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RECOMBINANT ANTIBODIES: A NATURAL PARTNER IN COMBINATORIAL ANTIFUNGAL THERAPY*

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SUMMARY

Monotherapy, in the form of amphotericin B or one of its liposomal derivatives, is the usual treatment for invasive fungal infections, due to lack of a safe, effective combination of antifungal drugs. Combination therapy is not necessarily beneficial – there may be mutual antagonism or indifference, increased toxicity or interference with concomitant medication. But the benefits of a well-tolerated, synergistic combination would be great – the enhanced efficacy would improve clinical outcome, reduce the need for prolonged courses of treatment and prevent the emergence of antifungal drug resistance. Antifungal antibodies would be a natural partner in a combinatorial approach to antifungal therapy. Analysis of the antibody response which occurs in patients with invasive candidiasis, being treated with amphotericin B, showed a close correlation between recovery and antibody to the immunodominant heat shock protein 90 (hsp90). The molecular chaperone hsp90 is essential for yeast viability. Mycograb® is a human recombinant antibody to hsp90 which shows intrinsic antifungal activity and synergy with amphotericin B both *in vitro* and *in vivo*. It is now the subject of a multinational, double-blind, placebo-controlled trial, in patients with culture-confirmed invasive candidiasis on liposomal amphotericin B.

INTRODUCTION

Invasive candidiasis is the most prevalent of the systemic fungal infections. It is a deep-seated, life-threatening form of the infection due to yeasts of the genus Candida. The commonest species is *Candida albicans,* but non-albicans species account for an increasing proportion of the infections. Almost any organ can become infected and the infection frequently disseminates, via the bloodstream, to multiple organs. At risk groups include immunosuppressed and neutropenic patients, organ transplant recipients, individuals receiving total parental nutrition or peritoneal dialysis, those with cerebrospinal fluid shunts and drug abusers. Today one of the most commonly affected patient groups is intensive care unit patients. These patients are debilitated, but not neutropenic, and are particularly at risk if the gastrointestinal tract is damaged, by disease, trauma or surgery, because the gut is a major harbinger of *Candida*.

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Reviewing the literature, *Fridkin* and *Jarvis* (1996) estimated that the mortality attributable to *Candida* was 38%, with crude mortality rates of 50% to 60%.

Treatment of candidiasis is dependent on three main classes of chemotherapeutic agents. The first of these is the polyenes, of which conventional amphotericin B (Fungizone®) is the parent compound. Lipid-based formulations of amphotericin B (Abelcet®, Ambisome® and Amphocil®) were developed to reduce the toxicity of amphotericin B. These are well-established, mainline therapeutics for the treatment of life-threatening, culture-confirmed, deep-seated infections. The second class is the onazoles, such as fluconazole and voriconazole. Fluconazole is the most commonly used antifungal drug for the treatment of superficial candidiasis (thrush). It is sometimes given to high-risk patients as prophylaxis against invasive candidiasis or as empiric treatment in suspected cases. It is not recommended for non-albicans species, many of which are intrinsically resistant to fluconazole (*Martins* and *Rex*, 1996) nor for neutropenic patients because it fungistatic, not fungicidal. Acquired resistance to fluconazole can occur among strains of *C. albicans,* and is particularly seen in patients with AIDS receiving long-term treatment for candidal oesophagitis. The echinocandins, such as caspofungin, are a new class of antifungals. In a recently published study in patients with invasive candidiasis (*Mora-Duarte* et al., 2002), caspofungin was found to be at least as effective as conventional amphotericin B, though differences in efficacy between the two groups were mainly a reflection of failures due to Fungizone toxicity. *Cryptococcus neoformans* is intrinsically resistant (because it lacks the target enzyme) and *Candida parapsilosis* shows relative resistance compared to other candidal species. Caspofungin recently received market authorisation, in the USA and Europe, for the treatment of invasive aspergillosis refractory to amphotericin B.

