Mycology newsletter

The ECMM/CEMM Mycology Newsletter is mailed to the members of the national societies affiliated to the European Confederation of Medical Mycology (about 3000 in 20 different countries)

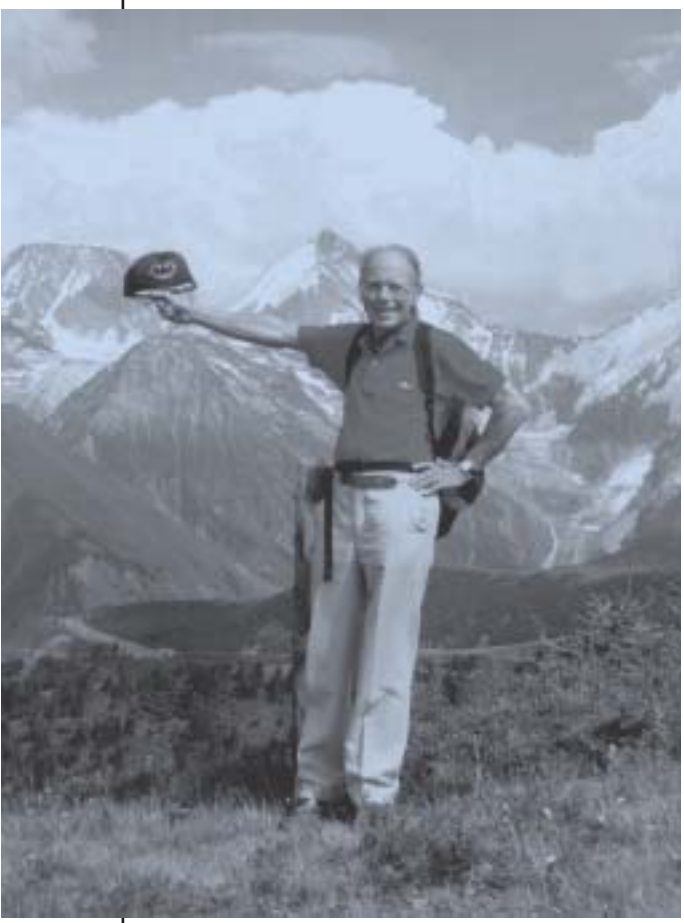
1/2000

ECMM
European Confederation of Medical Mycology
CEMM
Confédération Européenne de Mycologie Médicale

Message from the President

For all who entered medical mycology between 1960 and the late 1990s a scientific meeting without Edouard Drouhet will be a quieter and less congenial gathering. Although small in height he was a towering presence in the field. His enquiring mind and his friendliness, coupled with natural charm, influenced a generation of mycologists. His great gift to us all was that he was always interested in his chosen field and loved to talk to those who had something to say and who, in turn, would listen to him.

All of us have favourite Drouhet stories which we will continue to exchange for many years in affectionate memory of a great man. One of mine concerns the occasion of a meeting where three of us were presenting data on ketoconazole. Edouard had his usual vast collection of slides arranged in well-organised chaos. When we arrived at the lecture theatre two problems were apparent immediately. The translator would only be able to translate after each sentence and the slide projector would only take two slides at a time. While two of us sat down to prune our lectures by removing over half the slides and shortening the script Edouard Drouhet looked around the imposing lecture hall, shrugged his shoulders and said that he wasn't going to change his talk and that no one would expect it. He was right, of course, and our shortened talks fell flat while his attracted



Edouard Drouhet

(continued on page 3)

Contents

ECMM/CEMM

Mycology Newsletter

Editorial Advisory Board
Bertrand Dupont
Renée Grillot
Donald W.R. Mackenzie
Malcolm Richardson
Maria Anna Viviani (Editor)

Editorial office
c/o Istituto di Igiene e Medicina Preventiva
Università degli Studi di Milano
via F. Sforza 35, 20122 Milano, Italy

Direttore responsabile
Ivan Dragoni

Art Director
Luigi Naro

Contributions from:
Christophe d'Enfert, Renée Grillot,
Roderick J. Hay, Peter-Michael Rath,
Malcolm Richardson, Reinhard Rüchel,
Markus Ruhnke, Axel Schmidt,
Esther Segal, Paul Verweij,
Maria Anna Viviani

Printed by:
Lasergrafica Polver
via Kramer, 17/19
Milano, Italy

Official Office: 105, Bld Murat
75016 Paris, France

Registrazione Tribunale di Milano
n. 749 del 25.11.1997

ECMM Council

Affiliated Societies

Associação Portuguesa de Micologia Médica (ASPOMM)
President: M. Rocha
Vicepresident: R.M. Velho
Secretary: M.L. Rosado (*ECMM delegate*)
Treasurer: M. Gardete
Membership 2000: 50
Newsletter

Asociación Española de Micología (AEM) Sección de Micología Médica
President: J.M. Torres Rodríguez
Vicepresident: J. Pontón San Emeterio
Secretary: S. Santamaría del Campo
Treasurer: A.J. Carrillo Muñoz
President Mycology Section: F.J. Cabañes Saenz (*ECMM delegate*)
Membership 2000: 95
National meeting: November 8, 2000, Barcelona (joint with ECMM Congress)
Journal: Revista Iberoamericana de Micología

British Society for Medical Mycology (BSMM)
President: R.J. Hay (*ECMM delegate*)
General Secretary: R.A. Barnes
Meetings Secretary: D. Sullivan
Treasurer: G.S. Shankland
Membership 2000: 242
National meeting: April 1-3, 2001, Imperial College, London
Newsletter

Bulgarian Mycological Society (BMS)
President: T. Kantardjiev (*ECMM delegate*)
Vicepresident: G. Mateev
Secretary: A. Kouzmanov
Treasurer: T. Velinov
Membership 2000: 41
National meeting: November 23-25, 2000

Czech Mycological Group
ECMM delegate: A. Tomsíková

Danish Society for Mycopathologia
President: N.A. Peterslund
Vicepresident: L. Larsen
Secretary: J. Stenderup (*ECMM delegate*)
Treasurer: J. Stenderup
Membership 2000: 45

Deutschsprachige Mykologische Gesellschaft e.V. (DMykG)
President: H. Chr. Korting
Vicepresident: H. Hof
Secretary: C. Seebacher
Treasurer: W. Fegeler
ECMM delegate: H. Bernhardt
Membership 2000: 1100
Journal: Mycoses
Newsletter: Mykologie Forum

Federazione Italiana di Micopatologia Umana e Animale (FIMUA)
President: R. Esposito
Vicepresident: E. Concia
Secretary: A. Persi
Treasurer: M.A. Viviani (*ECMM delegate*)
Membership 2000: 160
National meeting: 2002, Modena
Newsletter: FIMUA news

Finnish Society for Medical Mycology
President: J. Issakainen (*ECMM delegate*)
Vicepresident: A. Kahanpää
Secretary: P. Kankaanpää
Treasurer: R. Voutilainen
Membership 2000: 59
National meeting: March 2001
Newsletter: Sienet ja Terveys (Fungi and Health)

Greek Mycological Group
ECMM delegate: O. Marcelou-Kinti

Hungarian Dermatological Society Mycology Section
President: G. Simon (*ECMM delegate*)
Secretary: G. Fekete
Membership 2000: 37

