The Fourth NIAID Workshop in Medical Mycology: Host Responses to Fungi

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PREFACE

The NIAID recognizes medical mycology as an area in need of development. An Institute-sponsored workshop on "Mycology Research in the 1990's" in Chicago, Illinois, 28-29 September 1991 addressed the increasing importance of medical mycology. Twenty medical mycologists from throughout the United States were invited to discuss the issues and to conceptualize and condense the active research areas into topic areas in need of development.

Five areas were targeted for focus. These were molecular mycology, diagnosis and treatment, immunology, antigen structure and function, and epidemiology. Each of these five topic areas was targeted for development into a separate workshop/minisymposium, co-sponsored by the NIAID and educational grants raised by the medical mycological community.

"Molecular Medical Mycology," the first workshop in the series, was held in Minneapolis, Minnesota on 24-26 June, 1993 and chaired by Dr. Paul T. Magee. One hundred and forty-seven mycologists attended and exchanged ideas. A key to the success of information exchange was the utilization of "break out" sessions that provided an informal setting for free exchange of ideas, an opportunity for a more active involvement for all of the participants, and an environment fostering new collaborations.

"Molecular and Immunologic Approaches to the Diagnosis and Treatment of Systemic Mycoses," the second workshop in the series, was held on the campus of Northern Arizona University, Flagstaff, Arizona, 8-11 June 1994. The workshop format was modeled after the first in the series, and was attended by 80 registrants. Drs. John Galgiani and Michael Pfaller chaired this event whose findings are still timely.

"Immunology in Medical Mycology (Part 1 of 2): Antigenic Peptides, Glycobiology and Vaccines," the third workshop in the series, was held at the Yellowstone Conference Center, Big Sky, Montana, 7-9 September 1995. The workshop format was modeled after the first two in the series and was attended by 90 registrants. Drs. Rebecca Cox, Jim Cutler, and George Deepe the workshop whose findings were summarized in ASM News 62;81-84, 1996.

"Immunology in Medical Mycology (Part 2 of 2): Host Responses to Fungi," the fourth workshop in the series, was held at Granlibakken Conference Center, Lake Tahoe, California, 20-23 August 1997. The workshop format was modeled after the previous three in the series and was attended by 75 registrants. Drs. Thomas Kozel and Juneann Murphy co-chaired the workshop along with session chairs Drs. Arturo Casadevall and Jack Sobel.

I believe the workshop series continues to be successful in accomplishing the stated goals, and that this success is representative of the field of medical mycology whose time has come. Finally, I would like to thank all those who contributed to the success of these workshops, including all of the participants, organizing and writing committees.

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INTRODUCTION

The workshop was designed to expand the network of investigators working with immunological systems in medical mycology; interface with investigators from other fields for interactive learning; identify the immune mechanisms/components preventing invasive fungal disease in humans; and, identify means of prevention and treatment of invasive fungal disease. Eleven theme and ten research presentations in five sessions reported representative research approaches in the field, and several presentations highlighted parallel developments in related fields. Seven "break out" sessions of approximately 9 participants each were led in discussion by facilitators who summarized the results in separate "at large" sessions.

Key Concepts

- Experimental results and clinical findings are often discordant
- The limitations of animal models must be realized
- Laboratory based advances must be translated into clinical advances
- Successful advance to clinical studies requires building upon initial findings
- There is a growing body of evidence to suggest that immune-based intervention and prevention strategies should be applied to the mycoses
- Fungal latency and reactivation of disease need more study
- The initial events in fungal infection need more study
- Fungal genome projects are needed for key fungal pathogens

Current Status

Induction of activated T cells is an important first step in adaptive protection. For development of activated T cells and a strong immune response, an antigen must either have adjuvant characteristics of its own or it must be introduced in association with an adjuvant. The immune response induced under the control of the adjuvant properties of a microorganism or its products can be protective or nonprotective. Consequently, understanding the adjuvant effects of the microbe can greatly influence understanding of the disease. The influence that each adjuvant has on the type and intensity of an immune response is critically important for vaccine development as well.

