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Consensus guidelines for the use of empiric and diagnostic-driven antifungal treatment strategies in haematological malignancy, 2014

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Key words
empiric antifungal therapy, diagnostic-driven antifungal treatment, haematological malignancy, invasive fungal disease, persistent or recurrent fevers of unknown origin.

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Abstract

Invasive fungal disease (IFD) causes significant morbidity and mortality in patients undergoing allogeneic haemopoietic stem cell transplantation or chemotherapy for haematological malignancy. Much of these adverse outcomes are due to the limited ability of traditional diagnostic tests (i.e. culture and histology) to make an early and accurate diagnosis. As persistent or recurrent fevers of unknown origin (PFUO) in neutropenic patients despite broad-spectrum antibiotics have been associated with the development of IFD, most centres have traditionally administered empiric antifungal therapy (EAFT) to patients with PFUO. However, use of an EAFT strategy has not been shown to have an overall survival benefit and is associated with excessive antifungal therapy use. As a result, the focus has shifted to developing more sensitive and specific diagnostic tests for early and more targeted antifungal treatment. These tests, including the galactomannan enzyme-linked immunosorbent assay and *Aspergillus* polymerase chain reaction (PCR), have enabled the development of diagnostic-driven antifungal treatment (DDAT) strategies, which have been shown to be safe and feasible, reducing antifungal usage. In addition, the development of effective antifungal prophylactic strategies has changed the landscape in terms of the incidence and types of IFD that clinicians have encountered. In this review, we examine the current role of EAFT and provide up-to-date data on the newer diagnostic tests and algorithms available for use in EAFT and DDAT strategies, within the context of patient risk and type of antifungal prophylaxis used.

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Introduction

Invasive fungal disease (IFD) is associated with significant mortality in patients undergoing haemopoietic stem cell transplantation (HSCT) or chemotherapy for haematological malignancies.[1–3] The high mortality rates (29–90%)[1–3] are not only related directly to the IFD itself but are also related indirectly to the negative impact IFD has in delaying subsequent conditioning or chemotherapy courses.[4] Delayed diagnosis, due to the poor sensitivity and specificity of traditional diagnostic tools (i.e. culture and histology), is a significant contributing factor to these high mortality rates.[5]

Until recently, the most common strategy used to manage suspected IFD in the setting of persistent or recurrent fevers of unknown origin despite broad-spectrum antibiotics was empiric antifungal therapy (EAFT).[6,7] However, the development of effective prophylactic and diagnostic-driven antifungal treatment (DDAT) strategies has changed the management paradigm. Here, we review the current roles of, and provide up-to-date algorithms for, EAFT and DDAT strategies. We also discuss the newer diagnostic tests (with increased sensitivity and specificity) that underpin the various DDAT strategies. We will focus on invasive aspergillosis (IA), as it is most commonly encountered in haematology populations and has generated the most evidence-based data.

Methodology

Questions asked

In preparing this update, we aimed to address the following questions:

1 What is the current role of EAFT in the management of febrile neutropenia?

2 Which diagnostic tools should clinicians use when employing a DDAT strategy?

3 How do data from studies of DDAT strategies help inform the development of new evidence-based empiric and DDAT algorithms?

Search strategy

We searched PubMed for publications between 1 June 2007 and 1 February 2014 (i.e. since the publication of the last guidelines) using the terms (singly or in combination) 'galactomannan', 'fungal PCR', '*Aspergillus* PCR', 'beta-D-glucan', 'high resolution computed tomography', '*Aspergillus* lateral flow', 'positron emission tomography', 'empiric antifungal therapy', 'pre-emptive antifungal therapy', 'allogeneic', 'invasive fungal disease', 'invasive aspergillosis', 'acute leukaemia', 'haematological malignancy', 'validity', 'sensitivity', 'specificity', 'bio-marker', 'prophylaxis', 'neutropenia' and 'treatment'. Each sourced article was read, as well as those contained in the bibliography of the sourced articles.

Use of EAFT – then and now

In the 1980s, EAFT became the standard of care based on the results of two randomised controlled trials (RCT), which showed that the addition of conventional amphotericin B deoxycholate (C-AmB) in patients with persistent fevers despite broad-spectrum antibiotics resulted in a non-significant decrease in IFD-related mortality.[6,7] The lack of overall survival benefit was initially attributed to the toxicity profile of C-AmB. However, after multiple comparisons with newer, less toxic and potentially more efficacious agents (Table 1),[8–13] the lack of overall survival benefit observed with EAFT is now thought to be due to the use of an insensitive and non-specific trigger for commencing EAFT, namely fever.

In the early 2000s, several RCTs comparing different antifungal agents for EAFT were published. However, since the publication of the 2008 guidelines, only one new comparative EAFT trial (performed in the paediatric population) appears in the literature (Table 1).[14] Though important for the management of the paediatric population, the study, overall, adds little new data regarding the utility of EAFT.

With the increasing use of effective voriconazole, posaconazole or liposomal amphotericin B prophylaxis, the current incidence of IFD has decreased,[15–19] yet EAFT is still widely used. This indicates that a substantial proportion of patients receive EAFT unnecessarily, resulting in significant unnecessary toxicity and costs. Studies have also demonstrated that an EAFT strategy still misses cases of IFD, particularly in afebrile and/or non-neutropenic patients.[20,21]

EAFT is still recommended by international guidelines as a standard of care, but now includes the caveat that it should be used in parallel with a diagnostic work-up (Fig. 1).[23–25] However, with the advent of effective DDAT strategies, discussion has shifted away from 'which agent is optimal for use as EAFT?' and towards whether EAFT should now be used at all. Given that the newer diagnostic tests are not readily available at all centres in Australasia, EAFT continues to have a role, but its use should now be restricted to particular clinical settings (see Table 3 and Fig. 1) (level II evidence; grade B recommendation).

The role of DDAT

Given that use of an EAFT strategy has never realised an overall survival advantage and results in excessive antifungal therapy use, research has focused on developing alternative strategies that incorporate newer diagnostic

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Table 1 Randomised trials comparing antifungal agents as empiric antifungal therapy (EAFT)

References	Study drugs	Primary endpoint	Sample size	Efficacy outcome	Safety outcome	Level of evidence
Walsh *et al*.[8]	AmB-D vs LAB	Efficacy: Composite of: survival for 7 days after starting study drug; resolution of fever during neutropenia; successful treatment of any baseline IFD; absence of breakthrough IFD during study-drug treatment or within 7 days of cessation and absence of premature discontinuation of study drug because of toxicity or lack of efficacy	702	Composite: Equivalence (49% vs 50%) Individual components: Significantly less breakthrough IFD with LAB (3.2% vs 7.8%; $P = 0.009$)	Favours LAB: Significantly fewer infusion-related fevers (17% vs 44%; $P < 0.001$) and nephrotoxicity (19% vs 34%; $P < 0.001$)	II
Winston *et al*.[9]	AmB-D vs Flu	Efficacy: Composite of: no fevers at EOT; absence of breakthrough IFD during therapy; no additional AFT required; no termination of study drug secondary to toxicity and alive at EOT	317	Composite: Equivalence (67% vs 68%)	Favours Flu: Significantly fewer adverse events (13% vs 81%; $P = 0.001$)	II
Wingard *et al*.[10]	LAB vs ABLC	Safety: Incidence of chills/rigors during day 1 of study drug	244		Favours LAB: Significantly fewer infusion-related chills/rigors (19% and 24% vs 80%; $P < 0.001$) and nephrotoxicity (14% and 15% vs42%; $P < 0.01$)	II
Boogaerts *et al*.[11]	AmB-D vs Itra	Efficacy: Resolution of fevers and neutropenia	384	Equivalence (38% vs 47%)	Favours Itra: Significantly fewer adverse events (5% vs 54%; $P = 0.001$) Significantly less discontinuation due to toxicity (19% vs 38%; $P = 0.001$)	II
Walsh *et al*.[12]	LAB vs Vori	Efficacy: Composite of: survival for 7 days post EOT; absence of breakthrough IFD; absence of premature discontinuation of study drug; resolution of fever during neutropenia and successful treatment of any baseline IFD	849	Composite: Favours LAB (31% vs 26%; 95% CI: –10.6 to 1.6) Individual components: Significantly less breakthrough IFD with Vori (1.9% vs 5%; $P = 0.02$)	Variable: Dependent on adverse event examined	II
Walsh *et al*.[13]	LAB vs CAS	Efficacy: Composite of: successful treatment of any baseline IFD; absence of breakthrough IFD during study-drug treatment or within 7 days of EOT; survival for 7 days post EOT; resolution of fever during neutropenia and absence of premature discontinuation of study drug because of toxicity or lack of efficacy	1123	Composite: Equivalence (34% vs 34%; 95% CI –5.6–6.0) Individual components: Significantly greater successful treatment of baseline IFD with CAS (51.9% vs 25.9%; $P = 0.04$)	Favours CAS: Significantly less discontinuation due to toxicity (10% vs 15%; $P = 0.03$) and nephrotoxicity (3% vs 12%; $P < 0.001$)	II
Maertens *et al*.[14]	LAB vs CAS	Efficacy: Composite of successful treatment of any baseline IFD (based on Adjudication Committee assessment); absence of breakthrough IFD during study-drug treatment or within 7 days of EOT (based on Adjudication Committee assessment); survival for 7 days post EOT; resolution of fever (defined as a temperature below 38°C for at least 48 hours) during neutropenia and absence of premature discontinuation of study drug because of toxicity or lack of efficacy	82 paediatric patients only	Composite: Equivalence: 32% (95% CI 13.7–50.3) vs 46% (95% CI 33.4–59.5)	Similar: Clinical: 46% (95% CI 26.6–66.6) vs 48% (95% CI 34.7–62.0). Laboratory: 19% (95% CI 6.6–39.4) vs 11 (95% CI 4.0–21.9)	II

