or exposures, antigen or antibody data and characteristic imaging findings; 3) to note, it is a mistake to make decisions for aspergillosis primarily on patient symptoms. The aphorism 'Treat the patient, not the X-ray' does aspergillosis patients a great disservice, as these disorders are so clinically quiet, symptoms

Test	Invasive	Chronic	Allergic	*Aspergillus* bronchitis
Respiratory samples				
Fungal culture	✓	✓✓	✓✓	✓✓✓
Microscopy	✓	✓	✓	✓
Antigen	✓✓✓	✓✓✓	—	—
Aspergillus PCR	✓✓✓	✓✓	✓✓	✓✓✓
Blood				
Antigen	✓✓	✓	—	—
IgG antibody	✓	✓✓✓	✓✓✓	✓✓
IgE antibody	—	✓✓	✓✓✓	-
Beta-D-glucan	✓✓	✓✓	—	—

Figure 3 Summary of diagnostic tests for pulmonary aspergillosis and their overall utility, excluding imaging and histopathology.* ✓ = <35% sensitive; ✓✓ = 35–75% sensitive; and ✓✓✓ = >75% sensitive. PCR = polymerase chain reaction; Ig = immunoglobulin.

alone are a poor guide to severity or improvement in most cases. A summary diagnostic approach by disease is shown in Figure 3.

TB clinics

CPA may 1) mimic TB (>20% of HIV-negative pulmonary TB);[80] 2) be a co-infection with TB during treatment (~10%);[81,82] or 3) follow TB, mostly in those left with cavities (6.5% per year, if residual cavity).[3,4,83] Haemoptysis, lack of fever, radiological features of pleural thickening or thick-walled cavities and surrounding infiltrates are all clues to the diagnosis of CPA (Figure 4). Empirical dual therapy with an azole and rifampicin is ill-advised because of the profound drug interactions. *Aspergillus* IgG testing is crucial,[84] and the diagnosis of CPA is

supported by positive microscopy, culture and PCR from sputum or antigen from bronchoscopy.

Asthma clinics

In patients who are struggling, recently had an exacerbation or who have bronchiectasis, the key test is the *Aspergillus* IgE or skin prick test,[16] supported by an *Aspergillus* IgG, a total IgE and respiratory fungal culture or PCR (Figure 4). The most common forms of aspergillosis in asthma are ABPA, severe asthma with fungal sensitisation and *Aspergillus* bronchitis.

Cystic fibrosis

This is very similar to asthma, except that all patients with cystic fibrosis have bronchiectasis, and *Aspergillus* bronchitis is as common as ABPA.[14,15,69] *Aspergillus* colonisation is also common. *Aspergillus* sensitisation alone is linked to worse lung function.