Israel Society for Medical Mycology
President: E. Segal
Secretary: I. Berdicevsky (*ECMM delegate*)
Treasurer: D. Elad
Membership 2000: 80

Polish Dermatologic Society Mycology Section
President: E. Baran
Vicepresident: Z. Gwiezdzinski and P. Ratka
Secretary: J. Szepietowski (*ECMM delegate*)
Treasurer: R. Bialynicki-Birula
Membership 2000: 98
Journal: Mikologia Lekarska (Medical Mycology)

Netherland Society for Medical Mycology (NVMy)
President: J.F.G.M. Meis (*ECMM delegate*)
Scientific Secretary: G.S. de Hoog
Secretary: E.P.F. Yzerman
Treasurer: R.W. Brimicombe
Membership 2000: 145
Newsletter: NVMy Newsletter

Russian Society of Mycology
President: S.A. Burova (*ECMM delegate*)
Vicepresident: V.B. Antonov
Secretary: N.M. Vasilyeva
Treasurer: I.V. Kurbatova
Membership 2000: 25

Société Belge de Mycologie Humaine et Animale/ Belgische Vereniging Voor Menselijke en Dierlijke Mycologie
President: D. Swinne
Vicepresident: N. Lateur, E. Van Hecke
Secretary: P.E. Lagneau, K. Lagrou
Treasurer: F. Symoens
ECMM delegate: N. Nolard
Membership 2000: 180

Société Française de Mycologie Médicale
President: Cl. de Bièvre
Vicepresident: D. Chabasse, O. Morin, N. Contet Audonneau
Secretary: B. Dupont (*ECMM delegate*)
Treasurer: P. Boiron
Membership 2000: 340
National meeting: May 25-26, 2001, Amiens
Journal: Journal de Mycologie Médicale

Swedish Society for Clinical Mycology
President: J. Faergemann
Vicepresident: T. Kaaman
Secretary: G. Pålsson
Treasurer: S. Johansson
ECMM delegate: L. Klingspor
Membership 2000: 105
National meeting: April 27, 2001, Stockholm
Newsletter

Swiss Mycological Group
ECMM delegate: M. Monod

Turkish Microbiological Society Mycology Section
President: Ö. Ang
ECMM delegate: E. Tümbay
Membership 2000: 21

(Information provided by the member Societies)

Message from the President

(continued from page 1)

all the questions. Edouard Drouhet will be missed by all of us.

The European Confederation of Medical Mycology also owes a debt of gratitude to Dr. Drouhet. His interest and enthusiasm provided the right catalyst for the development of our society and his support for the first committee was of critical importance in galvanising the society. He had a keen sense of European tradition and he was confident that, despite the worst efforts of politicians, a renaissance in European scientific thought and endeavour was eminently possible through well constructed programmes and the blossoming of societies such as ours.

Over the last few months we have given a large amount of thought as to how we should commemorate Dr. Drouhet in an appropriate way. After some deliberation we have decided to recommend the following proposal to you: the establishment of an annual Drouhet Lecture. It would be given every year on the occasion of the ECMM meeting and the lecturer would be chosen by a small subcommittee of the society together with the organisers of the annual meeting. In order to finance this initiative we are proposing to launch an appeal in this issue for funds. While the immediate purpose would be to cast a medallion that would be presented to the Drouhet lecturer on the occasion of the lecture, if we exceed the target, together with members of the ECMM, we would consider other ways in which to honour his name.

The members of the executive group are very enthusiastic about this proposal as we believe that it would be a fitting tribute to a great man. I hope that you will all agree with the idea and help us get the annual Drouhet Lecture off to a flying start.

Roderick J. Hay

Edouard Drouhet

Edouard Drouhet, a key figure in the field of Medical Mycology during the last century, died on January 1, 2000, in Paris. He was born in Barlad, Romania, in 1919 from a family of French descent. His grandfather was a medical doctor born in Blaye, in the Gironda region of France. Prof. Drouhet was very proud of his French ancestry. In his writing he recalls with great emotion a visit to Blaye made in 1997, together with Marianna Viviani, following a meeting of the French Society of Medical Mycology held in Arcachon. That day he discovered in the cemetery of this small town the grave of his great-grandmother.

Professors Drouhet, Mariat and Segretain: the elder statesmen of the Mycology Unit of the Pasteur Institute at the XIII Congress of ISHAM in Salsomaggiore, 1997. From left to right: Edouard Drouhet, Mrs Segretain, Gabriel Segretain, François Mariat, Mrs Mariat (courtesy E. Gueho).

In Romania, Drouhet began his studies in medicine and in 1944 he obtained his degree in medicine. Twelve years later, he also obtained a degree in medicine at the University of Paris. Soon after the end of the Second World War, he moved to Paris, together with his wife Victoria having earned a grant from the French Government. In 1946, he joined Joseph Magrou, a physician and botanist and the director of the Service de Physiologie Végétale-Mycologie of the Pasteur Institute.

Prof. Drouhet decided to settle in Paris and in 1948 he was appointed as "attaché de recherches" at the Centre National de la Recherche Scientifique (CNRS) under the direction of Prof. André Lwoff, Noble Prize winner for Medicine and Physiology. In Paris, he specialized in Microbiology at the Pasteur Institute (1947), in Serology at the Fournier Institute (1948), and in 1954 he definitely joined the Pasteur Institute as "Chef de Laboratoire" in the Service of Prof. Magrou. There he met Gabriel Segretain and François Mariat and the mutual understanding of the three scientists gave rise to the team that played a leading role in the development of Medical Mycology in France and in Europe. Under the influence of Drouhet, the Service modified the direction of its research programs turning from botany to Medical Mycology. It focused on human and animal pathogenic fungi and on the study of effective antifungal treatments for mycotic diseases.

In 1953, Prof. Drouhet together with Drs. Segretain and Mariat founded the "Cours supérieur de mycologie médicale" at the Pasteur Institute, a post-graduate Course that in 48 years trained about a thousand mycologists from France and other European and extra-European countries. In a short period of time the Course gained importance representing probably the most important result of the teaching activities of Drouhet brought indefatigably, to France and overseas, until recent years. In 1953, Drouhet was also a co-founder of the International Society of Human and Animal Mycology (ISHAM) and in 1956, together with Segretain and Mariat, he founded the Société Française de Mycologie Médicale. These three Institutions promoted and significantly contributed to the great development of the Medical Mycology in the ensuing years. Forty years later, Prof. Drouhet supported with great enthusiasm the creation of the European Confederation of Medical Mycology. He was convinced that future developments in European Medical Mycology would be achieved through the creation of networks on a continental level, dealing with epidemiological studies, on the standardization of diagnostic procedures, on educational programs and in training.

In 1972, Drouhet was given the rank of Professor at the Pasteur Institute and there he established the Centre National de Référence des Mycoses et des Antifongiques in Paris. In 1981, he succeeded Gabriel Segretain as Director of the "Unité de Mycologie" of the Pasteur Institute, a position that he held until his retirement in 1987. After his retirement, Drouhet actively participated in the scientific community's life and from 1990 he served as Chief Editor for the «Journal de Mycologie Médicale», which had replaced the «Bulletin de la Société Française de Mycologie Médicale». Through his guidance, the journal became one of the most authoritative in the field of

Medical Mycology.