AmB-D, amphotericin B deoxycholate or conventional amphotericin B; LAB, liposomal amphotericin B; IFD, invasive fungal disease; Flu, fluconazole; EOT, end of therapy; AFT, antifungal therapy; ABLC, amphotericin B lipid complex; Itra, itraconazole; Vori, voriconazole; CAS, caspofungin.

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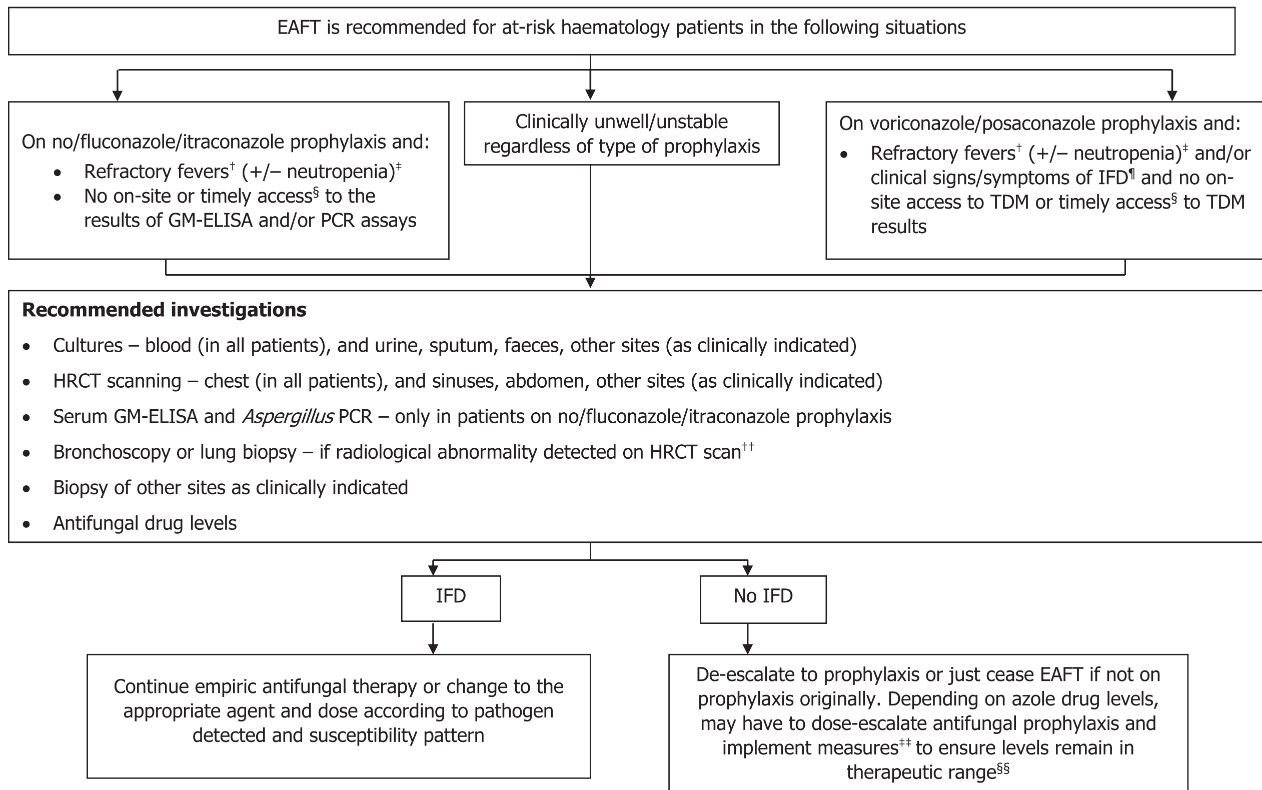

Figure 1 Empiric antifungal therapy strategy (EAFT). IFD, invasive fungal disease; GM-ELISA, galactomannan enzyme-linked immunosorbent assay; DDAT, diagnostic antifungal treatment; TDM, therapeutic drug monitoring; HRCT, high-resolution computed tomography scan. †Refractory fevers – persistent (daily for 3–5 days) or recurrent (after an afebrile period of 48 hours) fevers despite broad-spectrum antibiotics and negative microbiological investigations; ‡Neutropenia = neutrophil count $<0.5 \times 10^9$/L; §Timely access to results – results available consistently on-site or from an off-site laboratory within 3–5 days of sampling; ¶Clinical symptoms/signs of IFD – cough, chest pain, haemoptysis, dyspnoea, pleural effusion or rub, rhinorrhoea, epistaxis, ulceration or eschar of nasal septum or hard palate, maxillary pain, periorbital swelling, focal neurological signs or symptoms, skin lesions consistent with fungal infection (e.g. nodules, ulceration, satellitism); ††The procedure of choice is dependent on-site and type of lesion, type of antifungal prophylaxis, patient's clinical status, local experience with each test and patient's capacity to tolerate any complications[22] (see Table 2 for indications for bronchoscopy as first-line investigation vis-à-vis lung biopsy). All bronchoscopies and biopsies should be performed within 72 hours of EAFT commencement for maximum yield. BAL should be sent for microscopy, culture, GM-ELISA, fungal PCR testing (*Aspergillus* or panfungal depending on degree of suspicion of a non-*Aspergillus* mould) and cytology. Biopsy tissue (lung and other sites) should be sent for all the same tests with the addition of histology but with the exception of GM-ELISA. Ensure that the cardiothoracic surgeons and/or radiologists take a fresh specimen in addition to a specimen in formalin to maximise microbiological diagnosis;‡‡Sub-therapeutic itraconazole levels – ensure drug is taken as oral solution on empty stomach (1 hour before or 1 hour after meals); voriconazole levels – ensure drug is taken on empty stomach (1 hour before or 1 hour after meals), add omeprazole; sub-therapeutic posaconazole levels – ensure drug is taken with a high-fat (meal containing >20 g of dietary fat), and/or commercially available nutritional supplement (180–240 mL), ascorbic acid (500 mg), acidic drink (120–180 mL of coke, ginger ale, orange juice); §§Target therapeutic range for: itraconazole prophylaxis = 0.5 to 2 mg/L; voriconazole prophylaxis = >1.0 to <5.5 mg/L; posaconazole prophylaxis >0.7 mg/L with no upper limit.