Chronic obstructive pulmonary disease

Currently, it is not routine practice to actively seek any form of aspergillosis in COPD outpatients. Knowledge of sensitisation and whether patients are colonised adds a layer of understanding as to why individual patients are either very breathless or frequent exacerbators.[5,17,18] There is no direct evidence base currently for treating such patients, unless *Aspergillus* bronchitis can be diagnosed, although patients with fungal asthma do respond to antifungal agents. Colonised COPD patients may be
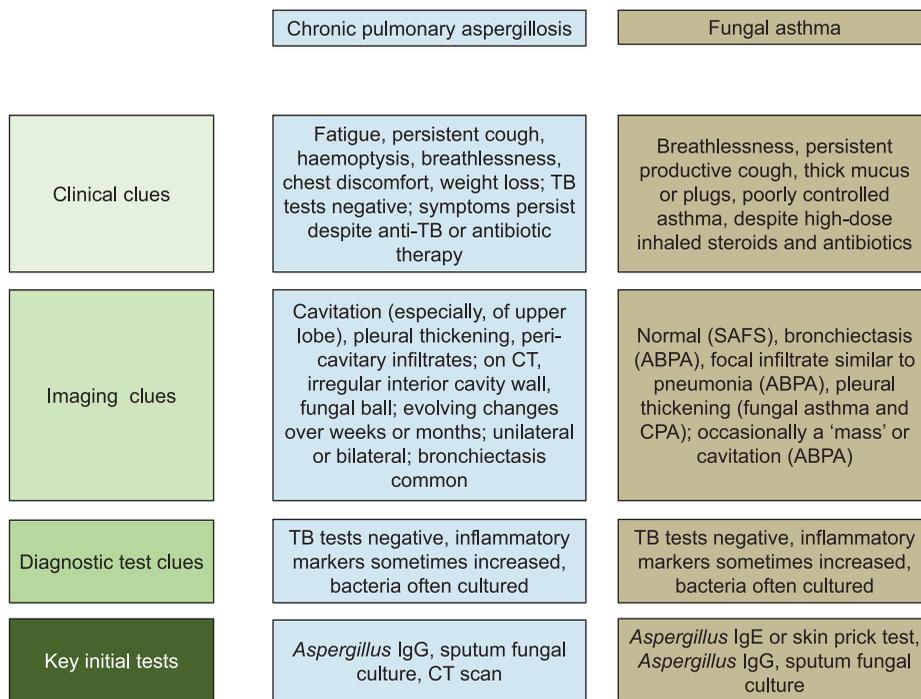
	Chronic pulmonary aspergillosis	Fungal asthma
Clinical clues	Fatigue, persistent cough, haemoptysis, breathlessness, chest discomfort, weight loss; TB tests negative; symptoms persist despite anti-TB or antibiotic therapy	Breathlessness, persistent productive cough, thick mucus or plugs, poorly controlled asthma, despite high-dose inhaled steroids and antibiotics
Imaging clues	Cavitation (especially, of upper lobe), pleural thickening, peri-cavitary infiltrates; on CT, irregular interior cavity wall, fungal ball; evolving changes over weeks or months; unilateral or bilateral; bronchiectasis common	Normal (SAFS), bronchiectasis (ABPA), focal infiltrate similar to pneumonia (ABPA), pleural thickening (fungal asthma and CPA); occasionally a 'mass' or cavitation (ABPA)
Diagnostic test clues	TB tests negative, inflammatory markers sometimes increased, bacteria often cultured	TB tests negative, inflammatory markers sometimes increased, bacteria often cultured
Key initial tests	*Aspergillus* IgG, sputum fungal culture, CT scan	*Aspergillus* IgE or skin prick test, *Aspergillus* IgG, sputum fungal culture

Figure 4 Diagnosis of CPA and fungal asthma. CT = computed tomography; SAFS = severe asthma with fungal sensitisation; ABPA = allergic bronchopulmonary aspergillosis; CPA = chronic pulmonary aspergillosis; Ig = immunoglobulin.

more at risk of developing invasive aspergillosis, as is the case with leukaemia and transplant patients, but again this is not proven.

Both invasive and chronic aspergillosis occur in patients admitted to hospital with COPD.[5] Positive culture(s) of *Aspergillus* is found in up to 20% on admission (with two samples submitted), and 22–67% reflect invasive aspergillosis, the remainder colonisation.[85,86] Clues to the diagnosis of aspergillosis include bilateral shadows, nodules, cavitation and recalcitrant wheeze (consider tracheobronchitis). Serum *Aspergillus* IgG and possibly glucan are useful in supporting a diagnosis of aspergillosis.

Intensive care units

Medical ICU patients are at risk of invasive aspergillosis for multiple reasons, including underlying respiratory disease, compensatory anti-inflammatory response syndrome (CARS) and its accompanying immune deficits,[87] and medications such as corticosteroids. The baseline incidence of invasive aspergillosis in the ICU is 1–12%, but severe influenza or Covid-19 can push the incidence up to 8–25%.[24–28] Cultures from bronchoscopy or tracheal aspirates are ~40% sensitive, and colonisation is as frequent as infection. BAL and *Aspergillus* antigen increases sensitivity to ~90%,[38] and in some studies up to 30% of serum antigen is positive.[88] Glucan may also be positive,[28] but not specific for aspergillosis, as it also indicates deep *Candida* infection and *Pneumocystis* pneumonia. Tracheobronchial aspergillosis is more common than is generally realised, requiring direct inspection for diagnosis.[61] Bronchoscopy in the first 48 h of admission with culture, microscopy and *Aspergillus* antigen testing, repeated after 7–10 days, is the optimal diagnostic approach. In Covid-19 and influenza patients, inspection of the airways and *Aspergillus* antigen on BAL are the most helpful diagnostic approaches.[24,27,28]

Leukaemia and other haematological disorders

Neutropenia (along with monocytopenia and thrombocytopenia), corticosteroid use and phagocyte dysfunction all contribute to the risk of invasive aspergillosis.[21] Prophylaxis with mould-active antifungals such as posaconazole is commonly used in the highest-risk patients. In those not on such prophylaxis, serum *Aspergillus* antigen and serum of blood PCR have a high sensitivity, but only PCR is sensitive in those on prophylaxis.[46,58] Presumptive infection is often highlighted by CT of the chest or sinuses. BAL is the best sample for diagnosis. Glucan is more useful in those without profound neutropenia.