Soluble peptide antigens such as ovalbumin given alone to mice will stimulate antigen-specific T cells to proliferate transiently, and the surviving antigen-specific T cells when restimulated with antigen are impaired in their ability to produce lymphokines. In contrast, the same soluble antigen given with the adjuvant, lipopolysaccharide (LPS), will stimulate a more intense proliferative response in the antigen-specific T cells than is seen with the antigen alone, and the T cells activated by the antigen-LPS combination acquire the capacity to transfer delayed-type hypersensitivity (DTH), produce IFN-g , and help antigen-specific B cells produce IgG2a antibodies. The adjuvant effects of LPS can be mimicked by the combination of IL-1 and IL-12 and can be inhibited by blocking CD28-B7 or CD40-CD40 ligand interactions. These findings suggest that peptide antigens given in combination with LPS induce a T helper 1 (Th1) response. Other adjuvants may influence the system differently and shift the immune response to a Th2 response.

It is clear that in order to develop a cell-mediated immune (CMI) response which is the key protective immune response against systemic fungal pathogens, adjuvants that set the stage for induction of a CMI response must be present as a part of the organism or in the immunogen preparation. Such adjuvants will have to induce proinflammatory cytokines (IL-1, IL-6, TNF- a) plus IL-12 and IFN- g as well as upregulate B7 molecules on antigen presenting cells (APC). Furthermore, such adjuvants may also need to enhance the expression of CD40 ligand on activated T cells and of CD40 on APC including B cells.

Pathogenic fungi are complex organisms with numerous chemical constituents that have the potential to act as adjuvants. We must define the protective immune responses against each mycotic agent and then gain an understanding of how the adjuvant characteristics of the organism influence the induction of that protective response. Once we have gained an understanding of the adjuvant characteristics of each fungal agent and learned how the organism's adjuvants affect the host's protective immune responses, we will be better able to apply immunomodulatory or immunoreplacement therapy in patients suffering from a mycotic disease and to develop fungal vaccines where practical.

Clearance of intracellular bacteria is dependent on conventional and unconventional T cells. Considering that clearance of fungal pathogens is also highly dependent on T cell responses, the lessons learned from studies with intracellular bacteria may be applicable to research on immune responses against mycotic agents. The location of the infectious agent or its antigens in the host's APC can direct the type of conventional T cells that are activated. For example, if the organism or antigen(s) is endosomal then $CD4^+$ T cells are typically activated because the microbial antigens are presented on Class II major histocompatibility complex (MHC) molecules to $CD4^+$ T lymphocytes. On the other hand, if the organism or its components gain entrance into the cytosol of the APC, then $CD8^+$ T cells are activated via interactions of the antigenic peptide on MHC Class I molecules on APC with the T cell receptor (TCR) on $CD8^+$ T cells. Unconventional T cells, such as $CD8^+$ a / b TCR, MHC Class I-like restricted; $CD4^-$, $CD8^-$, a /b TCR, CD1-restricted; $CD4^-$, $CD8^-$, g /dTCR, direct recognition; and $CD4^+$, $NK1^+$, CD1-restricted T cells, can be induced and may have important roles in the protective immune network that functions in clearance of a pathogen.

Although the general dogma has been that MHC Class II-restricted $CD4^+$ Th cells protect against bacterial and mycotic pathogens and MHC Class I-restricted $CD8^+$ T cells control viral infections and malignancies, it is now well established that MHC Class I-restricted $CD8^+$ T cells provide protection against intracellular bacteria. Moreover, the previous thinking was that T cell receptors recognized peptides in MHC molecules but not carbohydrates, lipids, or oligonucleotides; however, now it is known that specialized MHC Class I-like molecules such as CD1 can present lipids and glycolipids of *Mycobacterium* to $CD4^-$, $CD8^-$, a /b TCR T cells. In addition, it has recently been shown that N-formylated peptides are presented by MHC Class I-like molecules to $CD8^+$ T cells. Peripheral g /d T cells expressing V g 9Vd 2, irrespective of their fine specificity, respond to microbial ligands such as nonproteinaceous molecules containing phosphate possibly coupled to alkyl derivatives or to certain carbohydrates or nucleotides. Another unconventional T cell that possibly biases the immune response toward a Th2 response is the $NK1^+$ T cell. This T cell subset has both NK cell markers and T cell markers and has been reported to produce IL-4 which favors Th2 cell differentiation and IFN- g which favors Th1 development. If IL-4 production predominates when $NK1^+$ T cells are induced as it does in many cases, then the protective immune responses against intracellular bacteria and fungal pathogens would not be induced.