There are increasing reports of clinically significant antifungal drug resistance (*Espinel-Ingroff*, 1997; *Hope* et al., 2002; *Krcmery* and *Barnes*, 2002; *Kontoyiannis* and *Lewis*, 2002). Combination therapy is therefore being suggested as a means of combating resistance and improving clinical outcome, just as it is for serious bacterial infections (*Kontoyiannis* and *Lewis*, 2002). Potential problems with this approach include: Antagonism between static and cidal drugs, as occurs between fluconazole and amphotericin B (*Arganoza* et al., 1997); indifference between drugs that have the same target; increased risk of side effects when several potentially toxic antifungal drugs are used in combination and greater risk of undesirable interactions with other drugs such as immunosuppressive agents. Ideally an antifungal partner should be synergistic (enhancing the efficacy of the other antifungal drug), broad spectrum (against all clinically significant *Candida* species) and safe (both in terms of intrinsic toxicity and lack of interactions with other drugs). Passive immunotherapy, in the form of a naturally occurring antifungal antibody, has the potential to be an ideal partner for combination therapy.

RATIONALE FOR ANTIBODY THERAPY

Candida species commonly inhabit the mucosal surfaces of the gut and oropharynx, without producing symptoms, being held in check by a wide va-

riety of innate and specific defence systems. But if *Candida* does gain access to the bloodstream, it can spread to set up foci of infection in one or more systemic organs, and thereby become a life-threatening infection. Moreover, *Candida* itself is immunosuppressive, predisposing the patient to additional infections by bacteria, such as *Staphylococcus aureus* (*Carlson*, 1982; *Vartivarian* and *Smith*, 1993). In the past there has been considerable debate over the relative importance of antibody-mediated versus innate and cell-mediated immunity (CMI) in defence against candidiasis. The importance of phagocytic cells such as neutrophils can be demonstrated both experimentally and clinically (candidiasis being associated with deficiencies in the number and function of neutrophils) (*Vartivarian* and *Smith*, 1993). Evidence for the importance of CMI comes from animal studies and the well-documented association between specific defects in CMI and chronic mucocutaneous candidiasis (*Vartivarian* and *Smith*, 1993). Similarly HIV infection is associated with oral and oesophageal candidiasis. However these defects in CMI predispose to superficial, mucocutaneous candidiasis not deep-seated invasive candidiasis (*Matthews* et al., 1988a). Numerous studies have shown immune sera to be protective in animal models of candidiasis involving systemic forms of the infection resulting from intravenous injection with *Candida* (*Casadevall*, 1995; *Matthews* et al., 1996). Many of these early experiments were conducted with immune sera in which the nature of the protective antibody was uncharacterised. Without knowing the titre or specificity of the antibody such experiments were difficult to reproduce. It is now possible not only to characterise an antibody and its target, defining its sequence and mapping epitopes reactivity, but also to bulk produce it to current Good Manufacturing Standards (camp) at an economically viable cost. Antibody-based therapeutics can now be significant contenders in the development of novel antifungal drugs. Unlike vaccines, they avoid the need for the recipient to be immunocompetent and provide an immediate benefit to the patient – as required for a life-threatening infection.

Analysis of the antibody response produced by patients, being treated with amphotericin B, who survived invasive candidiasis, showed that such patients produced a strong, sustained antibody response to the 47 kilodalton antigen (*Matthews* et al., 1984,1987). On sequencing this was identified as the carboxy end of the stress protein heat shock protein 90 (hsp90) (*Matthews* and *Burnie*, 1989; *Panaretou* et al., 1999; *Swoboda* et al., 1995). Fatal cases produced no or falling antibody titres to this antigen. Antibody to this antigen is significantly ($p<0.05$) commoner in patients with systemic candidiasis than those colonised with *Candida* (*Porsius* et al., 1990), and common in patients with AIDS and chronic mucocutaneous candidiasis (*Matthews* et al., 1988a). Epitope mapping (*Matthews* et al., 1991a) defined the immunodominant antibody binding site of hsp90 and this epitope was used to raise both mouse monoclonal and human recombinant antibodies, which were protective in mouse models of invasive candidiasis (*Matthews* et al., 1991b,1995). Homologous epitopes have been identified in both yeasts and filamentous fungi, namely *C. albicans, C. parapsilosis, Torulopsis glabrata, Candida tropicalis, Candida krusei* and *Aspergillus fumigatus* (*Matthews*, 1991; *Santhanam* and *Burnie*, 2000; *Burnie* and *Matthews*, 1991; *Kumar* et al., 1993). Mycograb® is a human genetically recombinant antibody ("grAb") against the immunodominant epitope of the *Candida* hsp90 antigen, which has been produced to

cGMP standards and is currently being assessed in clinical trials in patients with culture-confirmed invasive candidiasis.