Since its beginning, Edouard Drouhet´s research work was characterized by its innovative nature. Before his era, the Pasteur Institute´s studies on fungal pathogens were restricted to the field of botany. Drouhet resolutely initiated the study of human and animal fungal pathogens, carrying out studies on the physiological and biochemical activities of these fungi in relation to their growth, morphogenesis and pathogenicity, and developing techniques for the rapid diagnosis of fungal diseases.

Drouhet greatly contributed to a better knowledge of diseases due to opportunistic fungi. He highlighted new aspects of *Candida albicans* infections developing studies in premature infants and children and discovering in 1980 a new cutaneous, ocular and osteoarticular septicemic syndrome in young heroin addicts. In 1953, he was the first to test in France such new antifungals as nystatin and amphotericin B, defining dosage and therapeutic indications and investigating their mechanisms of action.

A constant interest of Drouhet´s researches was the host´s immunological response to fungal infections. By 1950, he had already shown the role of a capsular polysaccharide (glucuronoxylomannan) of *Cryptococcus neoformans* in fungal virulence, and identified it as the cause of the immunological paralysis during cryptococcosis. Finally, he prepared standardized fungal antigens for the qualitative and quantitative study of serum antibodies in patients with deep-seated mycoses.

On 5 June 1999, at the ECMM Congress in Dresden, Edouard Drouhet took his leave from the scientific community. He was very ill and suffering a great deal, but he forced himself to fulfil his commitment and gave a remarkable lecture on *Emmonsia pasteuriana*, the new species he had identified and named to commemorate his beloved Pasteur. The audience witnessed a great lesson of scientific discipline and style and acknowledged Drouhet as a world class medical mycologist.

Maria Anna Viviani

Medical Mycology in Israel

Call for the 7th ECMM Congress, Tel-Aviv, Israel, June 16-19, 2001 - Deadline for Submission of Abstracts: March 16, 2001

The Israel Society for Medical Mycology (ISMM) will host the next congress of the European Confederation of Medical Mycology (ECMM), the 7th ECMM Congress, which will be held in the David Inter-Continental, in Tel-Aviv, during June 16-19, 2001. The ISMM is a member Society of the ECMM. The ISMM has about 70 registered members and an elected 3-member council. It is affiliated with the Israel Society of Microbiology (ISM). The ISMM members meet once a year, and in addition hold a scientific session in the frame of the annual meeting of the ISM. Two major international congresses in the field of Medical Mycology have been held in Israel: the VIIth ISHAM Congress in 1979 and the 1st *Cryptococcus* Conference in 1989, both being successful and remembered in the Medical Mycology Community.

The ISMM, the Local Scientific-Organizing Committee and the International Committee of the 7th ECMM Congress cordially invite all ECMM members to participate in the Congress.

The scientific program of the Congress will focus on most of the currently relevant topics in Medical Mycology, including emerging pathogens, infections in the compromised host, virulence factors, host-pathogen interactions, recent approaches to diagnosis and anti-fungal therapy. The program consists of two plenary sessions, nine symposia sessions, three round-table sessions with invited speakers, as well as poster and oral contributed presentations. A commercial exhibition and social events will be included as well.

We trust that the Congress participants will enjoy, in addition to the interesting and stimulating scientific program, the sites and sounds of Tel-Aviv, a vibrant Mediterranean city and look forward to see you all with us in Tel-Aviv next year.

Esther Segal
Congress Chairperson

Special report on...

Meetings 2000: Focus on *Aspergillus* and aspergillosis

by
**Christophe d'Enfert,
Renée Grillot,
Peter-Michael Rath,
Malcolm Richardson,
Reinhard Ruechel,
Markus Ruhnke,
Axel Schmidt,
Paul Verweij**

For those members who are working with *Aspergillus* or those with a clinical interest in aspergillosis there has been a wealth of information this year presented at conferences around Europe and further field. For this issue of the ECMM Newsletter we have commissioned a series of conference reports to bring the flavour of these meetings to a wider audience.

Focus on *Aspergillus*

Bagshot, Surrey, UK, 9 September 2000

Controversies in Fungal Infections

On September 9th a symposium was held in the United Kingdom entitled «Controversies in Fungal Infections». For the seventh time a broad range of problems related to the management of patients with invasive fungal infections was discussed in an interactive manner.
A wide variety of specialists were present among the participants although the majority worked in the field of medical microbiology.

One session entitled «Aspergillosis: continuing controversies» addressed problems related to the management of patients with invasive aspergillosis. The chairman, Chris Kibbler (London), presented several case studies to the audience and an expert panel. The panel consisted of Les Berger (London), James Burnie (Manchester), Archie Prentice (Plymouth), and Paul Verweij (Nijmegen). The presented cases gave rise to lively discussions. It is clear that clinical practice is quite variable between different institutes and between different countries. For instance, the use of chemoprophylaxis for the prevention of invasive aspergillosis in high risk patients is standard in some institutes while others use only fluconazole or no prophylaxis at all. Other issues that gave rise to discussions included the management of patients that fail to respond to first line treatment. Is there any merit in dose escalation or should we switch to another drug or do we need to combine antifungal drugs? Evidence for many decisions is lacking which complicates management strategies. Furthermore, the increasing number of agents available for use will only further complicate these decisions. The management of patients with invasive aspergillosis requires a systematic (and often multidisciplinary) approach. Also it was apparent that a general management strategy in many cases cannot be followed since many details need to be taken into consideration resulting in an individual approach.

Paul Verweij

Focus on *Aspergillus*

Paris, France, 21 March 2000

Consensus conference on prevention of aspergillosis in immunocompromised patients

Despite the recent progress in diagnosis and treatment of invasive aspergillosis (such as the development of antigenemia tests and amphotericin B formulations) the mortality associated with this infection remains high (60 to 90%). To minimize the exposure of high risk patients to *Aspergillus* conidia a strict surveillance programme in the hospital setting is required. The difficulties that face this problem and the absence of guidelines in this field prompted the Société Française d'Hygiène Hospitalière to organise a consensus conference on the prevention of aspergillosis in cancer patients and transplant recipients, that was chaired by Jacques Fabry. The conference was held in Paris on March 21, 2000, with 250 French delegates and the participation of the French Societies of Haematology, Bone Marrow Transplant, Organ Transplantation, Oncology, Mycology, and Infectious Diseases.

The following questions were addressed:
1. In what situations cancer and transplanted patients are at risk of invasive aspergilllosis?
2. Which preventive measures (primary or secondary) have been proved to be effective, for which patients and in which conditions?
3. What is the most effective surveillance strategy?
4. Which are the prevention strategies?

A review of the literature was carried out, before the conference, by two haematologists, one infectious diseases specialist and one epidemiologist. Then, during the meeting of March 21, 16 specialists from various disciplines presented the state of art of different aspects of the addressed questions. Finally a panel of 14 members drew up the recommendations concerning each of the addressed questions, taking into consideration the experts' reports and the discussion raised dur-

ing the conference.

The recommendations have been presented in two forms: an "extended text" (27 pages) intended for physicians and scientists involved in the management of the patients and their environment, and a "short text" designed for administrative or technical staff who are involved in political, financial or building design decisions concerning the patient at risk.