tests to enable earlier, and more targeted, antifungal therapy. These have been variably called 'pre-emptive', 'bio-marker driven' or 'diagnostic-driven' strategies. We recommend and use the term 'diagnostic-driven antifungal treatment' (DDAT) (level III evidence; grade C recommendation) because the terms 'pre-emptive' and 'bio-marker driven' are not wholly precise. A 'pre-emptive' strategy for IFD is not pathogenically analogous to the 'pre-emptive' strategy used to detect cytomegalovirus viraemia and end-organ disease,[26] and a 'bio-marker' strategy excludes chest HRCT scans that are

Table 2 Indications for performing a bronchoscopy or lung biopsy as first-line investigation of an abnormal high-resolution computed tomography scan (HRCT)

Bronchoscopy as first-line (vs lung biopsy)
Centrally located lesion
Diffuse infiltrates
Lung biopsy† as first-line (vs bronchoscopy)
Peripherally located lesion
Focal/nodular lesion

†Open lung biopsy (either thoracotomy or video-assisted) is the preferred diagnostic procedure, as its yield is greater due to the ability to get a larger volume of tissue. As a result, we recommend an open-lung biopsy over a CT-guided biopsy as the preferred investigation.[22]

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Table 3 Summary of recommendations for use of EAFT, diagnostic tests for IFD and DDAT strategies

Recommendation	Level of evidence/ Grade of recommendation
EAFT should only be used in the following settings: • Refractory fevers (+/– neutropenia) and no access or timely access to GM-ELISA and/or fungal PCR in those on no, fluconazole or itraconazole prophylaxis • Clinically unwell patients, regardless of type of antifungal prophylaxis • Refractory fevers (+/– neutropenia) and no access or timely access to TDM in those on voriconazole or posaconazole prophylaxis	II/B
Strategies that use GM-ELISA +/– PCR +/– HRCT scans should be labelled as diagnostic-driven antifungal treatment strategies	III/C
Allow a minimum of 2 weeks between initial and follow-up HRCT scans of chest	III-2/C
Non-specific signs on HRCT scan in combination with positive GM-ELISA and/or PCR results are consistent with diagnosis of IA	III/C
An HRCT scan should not be used as the only diagnostic test in a DDAT strategy	III/C
A positive GM-ELISA result should be interpreted with caution in very young children	III/C
We recommend twice-weekly testing with GM-ELISA in high-risk paediatric haematology populations	II/C
We do not recommend that body fluids other than serum or BAL be routinely screened using GM-ELISA in all haematology populations	III-2 to IV/D
To optimise the ability to diagnose IA, we recommend that BAL fluid be tested by both GM-ELISA and PCR	III/C
Pleural fluid should not be tested by fungal PCR assays, given the poor sensitivity in this specimen	III-1/C
Panfungal PCR can be used as part of a clinically driven DDAT strategy	III/C
Fungal PCR assays should be used only in combination with other fungal diagnostic tests (e.g. GM-ELISA, HRCT scan)	II/B
Based on current evidence, we cannot recommend the use of BDG either alone or as part of a DDAT for the diagnosis of IA	III/C
Based on current evidence, we cannot recommend the use of PET/CT either alone or as part of a DDAT for the diagnosis of IA	III/C
Based on current evidence, we cannot recommend the use of *Aspergillus* LFA either alone or as part of a DDAT for the diagnosis of IA	III/C
We do not recommend the use of a surveillance-driven DDAT strategy in patients receiving voriconazole or posaconazole prophylaxis	II/B
DDAT strategies can be used in routine clinical practice to diagnose IFD	II/B
Based on current evidence, we cannot recommend a DDAT over EAFT in the paediatric population	III/C
We recommend the use of azole TDM as part of DDAT strategies	III/C

EAFT, empiric antifungal therapy; GM-ELISA, galactomannan enzyme-linked immunosorbent assay; TDM, therapeutic drug monitoring; HRCT, high-resolution computed tomography; IA, invasive aspergillosis; DDAT, diagnostic-driven antifungal therapy; AFT, antifungal therapy; BAL, bronchoalveolar lavage; BDG, β(1,3)-D-glucan; PET, positron emission tomography; LFA, lateral flow assay; IFD, invasive fungal disease.

known to be a critical tool for IFD diagnosis. In essence, what is most important is not the name but its components, the patient populations in which it is used and how it is applied (i.e. timing of implementation, duration of use).[21]

DDAT can be used either in a surveillance-driven or clinically driven setting. The surveillance-driven DDAT strategy uses laboratory markers of infection at regular intervals throughout the entire at-risk period for the early detection of IFD (Fig. 2).[23] A clinically driven strategy is implemented only in the presence of persistent/recurrent fevers or other clinical manifestations of IFD, and uses the traditional tools of culture and histology, together with newer, more sensitive tools (Fig. 3).[23] Types of antifungal prophylaxis and ease of access to newer diagnostic tools, including therapeutic drug monitoring (TDM), will ultimately determine the choice of strategy for any given centre.

Diagnostic tests used in studies evaluating DDAT strategies

Imaging tools

Chest HRCT scans are used in most Australasian centres for IFD diagnosis based on their high sensitivity (89%), ease of access and the fact that their systematic use is associated with improved outcomes.[27–30] HRCT scans are also used to monitor responses to antifungal therapy. However, lesions may grow in size and number, or develop cavitation before resolution,[31] likely related to immune reconstitution. This is mostly seen if a repeat scan is performed within 2 weeks.[24] Thus, we recommend that a follow-up HRCT scan be performed no sooner than 2 weeks after the initial scan (level III-2 evidence; grade C recommendation).

Non-specific signs on HRCT, together with positive results on *Aspergillus* galactomannan (GM) enzyme-linked immunosorbent assay (ELISA), may be consistent with probable IA,[32–34] and HRCT scan patterns can vary according to neutrophil count and prior corticosteroid therapy.[34] Thus, we recommend that non-specific signs (i.e. radiological features other than a dense, well-circumscribed lesion(s) (>1 cm) with or without a halo sign; air-crescent sign or cavity in an area of consolidation), in addition to positive GM-ELISA or *Aspergillus* PCR, is compatible with IA diagnosis and should trigger antifungal therapy (level III; grade C recommendation).

The advantages of HRCT scans include their non-invasive nature and accessibility in most centres. The disadvantages of HRCT scanning include its limited ability to differentiate between various fungi and other aetiologies.[35–37] HRCT scanning has been incorporated into DDAT strategies, evaluated (Table 4) and shown to reduce antifungal therapy use without adverse effects on survival. As HRCT scans provide no information on fungal aetiology, they should be used only in combination with other diagnostic tools in DDAT strategies (Figs 2 and 3) (level III; grade C recommendation).

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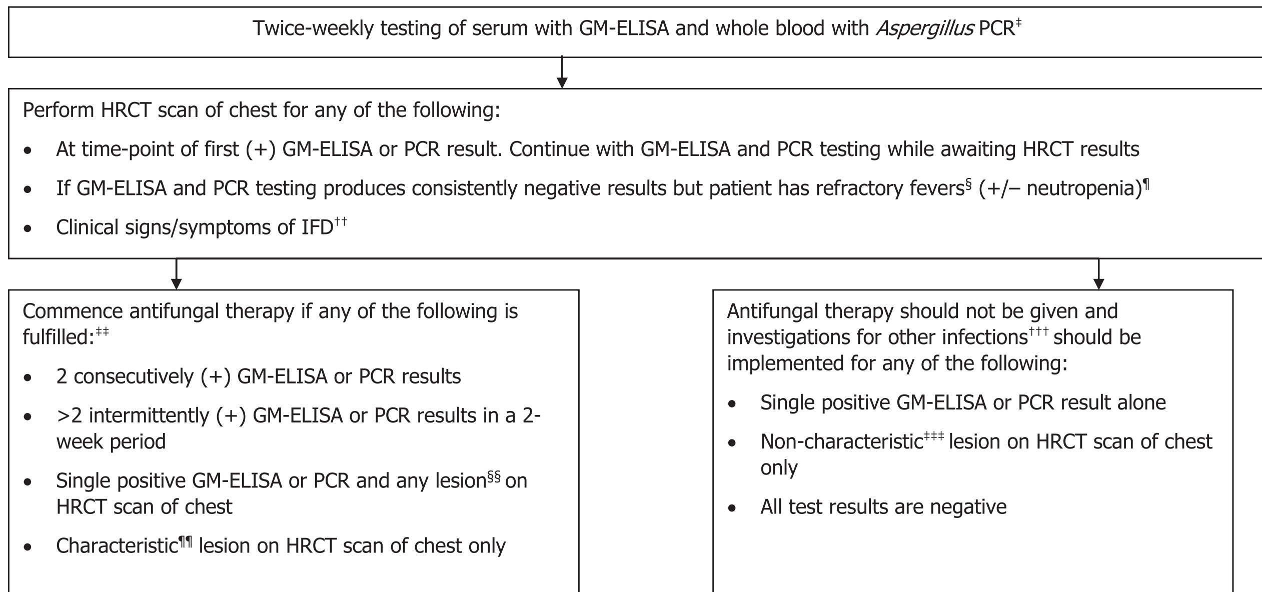