HEAT SHOCK PROTEIN 90: AN ANTIGEN TARGET

Heat shock proteins (also known as stress proteins) are ubiquitous families of proteins, produced both constitutively and inducibly, in response to a wide variety of stressful stimuli. The hsp90 family plays an essential role in cell physiology (*Csermely* et al., 1998; *Matthews* et al., 1998) acting as molecular chaperones for a variety of cellular proteins, including steroid receptors, protein kinases involved in signal transduction and endothelial nitric-oxide synthase. Their induction in response to stressful stimuli is a means of helping the cell to protect its components from the degradative effects of stress. When an organism invades the host, its environment becomes highly stressful - temperature, pH, ionic strength and nutritional composition all abruptly change - and it comes under attack from the host's immune system. In response, hsp levels rise in the invading pathogen which, by chaperoning key cellular proteins, helps to counter-balance the degradative effects of this adverse environment. In turn, the hsps themselves have become abundant targets against which the host directs its immune response. There are many bacterial, parasitic and fungal infections in which hsps have been identified as immunodominant antigens, and in some cases it has now been established that this immunity is protective. For example, in animal models of tuberculosis and histoplasmosis, vaccination with hsp65 and hsp60, respectively, induced protective cell-mediated immunity (*Bonnefoy* et al., 1994; *Matthews* et al., 1998). Vaccine-induced antibody to hsp90 has been correlated with protection against malaria in a squirrel monkey model (*Bonnefoy* et al., 1994).

Hsp90 is essential for yeast viability. In the relatively non-virulent yeast *Saccharomyces cerevisiae*, deletion of the two genes encoding hsp82 (the homologue of *C. albicans* hsp90) leads to cell death, while deletion of one gene leaves the yeast viable but unable to grow at higher temperatures (*Borkovich* et al., 1989). Higher concentrations of hsp82 are required for growth at temperatures above the optimal growth temperature. The hsp90 of *C. albicans* can confer hsp90 functions in *S. cerevisiae* (*Panaretou* et al., 1999). Overexpression of hsp90 by a transformant of *S. cerevisiae* was associated with a significant increase in virulence in mice compared to the parent strain, producing an infection more like that seen with *C. albicans* (*Hodgetts* et al., 1996). Therefore, hsp90 appears to be a virulence factor and overexpressed hsp90 may play a key part in helping the yeast adapt to its new stressful environment at higher body temperatures.

Protein extracts from exponentially growing *C. albicans* or *S. cerevisiae* yield not only full length hsp90 but also subfragments of 72-76 kDa and 47 kDa, which are the result of partial degradation within viable yeast cells (*Panaretou* et al., 1999). Mice infected with candidal protoplasts failed to produce an antibody response to hsp90 or its subfragments (*Pitarch* et al., 2001), compatible with loss of this antigen family during removal of the yeast cell wall. Immuno-electronmicroscopy studies suggested partial localisation of the 47 kilodalton antigen in the cell wall (*Matthews* et al., 1988b), and immunohistochemical staining of infected mice kid-

ney sections suggests binding of Mycograb® around yeast cells. Likewise, surface-expressed hsp90 serves as an antigen in Chagas' disease, ascariasis, leishmaniasis, toxoplasmosis and infection due to *Schistosoma mansoni* (*Johnson* et al., 1989; *Kumari* et al., 1994; *Dragon* et al., 1987; *Rojas* et al., 2000; *Streit* et al., 1996).

These features make an antibody-based hsp90 inhibitor, replicating a naturally occurring antibody response to candidal hsp90, an obvious candidate for combination antifungal therapy.

