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The career that this symposium honours, that of Ben E. de Pauw, MD, PhD, could be said to have begun in 1970 upon his graduation from the University of Amsterdam. However, it was his move to Nijmegen in 1975 to finish his PhD studies that really began the career in which he forged expertise in haematology, oncology, immunology and infectious diseases into one spectacular career that resulted in more than 200 publications.

Keywords: fungi, cancer, infections

Introduction

To put Ben de Pauw’s career into perspective, this review begins with the period just prior to his entry into the field (the 1960s). Table 1 is a simplified overview of the major antifungals introduced from that time onwards for the treatment and prophylaxis of neutropenic patients. Antibacterials are also included in the table because survival despite bacterial infections has been a major factor in the emergence of mycoses in these patients.

The 1960s

The recent addition of cephalothin and carbenicillin to the antibacterials used to treat neutropenic patients, along with gentamicin, did improve management; however, these drugs were usually used to treat established infection, not empirically. Mortality remained high. These antibiotics were administered through short plastic intravenous catheters or so-called ‘butterfly’ needles that could be taped down against the skin. Neither intravenous access method was a serious cause of sepsis because neither lasted more than a few days since local phlebitis or extravasation led to replacement.

Blood cultures were Gram-stained and subcultured when the broth became turbid upon once-daily observation, a process that required several days. Often, the septic patient was either dead or recovering before blood cultures became positive. Chest X-rays were known to be insensitive for the diagnosis of early pneumonia in the immunosuppressed patient but were a mainstay of diagnosis.

Conventional amphotericin B remained the only useful systemic antifungal drug. Rigors, hypotension and hypoxia often accompanied the first dose, frightening the patient and nursing staff and creating a powerful disincentive against empirical therapy in unstable patients. A nearly universal development of azotaemia led to interrupted and low-dose treatment. Alternate-day treatment was sometimes used but not in the double doses that such a regimen would require.

Flucytosine (5-fluorocytosine) was first introduced into clinical trials in 1967. The drug had been synthesized in 1957 as an antimetabolite for the treatment of cancer, like 5-fluorouracil, but flucytosine failed to have that property. It was not until 1963 that Emanuel Grunberg at Roche reported activity in experimental murine candidiasis. This study was a triumph of faith over reason, in that the drug had no antifungal activity in vitro screening assays; the inactivity was later found to be due to inhibitors in the culture medium. Flucytosine was primarily used orally, with an intravenous formulation available for use in some localities. Oral use was difficult or impossible in patients with severe mucositis. Early appreciation of flucytosine’s bone marrow toxicity and frequency of secondary drug resistance in Candida, combined with the pharmaceutical company’s low level of interest in this drug, essentially stalled development.

Autopsies were usually done and all-too-frequently revealed disseminated candidiasis or, less commonly, mould infection. Reports from this decade of autopsied acute leukaemia patients showed invasive fungal infections in 13% to 28%. The majority of infections were candidiasis, particularly gastrointestinal candidiasis. The probable reason for fewer mould infections was the earlier death from bacterial or candidal sepsis. Transfer to intensive care units was not commonly done except for respiratory failure because medical and oncology wards were prepared to offer fluid, blood products and pressor support. Monitoring of vital signs was by a diligent nursing staff, not by electronic monitors.

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Table 1. Major antifungals and antibacterials in use for neutropenic patients 1960–2008