Figure 2 Surveillance-driven diagnostic antifungal treatment strategy[†]. GM-ELISA, galactomannan enzyme-linked immunosorbent assay; HRCT, high-resolution computed tomography scan; IFD, invasive fungal disease. [†]Use a surveillance-driven strategy only if GM-ELISA and PCR testing are consistently available on-site or from an off-site laboratory within 3–5 days of sampling; [‡]Testing begins at start of cycle 1 of chemotherapy and continues until no longer at high risk. If the patient enters another high-risk period, then the surveillance strategy is recommenced and continued for the entire duration of the subsequent high-risk period; [§]Refractory fevers – persistent (daily for 3–5 days) or recurrent (after an afebrile period of 48 hours) fevers despite broad-spectrum antibiotics and negative microbiological investigations; [¶]Neutropenia = neutrophil count $<0.5 \times 10^9$/L; [††]Clinical symptoms/signs of IFD – cough, chest pain, haemoptysis, dyspnoea, pleural effusion or rub, rhinorrhoea, epistaxis, ulceration or eschar of nasal septum or hard palate, maxillary pain, periorbital swelling, focal neurological signs or symptoms, skin lesions consistent with fungal infection (e.g. nodules, ulceration, satellitism); [‡‡]Irrespective of the presence or absence of refractory fevers; [§§]Any lesion includes characteristic and non-characteristic lesions; [¶¶]Characteristic lesions include dense, well-circumscribed lesion(s) (>1 cm) with or without a halo sign; air-crescent sign; cavity; [†††]As clinically indicated, perform the appropriate investigations to diagnose/exclude other infections, including bacterial (e.g. *Legionella*, *Mycobacterium*, *Clostridium difficile* (bronchoscopy specimens, faecal specimen, urine for Legionella urinary antigen etc.)), viral (e.g. cytomegalovirus, respiratory viruses (PCR testing of plasma or bronchoalveolar lavage fluid etc.)) or non-infectious causes (e.g. GVHD (colonoscopy with biopsies)) or appropriate biopsies if not responding to directed antifungal therapy; [‡‡‡]Non-characteristic lesions include any other radiological abnormality except for dense, well-circumscribed lesion(s) (>1 cm) with or without a halo sign; air-crescent sign; cavity (e.g. consolidation, patchy ground glass opacification).

Paediatric at-risk populations While CT scans in children carry a risk of causing cancer, the mortality risk from IA is higher. Therefore, the benefits of routine HRCT scanning outweigh the risks and should be performed for the early diagnosis of possible IA. Non-specific signs (e.g. diffuse opacities) are more commonly seen, particularly in those 0–5 years of age. Nodules are observed in only 59% of children with confirmed pulmonary aspergillosis but only in 39% of children 0–5 years of age. Halo and air-crescent signs are rarely seen in children.[48]

Aspergillus GM-ELISA assay

GM is a polysaccharide found in the cell wall of most *Aspergillus* species. It is released from growing hyphae. Its presence in blood or body fluids is indicative of active infection. A commercial kit by Bio-Rad (Marnes-la-Coquette, Paris, France) is available (Platelia *Aspergillus* GM EIA). The current recommended cut-off value in serum for a positive result is an optical density index (ODI) of ≥0.5.[49,50]

The GM-ELISA assay has been extensively examined for sensitivity and specificity in patients undergoing allogeneic HSCT or chemotherapy for acute leukaemia. The sensitivity in serum samples has varied between studies from 33% to 100%,[49] likely related to differences in study design. A meta-analysis has reported an overall sensitivity of 71% and a specificity of 89% in serum.[51] The positive predictive value is low at 26–53% but the assay has an excellent negative predictive value (NPV) at 95–98%, indicating that it is most useful as a screening tool to exclude IA. Studies have reported that GM can be detected a median of 5–8 days before culture positivity and that the ODI values over time correlate with treatment outcomes.[52–54] Consequently, GM-ELISA has been included as a microbiological criterion in the revised European Organization for Research and Treatment of Cancer (EORTC)/Mycoses Study Group (MSG) definitions.[33]

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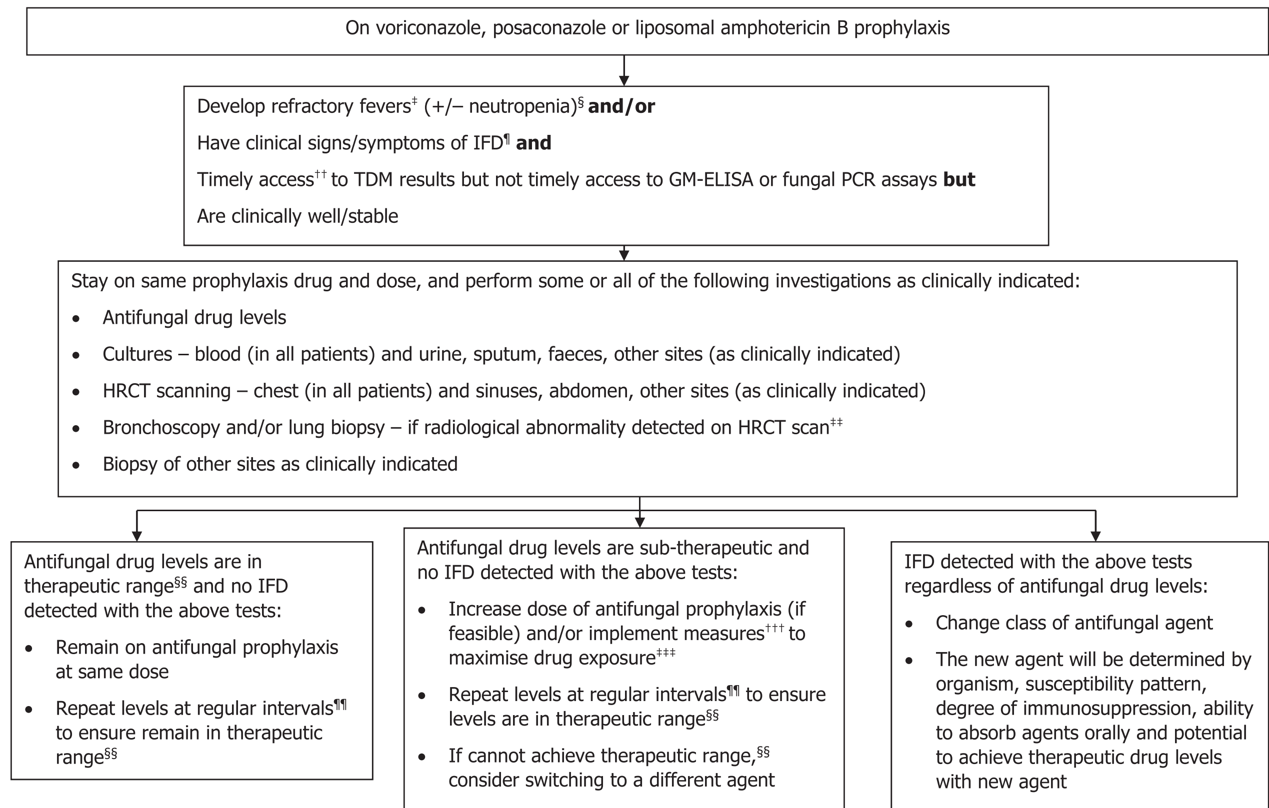