The cells and cytokines present during the development of a T cell response influence the developmental pathway that the T cells take. For instance, in the host response against *Listeria monocytogenes*, a CMI response is induced and the response can be separated into three stages in which the cells can provide effector and regulatory functions. The early or initial stage of the response involves neutrophils, macrophages and dendritic cells along with chemokines and proinflammatory cytokines such as IL-1, IL-6, TNF-a and IL-12. At the intermediate phase of induction of the Th1 cells, NK and g /d T cells can be induced to produce IFN-g. These latter cytokines along with the appropriate presentation of antigen to the T cells and the ligation of costimulatory molecules such as CD28-B7 will promote the induction of a strong Th1 response. The late phase or expression phase of the CMI response would be characterized by increased production of IL-2, IFN-g , TNF-b and possibly cytotoxic T cells.

Coccidioides immitis, the etiological agent of valley fever, is an excellent example of a systemic fungal pathogen that induces a protective CMI response. With the murine model, it is clear that

the host's immune response to *C. immitis* can vary depending upon the genetic characteristics of the host. For example, if the susceptible BALB/c and the resistant DBA/2 mouse strains are infected with 200 *C. immitis* arthroconidia by the pulmonary route, both strains become infected and an anti-*Coccidioides* DTH response can be detected by day 12 of the infection. The DTH response levels in the BALB/c mice fall after day 12 of infection, but the levels of DTH responsiveness of the DBA/2 mice remain high. Also at day 12, lung homogenates from DBA/2 mice have increased concentrations of IFN-g ; whereas similar homogenates from BALB/c mice have elevated IL-4 levels as compared to levels of the respective cytokine at day 6 of infection. In the DBA/2 mice there is a well-defined granulomatous response in the lungs and an increase in T cells with a g /d TCR and in $CD11c^{+}$ $DX5^{+}$ (NK) cells. In contrast, the BALB/c mice have less well-defined cellular responses in the lungs with an increase in B cell numbers as compared to DBA/2 mice. Both mouse strains infected with the same *C. immitis* isolate develop an anti-*Coccidioides* CMI response, but the early down-regulation of that protective CMI response and shift to a Th2 response in the BALB/c mice are the most likely predisposing changes responsible for the concomitant reduction in leukocyte influx into infected lungs and for the diminished protection. These findings emphasize the importance of genetic background of the host in development and maintenance of a protective immune response.

One approach to gaining an understanding of the mechanisms of adaptive antimicrobial immunity for the purpose of devising appropriate immunotherapeutic strategies has been to identify and characterize the immunodominant antigens. In candidiasis, the immunodominant antigens have rarely been the antigens that induce protection. For example, enolase, HSP70, and mannoprotein are immunodominant antigens of *C. albicans*, but do not induce protection. On the other hand, the yeast killer toxin receptor (YKTR) is a low-dominance antigen that induces a protective immune response that is primarily due to candidacidal antibodies. A seldom considered but possibly important component of protection against *C. albicans* is the activation exerted by some polysaccharides of *C. albicans* on neutrophils to produce cytokines that influence T cell responses. Like many other fungi, the immune responses to *Candida* are complex, and to gain a thorough understanding of the protective components, some unconventional thinking and novel approaches will be required.

Although it is recognized that T cells are critical for protection against *Cryptococcus neoformans*, antibodies (IgM, IgA, and IgG2) that bind to cryptococcal glucuronoxylomannan (GXM) have been identified in both HIV-infected and uninfected individuals with cryptococcosis. The role of these antibodies in the pathogenesis of human cryptococcosis is unknown. In serum from individuals immunized with GXM conjugated to tetanus toxoid (GXM-TT), the majority of the antibodies to GXM express VH3 idiotypes that bind the superantigen, staphylococcal protein A (SPA). Anti-GXM antibodies from HIV-infected individuals may have different epitope specificity than naturally occurring (pre-immune) and GXM-TT-elicited GXM antibodies. Sera from GXM-TT vaccinees promote phagocytosis of *C. neoformans* by human mononuclear cells. This phenomenon is mediated by human Fcg RIIa and IgG2 antibodies. These findings suggest that the status of the immunoglobulin repertoire may play a critical role in human antibody responses to GXM. Enhancement of effector cell antifungal activity by human GXM antibodies suggests that some antibodies might alter the course of human cryptococcosis. Taken together, these observations indicate that human antibodies might play a role in preventing infection in immunocompetent hosts, and represent useful therapeutic reagents to add to our armamentarium against cryptococcosis in immunocompromised patients.