Lung cancer

The optimal approach for diagnosis in these patients

has not been well studied. As many cases are probably sub-acute invasive, *Aspergillus* IgG testing may alert clinicians to infection, and prompt a bronchoscopy and BAL testing for microscopy, culture and *Aspergillus* antigen and PCR. Biopsy of abnormal areas is likely to be diagnostic.

Outpatient bronchoscopy

Bronchoscopy sometimes yields surprise findings, and all forms of aspergillosis can be diagnosed using bronchoscopy. Where it is possible, biopsy with fungal tissue stains are diagnostic of invasive and tracheobronchial disease. Thick mucus should be submitted for histopathology or cytology, as well as direct microscopy and culture, and additional samples sent for *Aspergillus* culture, antigen and PCR.

FUTURE PERSPECTIVES

Aspergillosis, like most fungal infections, is an unusual infection, but extremely important for the patient's survival and well-being. A high level of awareness is critical, especially as 'routine microbiology' is so often negative, and imaging is only occasionally specific. For life-threatening, invasive aspergillosis, speed is of the essence. Time to result should be less than 48 h, and preferably shorter. Send away tests for this disease should all be 'stat'. This urgency is not as important for chronic and allergic aspergillosis, but again 'routine microbiology' is of little help.

In future, better risk assessment tools may support early empirical therapy and confirmatory diagnosis. Such tools could include genetic assays in, for example, well-recognised, high-risk groups such as acute myeloid leukaemia, haematopoetic stem cell transplant recipients,[89] TB and severe asthma. Recent major advances in diagnostic test performance and speed need to fully leveraged by clinicians to improve the currently poor outcomes for too many patients globally.

Canada) and Cipla (Mumbai, India) and Pfizer (Groton, CT, USA). He sits on the Data and Safety Monitoring Board for a SARS CoV2 vaccine trial. In the last 3 years, he has been paid for talks on behalf of Dynamiker (Tianjin, China), Hikma (Amman, Jordan), Gilead (Foster City, CA, USA), Merck (Kenilworth, NJ, USA), Mylan (now Viatris) (Mumbai, India) and Pfizer . He is a longstanding member of the Infectious Disease Society of America Aspergillosis Guidelines Group, the European Society for Clinical Microbiology and Infectious Diseases Aspergillosis Guidelines Group.

References

1 Lass-Flörl C, et al. Pulmonary *Aspergillus* colonization in humans and its impact on management of critically ill patients. Br J Haematol 1999; 104(4): 745–747.

2 Denning DW. Community acquired *Aspergillus* pneumonia and/or pneumonitis. The Aspergillus Website. Macclesfield, UK: Fungal Infection Trust, 2015. https://www.aspergillus.org.uk/new_treatment/community-acquired-aspergillus-pneumonia-and-or-pneumonitis Accessed January 2021.

3 Denning DW, Pleuvry A, Cole DC. Global burden of chronic pulmonary aspergillosis as a sequel to tuberculosis. Bull World Health Organ 2011; 89: 864–872.

4 Bongomin F. Post-tuberculosis chronic pulmonary aspergillosis: an emerging public health concern. PLoS Pathog 2020; 16(8): e1008742.

5 Hammond EE, et al. The global impact of *Aspergillus* infection on COPD. BMC Pulm Med 2020; 20: 241.

6 Denning DW, Pleuvry A, Cole DC. Global burden of chronic pulmonary aspergillosis complicating sarcoidosis. Eur Respir J 2013; 41(3): 621–626.

7 Uzunhan Y, et al. Chronic pulmonary aspergillosis complicating sarcoidosis. Eur Respir J 2017; 49(6): 1602396.

8 Shin SH, et al. Incidence and risk factors of chronic pulmonary aspergillosis development during long-term follow-up after lung cancer surgery. J Fungi (Basel) 2020; 6(4): 271.

9 Lowes D, et al. Progression from asthma to ABPA to chronic pulmonary aspergillosis. Resp Med 2015; 109: 1509–1515.

10 Smith N, Denning DW. Underlying pulmonary disease frequency in patients with chronic pulmonary aspergillosis. Eur Resp J 2011; 37: 865–872.