Figure 3 Clinically driven diagnostic antifungal treatment strategy†. IFD, invasive fungal disease; TDM, therapeutic drug monitoring; GM-ELISA, galactomannan enzyme-linked immunosorbent assay; HRCT, high-resolution computed tomography scan. †Use a clinically driven strategy if GM-ELISA or PCR testing are not consistently available on-site or from an off-site laboratory within 3–5 days of sampling; ‡Refractory fevers – persistent (daily for 3–5 days) or recurrent (after an afebrile period of 48 hours) fevers despite voriconazole, posaconazole or liposomal amphotericin B and negative microbiological investigations; §Neutropenia = neutrophil count $<0.5 \times 10^9$/L; ¶Clinical symptoms/signs of IFD – cough, chest pain, haemoptysis, dyspnoea, pleural effusion or rub, rhinorrhoea, epistaxis, ulceration or eschar of nasal septum or hard palate, maxillary pain, periorbital swelling, focal neurological signs or symptoms, skin lesions consistent with fungal infection (e.g. nodules, ulcerations, satellitism); ††Timely access to results – results available consistently on-site or from an off-site laboratory within 3–5days of sampling; ‡‡The procedure of choice is dependent on site (central vs peripheral) and type of lesion (nodular vs diffuse infiltrates), type of antifungal prophylaxis (voriconazole, posaconazole or liposomal amphotericin B vs not), patient's clinical status, local experience with each test and patient's capacity to tolerate any complications[22] (see Table 2 for indications for bronchoscopy as first-line investigation vis-à-vis lung biopsy). All bronchoscopies and biopsies should be performed within 72 hours of EAFT commencement for maximum yield. BAL should be sent for microscopy, culture, GM-ELISA, fungal PCR testing (*Aspergillus* or panfungal, depending on degree of suspicion of a non-*Aspergillus* mould) and cytology. Biopsy tissue (lung and other sites) should be sent for all the same tests with the addition of histology but with the exception of GM-ELISA. Ensure that the cardiothoracic surgeons and/or radiologists take a fresh specimen in addition to a specimen in formalin to maximise microbiological diagnosis; §§Target therapeutic range for voriconazole prophylaxis = >1.0 to <5.5 mg/L; posaconazole prophylaxis >0.7 mg/L with no upper limit; ¶¶Voriconazole – weekly until in therapeutic range and then monthly; posaconazole fortnightly until in therapeutic range and then only if initiate/cease/change in dose of an interacting agent; †††Sub-therapeutic voriconazole levels – ensure drug is taken on empty stomach (1 hour before or 1 hour after meals), add omeprazole; sub-therapeutic posaconazole levels – ensure drug is taken with a high-fat (meal containing >20 g of dietary fat), and/or commercially available nutritional supplement (180–240 mL), ascorbic acid (500 mg), acidic drink (120–180 mL of coke, ginger ale, orange juice); ‡‡‡Drug exposure = drug concentration in blood.

The GM-ELISA assay has recently been validated for use in bronchoalveolar lavage (BAL) fluid. Three meta-analyses/systematic reviews have examined the performance of BAL-GM in patients with haematological malignancies or mixed comorbidities and concluded that GM-ELISA was more sensitive in BAL relative to GM detection in serum.[55–57] This is likely related to the greater fungal burden within the respiratory tract during active IA.[58] The ability to detect GM in BAL appears to be less affected by voriconazole, posaconazole and liposomal amphotericin B prophylaxis as compared with GM detection in serum.[57] However, the optimal ODI cut-off for positivity in BAL samples requires further clarification.

Piperacillin/tazobactam therapy has previously been associated with false-positive GM results, related to the presence of GM in the product itself.[59,60] Recent

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Table 4 Summary of studies evaluating diagnostic-driven antifungal therapy (DDAT) strategies for invasive fungal disease

References	Study design	Population	Tests evaluated	Endpoints	Results	Limitations of study	Level of evidence
Maertens *et al.*[20]	Prospective cohort, single centre	Allogeneic HSCT recipients, AML, ALL and MDS, n = 88 patients/ 136 episodes of neutropenia	GM-ELISA (daily) and HRCT scan of thorax	Number of patients who received DDAT versus EAFT.	DDAT strategy: Less AFT versus EAFT strategy (7.7% 35%). Decreased time to initiation of AFT in 10%. 10 afebrile patients were GM-ELISA positive. One case of Zygomycetes and two cases of invasive candidiasis missed.	An indirect estimate of diagnostic value of GM-ELISA and HRCT scan as study not randomised.	III-2
Oshima *et al.*[38]	Retrospective (2001–2005), single centre, observational	Allogeneic HSCT; n = 114	GM-ELISA, BDG and CXR (weekly) +/– HRCT scan	Primary: Incidence of early IFD (from date of HSCT until 7 days post engraftment). Secondary: EAFT and DDAT in retrospective cohort.	IFD incidence 15%. EAFT in 13/73 (17%) with persistent febrile neutropenia versus 4/60 (6%) with PFUO and positive diagnostic tests.	Not randomised. Retrospective. No concurrent controls.	III-3
Dignan *et al.*[39]	Retrospective, single centre, observational	Allogeneic HSCT; n = 99	HRCT scan (3 days PFUO or if clinically indicated)	EAFT versus DDAT guided by HRCT findings.	53/99 (54%) with PFUO only. HRCT guided therapy in 17/99 (17%). Estimated reduction of 68% in use of EAFT.	Non-randomised. No strict HRCT criteria defined for DDAT.	III-3
Hebart *et al.*[40]	RCT, multicentre, 1998–2001	Allogeneic HSCT recipients; n = 403	Surveillance panfungal PCR assay (twice weekly to day 30 post HSCT; then weekly from day 30–90)	Primary: Incidence of IFD and IFD-related mortality in patients after allogeneic HSCT. DDAT arm: Antifungal therapy if single positive PCR on surveillance or 120 h fever and neutropenia (hybrid empiric/DDAT strategy). EAFT arm: EAFT if refractory febrile neutropenia after 120 hours	DDAT arm: Proven/probable IFD 14.3% versus 15.8%. Increased use of L-AMB (57.1% vs 36.7%; $P < 0.05$). Decreased mortality at day +30 (1.5% vs 6%; $P = 0.015$). No difference in mortality at day +100.	Short duration of follow-up. Poor compliance with PCR testing beyond day 30. Evaluated PCR only. Study underpowered.	II
Cordonnier *et al.*[41]	RCT, multicentre, unblinded, non-inferiority	Autologous HSCT recipients and haematological malignancy; n = 293	GM-ELISA (twice weekly), +/– radiological or clinical signs of suspected IFD	Primary: Percentage of patients alive 14 days after neutrophil recovery or 60 days from randomisation if severe complication.	DDAT arm: Survival non-inferior (95.1% vs 97.3%). No difference in IFD-related mortality (2.1% vs 0%). Significantly less use of antifungal agents (39.2% vs 61.3%; $P < 0.001$). Probable/proven fungal infections more common (9.1% vs 2.7%; $P < 0.02$). Significantly lower mean antifungal drug costs ($P < 0.001$). No significant differences in serious adverse events.	Evaluated GM-ELISA only. Short duration of screening. High-risk patients under-represented. Reduced sensitivity of GM-ELISA as high optical density index cut-off used (ODI ≥1.5).	II
Barnes *et al.*[42]	Prospective cohort, single centre (Oct 2005–Mar 2006)	Allogeneic HSCT recipients, acute leukaemia and chemo-refractory disease receiving intensive therapy	GM-ELISA and M-EIA; PCR for *Candida* and *Aspergillus* (twice weekly) during febrile neutropenia or with chronic GVHD	Impact of PCR and GM-ELISA on IFD diagnosis, antifungal usage and clinical outcomes. Used in an integrated care pathway.	8/125 (6%) subjects with proven/probable IFD. Estimated net savings from DDAT (reduction in antifungal drug costs minus cost of diagnostics) was £52, 839 over 6 months. Improved diagnostic accuracy when multiple PCR tests positive (Diagnostic OR = 30).	An indirect estimate of diagnostic value of GM-ELISA and PCR as study was not randomised. Impact of mould-active prophylaxis (itraconazole) on diagnostic test performance not able to be assessed as no comparator group.	III-2
Girmenia *et al.*[43]	Prospective (Mar 2006–Feb 2007)	Haematological malignancy, autologous HSCT, n = 146 patients	GM-ELISA (daily for 3 days) in selected patients (PFU0 >4 days' duration or clinical features suggestive of IFD)	Feasibility of DDAT strategy where GM-ELISA used for diagnosis rather than screening.	Empiric antifungal use reduced by 43%. GM-ELISA used in 40% of neutropenic episodes. GM positive in 24/27 with proven/probable disease.	Single centre. Not randomised.	III-3
Aguilar-Guisado *et al.*[44]	Prospective cohort (Nov 2002–Feb 2005)	Patients with febrile neutropenia following chemotherapy or post HSCT, n = 66 patients	Selected use of EAFT in patients with PFUO and additional clinical criteria, including severe sepsis, septic shock or with localised infection (lung, sinus, abdomen, skin or central nervous system) or individualised decision in high-risk patient	IFD incidence and mortality in patients with PFUO according to whether EAFT indicated on clinical criteria.	IFD incidence 9%. Rates of PFUO 66/347 (19%) of which 26/66 (39.4%) received EAFT. IFD incidence in EAFT = 11.5% versus 0% in non-treated arm.	Increased mortality in EAFT group may be explained by a higher proportion of high-risk patients (AML and allogeneic HSCT) in this group. Lack of availability of GM-ELISA in diagnostic work-up. Non-randomised.	III-3