<table>
<thead>
<tr>
<th>Decade</th>
<th>1960s</th>
<th>1970s</th>
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<td>amphotericin B</td>
<td>amphotericin + flucytosine</td>
<td>amphotericin B</td>
<td>lipid amphotericin B</td>
<td>iv itraconazole</td>
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<td>flucytosine</td>
<td>miconazole</td>
<td>ketoconazole</td>
<td>itraconazole capsules</td>
<td>voriconazole</td>
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<td></td>
<td>clotrimazole</td>
<td>itraconazole capsules</td>
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<td>caspofungin</td>
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<td><strong>Major antibacterials for treatment or prophylaxis in neutropenic patients</strong></td>
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<td>cephalothin</td>
<td>cephalothin</td>
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<td>carbenicillin</td>
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<td>piperacillin</td>
<td>carbapenems</td>
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The major diagnostic test for candidiasis during life was blood culture, an admittedly slow and insensitive tool for patients seeding from the gastrointestinal tract rather than, as is now often the case, seeding from a central venous catheter. Even when *Candida* was isolated from the bloodstream, some considered that transient candidaemia did not require treatment. To add to the confusion, *Candida* isolated from the sputum was thought by many to indicate *Candida* pneumonia. Diagnoses of aspergillosis and other mould infections were quite controversial. Isolation of *Aspergillus* from the sputum was not uncommon in adults but lacked specificity and sensitivity. Only 4 (13%) of the 25 patients in Young’s NIH series of invasive pulmonary aspergillosis who had a sputum culture for fungus had one positive culture, and only 2 had more than one positive culture.³ The distinction between invasive and non-invasive aspergilloses was blurred. Patients with chronic cough and *Aspergillus* in the sputum were considered by many to have aspergillosis of the bronchi or lung. The clinical, pathological and radiological consequences of vascular invasion in the neutropenic patient with aspergillosis were not recognized by many, if not most, oncologists and infectious disease practitioners at this time. The appellation ‘infectious disease specialist’ was not common coinage at the time. The first infectious disease subspecialty examination was not given in the USA until 1966 and the subspecialty was not recognized much beyond the continental USA during this decade.

The 1970s

As more effective drugs for leukaemia and more active antibacterial drugs made their way into oncology wards, mycoses began to be noticed as a serious problem. One approach was to filter mould spores out of the air and attempt to reduce intestinal colonization with *Candida*. Laminar flow units were credited with decreasing aspergillosis in patients with prolonged neutropenia but oral administration of nystatin or amphotericin B tablets, given in laminar flow units or on the wards, did not decrease the incidence of invasive candidiasis. Although intravenous amphotericin B was tried occasionally for prophylaxis, a less toxic drug was clearly needed for patients not able to tolerate this toxic drug. Azoles, a new class of antifungals, appeared promising because of low toxicity.⁴ *Miconazole*, the first systemic azole, was only available intravenously. The polyethoxylated castor oil excipient caused severe phlebitis with the short intravenous catheters still in use at that time. The next azole, clotrimazole, was poorly tolerated orally at doses that produced a measurable blood level and also induced its own metabolism, with progressively falling blood levels.⁵ The advent of ketoconazole was recognized as a breakthrough for its oral bioavailability and low toxicity. This azole, despite early enthusiastic claims of success, was not significantly effective for treating candidiasis or aspergillosis in immunosuppressed patients. The absence of an intravenous formulation and the necessity of gastric acid for gastrointestinal absorption impaired usage except for prophylaxis, a usage that received mixed reports of efficacy. It was only later, when itraconazole and fluconazole were developed, that successful prophylaxis with azoles began to be reported convincingly.

Intravenous plastic catheters, mostly short but some implanted, were attracting attention as a source of sepsis, including candidaemia. The report of sight-threatening *Candida* endophthalmitis in 76 patients by Edwards et al.⁶ helped to put into focus the dangers of candidaemia, even when transient.

The 1980s

The study of empirical antibacterial and antifungal therapy in febrile neutropenic patients by Pizzo et al.⁷ led to the increasing use of amphotericin B for empirical therapy. A major limitation of this approach was the substantial toxicity of this antifungal. Better azole antifungals were needed to change the entire paradigm. Kenneth Richardson, working at Pfizer in Sandwich, UK, successfully modified the azole molecule to synthesize fluconazole. Peter Troke, also at Pfizer, confirmed the antifungal spectrum...
and efficacy of fluconazole in mice experimentally infected with *Candida albicans*. This drug was to revolutionize the treatment and prophylaxis of candidiasis.\(^{8,9}\) Clear evidence of efficacy in candidaemia of non-neutropenic patients and prevention of disseminated candidiasis in stem cell transplant recipients was obtained for the first time with an azole. The safety profile and efficacy of this agent were, and are, remarkable, sufficient to see Queen Elizabeth II bestow on Kenneth Richardson the honour of an being an Officer of the Order of the British Empire. Dr Richardson was later inducted into the National Inventor’s Hall of Fame (http://www.invent.org).