Both texts, extended and short, are available in French at the Agence Nationale d'Accréditation et d'Evaluation en Santé (ANAES), Service Communication et Diffusion, 159 rue Nationale, 75640 Paris Cedex 13, France. They are also available at the ANAES website: <www.anaes.fr>.

Renée Grillot

Focus on *Aspergillus*

Toronto, Canada, 17-20 September 2000

40th Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC)

The American Society for Microbiology's (ASM) ICAAC is a truly international meeting. This year the meeting was held in Toronto. With over 12,000 attendees and in excess of 2000 presentations the meeting requires staminar. Increasingly, this is the conference where the latest information on new and not so new antifungal drugs, and the diagnosis of fungal infections is presented. The theme of this issue of the ECMM Newsletter is *Aspergillus* and aspergillosis. This conference report will focus on the same theme. A very useful, fully searchable CD-ROM form of the abstract tome identified 77 abstracts using the search term *Aspergillus*. As to be expected the majority of presentations dealt with antifungals and antifungal treatment. Given the success of antibacterial agents that target bacterial cells wall synthesis, a similar strategy for fungi has long been thought attractive. Pioneering work performed over 25 years ago identified the echinocandins as antifungal agents that interfered with fungal cell wall synthesis. Echinocandin B and some its derivatives have been and gone. More recent discovery of both echino- and pneumocandins with a broader antifungal spectrum, including impressive activity against *Aspergillus* species, than previous has ignited a renewed interest in these compounds. An useful overview of the candins was given by John Rex. Three compounds are currently undergoing clinical trials: MK991 (caspofungin, Cancida: Merck), FK463 (Fujisawa), and LY303366 (VER-002, V-Echinocandin: Versicolor). A variety of presentations and posters provided the latest findings. All three candins share some common properties. In addition, there were presentations showing that the combination of a candin with amphotericin B might be useful. Some exciting developments indeed but John R Graybill posed the question: «Will the promise of the candins be realised?».
Maybe we'll hear some answers at next years ICAAC.

New azoles with activity against *Aspergillus* were reviewed by David Stevens. Scattered throughout the conference were a few presentations on itraconazole, voriconazole, posaconazole and ravuconazole. Frank Odds posed the question: «Have we seen the best of the triazole anti-fungals?» emphasising the view that it is too early to judge to what extent these compounds represent substantial improvements on older members of the chemical class.

A few clinical studies and surveys were presented, as well as posters and slide sessions confirming the value of galactomannan detection and PCR in the early diagnosis of invasive aspergillosis.

Another topical theme was the isolation of *Aspergillus* and other moulds from environmental samples in hospitals: air and water, and from tobacco and marihuana. These studies are of considerable interest to epidemiologists and although it has not been possible to prove transmission to patients, the concordance of genotypes of the isolates from these studies strongly suggests nosocomial transmission. However, the debate on nosocomial versus community-acquired aspergillosis goes on.

In conclusion, a wealth of information which fortunately can be accessed by everyone on a dedicated ICAAC web site which is fully searchable (www.icaac.org) and available to ASM members and non-members alike.

Malcolm Richardson

Focus on *Aspergillus*

The Hygiene Institute of the University of Göttingen.

Göttingen, Germany, 7-8 April 2000

An international workshop on *Aspergillus* and aspergillosis was held at the Department of Bacteriology, Institute of Hygiene, University of Göttingen. The workshop was organized by the Deutschsprachige Mykologische Gesellschaft (DMykG) and the Deutsche Gesellschft für Hygiene und Mikrobiologie (DGHM)

Interdisciplinary Forum on Aspergillosis

Session 1

Molecular epidemiology of *Aspergillus* and fungus-host interaction

Two contributions focused on the molecular epidemiology of aspergilli. Renée Grillot (Grenoble, France) presented data from the European Study Group for Research on Biotypes and Genotypes of *Aspergillus* (EBGA-Network). Serial *A. fumigatus* isolates obtained from lung transplant patients were genotyped by random amplification of polymorphic DNA (RAPD), sequence-specific DNA primers (SSCP), and multi-locus enzyme electrophoresis (MLEE). Combining the results of the three methods resulted in a higher discriminatory power. In the follow-up different genotypes were found in the same patient. Although most of the patients were infected by unique types, two genotypes were found in different patients. Since the patients were not protected by laminar air flow systems the occurrence of the same genotypes in different patients may indicate that these types were predominant in the hospital environment. Susceptibility testing of strains obtained from patients under long-term treatment revealed no increase in resistance to amphotericin B and itraconazole.

The second contribution (Peter-Michael Rath, Essen, Germany) focused on the genetic diversity of *Aspergillus* spp. other than *A. fumigatus*. Whereas in a number of studies the genetic diversity of *A. fumigatus* has been shown by using different techniques, data on the diversity of *Aspergillus* spp. other than *A. fumigatus* is scanty. By using RAPD technique it could be shown that a high genetic diversity exists also in *A. flavus*, *A. niger*, and *A. terreus*. When analyzing an outbreak of invasive *A. terreus* infections, environmental isolates were found to be identical with isolates of some but not of all patients, indicating a possible environmental source.

Summing up the two contributions it is evident that a number of different techniques have been used for fingerprinting aspergilli. Up to now it is unclear which method or combination of methods is the most appropriate for studying the molecular epidemiology of these opportunistic pathogens. All typing studies have revealed a high genetic diversity of aspergilli, but it has not been possible to identify types with a higher ability to induce infections up to now. Furthermore, studies on the interlaboratory reproducibility of typing results are crucial.

Further topics were the binding of *A. fumigatus* conidia to tissue cells, virulence factors and host defense mechanisms. Principally, the first step of an infection is the adherence of the pathogen to host cells after ingestion or inhalation. Jean-Philippe Bouchara

(Angers, France) reviewed the interactions between host proteins and *A. fumigatus* conidia. *In vitro* studies by using flow cytometry and adherence assays revealed that the conidia bind specifically to lung cells. At least two distinct adherence systems localised on the echinulations of the outer cell wall layer of the conidia are involved: firstly, the binding to the tripeptide sequence RGD (arginine-gylcine-aspartic acid) of fibronectin mediated by two fungal polypeptides; secondly, binding to laminin and fibrinogen mediated by a sialic acid-specific lectin of the conidial wall. Furthermore, hydrophobic interactions may also contribute to adherence. The ability to bind appears to increase with maturation of the conidia.

Jean-Paul Latgé (Paris, France) pointed out that despite the large number of putative virulence factors found in *A. fumigatus* (for example pigments, RNase, hemolysin, gliotoxin, proteases, phospholipases), with the exception of melanin, no other virulence factors have been identified in animal experiments with single-gene knock-out mutants. However, when inoculating several wild-type strains together into immunocompromised mice differences in the aggressiveness of the strains were found, indicating that several factors play together or factors are only produced *in vivo* but not *in vitro*. Furthermore, the resistance to host defense mechanisms might be more important than the production of disease-specific factors.

The host defense mechanisms include the mechanical clearance of the conidia by ciliary action, the activation of the complement system, the ingestion and killing of conidia by macrophages, the killing of hyphae and conidia by neutrophils, and the activation of platelets by attachment to hyphae.

Emmanuel Roilides (Thessaloniki, Greece) reviewed acquired immunity in aspergillosis. Resistance to fungal infections is associated with increased levels of IL-2, IL-12, and gamma interferon (type 1 response), but the exact mechanisms are unknown. Antibodies show no protective activity.