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Table 4 Continued

References	Study design	Population	Tests evaluated	Endpoints	Results	Limitations of study	Level of evidence
Blennow *et al*.[45]	RCT, single centre	Reduced intensity allogeneic HSCT, n = 99 patients	Panfungal PCR (weekly surveillance to Day 100) then only on clinical suspicion of IA	Primary: Reduction in proven/probable IFD in PCR-guided antifungal treatment. Patients with PCR positive on screening randomised to antifungal treatment or no antifungal treatment. Secondary: Survival at 100 days. Survival at 1 year. I year incidence IFD. Risk factors for IFD.	DDAT arm: No significant difference in incidence of IFD in those randomised (0/8 vs 1/13). A single positive PCR on screening poorly predictive of IFD. Low sensitivity of PCR in diagnostic work-up of patients with clinical suspicion of IA (5/6 with proven/probable IA yielded negative PCR).	Only 21 of patients randomised. Low incidence of IFD in first 100 days, impacting predictive value of PCR.	III-3
Pagano *et al*.[46]	Prospective, multicentre	Haematological malignancies receiving chemotherapy, n = 397 patients	Laboratory (unspecified) or clinical signs of localising infection (no protocol)	Primary: Impact of empiric versus DDAT strategy on patient outcomes.	DDAT strategy: Increased incidence of IFD (23.7% vs 7.4%; $P < 0.05$). Increased IFD-related mortality (22.5% vs 7.1%). Increased overall mortality at 80 days (15.9% vs 6.3%). Younger age and empiric antibiotics independent predictors of reduced mortality on multivariable analysis.	Non-randomised. Administration of DDAT discretionary and not protocolised. Biomarkers not routinely incorporated into care pathways.	III-2
Tan *et al*.[47]	RCT, single centre	High-risk AML/MDS, Auto/allogeneic HSCT, n = 47 patients	GM-ELISA (twice-weekly), CT chest with positive marker or clinical suspicion	Primary: Proportion of patients receiving empiric antifungal therapy.	DDAT arm: Trend towards less empiric antifungal use (9/27 (33%) 11/25 (44%); $P = 0.57$). No significant difference in survival at 12 weeks.	Underpowered to detect difference in primary endpoint. Single-centre study wherein patients treated with itraconazole prophylaxis; therefore, results may not be generalisable to other settings.	II
Morrissey *et al*.[21]	RCT, multicentre (30 Sept 2005–19 Nov 2009)	Allogeneic HSCT, AML and ALL, n = 240	DDAT guided by GM-ELISA and *Aspergillus*-specific nested PCR assay (twice weekly) versus standard diagnostics	Primary: Proportion of patients who received at least one course of EAFT Secondary: All cause mortality. IA-related mortality. IFD- (not IA-) related mortality. Incidence proven/probable and possible IFD. Toxicity (hepatoxicity/nephrotoxicity). Number of courses of EAFT.	DDAT arm: Significant reduction in EAFT (15% vs 32%; $P = 0.002$). Significant increase in diagnosis of probable IA (16% vs 0%; $P < 0.0001$) and possible IA (5% vs 0%; $P = 0.013$). Significant reductions in EAFT use and improved IA diagnosis observed in subjects on first-generation azole (fluconazole or itraconazole) prophylaxis but not in patients on second-generation azole (voriconazole, posaconazole) prophylaxis. No significant difference in diagnosis of IFD other than IA. No significant difference in toxicity or mortality outcomes.	Small number of patients with acute leukaemia. Small but significant centre effect limiting the generalisability of study results. DDAT strategy directed at detection of IA and not other mould infections.	II

ALL, acute lymphoblastic leukaemia; L-AMB, liposomal amphotericin B; AML, acute myeloid leukaemia; EAFT, empirical antifungal therapy; GM-ELISA, galactomannan enzyme-linked immunosorbent assay; HRCT, high-resolution computed tomography; HSCT, haemopoietic stem cell transplantation; IA, invasive aspergillosis; IFD, invasive fungal disease; MDS, myelodysplastic syndrome; M-EIA, Mannan enzyme-linked immunosorbent assay; RCT, randomised controlled trial; DDAT, diagnostic-driven antifungal therapy; PCR, polymerase chain reaction; PFUO, persistent or recurrent fever of unknown origin; GVHD, graft versus host disease; OR, odds ratio.

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manufacturing processes have addressed this issue,[61] with no GM detected in any vials containing the newer formulation.[62] Generic formulations, however, may still contain GM.[63]

The performance of the GM-ELISA assay is not technically demanding. Costs can be reduced by batching samples. Results can be available in 4 h, although confirmation (repeat testing) of a positive result is recommended. Diagnostic microbiology laboratories that routinely perform serology tests are well positioned to incorporate the GM-ELISA into their test menu, pending adequate funding for human resources and consumables. Thus, GM-ELISA is likely to become increasingly available in Australasia.

Paediatric at-risk populations Higher rates of false-positive results are seen in children than in adults, particularly in those without significant risk factors for IA;[64] thus, caution is advised in interpreting positive GM results in very young children (level III evidence; grade C recommendation). Twice-weekly screening with confirmation testing of positive GM results may aid early detection of IA in high-risk patients but is not recommended in lower risk populations (level II evidence, grade C recommendation). Performance of GM in other body fluids remains predominantly anecdotal, usually performed in cases with a high pre-test probability. Recent paediatric data suggest that an acceptable correlation may exist between GM values in serum and urine.[65] Currently, routine screening of body fluids other than serum or BAL is not recommended (level III2–IV evidence, grade D recommendation).

Nucleic acid assays

PCR assays have the potential for rapid and early detection of IFD, with increased sensitivity and specificity. Critical to the sensitivity and specificity of fungal PCR assays are the specimen type, specimen processing and storage methods, DNA extraction methods and PCR assay design. A detailed discussion concerning PCR assays can be found in the review by Lau *et al*.[66]

Aspergillus PCR has been evaluated in a meta-analysis (level I evidence),[67] with sensitivity of 75% to 88% reported, depending on whether two positive PCR results or a single positive PCR result, respectively, were required to diagnose IA.[67] It was also reported that several assays had excellent NPV and were suitable for use in surveillance-driven DDAT strategies.[67] Subsequent studies evaluating surveillance-driven DDAT strategies using one of these recommended PCR assays have confirmed these findings.[21]

The greatest limitation to the widespread use of PCR for IFD diagnosis and its inclusion in the EORTC/MSG criteria is the lack of standardisation.[33] Consequently, the European *Aspergillus* PCR Initiative has developed standardised procedures for *Aspergillus* PCR, and a calibrator in collaboration with the *Aspergillus* Technology Consortium and Invasive Aspergillosis Animal Model group.[68–70] The aim of this work is to improve the accuracy of the assay and reduce interlaboratory variability. A commercial *Aspergillus* PCR assay (Myconostica, MycAssay *Aspergillus*, Manchester, UK) has been shown to have comparable sensitivity and specificity to a well-validated in-house assay.[71] The advantages of this assay are that it is standardised and that its components are quality controlled.