MYCOGRAB®: AN ANTIFUNGAL ANTIBODY AGAINST HSP90

Mycograb® was derived from the anti-hsp90 antibody cDNA of patients recently recovered from invasive candidiasis (*Matthews* and *Burnie*, 2001; *Matthews* et al., 2003). It consists of the antigen-binding variable domains of antibody heavy and light chains linked together to create a recombinant protein which is expressed in *Escherichia coli*. It does not have an Fc component and therefore its activity is not dependent on recruitment of white blood cells or complement. It is simply dependent on its ability to bind to and inhibit hsp90. Its antifungal activity *in vitro* can be demonstrated using assays designed to assess conventional antifungal drugs (*Matthews* et al., 2003). It has shown a broad range of activity against all yeasts tested – compatible with the conserved nature of the target antigen in different yeast species. Using these same assays it is possible to demonstrate synergy with amphotericin B (*Matthews* et al., 2003). In contrast, for all strains examined to date, it has usually shown indifference when used in combination with fluconazole - the exception being a fluconazole-sensitive strain of *C. albicans* with which it showed synergy. This mutual enhancement of activity when combined with amphotericin may simply reflect the effect of combining two drugs directed against two different targets within the fungus or it may have as its basis the increased leakiness of yeast cells in the presence sub-lethal doses of amphotericin, thereby giving Mycograb® greater access to intracellular hsp90 (*Matthews* et al., 2003).

Serum levels of amphotericin B in patients are 1 to 2 μg/ml (*Bekersky* et al., 2002; *Bindschadler* et al., 1969; *Groll* et al., 1998), consistent with a therapeutic response occurring when the minimum inhibitory concentration (MIC) of the *Candida* isolate is equal or less than 0.5 μg/ml (*Rex* et al., 2001), but therapeutic failure when the MIC is greater than to 1 μg/ml (*Nguyen* et al., 1998). Mycograb®, at levels readily achievable in the serum, is able to significantly reduce the MIC of amphotericin B to 0.5 μg/ml or less, even for strains which previously had an MIC > 1 μg/ml (*Matthews* et al., 2003).

In a mouse model of systemic candidiasis, intravenous administration of Mycograb® alone produced a statistically significant improvement in the infections caused by each species examined (*Matthews* et al., 2003). Amphotericin B alone cleared the *C. tropicalis* infection, but failed to clear infections caused by *C. albicans, C. krusei, C. glabrata* or *C. parapsilosis* from one or more organs. By combining Mycograb® with amphotericin B, complete resolution of infection was achieved for *C. albicans, C. krusei* and *C. glabrata*; for *C. parapsilosis* the liver and spleen were cleared, but renal counts were unaltered by either drug alone or in combination (*Matthews* et al., 2003).

The immunological reactivity of Mycograb® with candidal hsp90 can be

demonstrated by immunoblot, immunohistochemistry and by ELISA (presenting the target epitope as a synthetic peptide). Following two dimensional gel electrophoresis of candidal extracts, the Mycograb® preferentially binds to the two truncated forms of hsp90, represented by the 40 and 47 kDa spots – indicating that the epitope is more accessible to antibody binding here than in the full-length hsp90 protein (*Matthews* et al., 2003). This is compatible with the observation that patients recovering from invasive candidiasis much more commonly had antibody against the 47 kDa antigen band on immunoblots of a one-dimensional gel of *Candida* than antibody to the full-length hsp90 protein (*Matthews* et al., 1984, 1987; *Matthews* and *Burnie*, 1989).

