MINIREVIEW

Recent developments in the immunology and biology of *Malassezia* species

Helen Ruth Ashbee

Skin Research Centre, Institute of Molecular and Cellular Biology, Faculty of Biological Sciences, University of Leeds, Leeds, UK

Correspondence: H. Ruth Ashbee, Skin Research Centre, Institute of Molecular and Cellular Biology, Faculty of Biological Sciences, University of Leeds, Leeds LS2 9JT, UK. Tel.: +44 113 3435616; fax: +44 113 3435638; e-mail: h.r.ashbee@leeds.ac.uk

Received 22 September 2005; revised 29 November 2005; accepted 29 November 2005. First published online 21 February 2006.

doi:10.1111/j.1574-695X.2006.00057.x

Editor: Willem van Leeuwen

Keywords
Malassezia; atopic dermatitis; dendritic cell; keratinocyte; inflammatory response.

Abstract

Malassezia spp. are members of the normal cutaneous flora, but are also associated with several cutaneous diseases. Recent studies of the interaction of *Malassezia* spp. with melanocytes, fibroblasts, keratinocytes and dendritic cells have highlighted their potential to modulate the immune response directed against them. In normal skin they may downregulate the inflammatory response, allowing them to live as commensals. In contrast, in atopic/eczema dermatitis syndrome and psoriasis, they may elicit an inflammatory response that contributes to the maintenance of lesions. Future research may define ways to influence this inflammatory cycle and hence to control or prevent exacerbations of these diseases.

Introduction

Yeasts of the genus *Malassezia* undergo asexual reproduction by monopolar or sympodial budding from a broad base (Chen & Hill, 2005). Some of the species are able to undergo a phase transition from yeasts to hyphae, although the factors that control this transition are poorly understood (Chen & Hill, 2005). Except for *M. pachydermatis*, the species require an exogenous source of lipid owing to their inability to synthesise C14–C16 saturated fatty acids, because of a block in the *de novo* synthesis of myristic acid (Shifrine & Marr, 1963).

The taxonomy of this genus has been very confused, with the yeast phase originally described as *Pityrosporum* and the hyphal phase as *Malassezia*, before they were unified in 1986 with both phases included in *Malassezia* (Cannon, 1986). Since then considerable further change has occurred. The number of species in the genus has increased from two in 1989 to 11 currently characterized species, with others tentatively named but not formally accepted. The definition of seven species (*M. furfur, M. pachydermatis, M. sympodialis, M. globosa, M. obtusa, M. restricta* and *M. slooffiae*) by Guillot & Gueho (1995) unified several previous taxonomic schemes. Since then, however, work by other groups has led to the definition of more species, including *M. dermatis* (Sugita *et al.*, 2002), *M. japonica* (Sugita *et al.*, 2003), *M. yamotoensis* (Sugita *et al.*, 2004) and *M. nana* (Hirai *et al.*, 2003). A further species, *M. equi*, has been described and tentatively named (Nell *et al.*, 2002), but not fully characterized. Other groups have found significant variation within some species (Sugita *et al.*, 2005), which may be sufficient to lead to their division into further species in the future (Cabanes *et al.*, 2005).

Malassezia in health and disease

Malassezia spp. are members of the normal human cutaneous flora. They occur mainly on the trunk and head (Leeming *et al.*, 1989), with the highest densities found in postpubertal adolescents and adults, before levels decrease in late middle age (Bergbrant & Faergemann, 1988). The description of the new species of *Malassezia* has resulted in a number of studies examining the distribution of the species at different body sites. The results are summarized in Table 1, and demonstrate the variability in the results obtained. These variations may be a result of the sampling methods and culture media used, as most of the studies have used only qualitative skin-sampling methods (contact