The glaring drawback in the efficacy of fluconazole was *Aspergillus*. A significant increase in the incidence of aspergillosis was noted during this decade in several centres.\(^{10}\) Although itraconazole had been discovered in 1980 and was known to have activity *in vitro* against *Aspergillus*, development of this drug lagged badly. For this decade and part of the next, itraconazole was not available intravenously and was known to have irregular absorption from the gastrointestinal tract. Clinical trials of prophylaxis in immunosuppressed patients remained controversial until the better-absorbed hydroxypropyl β-cyclodextrin solution was introduced into clinical trials in the 1990s.

As mycoses in neutropenic patients received increasing attention, attempts were made to develop a diagnostic test that could be used on blood. The most notable were assays for arabinotol and mannan and antibody to *Candida* antigens, including enolase. The only commercial venture was the marketing of the Ramco latex kit, called Cand-Tec. This test detected an unidentified metabolic product in the blood of patients with deeply invasive candidiasis. After extensive evaluation, this test was found to be inadequately sensitive. Early experience with detecting galactomannan in patients with aspergillosis was encouraging enough for a commercial test for this antigen to be developed in the next decade.\(^{11}\)

### The 1990s and beyond

Advances in medicine resulted in better support for the critically ill patient, better diagnostic techniques, a larger array of antibacterial and antiviral drugs, improved chemotherapy for cancer and improved antifungal agents. Among the noteworthy technical advances was computed tomography, which dramatically improved the early detection of invasive pulmonary aspergillosis.\(^{12}\)

Attempts to decrease amphotericin B toxicity with alternative formulations began with a particulate suspension and methyl esters back in the 1970s. Experiments by Lopez-Berestien and colleagues with lipid formulations in the 1980s eventually led to the first marketed lipid preparation, ABLC, a microparticulate lipid complex. This formulation was followed by the colloidal dispersion (ABCD) and a liposomal formulation (AmBisome).\(^{13}\) Only the liposomal formulation had toxicity sufficiently low for it to be employed in prospective clinical trials for empirical use, though all three formulations were used for the treatment of deep mycoses.

Pfizer’s discovery of voriconazole in the 1980s led to a development plan that brought together investigators from both Europe and the USA to design a clinical trial for the primary treatment of invasive aspergillosis. When the design was agreed upon, there began the largest and certainly the most expensive study of its time. The results of this trial, which enrolled patients between 1997 and 2000, not only showed the efficacy of voriconazole, but also created a consensus on diagnostic criteria for invasive aspergillosis.\(^{14,15}\) This consensus was published in 2002 and became a guide for subsequent trial design.

Development of newer antifungals continued during this period, with the introduction of posaconazole and three members of a newer class of antifungals, the echinocandins.\(^{16}\) The low toxicity of these newer agents has created attractive options for the prophylaxis and treatment of patients with prolonged neutropenia.

Ben de Pauw was the chairman between 1995 and 2001 of the Invasive Fungal Infections Group of the European Organization for Treatment of Cancer (EORTC) and played a pivotal role in both the design of the voriconazole trial and production of the consensus document. Through Dr Donnelly’s and Dr de Pauw’s persistence and guidance, the first revision of the consensus document is now published.\(^{17}\) Nor should this be the last revision, as science and medicine continue to advance, built upon the foundation stones laid by dedicated and gifted physicians such as Ben de Pauw.

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### Transparency declarations

None to declare.

### References