At the primary structure level, hsp90 is composed of three domains: The N-terminal region (Met^{1}-Arg^{400}), the middle region (Glu^{401}-Lys^{615}) and the C-terminal region (Asp^{621}-Asp^{732}) (*Matsumoto* et al., 2002). The assembly of the hsp90-substrate protein complex requires ATP and involves a conformational change in the hsp90. Hsp90 has two ATP binding sites, one in the C-terminal domain and one in the N-terminal domain. The C-terminal ATP binding site is the first example of a cryptic chaperone nucleotide-binding site, which is opened by occupancy of the N-terminal site (*Soti* et al., 2002). This process requires communication between these two sites through the middle domain, which has a γ-phosphate-binding motif similar to other GHKL family members involving QQSKILKVI, which overlaps the N-terminal end of the peptide recognised by Mycograb® (*Matthews* et al., 1991a; *Dutta* and *Inouye*, 2000). The importance of the middle region in yeast hsp90 is illustrated by the finding that point mutations in this domain caused deficient binding to the N-terminal region which in turn was associated with the yeast cells being unable to grow higher temperatures (37°C) (*Matsumoto* et al., 2002). Since the interaction between the N-terminal and middle regions is essential for the *in vivo* function of hsp90 in yeasts (*Matsumoto* et al., 2002), this could explain the antifungal activity of both Mycograb® (which binds the middle region) and radicicol (which binds the N-terminal region) (*Schulte* et al., 1999).

Hsp90 is part of the steroid hormone receptor superfamily of proteins (*Pratt*, 1993). In the water mold *Achlya ambisexualis*, sexual reproduction involves branching in the opposite mating type, which is induced by the binding of a steroid hormone to a steroid hormone receptor complexed with hsp90 (*Brunt* and *Silver*, 1986). Induction of hsp90 by steroids may be responsible for the upregulation of the stress response in *C. albicans* observed following treatment with 17-β-oestradiol (*O'Connor* et al., 1998). The frequency of *Candida* infections in pregnant women and the oestrogen-dependence of *Candida* colonisation in the rat model, could be linked to this steroid-induced enhanced protection, which results in the treated yeasts becoming resistant to an otherwise lethally high temperature (48.5°C) and oxidative stress (menadione exposure). Exposure to the steroid also induced yeast-to-hyphal transformation (which is thought to be linked to virulence) and increased colony size.

There may be additional means by which Mycograb® achieves benefits in the infected human host, since the epitope recognised by this antibody is highly conserved and present in human hsp90 (*Hickey* et al., 1986; *Matthews* et al., 1991a). The binding of hsp90 to endothelial nitric oxide synthase leads to the release of nitric oxide (*Garcia-Gardena* et al., 1998) which in turn regulates cardiovascular haemodynam-

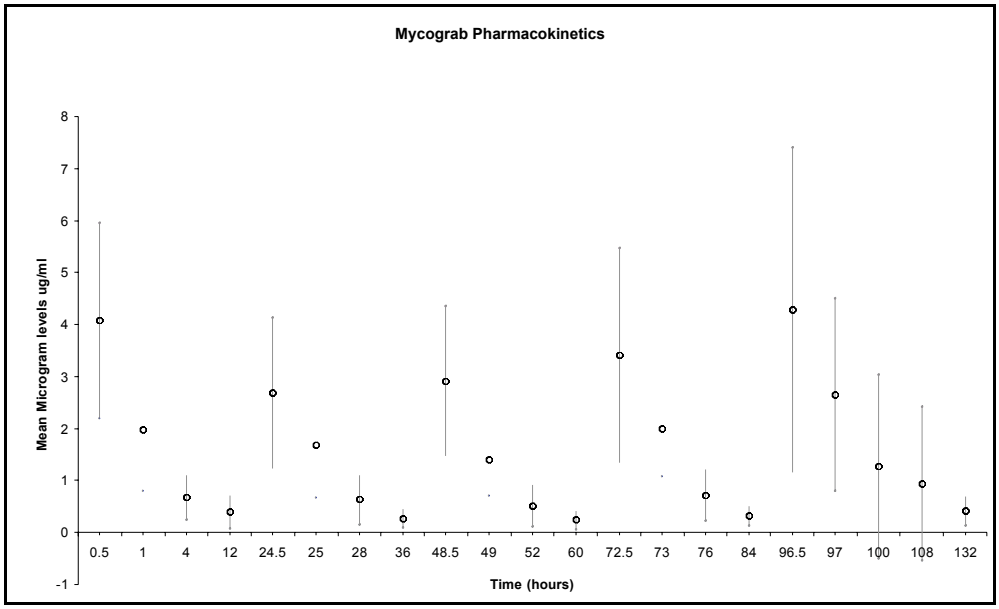

Figure 1: Mean serum levels (with standard deviation) of Mycograb® in 10 patients given Mycograb® at a dose of 1 mg/kg body weight b.d. for 5 days.