Table 1. The species of *Malassezia* isolated from healthy and diseased skin

	Healthy skin			Pityriasis versicolor			Seborrhoeic dermatitis			Atopic dermatitis		
Ref.	Species (% of subjects colonized; site)	*N*	Sampling method/ culture medium	Species (% of subjects colonized; site)	*N*	Sampling method/ culture media	Species (% of subjects colonized; site)	*N*	Sampling method/ culture media	Species (% of subjects colonized; site)	*N*	Sampling method/ culture media
Crespo-Erchiga *et al*. (1999)	*M. sym* (93; trunk)	136	Swabs/ mDixons*	*M. glob* (87) *M. sym* (34)	100	Tape strip/ mDixons	*M. res* (43; head/face) *M. glob* (34; head/face) *M. sym* (19; head/face)	75	Tape strip/ mDixons			
Aspiroz *et al*. (1999)	*M. glob* (60; trunk) *M. res* (51; scalp)	38	Swabs/ L&N									
Crespo-Erchiga *et al*. (2000)	*M. glob* (12; forehead) *M. sym* (42; shoulders)		Tape strip/ mDixons	*M. glob* (77) *M. sym* (32)	96	Tape strip/ mDixons						
Nakabayashi *et al*. (2000)	*M. glob* (22) *M. sym* (10) *M. fur* (3)	105	Brush or tape strip/ mDixons	*M. glob* (55) *M. sym* (9)	22	Swab, Dixons	*M. glob* (21) *M. fur* (21)	42	Swab, Dixons	*M. fur* (21; face/ neck) *M. glob* (14; face/ neck) *M. sym* (7; face/ neck)		Swab, Dixons
Gupta *et al*. (2001a)				*M. sym* (59) *M. glob* (25) *M. fur* (11)	111	Skin scrape, L&N†						
Gupta *et al*. (2001b)	*M. sym* (50; forehead)	20	Contact plates/L&N	*M. sym* (63)	23	Contact plates/L&N	*M. glob* (45)	28	Contact plates/L&N	*M. sym* (51)	31	Contact plates/L&N
Sugita *et al*. (2001)	*M. res* (61; scalp/ neck) *M. sym* (50; scalp/ neck) *M. glob* (44; scalp/ neck)	18	Adhesive/DNA extraction							*M. glob* (94) *M. res* (88) *M. sym* (41) *M. fur* (41)	32	Adhesive/DNA extraction
Lee *et al*. (2001)							*M. res* (75; face) *M. glob* (60; face)	20	?/ L&N			
Aspiroz *et al*. (2002)	*M. glob* (82) *M. sym* (35)		Swab/L&N	*M. glob* (90) *M. sym* (41)	79	Swab/ L&N						

Table 1. Continued.

Ref.	Healthy skin			Pityriasis versicolor			Seborrhoeic dermatitis			Atopic dermatitis		
	Species (% of subjects colonized; site)	*N*	Sampling method/ culture medium	Species (% of subjects colonized; site)	*N*	Sampling method/ culture media	Species (% of subjects colonized; site)	*N*	Sampling method/ culture media	Species (% of subjects colonized; site)	*N*	Sampling method/ culture media
Faergemann (2002)	*M. sym* (40) *M. obt* (25) *M. glob* (23)		Contact plates/L&N	30			*M. obt* (30) *M. sym* (30) *M. res* (64) *M. glob* (41)	16	Contact plates/L&N 70 Swabs/DNA extraction	*M. sym* (40) *M. obt* (25) *M. glob* (23)	125	Contact plates/L&N
Gemmer *et al.* (2002)												
Gupta & Kohli (2004)	*M. sym* (80+; head) *M. glob* (25; head)	157	Contact plates/L&N									

N, number of subjects – there may be multiple samples per subject.
M. sym: Malassezia sympodialis; M. glob: Malassezia globosa; M. res: Malassezia restricta; M. fur: Malassezia furfur; M. obt: Malassezia obtusa.
*mDixons, modified Dixons agar (Van Abbe, 1964).
†L&N, Leeming & Notman agar (Leeming & Notman, 1987).

plates, swabs, sellotape strips) and so cannot accurately determine the relative proportion of species on the skin. The medium on which the samples are plated will have a significant impact on the range of species recovered. However, it may be that the differences between studies and from country to country represent genuine geographic variations in the predominant *Malassezia* species on normal skin. Until there is a systematic survey, using quantitative sampling methods and optimized culture medium, this cannot be determined.

The carriage rates found on children have varied significantly between studies, with one study finding no *Malassezia* on the skin of 60 healthy children (Abraham *et al.*, 1987) whilst others found *Malassezia* present on up to 98% of the children sampled (Bergbrant & Broberg, 1994; Ashbee *et al.*, 2002; Bernier *et al.*, 2002).