Recommendations

- Define the adjuvant properties of fungal agents and their products.

- Efforts should be directed toward gaining a more detailed understanding of the cells, cytokines, chemokines and costimulatory molecules involved in protective and non-protective immune responses against each of the fungal pathogens. Emphasis on

Th1/Th2 dichotomy seems too great and more attention should be directed toward gaining an understanding of the full complexity of the protective mechanisms.

- Establish how genetics of the host and genetics of the organism influence the protective responses.
- Acquire knowledge of how the host recognizes or responds to various fungal proteins, glycoproteins, polysaccharides and glycolipids.
- Relate findings from normal host to the compromised host.
- Define how fungal pathogens evade the immune responses.

INFLAMMATION AND GRANULOMA

Current Status

Inflammation and granuloma formation are two host tissue responses that are characterized by infiltrating leukocytes and that are necessary for clearance of microbes from tissues. Cytokines that play major roles in cellular infiltration into tissues are referred to as chemotactic cytokines or chemokines. Chemokines are grouped into 4 subfamilies based on structure-function relationships. The subfamilies are: 1) C-X-C chemokines which have the first two cysteines on their N-terminus separated by one other amino acid, 2) C-C chemokines which have two adjacent cysteines, 3) C-chemokines that have at present two members lymphotactin and SCM-1, and 4) CX_3C chemokines with fractalkine as a single member of the subfamily. Besides being chemotactic for certain leukocyte subsets, chemokines have other functions. They can cause degranulation of certain leukocytes, they can activate natural effector cells, and they can stimulate angiogenesis or angiostasis. Indirectly, chemokine activity can result in modulatory effects on the immune responses. With such an array of capabilities, chemokines are prime candidate molecules for being involved in pathogenesis of infectious diseases. The function of chemokines is dependent on receptors for the chemokine being on the cells they stimulate. Consequently, modulation of the chemokine receptors can affect the function of the chemokine. Chemokine receptors also have been found to be docking sites for viruses such as HIV. In fact, HIV can gain entrance into cells through certain chemokine receptors such as CXCR4 on lymphocytes and CCR2B, CCR3, and CCR5 on monocytes.

Since chemokines and chemokine receptors play important roles in innate as well as in acquired immune responses, it seems reasonable to predict that chemokines and their receptors would be essential in host defenses against fungi. Recently published studies have shown that indeed this is the case. Three C-C chemokines, MCP-1, MIP-1a , and TCA3 have been studied in a murine cryptococcosis model. MCP-1 and MIP-1ahave been shown to affect migration of leukocytes in *C. neoformans* infections and also modulate clearance of the organism, and TCA3 has been shown to be elevated in the anticryptococcal CMI response. Like TCA3, MIP-1a levels are increased at the site of an anticryptococcal DTH reaction. TCA3 and MIP-1a recruit neutrophils and lymphocytes into an anticryptococcal DTH reaction site. Mice that have anticryptococcal CMI responsiveness show reduced ability to clear *C. neoformans* when MIP-1a is neutralized. After an intratracheal infection with *C. neoformans,* MCP-1 is involved in recruitment of CD4± T cells and monocytes into the infected lungs of mice; whereas, MIP-1a recruits neutrophils and monocytes/macrophages. Neutraliza-tion of MCP-1 significantly decreases MIP-1alevels in the lungs; however, neutralization of MIP-1a has no significant effect on MCP-1 suggesting that MIP-1a is largely dependent on MCP-1 production. Animal models which have been so valuable in defining and understanding the role of a limited number of chemokines in host defense against fungal agents are expected to be useful in future investigations on chemokines and chemokine receptors in protection against infectious organisms.