ics and causes large vessel vasodilatation. Other pathways catalysed by hsp90 include activation of the prekallikrein-kininogen complex, leading to release of bradykinin, another active biological – responsible, for example, for the angio-oedema seen with ACE inhibitors (*Kusukam* et al., 2002). Inhibition of such pathways, which could be activated by release of endogenous human hsp90 from damaged tissues or candidal hsp90 from lysed yeasts, may be of benefit in counteracting many of the signs of septic shock.

CLINICAL TRIALS

Mycograb® was first assessed in an open-labelled, tolerance and pharmocokinetic study, carried out in the UK, involving five patients with invasive candidiasis receiving liposomal amphotericin B (Abelcet®). After a test dose of Mycograb® (0.1 mg/kg), a single dose of Mycograb® was given (1 mg/kg) followed, at least 24 hours later, by two doses (1 mg/kg) given 8 or12 hours apart. All patients were closely monitored both clinically and by a wide range of laboratory parameters (blood chemistry, haematology, coagulation profile). No treatment-related adverse effects were observed. Mycograb® was undetectable at the 0.1 mg/kg dose. Blood samples taken 30 minutes after receiving a single i.v. bolus of 1 mg/kg, gave serum levels ranging from 1.5 to 4.0 mg/l. Serum levels become undetectable by 8 hours. When two doses were given, with an interval of 8 hours or 12 hours, a slight increase in the levels occurred following the second dose indicating some tissue accumulation. This first study was designed to obtain preliminary data on safety and pharmacokinetics, and not for the assessment of efficacy, but among the three patients who

Table 1: The serum pharmacokinetic data from the same 10 patients receiving Mycograb® over a 5-day period. The data was interpreted by non-compartmental pharmacokinetic analysis PK Solutions 2.0 for C_{max} (μg/ml), AUC_{0-t}, $AUC_{0-\infty}$, (μg (min/ml), $t_{1/2\alpha}$ (min), $t_{1/2\beta}$ (hours) and Mean Residence Time (MRT) (hours)

	Day number (n = 10)				
	1	2	3	4	5
C_{max} (μg/ml)	9.9	4.9	7.4	6.6	8.3
AUC_{0-t} (μg (min/ml)	13.2	10.0	9.5	12.1	27.7
$AUC_{0-\infty}$ (μg (min/ml)	19.2	12.6	12.1	15.3	34.8
$t_{1/2\alpha}$ (min)	18	24	18	24	24
$t_{1/2\beta}$ (hours)	10.7	6.5	7.4	6.8	12.0
MRT (hours)	10.8	6.8	6.9	6.9	14.4

received the full dose of 1mg/kg b.d., albeit only for 24 hours, there was an association with cultures becoming negative and improvement in one or more clinical parameters.

Mycograb® is now the subject of a double-blind, placebo-controlled efficacy and safety study involving over 30 centres in 10 countries. All patients have culture-confirmed invasive candidiasis and are being treated with liposomal amphotericin B (Abelcet® or Ambisome®), in combination with a 5 day course of Mycograb® (1 mg/kg b.d.) or placebo (saline). Patients are carefully monitored both clinically and by laboratory parameters, including fungal cultures. Assessment of efficacy is based on clinical response, mycological response, overall mortality and *Candida*-associated mortality. In addition, the test and control arm will be compared to determine whether the need for prolonged courses of amphotericin B (> 10 days) is less in the Mycograb®-treated group – the aim being to develop a shorter, more effective course of treatment, using this combination of antifungals, in place of a prolonged course of monotherapy with its associated increased cost and risk of toxicity.