Malassezia spp. are associated with several cutaneous and systemic diseases, either as the causative agent of these conditions or associated with lesions. Diseases with which *Malassezia* is associated include pityriasis versicolor, seborrhoeic dermatitis, dandruff, folliculitis, atopic dermatitis [recently renamed atopic eczema/dermatitis syndrome (AEDS); Johansson *et al.*, 2001], psoriasis, confluent reticulate papillomatosis and seborrhoeic blepharitis. *Malassezia* spp. have also been shown to cause more deep-seated infections, including mastitis, sinusitis, septic arthritis, malignant otitis externa, fungaemia, pulmonary vasculitis, peritonitis and meningitis (Ashbee & Evans, 2002).

Malassezia spp. are the aetiological agents of pityriasis versicolor (PV), with lesions most commonly seen on the trunk (Gordon, 1951). Scrapings from lesions examined microscopically demonstrate the transition of the organism from the yeast to the hyphal phase, producing a characteristic 'spaghetti and meatballs' appearance. One of the characteristics of PV, and often the factor that causes patients to seek diagnosis and treatment, is alteration in the pigmentation of the skin. Lesions may be either hypo- or hyper-pigmented, depending on the normal skin colour. Many reasons have been suggested for the disturbance in pigmentation of the skin, including a block in the transfer of melanosomes to keratinocytes and inhibition of melanin production either by azelaic acid or lipoxygenase (Charles *et al.*, 1972; Karaoui *et al.*, 1981; Galadari *et al.*, 1992). Recently, tryptophan metabolites of *M. furfur* have been shown to induce apoptosis in melanocytes, with concomitant decreases in melanin synthesis that may account for depigmentation associated with one form of PV (Kramer *et al.*, 2005). Several groups have examined the species of *Malassezia* isolated from the lesions of PV to determine whether specific species were associated with the disease (see Table 1). *M. globosa* has been suggested to be the 'causative' agent (Crespo-Erchiga *et al.*, 2000), and several authors have

Table 2. Evidence that *Malassezia* is a contributory factor in the aetiology of atopic/eczema dermatitis syndrome (AEDS)

Finding	References
More patients with AEDS lesions on the head and neck are skin-prick-positive to *Malassezia* than patients with lesions in other areas	Waersted & Hjorth (1985)
Antifungal treatment results in clinical improvement in AEDS patients with lesions on the head and neck	Clemmensen & Hjorth (1983)
Antifungal treatment results in clinical improvement in AEDS lesions at any site	Back *et al*. (1995)
Increased prevalence of Type I hypersensitivity to *Malassezia* in AEDS patients	Young *et al*. (1989)
AEDS patients with lesions of the head and neck are more likely to have *Malassezia*-specific IgE responses	Devos & van der Valk (2000)
Prevalence of IgE specific to *Malassezia* is higher than IgE to other fungi in AEDS patients with head and neck lesions	Lindgren *et al*. (1995)
AEDS patients have Th_2 cells specific to *Malassezia*	Tengvall Linder *et al*. (1996)
Direct contact with *Malassezia* induces eczematous skin reactions in sensitized AEDS patients ('atopy patch test')	Rokugo *et al*. (1990)

supported this view (Nakabayashi *et al*., 2000; Aspiroz *et al*., 2002), although others have found *M. sympodialis* to be the predominant species associated with the lesions (Gupta *et al*., 2001a).

The role that *Malassezia* spp. play in seborrhoeic dermatitis (SD) and dandruff is controversial and has been the subject of much debate. There is also disagreement in the literature over whether they are different conditions, or extremes of the same condition, but in this review they will be considered as a single condition. The association of the organism with lesions, the finding that antifungals improve the condition, and that on withdrawal of treatment there is an increase in the population densities of *Malassezia* and consequent relapse in the disease have all supported the role of *Malassezia* yeasts as the aetiological agent. As with other diseases, researchers have attempted to determine which of the new species of *Malassezia* can be detected on lesions of dandruff (see Table 1). Gemmer *et al*. (2002) used a novel method of nested PCR of the intergenic transcribed spacer region after sampling using scalp swabs, without the use of culture to recover organisms from patients with dandruff. This study found that *M. furfur* was not present, but that *M. restricta* and *M. globosa* were the predominant species associated with the scalp, in both health and disease. More patients with dandruff were colonized than those without disease. These findings are similar to those of other groups, who used traditional sampling and culture methods to enumerate yeasts present in SD or dandruff (Crespo-Erchiga *et al*., 1999; Lee *et al*., 2001).