An important focus in infectious disease research is the antimicrobial capabilities of the leukocytes within tissues. The ability of any given leukocyte population to kill or inhibit the growth of a fungus is dependent to some degree on: i) the fungus, ii) the form of the organism, iii) the anatomical site of origin of the leukocyte, iv) the local environment including other types of infiltrating cells, cytokines, and chemokines in the tissues where the effector cells are found, v) the health or disease state of the host, vi) the genetic background of the host, and vii) the immune status of the host. When studying various effector cells and their abilities to inhibit fungal growth, it is important to consider how species differences in the source of the effector cells may influence the results. If studying the effector cells *in vitro*, one must consider the matrix on which the effector cells are found in the body and attempt to simulate that *in vitro*. When studies are done *in vitro*, one must consider the influence of other leukocytes or host cell types on the activity of the effector cells being studied. Natural effector cells from patients with an underlying disease or a concomitant infection with another agent such as HIV may be compromised in some of their activities against fungal pathogens and that must be considered when trying to draw conclusions from data obtained with effector cells from normal or control subjects. Learning how to augment the antifungal activity of effector cells such as monocytes could be valuable for immunotherapy. Choloroquine, a drug that raises the pH of phagolysosomes, looks promising for this purpose because it has been shown to enhance the anticryptococcal activity of mononuclear phagocytes from both HIV-infected and uninfected individuals. Effector cell function is a critical element in host defense, so consequently deserves much further attention.

Endothelial cells lining the blood vessels serve to prevent or enhance movement of leukocytes or blood-borne organisms from entering tissues. Consequently, endothelial cells are important gatekeepers in inflammation, granulomas and entrance of microorganisms into the tissues. In the region of infection, the endothelial cells change their surface receptors and are referred to as inflamed endothelium. The surface markers on leukocytes interacting with the surface of inflamed endothelium along with chemokines in the infected tissues encourage the migration of leukocytes into the infected tissues. During the process of hematogenous dissemination of fungal pathogens, the organisms most likely interact with endothelial cells. These interactions can stimulate the endothelial cells to produce chemokines such as IL-8 and express adhesion molecules such as E-selectin, VCAM-1 and ICAM-1 which are necessary for extravasation of the leukocytes into tissues. In the case of *Candida albicans*, the process of stimulation of the endothelial cells requires phagocytosis of live germinating *C. albicans*. The mechanism whereby *C. albicans* induces endothelial cells to produce IL-8 and express E-selectin is different than the mechanisms for *Candida*-induced expression of VCAM-1 or ICAM-1. It is clear that these mechanisms impact on the pathogenesis of the disease, so it is necessary to fully understand the different mechanisms involved in leukocyte and organism movement from the blood stream to tissues. Once the understanding is available it may be possible to selectively alter the endothelial cell responses to enhance host defenses.

Recommendations

- Gain an understanding of mechanisms of effector cell trafficking into tissues infected with the fungal pathogens in normal compared to immunocompromised hosts.
- Determine how the organism modulates trafficking of the various leukocyte populations.
- Establish how effector cells are modulated for intra- and extra-cellular killing of the various fungi.
- When using animal models, it is necessary to show that the model relates to the human infection.

MUCOSAL AND LUNG RESISTANCE IN INFECTIOUS DISEASE

Current Status

Most systemic fungal infections are acquired through the respiratory route, then the organism moves to extrapulmonary sites in the body. Frequently the organisms localize in certain tissues where they remain until cleared by the host. Consequently, understanding the protective mechanisms that function in the tissue where the organisms reside and how those mechanisms can be modulated to protect the host are of special interest.

Many organisms enter the body through mucosal surfaces, so it is understandable that the innate and immune defenses of the mucosa are key to whether or not establishment of disease is prevented and how the disease is limited once established. Recent studies have shown that intranasal administration of vaccines is a particularly effective way to induce mucosal immunity in the airways as well as in the female reproductive tract. Delivery of protein antigens along with a mucosal adjuvant via the oral or intranasal routes can induce different types of immune responses depending on the adjuvant used. Cholera toxin as the adjuvant with the antigen of interest induces Th2 type responses resulting in mucosal IgA and serum IgG1 and IgE antibodies. In contrast, other adjuvants such as C fragment (Tox C) of tetanus toxoid or *E. coli*-labile toxin (LT) in combination with antigen induce a brisk CMI response, serum IgG2a antibody, and a mucosal IgA response. Careful selection of mucosal delivery systems and adjuvants have the potential to develop the type of protective immunity necessary for good host defense against many microbes and should be considered in future studies directed at protection against fungal agents.