11 Denning DW, Pleuvry A, Cole DC. Global burden of ABPA in adults with asthma and its complication chronic pulmonary aspergillosis. Med Mycol 2013; 51: 361–370.

12 Agarwal R, Denning DW, Chakrabarti A. Estimation of the burden of chronic and allergic pulmonary aspergillosis in India. PLoS One 2014; 9: e114745.

13 Kumari J, et al. Prevalence and risk factors of allergic bronchopulmonary aspergillosis and *Aspergillus* sensitization in children with poorly controlled asthma. J Trop Pediatr 2020; 66(3): 275–283.

14 Manti S, et al. Allergic bronchopulmonary aspergillosis in children. Pediatr Allergy Immunol 2020; 31 (Suppl 26): 20–22.

15 Lattanzi C, et al. Allergic bronchopulmonary aspergillosis in children with cystic fibrosis: an update on the newest diagnostic tools and therapeutic approaches. Pathogens 2020; 9(9): 716.

16 Rapeport WG, Ito K, Denning DW. The role of antifungals in the management of patients with severe asthma. Clin Transl Allergy 2020; 10: 46.

17 Tiew PY, et al. Environmental fungal sensitisation associates with poorer clinical outcomes in COPD. Eur Respir J 2020; 56(2): 2000418.

18 Wu Y, et al. Respiratory *Aspergillus* colonization was associated with relapse of acute exacerbation in patients with chronic obstructive pulmonary disease: Analysis of data from a retrospective cohort study. Front Med May 2021; 8: 640289.

19 Prats JAGG, Denning DW. *Aspergillus* bronchitis. The Aspergillus Website. Macclesfield, UK Fungal Infection Trust, 2016. https://www.aspergillus.org.uk/new_treatment/aspergillus-bronchitis/ Accessed January 2021.

20 Al-Alawi A, Ryan CF, Flint JD, Müller NL. *Aspergillus*-related lung disease. Can Respir J 2005; 12(7): 377–387.

21 Danion F, et al. Why are so many cases of invasive aspergillosis missed? Med Mycol 2019; 57(Suppl): S94–S103.

22 Chen CY, et al. Clinical characteristics and treatment outcomes of pulmonary invasive fungal infection among adult patients with hematological malignancy in a medical centre in Taiwan, 2008–2013. J Microbiol Immunol Infect 2020; 53(1): 106–114.

23 Ghez D, et al. Early-onset invasive aspergillosis and other fungal infections in patients treated with ibrutinib. Blood 2018; 131: 1955–1959.

24 Schauwvlieghe AFAD, et al. Invasive aspergillosis in patients admitted to the intensive care unit with severe influenza: a retrospective cohort study. Lancet Respir Med 2018; 6: 782–792.

25 Loughlin L, et al. Pulmonary aspergillosis in patients with suspected ventilator-associated pneumonia in UK intensive care units. Am J Respir Crit Care Med 2020; 202: 1125–1132.

26 Chen L, et al. Invasive pulmonary aspergillosis in immunocompetent patients hospitalised with influenza A-related pneumonia: a multicenter retrospective study. BMC Pulm Med 2020; 20(1): 239.

27 Waldeck F, et al. Influenza-associated aspergillosis in critically-ill patients-a retrospective bicentric cohort study. Eur J Clin Microbiol Infect Dis 2020; 39(10): 1915–1923.

28 Armstrong-James D, et al. Confronting and mitigating the risk of COVID-19 associated pulmonary aspergillosis (CAPA). Eur Resp J 2020; 56: 2002554.

29 Yan X, et al. Clinical characteristics of 45 patients with invasive pulmonary aspergillosis: Retrospective analysis of 1711 lung cancer cases. Cancer 2009; 115(21): 5018–5025.

30 Hung ML, et al. Invasive aspergillosis in patients with systemic lupus erythematosus: a retrospective study on clinical characteristics and risk factors for mortality. Lupus 2018; 27(12): 1944–1952.

31 Wang LR, et al. Invasive fungal disease in systemic lupus erythematosus: a systematic review of disease characteristics, risk factors, and prognosis. Semin Arthritis Rheum 2014; 44(3): 325–330.

32 Chen J, et al. Risk factors for invasive pulmonary aspergillosis and hospital mortality in acute-on-chronic liver failure patients: a retrospective-cohort study. Int J Med Sci 2013; 10(12): 1625–1631.