Two recent studies have examined the stratum corneum from patients with dandruff and found that the structure is altered compared with normal skin. The corneocytes of skin affected by dandruff are loosely associated and desmosome numbers are reduced or absent (Warner *et al*., 2001). The lipids present in the stratum corneum are also decreased, which affects the epidermal water barrier, and the itch response to histamine has been shown to be increased in dandruff patients, probably as a result of the impaired barrier function of the skin (Harding *et al*., 2002). These defects may allow *Malassezia* to penetrate the skin, elicit inflammation and hence contribute to SD.

A mouse model of SD has recently been described in which the mice spontaneously develop inflammatory lesions that resemble SD (Oble *et al*., 2005). The mice have a defect in the T-cell receptor and severe lymphopenia, and develop disease once they reach sexual maturity. Histological examination of the skin demonstrated the presence of 'small ovoid structures' in the lesions, which the authors believed to be *Malassezia* yeasts. The authors also showed that the use of fluconazole to treat the mice resulted in clinical improvement, and they suggested that this mouse model might be useful to study SD. However, the mice spontaneously recovered from the disease once their T-cell count increased, and their lymphopenia resolved. It has previously been shown that *Malassezia* is not found normally on the skin of mice (Guillot *et al*., 1994), so the interpretation of the ovoid structures as yeasts may not be correct. Thus, the utility of this model in studying SD will need further evaluation.

Atopic eczema/dermatitis syndrome is a common skin disease, the aetiology of which is not fully understood. However, since a study in 1983 found that treatment of patients, who had lesions on the head and neck, with ketoconazole resulted in clinical improvement, there has been growing interest in the role of *Malassezia* as a contributory factor in the disease (Clemmensen & Hjorth, 1983). The various findings that support the involvement of *Malassezia* in AEDS are detailed in Table 2. Patients usually have an increased sensitization to *Malassezia*, via both the cellular and humoral immune responses. The cellular responses tend to be of Th2 type (Tengvall Linder *et al*., 1996), with higher levels of IL4 and IL5 production by the T cells of patients than of healthy controls (Tengvall Linder *et al*., 1998). These cytokines are associated with immunoglobulin E (IgE)-mediated reactions and so may drive the continued production of IgE (Wollenberg & Bieber, 2000). Allergen-specific IgE on mast cells may result in histamine release, and the itching associated with this would encourage scratching and so provide an opportunity

for ongoing sensitization to *Malassezia*. Treatment with antifungals reduces the population of yeasts on the skin, and this decrease in the antigen load would help to break the inflammatory cycle.

As with other diseases, there have been several studies attempting to define whether one particular species of *Malassezia* is associated with AEDS (see Table 1). There has been no consensus about which species is/are associated with lesions, with *M. globosa, M. restricta* (Sugita *et al.*, 2001), *M. sympodialis* (Gupta *et al.*, 2001b; Faergemann, 2002) and *M. furfur* (Nakabayashi *et al.*, 2000) all commonly isolated from lesions.