It is well known that immunosuppressive drugs like corticoids increased the risk of acquiring aspergillosis. Macrophages treated with glucocorticoids showed an impaired killing activity to ingested conidia. However, M-CSF, GM-CSF, and gamma-interferon restore the dexamethasone-induced suppression of monocyte activity against *A. fumigatus*.

In summary, our knowledge on pathogen-specific virulence factors as well as on the defence mechanisms of the host is incomplete. New approaches such as the analysis of differential gene expression *in vivo*, and the study of T-cell immunity in infected patients should be undertaken.

Peter-Michael Rath

Session 2

Molecular biology of *Aspergillus*

The second part of the Interdisciplinary Forum was mostly focused on the molecular biology of *Aspergillus fumigatus* and its application to improved diagnosis of *A. fumigatus*-related diseases and a better understanding of *A. fumigatus* virulence.

Various molecular tools have been developed to investigate gene functions in *A. fumigatus*. For transformation of the fungus usually a protoplast fusion method is used, if homologous integration of the transforming DNA is the desired event. Electroporation is even more efficient but results in a higher degree of ectopic integrations. The transformation system usually takes advantage of dominant selectable markers such as the antibiotics phleomycin or more frequently hygromycin. As an alternative, an auxotrophic marker system based on OMP decarboxylase (pyrG)-negative mutants, which require pyrimidines for growth, has been developed. The advantage of this latter system primarily lies in its counterselectability thus allowing various techniques to modify the genome to be developed.

One of the major limitations for reverse genetics in *A. fumigatus* is the high frequency of ectopic integration of the transforming DNA. A novel technique that I presented during this meeting now allows the construction of modified cosmids by *in vivo* recombination in *Escherichia coli* and the subsequent use of these cosmids to introduce knock-outs in the *A. fumigatus* genome at high frequency (up to 50%), thanks to large homology regions between the modified cosmid and the target locus.

On the other hand, insertional mutagenesis is likely to provide a novel mean to understand the basis of *A. fumigatus* pathogenicity. As mentioned above, electroporation which results in high transformation efficiency and ectopic integration has been used successfully to generate collections of *A. fumigatus* insertional mutants. However, transformation-mediated insertional mutagenesis appears to promote genomic rearrangements that hamper the molecular analysis of the insertional mutants. Recently, we succeeded in developing a transposon-based insertional mutagenesis system for *A. fumigatus* that is likely

to represent a useful tool for the study of this fungus.

Two specific examples of the application of targeted gene inactivation were presented during this part of the meeting.

Axel Brakhage (Darmstadt, Germany) and Bernhard Jahn (Mainz, Germany) presented their collaborative efforts to define the function of pigment biosynthesis in the virulence of *A. fumigatus*. They have cloned a gene cluster involved in the biosynthesis of a secondary metabolite similar to dihydroxynaphtalene (DHN) melanin. One of the genes in this cluster encodes a polyketide synthase required for both pigmentation and ornamentation of the conidia. Interestingly, inactivation of the PksP polyketide synthase also impairs virulence of the fungus in a mouse model of aspergillosis. This appears to be linked to a higher susceptibility of mutant conidia to reactive oxygen species that are part of host defence mechanisms. Inactivation of other genes in the DHN melanin biosynthesis pathway impairs conidial pigmentation but does not influence *A. fumigatus* virulence suggesting that PksP is the sole virulence factor in this pathway. Comparisons with *A. nidulans* which is non pathogenic would suggest that the contribution of PksP to *A. fumigatus* virulence occurs not only by the production of the protective DHN melanin but also by the production of other secondary metabolites that could interfere with the host response.

Utz Reichard (Göttingen, Germany) has used a dominant selectable marker system based on hygromycin for knocking out two different endoproteinase genes, *PEP2* and *ALP2*, the expressed proteins of which had been purified from an *A. fumigatus* cell wall fraction. Interestingly, PEP2 as an aspartic proteinase and ALP2 as a subtilisin-related proteinase showed homology to the vacuolar proteinases of *Saccharomyces cerevisiae*, proteinase A and B, respectively. The *PEP2*-negative mutant did not reveal a morphological or developmental phenotype, however, the *ALP2*-negative mutant showed up to 80% inhibition in sporulation which was most likely due to a reduction of the mean and median diameter of the mutant conidiophore vesicles. Thus, in contrast to yeasts where functional proteinase A is a major key enzyme and normally required for proteinase B activity, in *Aspergillus* the vacuolar subtilisin seems to be more independent and crucial for development. In addition, ALP2 raises further clinical interest, since it has been recently identified to be the major allergen of *A. fumigatus* in patients suffering from allergic bronchial asthma. In this respect, producing substantial amounts of defined antigens has become an important field of research in aspergillosis.

Michel Monod (Lausanne, Switzerland) presented a novel approach for the efficient production and purification of *A. fumigatus* antigens that can be used for diagnostic purposes. This takes advantage of an expression system in the methylotrophic yeast *Pichia pastoris* where antigen-coding genes are expressed under the control of the strong promotor of the *P. pastoris* alcohol oxidase gene, thus resulting in high production yields. Several antigens have been purified using this approach, including proteases, peptidases, catalases and a ribonuclease. These purified antigens are now evaluated for their use in the diagnosis of aspergilloma in collaboration with the group of Jean-Paul Latgé (Institut Pasteur, France) and several hospital laboratories in France.

These different presentations therefore demonstrated that more and more technologies are becoming available to reach a better understanding of *A. fumigatus* biology and virulence. Although there is probably still a long way before we fully understand the molecular mechanisms underlying *A. fumigatus* virulence, significant progress is likely to be made in the next few years by a combination of molecular approaches and appropriate cellular and animal models.

Christophe d'Enfert

Session 3

Clinical aspects of aspergillosis

In the third session clinical aspects of aspergillosis including allergy were treated.

Aspects of invasive aspergillosis were treated by Reinhard Rüchel (Göttingen, Germany). The foremost clinical features of invasive and disseminated aspergillosis were briefly reviewed. Emphasis was placed on the influence that the different degrees of immunosuppression of the host has on the appearance of pulmonary aspergillosis in the roentgenographic image and on the morphology of fungal elements in respiratory secretions. Among others, encephalitis as the most dangerous complication of invasive aspergillosis was exemplified by roentgenographic images and evidence from necropsy.

The role of aspergillosis as an opportunistic infection in solid organ transplant patients was reviewed by Thomas Lorf (Göttingen, Germany). It was shown, that aspergillosis represents the major mycological threat to these patients; and is not exclusively an early onset disease. Lethal infections may also arise more than two month after transplantation. The performance of parenteral antimycotic prophylaxis using amphotericin B or liposomal amphothericin B in a reduced dosage regimen (1 mg/kg/d) was discouraged. A full therapeutic dosage (e.g. AmBisome 3 mg/kg/d) should be recommended to suppress invasive aspergillosis particularly in transplant patients at risk (e.g. retransplantation, haemofiltration, primary graft dysfunction).