The performance of *Aspergillus* PCR has been examined in BAL specimens and, like the GM-ELISA assay, its sensitivity and specificity observed to be superior than in blood specimens. Two meta-analyses have evaluated the performance of *Aspergillus* PCR in BAL, and have reported sensitivities of between 79% and 91%, respectively, with specificities of between 92% and 94%, respectively.[72,73] Another meta-analysis examined GM in BAL either alone or in combination with *Aspergillus* PCR and determined that GM-ELISA was more sensitive than *Aspergillus* PCR, but both were equally specific.[57] Combining both GM-ELISA and *Aspergillus* PCR in BAL fluid to diagnose IA optimises sensitivity and specificity. Positive and negative results when both tests are applied to BAL are highly predictive of the presence or absence of IA, respectively.[74–76] We recommend that BAL fluid should be tested by both GM-ELISA and *Aspergillus* PCR to optimise the ability to detect IA (level III evidence; grade C recommendation). The use of two or more antifungal agents may reduce the performance of *Aspergillus* PCR in BAL fluid,[74] or, alternatively, repeatedly negative results may indicate successful clinical response.[21]

Recently, a study reported that the testing of pleural fluid using a well-validated *Aspergillus* PCR assay had reduced sensitivity compared with fresh tissue,[77] indicating that pleural fluid should not be used for PCR testing to aid the diagnosis of IA (level III-1 evidence; grade C recommendation). Another study evaluated the performance of the same *Aspergillus* PCR assay in cerebrospinal fluid (CSF) samples from immunocompromised patients with suspected central nervous system IA,[78] and demonstrated a sensitivity and specificity of 100% and 93% respectively.[78] PCR testing of CSF may be useful to confirm/exclude the diagnosis of IA in at-risk patients, particularly if operative procedures (e.g. obtaining a brain biopsy) cannot be safely performed.

With increasing use of voriconazole, posaconazole and liposomal amphotericin B antifungal prophylaxis and more intensive immunosuppressive therapies, non-*Aspergillus* mould species are emerging. In response, panfungal PCR assays have been developed. Most

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panfungal PCR studies have analysed tissue samples,[66,79,80] with one study performed on whole blood.[80] Overall, panfungal PCR was more sensitive than culture and histology, with positive PCR results from culture-negative samples.[66,79,80] Panfungal PCR, however, is disadvantaged by the need for DNA sequencing of any amplicon, with consequent increased turn-around times (TAT). Panfungal PCR assays appear useful as an adjunct to culture and histology, and are recommended for use in a clinically driven DDAT strategy (level III evidence; grade C recommendation).

Despite the technical difficulties associated with fungal PCR assays, they are not overtly problematic to implement if subject to appropriate care and methodological guidance. Conversely, their interpretation in the clinical setting requires significantly more expertise. Fungal PCR assays are already implemented in a number of laboratories in Australasia. Based on the above discussion, we recommend that PCR should be used only in combination with other tests, as part of a DDAT strategy (level II evidence; grade B recommendation).

β(1–3)-D-glucan (BDG) assay

The BDG is a component of the cell wall of many fungi, and it can be detected in serum by four commercially available tests. However, the assays have not been extensively evaluated in the haematology populations, particularly in BAL or as part of DDAT strategies.[38,81] It is also associated with high rates of false positivity (in both adults and children),[82,83] high expense (US $1279 for 43 specimens) and the requirement to test in duplicate. This assay has not been evaluated in the Australasian setting and no commercially available proficiency-testing panel is available for quality control purposes. No diagnostic laboratory in Australasia currently performs BDG testing. Given the above, we cannot recommend its use as a part of a DDAT strategy (level III evidence; grade C recommendation).

Potential for other tests to be used in DDAT strategies in the future

Positron emission tomography scans

18-fluoro-2-deoxy-D-glucose positron emission tomography is taken up by active cells with high glucose consumption, including neutrophils, lymphocytes and activated macrophages.[84] Hence, it accumulates at sites of infection/inflammation. Recently, positron emission tomography/computed tomography (PET/CT) was evaluated in comparison to conventional testing in haematology patients with persistent febrile neutropenia at high risk for IFD. PET/CT was more sensitive.[85–87] However, larger prospective studies need to be performed to better determine its precise role; currently, we cannot recommend the inclusion of PET/CT in DDAT strategies (level III evidence; grade C recommendation).

Aspergillus lateral flow assay (LFA)

This point-of-care immunochromatographic device has many advantages, including its ease of use, no need for expensive equipment or to be performed in a reference laboratory by trained staff, rapid TAT from sampling to result (<1 h) and low cost. The *Aspergillus* LFA has been evaluated in comparison with GM-ELISA and *Aspergillus* PCR. A retrospective case–control study found that *Aspergillus* LFA was not as sensitive as *Aspergillus* PCR (81.8% vs 95.5%) but more sensitive than GM-ELISA (81.8% vs 77%) when used in serum.[88] In tandem with PCR, sensitivity and specificity were 100%. Further evaluation in larger prospective studies is still required; currently, we cannot recommend the inclusion of the *Aspergillus* LFA in DDAT strategies (level III evidence; grade C recommendation).

Results of DDAT studies performed to date

A number of studies have been published since the last guidelines (Table 4), examining various DDAT strategies.[20,38,39,42–44] Collectively, the non-comparative studies indicate that a DDAT strategy can safely and effectively guide antifungal therapy use.

Subsequently, RCTs of DDAT strategies have been performed. Hebart *et al*. compared a traditional EAFT strategy with a hybrid PCR-/fever-directed strategy in 403 allogeneic HSCT recipients.[40] Significantly more antifungal therapy was administered to patients in the PCR-/fever-directed arm (57.1% vs 36.7%; $P < 0.001$),[40] likely due to the initiation of antifungal therapy for persistent fevers and/or high rates of PCR false-positivity. Thirty-day mortality rates were significantly better in the PCR-/fever-directed arm (1.5% vs 6.3%; $P = 0.015$), but this significant difference was not sustained to day 100 (6.3% vs 13.2%; $P = 0.106$),[40] likely due to less frequent PCR testing after day 30.

Cordonnier *et al*. performed a non-inferiority RCT comparing an EAFT strategy to a hybrid DDAT strategy in patients undergoing autologous HSCT or chemotherapy for haematological malignancies.[41] The primary outcome was survival to 14 days after recovery from neutropenia or to day 60 post-enrolment if patients had persistent neutropenia.[41] In the DDAT arm, patients were administered antifungal therapy based on a number of clinical/radiological manifestations or positive GM-ELISA (ODI ≥1.5) results detected after 4 days of persistent fevers.[41]

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DDAT was determined to be non-inferior for the primary outcome (95.1% vs 97.3%; 95% confidence interval for difference: −5.9% to 1.4%; $P = 0.31$) and was associated with a 35% reduction in antifungal therapy use despite significantly more IFD diagnoses (9.1% vs 2.7%: $P = 0.02$).[41] However, on subgroup analysis, the survival rates for those assigned to the DDAT strategy during induction chemotherapy indicated that inferiority could not be excluded.[41] Subsequently, the authors recommended EAFT as the preferred strategy for patients undergoing induction chemotherapy.[41] Notably, the potential inferiority of the DDAT strategy in those receiving induction chemotherapy may have been due to the 7-day delay in the administration of antifungal therapy,[41] indicating that the time-point at which screening with these assays is started is critically important. Moreover, the use of a high ODI cut-off (≥1.5) for GM-ELISA positivity may have reduced the effectiveness of the DDAT strategy used in this study.