So far, blood samples taken for pharmacokinetics (Figure 1) have suggested serum levels are not affected by varying degrees of renal insufficiency, liver failure or the patient receiving haemodialysis. Mycograb® was not detectable in urine samples The data was interpreted by non-compartmental pharmacokinetic analysis PK Solutions 2.0 for C_{max} (μg/ml), AUC_{0-t}, $AUC_{0-\infty}$, (μg (min/ml), $t_{1/2\alpha}$ (min), $t_{1/2\beta}$ (hours) and Mean Residence Time (MRT) (hours). This showed (Table 1) that the C_{max} levels obtained were generally in the range required to achieve demonstrable synergy with amphotericin B *in vitro* for the strains of *C. albicans, C. krusei* and *C. tropicalis* (4 μg/ml) and *C. glabrata and C. parapsilosis* (8 μg/ml). Mouse pharmacokinetic studies suggest that tissue levels may be sustained for longer periods than serum levels (*Matthews* et al., 2003).

OTHER DISEASE TARGETS

Other infectious diseases in which hsp90 plays a key role in the physiology of the organism and its ability to meet the challenge of survival in the human

host, could benefit from treatment with an hsp90 inhibitor such as Mycograb®. Hsp90 is an immunodominant antigen in *Aspergillus fumigatus* (*Burnie* and *Matthews*, 1991; *Kumar* et al., 1993). The role of humoral immunity in host defence against aspergillosis is uncertain, but Mycograb®, given in combination with a cell-wall active antifungal such as amphotericin B or an echinocandin, may be able to reach the target hsp90 and inhibit it, and thereby be of benefit in the treatment of invasive aspergillosis. Since invasive aspergillosis is relatively refractory to treatment, it is likely that a more prolonged course, and possibly higher doses, of Mycograb® would be required in such cases.

Several different families of hsps play important roles in parasitic infections, being involved in differentiation, protection from the host cell's killing mechanisms and virulence (*Polla*, 1991). The importance of humoral immunity to malaria was demonstrated by *Cohen* et al. (1961), who showed clinical improvement in African children suffering from severe malaria following passive transfer of immunoglobulin from immune adults. *Bonnefoy* et al. (1994), in a squirrel monkey vaccination trial found a close correlation between antibody response to hsp90 and resistance to heavy challenge from highly virulent *Plasmodium falciparum*. Analysis of the antibody responses to hsp90, hsp70 and hsp65 in Thai patients with malaria showed that antibody titres to hsp90 were particularly high (*Zhang* et al., 2001). Recently it has been shown that hsp90 is essential for *P. falciparum* growth in human erythrocytes, suggesting hsp90 as a potential drug target for antimalarials (*Banumathy* et al., 2003).

Other candidal antigens which could be used as targets for the development of therapeutic antifungal antibodies include cell-surface adhesins, antibodies to which can prevent binding of the yeast to host cell receptors (*Lee* et al., 1996), heat shock mannoproteins (*Polonelli* et al., 1994a) and yeast killer-toxin-like anti-idiotype antibodies (*Polonelli* et al., 1994b). The close association between recovery from cryptococcosis and the host's antibody response to the polysaccharide capsule of this yeast makes antibody therapy an attractive goal, which is being explored by *Casadevall* and co-workers. Passive administration of monoclonal antibodies against the capsular polysaccharide of *C. neoformans* can prolong the survival of lethally infected mice (*Shapiro* et al., 2002), provided the murine antigen-binding V regions are paired with human C regions of the correct isotype, in these mouse-human chimeric antibodies (*McLean* et al., 2002).

CONCLUSION

Antifungal antibodies offer a new approach to the treatment of these important, life-threatening infections. They provide a means of directly applying our growing knowledge of the immunology and pathogenesis of candidiasis to the development of completely novel therapeutics. By using passive antibody therapy rather than vaccines, they avoid the need for the recipient to be immunocompetent and provide an immediate benefit to the patient. They are a natural partner for combination therapy. Mycograb® has been primarily designed for use in combination with existing cell-wall active antifungal drugs, which facilitate its penetration to the target hsp90 antigen. It is believed that the

synergy between Mycograb® and amphotericin B will provide a much more effective therapeutic combination, which treats the infection relatively quickly, thereby reducing cost and risk of toxicity. Future potential applications lie in the treatment of other infectious diseases in which hsp90 plays a key role in the survival of the pathogen in the host, and may include invasive aspergillosis and malaria.

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