Reto Crameri (Davos, Switzerland) referred to the difficulties of differential diagnosis of the allergic diseases, particularly allergic bronchopulmonary aspergillosis (ABPA) and sensitization only to aspergilli. Since ABPA only requires consequent therapy with corticosteroids, a precise differential diagnosis is important but cannot be achieved using tests which are based on crude allergens, which have been prepared from cultures of *Aspergillus fumigatus*. The outcome of such tests is largely affected by the varying content of the numerous proteinaceous, potentially allergenic constituents in preparations which are marketed by different producers. A solution for this problem of reproducibility is the generation of the most important fungal allergens as highly defined recombinant proteins, using a phage surface display technology for cloning of specific DNAs and subsequent generation of the recombinant allergens.

Reinhard Rüchel

Session 4

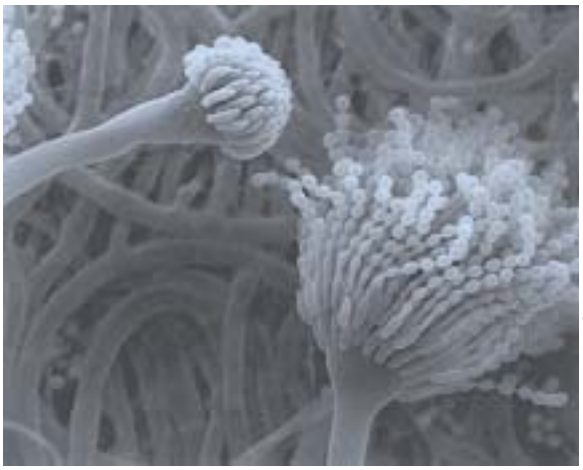
New developments in the diagnosis of invasive aspergillosis

The second day of the conference was dedicated to clinical and diagnostic aspects of invasive aspergillosis.

Reinhard Kappe (Heidelberg, Germany) presented recent data on laboratory diagnosis of aspergillosis from literature sources. The laboratory diagnosis of invasive aspergillosis largely depends on conventional methods like microscopy, including Calcofluor white staining, and culture of specimens from the lower respiratory tract. Definitive diagnosis by finding tissue invasion often depends on histological examination of biopsies stained with Gomori's methenamine silver stain. Antigen testing twice weekly of serum samples by enzyme immunoassay is recommended for monitoring patients at risk. Antibody detection may be useful for monitoring successful therapy and recovery rather than providing an early diagnosis. The detection of *Aspergillus* DNA in bronchoalveolar lavage fluid complements conventional methods. The clinical usefulness of enzyme immunoassays for the detection of circulating β-glucan in plasma and that of PCR assays for the detection of *Aspergillus* DNA in EDTA blood or in serum is under investigation. Dr. Kappe concluded that the detection of *Aspergillus* antigen (primarily galactomannan) may be helpful in establishing the diagnosis, but problems still remain with low sensitivity of most essays.

Hermann Einsele (Tübingen, Germany) summarised the experience with PCR-based diagnosis of *Aspergillus* infections from his laboratory. The early diagnosis of invasive fungal infections remains difficult. Culture and histopathology for the diagnosis of *Aspergillus* and other fungal infections have limited sensitivity and specificity. Cultures from blood are rarely positive in patients with invasive aspergillosis, and cultures from bronchoalveolar lavage fluid only become positive at advanced stages of disease. According to Dr. Einsele polymerase chain reaction (PCR) assays have emerged as powerful tools with high sensitivity and specificity for the diagnosis of a broad variety of fungal pathogens. In one study in 134 recipients undergoing allogeneic bone-marrow transplantation (allo-BMT), 261 of the 268 scheduled bronchoalveolar lavages were done, and each patient had at least one lavage at the time of

transplant. In seven of the 134 patients, lavage samples were positive on PCR for *Aspergillus*, whereas microscopy and culture were negative. Five of these seven patients developed invasive pulmonary aspergillosis at a median of 64 days after transplantation, confirmed by culture assay and histopathology. In a second study, to assess prospectively the potential value for early diagnosis of invasive aspergillosis after allo-BMT, blood samples were collected 2-4x/week and screened for the presence of Aspergillus. Out of 1193 whole blood samples, 169 were found PCR positive and 1024 PCR negative. Seven patients developed proven and one probable invasive pulmonary aspergillosis, all of whom were found PCR positive a mean of 21 days (range 2-45 days) prior onset of clinical symptoms. The sensitivity of the assay based on any positive PCR result was 100% and the specificity 64.6%. According to these data, PCR-based diagnosis for invasive pulmonary aspergillosis shows promise for further investigations. Moreover, these data have to be confirmed by other groups as well as there is a need for better standardisation of the procedures.



Courtesy E. Gueho

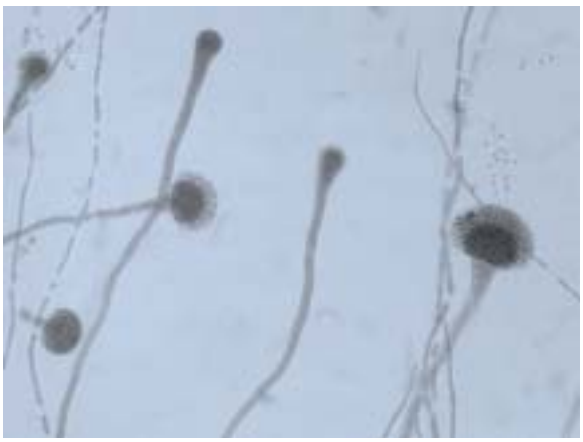
The usefulness of recombinant proteins for improving the serodiagnosis of *A. fumigatus*-associated diseases was shown by Michael Weig (Göttingen, Germany). Commercially available test systems are based on crude and undefined antigen mixtures of conidial, mycelial, cytosolic, metabolic, or cell wall fractions of *A. fumigatus* to detect specific antibodies in aspergillosis patients. Mitogillin is a small 18 kDa immunodominant protein that is secreted by *A. fumigatus*. This protein, which is a member of the ribotoxin family, was found in urine of patients who suffer from invasive aspergillosis. The gene encoding for mitogillin was amplified by PCR, subcloned into high-level expression plasmid pQE30 and used to produce large scale hexahistidine-tagged proteins. After affinity chromatography, the purified recombinant protein was used as a standardised antigen to detect IgG, IgM and IgA antibodies in serum samples of patients who suffer from proven aspergilloma, invasive pulmonary aspergillosis and invasive disseminated aspergillosis. A high correlation of IgG antibody production to mitogillin and the clinical course of disease in the respective patients was observed, indicating that recombinant technology might be useful for improving serological diagnosis.

The topic of prophylaxis against invasive fungal infections was taken up by Jörg Beyer (Marburg, Germany). Fungal infections are of increasing importance in severely neutropenic and immunosuppressed patients because of their high incidence and their high mortality once systemic dissemination has occurred. Various prophylactic strategies have been developed that include environmental measures as well as topical and systemic antimycotic prophylaxis. Specific strategies were to discussed for neutropenic patients in detail. Autopsy data did show that up to 25% of patients with leukaemia or undergoing allogeneic BMT have histological evidence of invasive fungal infections. However, all neutropenic patients with solid or haematological malignancies do not have the same risk for developing a fungal infection, which depends on the severity and duration of the neutropenic phase. Patient with short duration of neutropenia (less than 7-10 days) do not require antifungal prophylaxis. The population which should be considered for prophylaxis are the high risk patients with prolonged neutropenia (e.g. more than 10 days, allogeneic BMT, history of previous fungal infection). Despite the fact that numerous studies have been performed in this area there is still no definitive consensus on preventive modalities in these patients. In neutropenic patients *Aspergillus* spp. and *Candida* spp. are the most common fungal pathogens isolated. Since most studies fail to show conclusively that both invasive *Candida* and *Aspergillus* infections can be prevented, careful barrier nursing and prevention from environmental exposure to spores is essential for all neutropenic patients. In addition, even when studies do show a decrease of muco-cutaneous fungal infections and sometimes a decrease in the incidence of invasive fungal infections as well, most if not all studies fail to show an effect on survival of the treated patient population.