Studies by Tan *et al*. and Blennow *et al*. are limited by premature termination and poor recruitment, respectively, hampering any further interpretation.[45,47]

Morrissey *et al*. compared a DDAT strategy, based solely on twice-weekly screening with *Aspergillus*-nested PCR and GM-ELISA, with the standard diagnostic strategy of culture and histology in patients undergoing allogeneic HSCT or remission-induction therapy for acute leukaemia. The primary outcome was the difference in the use of EAFT at 26 weeks of follow-up.[21] Significantly fewer patients in the DDAT strategy arm received EAFT (15% vs 32%; $P = 0.002$) although this arm exhibited a higher rate of probable IA (16% vs 0%; $P < 0.0001$).[21] No significant difference in mortality was observed between the two arms, although the trial was not powered for this endpoint.[21]

The trial by Morrissey *et al*. provided insight into how GM-ELISA and *Aspergillus* PCR are best used in the setting of different types of antifungal prophylaxis.[21] The significant reduction in EAFT use and improved IA diagnosis in the DDAT strategy arm was observed only in those on fluconazole or itraconazole and not in those on voriconazole or posaconazole prophylaxis.[21] Therefore, we do not recommend that a surveillance-driven DDAT strategy be used in patients taking voriconazole or posaconazole prophylaxis (level II evidence; grade B recommendation).

Considered collectively, these studies allow us to draw some conclusions regarding the use of DDAT strategies (see Table 5 for a summary). We conclude from the available data that surveillance-driven and clinically driven DDAT strategies can be recommended for use in clinical practice, but the choice of diagnostic tools and type of DDAT strategy should be adapted to local resources and type of antifungal prophylaxis used (level II evidence; grade B recommendation).

Table 5 Summary conclusions from studies evaluating diagnostic-driven antifungal therapy (DDAT) strategies

Reduced use and costs of antifungal therapy
Improved ability to diagnose IA
Enable an earlier diagnosis of IA compared with culture and histology
At a minimum, DDAT strategies have no adverse effect on survival and if intensive screening is performed (minimum of twice weekly), may improve survival
Optimal performance when used to screen high-risk patients on no, fluconazole or itraconazole prophylaxis

DDAT, diagnostic-driven antifungal therapy; IA, invasive aspergillosis.

Paediatric at-risk populations There is currently insufficient evidence to recommend a DDAT strategy over EAFT (level III; grade C recommendation).

Other considerations for the successful implementation of DDAT strategies

Use of TDM in the setting of suspected IFD

A detailed summary of the evidence for azole TDM has been provided in the optimising drug therapy guidelines by Chau *et al*. 2014, appearing elsewhere in this supplement. Chau and colleagues recommend that azole drug levels be measured if breakthrough infection is suspected. Patients on voriconazole or posaconazole prophylaxis who develop persistent fevers or other clinical manifestations of suspected breakthrough IFD should have TDM included in a clinically driven DDAT strategy (Fig. 3). Measurements of voriconazole/posaconazole levels are important, as the results may have utility in therapeutic decision making (Fig. 3). Thus, we recommend that azole TDM be used as part of any DDAT strategy (level II evidence; grade C recommendation).

Stakeholder involvement

It is critical to the success of a surveillance-driven DDAT strategy that GM-ELISA and PCR assays are available with twice-weekly testing (minimum) at a maximum TAT from sampling to results of 3–5 days. Timely access to HRCT scanning (i.e. within 48 h of request) is also necessary.[24] For a clinically driven DDAT strategy, where EAFT can be used initially to buy time while investigating, timely access to GM-ELISA, PCR and TDM assays results (i.e. within 3–5 days of sampling) is not as critical; however, reliable and timely access to bronchoscopy and biopsy (either performed by radiologist or surgeons) services (including histopathological reporting) is necessary

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Table 6 Antifungal agents for empiric antifungal therapy (EAFT)

Drug	Dose, frequency, route (adults)	Dose, frequency, route (children and adolescents)	Indication (children and adolescents)	Limitations
AmB-D	0.7–1.0 mg/kg IV, daily	0.7–1.0 mg/kg IV, daily	As a second-line alternative in patients on no, fluconazole or itraconazole prophylaxis	Toxicity: nephrotoxicity, infusion-related reactions (60–80%) Lack of availability in many Australasian centres
LAB	3 mg/kg IV, daily†	3 mg/kg IV, daily†	As first-line in patients on voriconazole or posaconazole prophylaxis	Toxicity (less than AmB-D): infusion-related reactions, nephrotoxicity Cost Not available at all centres
ABLC	5 mg/kg IV, daily	5 mg/kg IV, daily	As a second-line alternative in patients on voriconazole or posaconazole prophylaxis	Toxicity: nephrotoxicity, infusion-related reactions (greater than LAB) Not available at all centres
CAS	70 mg loading dose on day 1, thereafter 50 mg IV, daily	70 mg/m^2 loading dose on day 1, thereafter 50 mg/m^2 IV, daily (maximum 70 mg/day)	As first-line in patients on no, fluconazole or itraconazole prophylaxis	Narrow spectrum of activity
Voriconazole	6 mg/kg loading dose IV, 12-hourly for 24 hours (two doses), thereafter 4 mg/kg IV, 12-hourly	Children 2 to <12 years or young adolescents (12–14 years) weighing <50 kg: 9 mg/kg loading dose IV, 12-hourly for 24 hours (two doses), thereafter 8 mg/kg IV, 12-hourly Older adolescents or young adolescents weighing >50 kg: as per adult dosing	As a second-line alternative to CAS or LAB[b]	Did not meet the non-inferiority criteria for the composite endpoint in comparison to LAB[12] In a meta-analysis (as per authors' pre-specified criteria), voriconazole was inferior to LAB as more patients stopped voriconazole prematurely‡[89] TDM required

†Increase to 5 mg/kg/day if mucormycosis is suspected; ‡Methodological issues: paucity and heterogeneity of data. AmB-D, amphotericin B deoxycholate or conventional amphotericin B; IV, intravenously; LAB, liposomal amphotericin B; ABLC, amphotericin B lipid complex; CAS, caspofungin; TDM, therapeutic drug monitoring.

for such an approach to be effective.[24] In any institution, a team of haematologists, infectious diseases specialists, microbiologists, respiratory physicians, histopathologists and pharmacists should be involved in the development and implementation of DDAT strategies, adapted to local needs and resources. The work of such a team is critical, and should include the involvement of senior management, as DDAT strategies are likely to impact on current infrastructure and service provision.[24]

Evidence-based algorithms to guide use of EAFT and DDAT strategies

The risk of IFD and the type of prophylaxis are determined at the commencement of the first cycle of chemotherapy (see antifungal prophylaxis guidelines by Fleming *et al*. 2014, appearing elsewhere in this supplement). This, in turn, determines what diagnostic and treatment approaches are used at various time-points in the patient's journey through chemotherapy and/or HSCT. Figures 1–3 integrate the data that have been presented here and outline the clinical situations in which EAFT should be administered, along with the subsequent work-up and the role of surveillance-driven and clinically driven DDAT strategies. Given that many centres in Australasia do not have on-site access to, or cannot get results in a timely manner for, GM-ELISA and/or PCR testing from another institution, a surveillance-driven DDAT strategy will not commonly be used. In the Australasian setting, a clinically driven DDAT strategy will be more common. Notably, all strategies discussed in the current guidelines are not mutually exclusive and some, or all, strategies may be used in the one centre for different patient populations and even in the same patient at different time-points. Table 6 outlines the antifungal agents and doses that are recommended as EAFT (level II evidence; grade B recommendation). Please refer to the treatment guidelines by Chen *et al*., 2014 (yeasts), Blyth *et al*., 2014 (moulds) and Cooley *et al*., 2014 (*Pneumocystis jirovecii*), appearing elsewhere in this supplement, for antifungal therapy guidance following a diagnosis of IFD using a DDAT strategy.

Conclusion

The management of IFD remains challenging. With the advent of effective antifungal prophylaxis and DDAT strategies, EAFT no longer represents the panacea it once

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was. An EAFT strategy now has specific indications and should be used only in tandem with a diagnostic work-up with the aim of diagnosing/excluding IFD or other aetiologies. The role and format of DDAT strategies have been well delineated and refined in recent years, providing evidence for their use in routine clinical practice. While there has been much discussion about the development of a consensus DDAT strategy,[90] differences in local epidemiology and resource availability render this debate redundant. DDAT strategies will invariably differ between institutions and be informed by the type of antifungal prophylaxis used, clinical manifestations, patient populations and local resources.

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