From studies with *Candida albicans*, it is apparent that systemic immunity does not necessarily relate to immunity or protection at the vaginal mucosa. Innate and immune responses in the vagina have some unique characteristics and clearly differ from host responses in other tissues. For instance, vaginal T lymphocytes are phenotypically different from peripheral T cells. Vaginal mucosa has a high percentage of g /d TCR+ cells, lacks detectable $CD8^+$ T cells, and has $CD4^+$ T cells that express atypical CD4 molecules. Vaginal epithelial cells from mice, primates and man can inhibit the growth of *C. albicans*; but in pseudoestrus conditions these inhibitory capabilities are diminished. The commensal relationship of *C. albicans* in the vagina is hormonally regulated and controlled by innate effector mechanisms and host cells with unique characteristics. Regional protective responses tend to have specialized cells and control mechanisms which often differ from the commonly defined systemic immune responses. The currently available data emphasize the need to explore the characteristics of the regions of the body which harbor the organisms and to determine the regional features that limit the growth of the organism.

The lung is another epithelial surface that interfaces with the environment and consequently is extremely important in protection against fungal diseases. The lung is also a regional tissue that has some specialized host defensive mechanisms. To protect the host from infection with the many microbes that are inhaled, the lung has developed multiple means to avoid infection. However, to prevent those defensive mechanisms from causing unnecessary tissue damage, both the nonspecific or innate responses as well as immune-mediated inflammation must be regulated. The desire to enhance protective pulmonary immune responses by vaccination and to prevent or control unwanted hypersensitivity responses underlie the need to understand basic innate and immune mechanisms in the lung.

The host responses to infectious organisms entering the lungs can be divided into three phases. The afferent phase that includes the uptake and processing of the organism or its antigens by regional cells (alveolar macrophages and dendritic cells) and leukocytes (neutrophils and monocyte/macrophages) that influx in response to the infection. The second or central processing phase may take place in the regional lymph nodes and is defined by antigen presentation by APC such as dendritic cells or macrophages to the T cells which clonally expand, differentiate, and

regulate B cells to expand and differentiate. The final phase, the efferent phase, encompasses both lymphocyte emigration into the site of unresolved infection, followed by activation of natural effector cells in the lungs and elimination of the infectious agent. A thorough understanding of each of these phases and how they relate to clearance of the different pulmonary fungal pathogens are necessary if immunomodulatory therapy is ever to be applied.

Pneumocystis carinii (PC) is an opportunistic fungal agent that causes PC pneumonia in immunocompromised hosts. Since this organism is primarily limited to the lungs, investigations on the host resistance mechanisms effective against PC allow us to dissect the various regional protective mechanisms. It is thought that PC is a highly adaptable organism which survives in and colonizes the immunocompetent host, but does not cause disease until the hosts' $CD4^+$ T cells become diminished or have reduced function. It is not clear how $CD4^+$ T cells, PC-specific antibodies or $CD8^+$ T cells contribute to resistance. Investigations with the murine model of PC pneumonia have shed some light on the key host responses and the deficiencies necessary for the establishment of infection. Although the mouse PC differs from human PC, it is anticipated that the studies on mouse PC will result in information useful in planning studies in humans.

Recommendations

- Future studies should be directed at gaining a better understanding of host protective mechanisms in the infected tissues.
- Establish means for measuring regional protective responses.
- Defining the relationship, if any, between regional protection and systemic immunity.
- Understanding compartmentalization of the protective immune response and how vaccines can direct the protective response to the appropriate compartment are challenges worthy of future investigations.

EMERGING TECHNOLOGIES FOR APPLICATION TO STUDIES IN HOST RESISTANCE

Current Status

The purpose of the session was to explore immunological technologies that have been developed in model systems and to examine the potential application of these technologies to studies of the host response to fungi.