Amphotericin B (AmB) remains the antifungal agent with the broadest spectrum of action available and is thus the standard treatment for immunocompromised patients with proven or suspected fungal infections, especially aspergillosis. This statement by Markus Ruhnke (Berlin, Germany) reflects still the current state of the art therapy for invasive fungal infections. However, its potential for nephrotoxicity limits its usefulness. Lipid formulations of amphotericin B may allow therapy to be administered with reduced renal toxicity. Three different lipid formulations of amphotericin B are currently available: amphotericin B lipid complex (ABLC), amphotericin B cholesteryl sulphate (ABCD), and liposomal amphotericin B (L-AmB). These compounds have different pharmacokinetics properties and usually achieve higher serum and/or tissue concentrations than amphotericin B. At present, there are no studies comparing the lipid formulations with each other and only a few randomised trials comparing them with conventional amphotericin B (D-AmB). However, a number of open

clinical trials and compassionate-use protocols suggest that lipid-based formulations of amphotericin B can achieve good response rates with minimal toxicity in patients with a variety of invasive mycoses, including those who have proved refractory or intolerant to previous therapy with conventional amphotericin B. Unfortunately, the cost of these compounds remains very high and may represent a limiting factor in their use. In addition, the optimum daily and total dose of the lipid formulations has not been established. At present, these lipid formulations are indicated for patients with systemic mycoses, primarily invasive aspergillosis, who are intolerant of or refractory to conventional amphotericin B, defined as follows: (i) development of renal dysfunction with serum creatinine ≥ 2.0 mg/dL during therapy with conventional amphotericin B; (ii) severe infusion-related adverse events despite premedication regimens, and (iii) disease progression under conventional amphotericin B. Data from non-comparative studies suggest that nephrotoxicity is markedly decreased for L-AmB and ABLC but less with ABCD. Infusion-related adverse effects were observed as followed: D-AmB ≥ ABCD > ABLC > L-AmB. Furthermore, a recently completed but not yet published study by Wingard et al. comparing two lipid formulations (Randomised double-blind Comparative Safety Trial of AmBisome and Abelcet in Febrile Neutropenic Patients) presented at the 9th Focus on Fungal Infections in March 1999 in San Diego, California suggested that AmBisome at 3 mg/kg/day or 5 mg/kg/day was more successful and significantly less toxic than 5 mg/kg/day Abelcet. The authors observed with AmBisome less nephrotoxicity, fewer discontinuations due to toxicity, fewer infusion-related reactions, and a reduced requirement for medications to treat and prevent infusion-related reactions.

The early morning session was concluded by a review from Frank-Michael Müller (Würzburg, Germany) on «Triazoles and new antifungal agents for the treatment of aspergillosis». The emergence of *Aspergillus* infections in patients undergoing high dose chemotherapy, allogeneic BMT, lung transplantation or AIDS with a mortality of up to 81% was outlined. The overall response rate of AmB in invasive aspergillosis is no more than 55%, but as stated by other referents AmB remains the agent of choice for initial therapy of invasive aspergillosis. Itraconazole may be an alternative option, but the lack of current data do not support the routine use. New potent compounds with a broader spectrum, fungicidal activity, reliable safety, tolerance and efficacy data are warranted. Several new systemic antifungals are currently in clinical trials such as 1) polyene antibiotics (Liposomal Nystatin: Nyotran™ from Aronex) 2) 1,3-β-Glucan-synthase-Inhibitors (LY 303366 from Versicor, MK-0991 from Merck, FK463 from Fujisawa) and 3) new triazoles (voriconazole = Vfend™ from Pfizer, SCH56592 from Schering-Plough, BMS-207147 from Eisai/Bristol-Myers Squibb). The liposomal formulation of Nystatin (Nyotran™) provides a broad antifungal spectrum with good *in vivo* activity in neutropenic mice and rabbits. High concentrations in spleen and kidney could be achieved with the drug and a trend towards reduced renal toxicity was observed in the animal model. The benefits over conventional AmB/liposomal AmB need to be established. The new triazoles voriconazole, SCH-56592 and BMS-207147 show a comparable or superior *in vitro* and *in vivo* activity to fluconazole, itraconazole, and AmB (SCH-56592 > voriconazole > BMS-207147). Of interest is the good oral bioavailability concerning voriconazole. At the moment, data are not clear concerning cross-resistance with other azoles as well as the pharmacokinetic and pharmacodynamic profile. Furthermore, safety, efficacy and tolerance data are lacking to determine actual benefits. The new drug group of the Echinocandins with the representatives LY-303366, MK-0991, and FK463 have a different mode of action, because they interfer with the cell wall. Excellent *in vitro* and *in vivo* activity against *Aspergillus* spp. (FK463 > LY-303366 > MK-0991) have been demonstrated. Favour-able pharmacokinetics with the iv. formulation has been shown, but the oral bioavailability is very poor. Phase II / III - studies are ongoing with these agents with MK-0991 (Cancidas™) as the most advanced studied agent. At the moment, an evaluation as single agents for proven or suspected invasive aspergillosis in the neutropenic host as well as for invasive candidosis is under way. At the moment, we do not know the safety, efficacy and tolerance data to determine actual benefits. Some new systemic acting antifungals are in preclinical or clinical studies such as:



1) P-Glykoprotein-Inhibitor = Aureobasidin A (LY 295337),
2) Protein-Synthesis (Inhibition of translation EF-2),
3) Synthetic Sordarine derivatives (e.g. GM 237354),
4) Virulence factors (Phospholipases, Aspartate-Proteinases), and
5) Antimicrobial Peptides (Cecropin, Defensin, Mycoprex™).

The clinical benefit and their role in the antimycotic armentarium of all these agents have to be determined. However, the times of limitation in the spectrum of antifungal agents are going to change soon.