33 Bae S, et al. Invasive pulmonary aspergillosis in patients with severe fever with thrombocytopenia syndrome. Clin Infect Dis 2020; 70(7): 1491–1494.

34 Antinori S, et al. Trends in the post mortem diagnosis of opportunistic invasive fungal infections in patients with AIDS: a retrospective study of 1,630 autopsies performed between 1984 and 2002. Am J Clin Pathol 2009; 132(2): 221–227.

35 Denis B, et al. Relevance of EORTC criteria for the diagnosis of invasive aspergillosis in HIV-infected patients, and survival trends over a 20-year period in France. Clin Infect Dis 2015; 61: 1273–1280.

36 Horvath JA, Dummer S. The use of respiratory-tract cultures in the diagnosis of invasive pulmonary aspergillosis. Am J Med 1996; 100: 171–178.

37 Tarrand JJ, et al. Diagnosis of invasive septate mold infections. A correlation of microbiological culture and histologic or cytologic examination. Am J Clin Pathol 2003; 119: 854–858.

38 Meersseman W, et al. Galactomannan in bronchoalveolar lavage fluid: a tool for diagnosing aspergillosis in intensive care unit patients. Am J Respir Crit Care Med 2008; 177: 27–34.

39 Vergidis P, et al. High-volume culture and quantitative real-time PCR for the detection of *Aspergillus* in sputum. Clin Microbiol Infect 2020; 26: 935–940.

40 Fraczek MG, et al. Volume dependency for culture of fungi from respiratory secretions and increased sensitivity of *Aspergillus* quantitative PCR. Mycoses 2014; 57: 69–78.

41 Chakrabarti A, et al. Eight-year study of allergic broncho-pulmonary aspergillosis in an Indian teaching hospital. Mycoses 2002; 45: 295–299.

42 Wrench C, et al. Reduced clearance of fungal spores by chronic obstructive pulmonary disease GM-CSF- and M-CSF-derived macrophages. Am J Respir Cell Mol Biol 2018; 58(2): 271–273.

43 Greub G, Bille J. *Aspergillus* species isolated from clinical specimens: suggested clinical and microbiological criteria to determine significance. Clin Microbiol Infect 1998; 4(12): 710–716.

44 Khot PD, et al. Development and optimization of quantitative PCR for the diagnosis of invasive aspergillosis with bronchoalveolar lavage fluid. BMC Infect Dis 2008; 8: 73.

45 Duthie R, Denning DW. *Aspergillus* fungemia. Two cases and review. Clin Infect Dis 1995; 20: 598–605.

46 Lass-Flörl C. How to make a fast diagnosis in invasive aspergillosis. Med Mycol 2019; 57(Suppl): S155–S160.

47 Schelenz S, et al. British Society for Medical Mycology: diagnostic standards for the management of serious fungal diseases. Lancet Infect Dis 2015; 15: 461–474.

48 Izumikawa K, et al. Bronchoalveolar lavage galactomannan for the diagnosis of chronic pulmonary aspergillosis. Med Mycol 2012; 50(8): 811–817.

49 Fayemiwo S, et al. Comparative performance of Aspergillus galactomannan ELISA and PCR in sputum from patients with ABPA and CPA. J Microbiol Method 2017; 140: 32–39.

50 Jenks JD, et al. The *Aspergillus* lateral flow assay for the diagnosis of invasive aspergillosis: an update. Curr Fungal Infect Rep 2020 Dec 4: 1-6. doi: 10.1007/s12281-020-00409-z. Online ahead of print.

51 Bouza Santiago E, et al. Comparison of the performance of two galactomannan detection tests: platelia galactomannan (Bio-Rad) and galactomannan virclia (Vircell) European Conference on Clinical Microbiology and Infectious Diseases, Paris, France, 1 July 2020. https://www.aspergillus.org.uk/conference_abstracts/comparison-of-the-performance-of-two-galactomannan-detection-tests-platelia-galactomannan-bio-rad-and-galactomannan-virclia-vircell/ Accessed January 2021.

52 Denning DW, et al. Chronic pulmonary aspergillosis – rationale and clinical guidelines for diagnosis and management. Eur Resp J 2016; 47: 45–68.

53 Ma X, et al. Prospective study of the serum *Aspergillus*-specific IgG, IgA and IgM assays for chronic pulmonary aspergillosis diagnosis. BMC Infect Dis 2019; 19: 694.