Two approaches to genetic analysis of the host response were considered. Gene deletion mouse mutants with defined deficiencies in the immune system are valuable tools for understanding the anti-infective immune response in an *in vivo* setting. Classical gene targeting approaches have allowed for stable deletion of genes responsible for cell surface molecules, cytokines or intracellular signaling molecules. Results from such knock-out mice have the potential for identifying the role of targeted genes in the host response but also must be interpreted with caution due to compensatory effects. Thus, as a general rule, infection experiments with gene deletion mutant mice reveal an essential rather than a conditional role of a given molecule or cell.

An alternative approach to genetic analysis of the immune response is use of recombinant congenic strains of mice. Recombinant congenic strains transform a multigenic difference between two inbred mouse strains into several sets of oligogenic differences which are amenable to analysis. Recombinant congenic strains have been used to map a number of genes controlling susceptibility to various types of cancer, infectious diseases, metabolic diseases, and T lymphocyte reactivity.

Identification of appropriate epitopes for immunologic targeting of fungal infections poses a particularly difficult challenge. Identification of protective monoclonal antibodies and a determination of the epitope recognized by the antibody offers particular promise. Peptide phage display libraries have been applied to identification of epitopes recog-nized by monoclonal anti-glucuronoxylomannan antibodies that are pro-tective in a murine model of cryptococcosis. Identification of peptide mimotopes is a valuable strategy because the peptide mimotopes are potential immunogens and can be used to analyze the crystal structure of the interaction of the peptide and the protective monoclonal antibody.

In many cases, epitopes that elicit protective immune responses to the fungi are unknown. One promising approach to the problem is the application of genetic and genomic vaccine technologies. Mice have been immunized with expression libraries containing the entire genome of a pathogen. Challenge of mice with the pathogen allows identification of libraries that contain plasmids encoding for protective antigenic epitopes. In this way, expression library immunization provides an unbiased, systemic approach for isolating vaccine candidates.

Finally, the cellular immune response to fungal pathogens is a critical component of host resistance. Emerging technologies used to define T cell determinants have the potential to greatly enhance our understanding of the immune response to pathogenic fungi and may facilitate vaccine design.

Recommendations

- Emphasize identification of immunoprotective antigenic epitopes, with special consideration of novel technologies that might simplify and clarify the importance of candidate immunogens in the protective response.

- Develop collaborations between immunologists who work with model systems and medical mycologists.

- Encourage immunologists who work with model systems to consider studies of complex microbial pathogens, including the pathogenic fungi.

TOPIC SUMMARY AND SPEAKERS

Induction and Expression of the Immune Response

Chairperson: Arturo Casadevall, M.D., Ph.D., Albert Einstein College of Medicine
Marc Jenkins, Ph.D., University of Minnesota
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Immunomodulation

Chairperson: Jack D. Sobel, M.D., Wayne State University
Judith E. Domer, Ph.D., Tulane University
Juneann W. Murphy, Ph.D., University of Oklahoma Health Sciences Center

Inflammation and Granuloma

Chairperson: Juneann W. Murphy, Ph.D., University of Oklahoma Health Sciences Center
Joost J. Oppenheim, M.D., National Cancer Institute
Stuart M. Levitz, M.D., Boston University
Scott G. Filler, M.D., Harbor-UCLA Research and Education Institute
Gary B. Huffnagle, Ph.D., University of Michigan

Mucosal and Lung Resistance in Infectious Disease

Chairperson: Juneann W. Murphy, Ph.D., University of Oklahoma Health Sciences Center
Jerry R. McGhee, Ph.D., The Immunology Vaccine Center, UAB
Paul L. Fidel, Jr., Ph.D., Louisiana State Medical Center
Mary F. Lipscomb, M.D., University of New Mexico
Allen G. Harmsen, Ph.D., The Trudeau Institute, Inc.

Emerging Technologies for Application to Studies in Host Resistance

Chairperson: Thomas R. Kozel, Ph.D., University of Nevada, Reno
Stefan H.E. Kaufmann, M.D., University of Ulm
Matthew Scharff, M.D., Albert Einstein College of Medicine
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