Markus Ruhnke

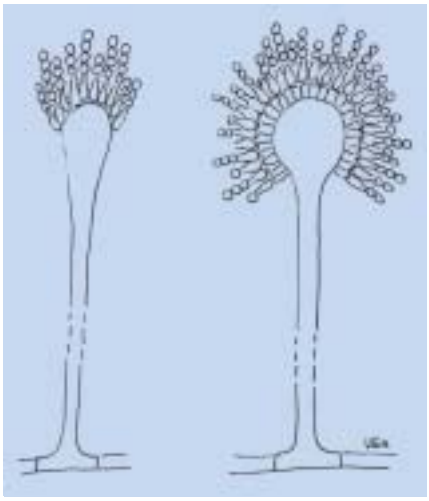
Session 5

New developments in the treatment of aspergillosis

As Mahmoud A. Ghannoum (Cleveland, USA), who was originally invited to give this lecture, was unfortunately not able to attend the conference, Jacques Bille (Lausanne, Switzerland) discussed in his talk «Antifungal *in vitro* testing - Current status» options and problems of *in vitro* antifungal testings. Central aspects such as solubility of the test substances, end points for MIC determinations, inoculum size, medium compositions and superficial sporulation of moulds in dilution assay methods were highlighted. In contrast to these experimental problems it was shown that there is an urgent clinical need for standardization of the antifungal *in vitro* testing methods especially for moulds such as *Aspergillus fumigatus*. For the *in vitro* testing of yeasts, standard methods such as the NCCLS-27 A standard have already been established. Even if standardized procedures have been described, the evaluation of valuable breakpoints for the susceptibility discrimination, and the assessment of the *in vitro*/*in vivo*-correlation will be further questions to be evaluated e.g. by animal testings and/or clinical studies.

In her talk «Components of the *Aspergillus* cell wall as potential drug targets», Veronica M. Hearn (Leeds, UK) pointed out that the major structural components of the cell wall of *Aspergillus*, namely glucans and chitin, are the target for many antifungal drugs. Much of the interest has focused on the enzymes involved in their synthesis i.e., chitin and glucan synthases. Nikkomycin treatment showed decreased chitin synthase activity and a decrease in cellular chitin, resulting in a change in fungal morphology of *A. fumigatus*. Combining nikkomycin with itraconazole increased inhibition of this enzyme. There is thought to be at least six different CHS genes in *A. fumigatus*. Only with the chsG- mutant is a delay seen in the onset of aspergillosis in neutropenic mice and there is a decrease in mortality, when compared with the wild type strain. Because of the functional redundancy of these enzymes it is difficult to entirely destroy their ability to synthesise chitin. Potentially more effective are the drugs which target glucan synthase. Cilofungin was the first echinocandin B derivative developed for clinical trials but, due to the toxicity of the carrier vehicle, its use is currently in abeyance. A new generation of these lipopeptide antifungal agents including LY303366 and MK-0991 have shown high potency against *Aspergillus* species. No significant differences in *in vitro* antifungal activity of LY303366 and MK-0991 have been found. *In vivo*, the therapeutic activities of these compounds were comparable to that of amphotericin B in animal models of aspergillosis, and MK-0991 is currently in use, using the Merck protocol, against patients with invasive aspergillosis. However, it has been reported that LY303366 showed lethal toxicity in mice pretreated with cortisone acetate. The clinical relevance of these findings in steroid treated patients to the clinical safety of this compound and other echinocandins needs to be determined. In recent studies, a new injectable echinocandin, FK463, has been shown to be effective in treating disseminated aspergillosis in a murine model. Echinocandins provoke remarkable quantitative changes in the main glucan associated protein constituents of the cell wall.

Axel Schmidt (Wuppertal, Germany) presented the topic «Animal models of aspergillosis - also useful for vaccination strategies?». Vaccination strategies against infections caused by *A. fumigatus* do not seem to be of real benefit. They will not have a setting in mycotoxicoses, allergic disorders or circumscribed infections/colonisations caused by *Aspergillus* species. In contrast, invasive aspergillosis is a disease of the severely immunocompromized host and in these cases, if however, only a passive immunisation with immunoglobulins would be taken into consideration. Until now there are no preclinical and/or clinical data available concerning the efficacy of specific immunoglobulins; animal testings could offer an approach for the assessment of this topic. Animal testings in general are under a high public pressure and FRAME (Fund for the Replacement of Animals in Medical Experiments) offers the 3R strategy: "Refinement", "Reduction" and/or "Replacement". These aspects have to be taken into consideration also for the performance of animal testings in infectiology. *A. fumigatus* is an opportunistic pathogen although birds show a relatively high susceptibility to infections caused by *A. fumigatus*; the reasons are almost not understood until now. In laboratory animal species, rabbits have the highest susceptibility towards *A. fumigatus* infections followed by mice, rats and guinea pigs. Mice are the animal species most often used since they are easy to handle. Infections in mice can be established by the intravenous, intranasal and intraabdominal route. All of these three infection routes result into an infection process which is almost restricted to the kidneys as main target organ. Intravenous infections show an excellent infection-dose/mortality ratio and do not require immunosuppression of the animals. These models are extremely valuable e.g. for the evaluation and development of investigational new antimycotics and/or vaccination strategies. Intranasal infections in mice are difficult to establish or standardize. Animals need a pre-



ceeding immunosuppression and different individual animals do not react reproducibly to these regimens. Further, lethal anaerobic septicaemia, mostly due to *Bacteroides* species, was observed under these immunosuppression regimens. Therefore, intranasal infections in mice can not be recommended as first choice in order to assess pathogenicity and/or virulence factors of *A. fumigatus* strains. Mice are primary resistant towards intraperitoneal application of ungerminated *A. fumigatus* conidia. However, it is a prerequisite, that *Aspergillus* conidia have to be pregerminated before intraperitoneal administration. Histopathological examinations are of great benefit in the study of these animal models as they give detailed information about the infectious process. Measuring colony forming units in tissues is a method of limited predictability as it cannot discriminate between infective tissue lesions and cavity infections, e.g. in the kidney pelvis. Quantitative methods for measuring fungal organ burdens, e.g. by chitin-ELISA methods, are also performed and are an alternative towards solely measuring cfu's in tissues.

In his talk «Genome Project *A. fumigatus*: When will it be done?», David W. Denning (Manchester, UK) gave an update of the current status of the *A. fumigatus* Genome Project. The significant participants in this project and their results and contributions to the project were detailed. Afterwards, estimated timelines for the continuation of the project and the corresponding responsibilities of the participating research units were highlighted. In conclusion, this project offers visible approaches for understanding pathogenicity factors of *A. fumigatus* and will also be a fundamental tool for defining new targets in antimycotic reasearch.

After the closing remarks from Hans C. Korting (München, Germany), President of the Deutschsprachige Mykologische Gesellschaft, a Round Table Discussion with Hans C. Korting, Reinhard Rüchel, Markus Ruhnke and Michael Weig - chaired by Uwe Gross – took place. The discussion was mainly focused on environmental questions concerning moulds as well as allergic disorders and food spoilage.

Axel Schmidt

Chronic mucocutaneous candidosis: an invitation to collaborate in establishing a European database

The best way of approaching an understanding of the genetic basis of chronic mucocutaneous candidosis (CMC) is to study a suitable cohort of families where there are both affected and unaffected individuals. The purpose of this enquiry is to set up a database on CMC in Europe in order to establish whether there are sufficient numbers of patients with CMC and associated disorders to justify a study of the genetic basis of the disease.

I would be very interested to hear from anyone who has any patient with CMC with or without a family history or who can pass on the request to a colleague. I can send them by e-mail (or fax) an appropriate short questionnaire in order to establish the numbers of patients in each clinical category who might form the basis for a pan-European study of susceptibility to CMC.

At this stage we would hope to establish whether there are sufficient numbers of patients and the interest to take the project further. Any expression of interest at this stage does not commit you to further involvement!

Roderick J. Hay
St John's Institute of Dermatology, St Thomas Hospital, London SE1 7EH, UK
e-mail: r.hay@umds.ac.uk
fax +44 171 960 5802