54 Stucky Hunter ES, Richardson MD, Denning MD. Evaluation of LD Bio *Aspergillus* ICT lateral flow assay for IgG and IgM antibody detection in chronic pulmonary aspergillosis. J Clin Microbiol 2019; 57: e00538-19.

55 Baxter CG, et al. Performance of two *Aspergillus* IgG EIA assays compared to the precipitation technique in chronic and allergic aspergillosis. Clin Microbiol Infect 2013; 19: e197–204.

56 Stucky Hunter E, et al. Effect of patient immunodeficiencies on the diagnostic performance of serological assays to detect *Aspergillus*-specific antibodies in chronic pulmonary aspergillosis. Resp Med 2021 [In Press].

57 Morton CO, et al. Determining the analytical specificity of PCR-based assays for the diagnosis of IA: what is aspergillus? Med Mycol 2017; 55(4): 402–413.

58 Egger M, et al. Blood *Aspergillus* PCR: the good, the bad, and the ugly. J Fungi (Basel) 2020; 6(1): 18.

59 Harrison E, et al. Aspergillus DNA contamination in blood collection tubes. Diagn Microbiol Infect 2010; 67: 392–394.

60 Langridge PJ, Sheehan RL, Denning DW. Microbial yield from physiotherapy assisted sputum production in respiratory outpatients. BMC Pulm Med 2016; 16: 23.

61 Yazıcıoğlu Moçin O, et al. Bronchoscopy as an indicator of tracheobronchial fungal infection in non-neutropenic intensive-care unit patients. Clin Microbiol Infect 2013; 19(3): E136–141.

62 Peghin M, et al. Unusual forms of subacute invasive pulmonary aspergillosis in patients with solid tumors. J Infect 2014; 69(4): 387–395.

63 Henzler C, et al. Diagnostic performance of contrast enhanced pulmonary computed tomography angiography for the detection of angioinvasive pulmonary aspergillosis in immunocompromised patients. Sci Rep 2017; 7(1): 4483.

64 Muldoon EG, et al. Aspergillus nodules; another presentation of chronic pulmonary aspergillosis. BMC Pulm Med 2016; 16: 123.

65 Lowes D, et al. Predictors of mortality in chronic pulmonary aspergillosis. Eur Resp J 2017; 49: 1601062.

66 Roberts CM, Citron KM, Strickland B. Intrathoracic aspergilloma: role of CT in diagnosis and treatment. Radiology 1987; 165(1): 123–128.

67 Kosmidis C, et al. Chronic fibrosing pulmonary aspergillosis: a cause of "destroyed lung" syndrome. Infect Dis (Lond) 2017; 49: 296–301.

68 Mou Y, et al. A retrospective study of patients with a delayed diagnosis of allergic bronchopulmonary aspergillosis/allergic bronchopulmonary mycosis. Allergy Asthma Proc 2014; 35(2): e21–26.

69 Delfino E, et al. Respiratory fungal diseases in adult patients with cystic fibrosis. Clin Med Insights Circ Respir Pulm Med 2019; 13: 1179548419849939.

70 Gamaletsou MN, et al. F508del CFTR gene mutation in patients with allergic bronchopulmonary aspergillosis. J Asthma 2018; 55: 837–843.

71 Asano K, et al. New clinical diagnostic criteria for allergic bronchopulmonary aspergillosis/mycosis and its validation. J Allergy Clin Immunol 2021; 147(4): 1261–1268.e5.

72 Agarwal R, et al. Pictorial essay: Allergic bronchopulmonary aspergillosis. Indian J Radiol Imaging. 2011; 21(4): 242–252.

73 Sehgal IS, et al. Sensitization to *A. fumigatus* in subjects with non-cystic fibrosis bronchiectasis. Mycoses 2021; 64(4): 412–419.

74 Bowyer P, Denning DW. Genomic analysis of allergen genes in *Aspergillus* spp: the relevance of genomics to everyday research. Med Mycol 2007; 45: 17–26.

75 Glaser AG, et al. Auto- and cross-reactivity to thioredoxin allergens in allergic bronchopulmonary aspergillosis. Allergy 2008; 63(12): 1617–1623.

76 Denning DW, et al. The link between fungi and asthma – a summary of the evidence. Eur Resp Journal 2006; 27: 615–626.

77 Denning DW, et al. Pulmonary aspergillosis in AIDS. N Engl J Med 1991; 324: 654–662.

78 Gago S, Denning DW, Bowyer P. Pathophysiological aspects of *Aspergillus* colonization in disease. Med Mycol 2019; 57 (Suppl 2): S219–S227.

79 Sterclova M, Vasakova M, Metlicka M. Significance of specific IgG against sensitizing antigens in extrinsic allergic alveolitis: serological methods in EAA. Rev Port Pneumol 2011; 17(6): 253–259.

80 Oladele RO, et al. Chronic pulmonary aspergillosis as a cause of smear-negative TB and/or TB treatment failure in Nigerians. Int J Tuberc Lung Dis 2017; 21: 1056–1061.

81 Setianingrum F, et al. Evaluation and comparison of automated and manual ELISA for diagnosis of chronic pulmonary aspergillosis (CPA) in Indonesia. Diagn Microbiol Infect Dis 2020; 98: 115124.

82 Kwizera R, et al. Elevated *Aspergillus*-specific antibody levels among HIV infected Ugandans with pulmonary tuberculosis. BMC Pulm Med 2017; 17: 149.

83 Page ID, et al. Chronic pulmonary aspergillosis commonly complicates treated pulmonary tuberculosis with residual cavitation. Eur Resp J 2019; 53: 1801184.

84 Denning DW, et al. Case definition of chronic pulmonary aspergillosis in resource-constrained settings. Emerg Infect Dis 2018; 24(8):e171312.

85 Guinea J, et al. Pulmonary aspergillosis in patients with chronic obstructive pulmonary disease: incidence, risk factors, and outcome. Clin Microbiol Infect 2010; 16(7): 870–877.

86 Xu H, et al. Invasive pulmonary aspergillosis in patients with chronic obstructive pulmonary disease: a case control study from China. Clin Microbiol Infect 2012; 18(4): 403–408.

87 Ward NS, Casserly B, Ayala A. The compensatory anti-inflammatory response syndrome (CARS) in critically ill patients. Clin Chest Med 2008; 29(4): 617–625, viii.

88 Dabas Y, Mohan A, Xess I. Serum galactomannan antigen as a prognostic and diagnostic marker for invasive aspergillosis in heterogeneous medicine ICU patient population. PLoS One 2018; 13(4): e0196196.

89 White PL, Parr C, Barnes RA. Predicting invasive aspergillosis in hematology patients by combining clinical and genetic risk factors with early diagnostic biomarkers. J Clin Microbiol 2017; 56(1): e01122-17.

── **R É S U M É**

Des innovations significatives au cours de la dernière décennie ont abouti à un diagnostic plus sensible et plus rapide de l'aspergillose pulmonaire, allergique, chronique et invasive ainsi que de la bronchite et des nodules à *Aspergillus.* La nouvelle cartographie des diagnostics a révélé que l'incidence et la prévalence de l'aspergillose étaient plus élevées qu'on ne le pensait auparavant. Le traitement antifongique oral et intraveineux offre des taux de réponse clinique élevés aux patients affectés. L'absence de diagnostic laisse les patients surtraités par des agents antibactériens, des corticoïdes et des anti-tuberculeux aboutissant à une poursuite de la maladie et souvent au décès. L'introduction de plusieurs tests de diagnostic très performants contribue à redéfinir la prise en charge du patient. L'antigène d'*Aspergillus* peut être détecté sur 70–95% des échantillons de bronchoscopie chez les patients ayant une aspergillose invasive et chronique en moins d'une heure. La recherche d'immunoglobuline G (IgG) *Aspergillus* (précipitines) a une sensibilité de >90% et une spécificité de 98% pour l'aspergillose chronique et allergique. Une culture fongique respiratoire de grand volume et la réaction de polymérisation en chaîne à *Aspergillus* ont une sensibilité 3–5 fois plus élevée que la culture bactérienne de routine. *Aspergillus* IgE (ou le test cutané) diagnostique la sensibilisation à *Aspergillus* dans l'asthme, la mucoviscidose, les bronchopneumopathie chronique obstructive et après traitement de la TB et il est bien corrélé à une altération de la fonction pulmonaire et/ou à des exacerbations. Dans le monde, les cliniciens et les techniciens de laboratoire doivent intégrer ces excellents nouveaux outils pour améliorer les résultats cliniques en rendant les résultats d'une manière plus rapide et exacte.