Immunology of *Malassezia*-associated diseases

The last five years have seen a dramatic increase in our understanding of the way in which *Malassezia* interacts with the immune system. Much of the impetus for this has been from studies attempting to elucidate the role of *Malassezia* in AEDS. Several studies have defined allergens present in *Malassezia* that are able to bind IgE (summarized in Ashbee & Evans, 2002). The use of crude extracts of *Malassezia* as allergens for clinical testing of AEDS patients has many problems, including instability and variability of the allergens. These problems may be overcome using purified allergens. Several allergens have now been defined and produced as recombinant proteins, of which 11 are now described (Mala f 1–4, Mala s 5–11; Andersson *et al.*, 2003, 2004). (The nomenclature of the allergens has recently changed from the original Mal, which is used to denote allergens from apple, to Mala.) The distribution of the allergens varies, with some specific to a particular species of *Malassezia* (Mala f 2 and Mala f 3 are found only in *M. furfur*) and others common to several species (e.g. Mala s 6; Andersson *et al.*, 2003), a finding that may partly explain the variation in the results found in the literature, with different studies using different species for their antigen source. Many patients with AEDS have circulating IgE to *Malassezia* spp. and most commonly to *M. globosa* (Koyama *et al.*, 2001; Zargari *et al.*, 2003), but, despite the presence of common antigens across the species, the use of more than one species in testing for *Malassezia*-specific IgE has been found to improve the sensitivity of tests (Zargari *et al.*, 2003). If the recombinant allergens are used in testing, IgE is most commonly present to Mala s 9, but again the sensitivity of testing increases with the use of more than one allergen (Zargari *et al.*, 2001). These allergens have been used to detect specific IgE in patients with AEDS, in the skin-prick test (SPT; to detect Type I IgE-mediated hypersensitivity) and the atopy patch test (APT; to detect Type IV delayed hypersensitivity). Skin-prick testing with *Malassezia* is positive in more patients with AEDS than in healthy individuals or in those with other *Malassezia*-associated diseases, with patients having lesions localized to the head and neck having the highest rate of positivity. Some authors have stated that the SPT is a more sensitive method for detecting reactivity to *Malassezia* in AEDS patients than the measurement of IgE (Johansson *et al.*, 2003), but others have called the utility of the SPT into doubt (Devos & van der Valk, 2000). The overall picture that emerges from the large number of studies examining the contributory role of *Malassezia* in AEDS is that, whilst the organism may not be responsible for the initiation of lesions, once the cycle of itch and scratch has been initiated, its antigens will contribute to the maintenance of lesions, especially in the head and neck area, where the highest population densities of the organism occur.

There have been large numbers of studies examining the cellular and humoral immune responses to *Malassezia* in patients with many *Malassezia*-associated diseases. For many of the diseases, the responses reported have varied widely, and the possible reasons for this have been reviewed (Ashbee & Evans, 2002). An important trend that has occurred over the last 5–7 years has been to move away from studying immune responses as measured in peripheral blood cells and to begin to dissect out the responses, specific

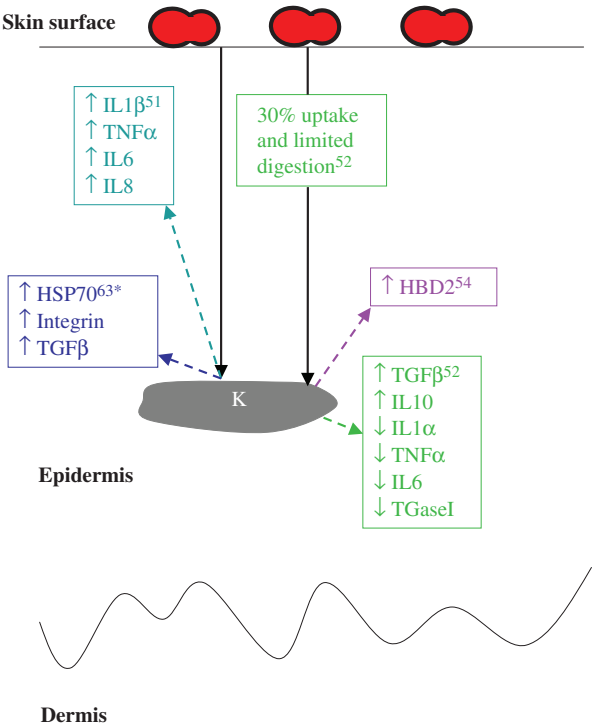

Fig. 1. A schematic illustrating the effects of *Malassezia* on keratinocytes (K) in normal human skin. The interaction of *Malassezia* with keratinocytes has been variously shown to upregulate or downregulate proinflammatory and other cytokine production. Uptake and digestion of *Malassezia* by keratinocytes appears relatively inefficient. Dashed lines indicate the resultant effects after interaction between the cell type and *Malassezia*. *This effect occurs in normal skin, but to a much greater extent in psoriatic skin. Red blobs = *Malassezia* Yeasts.

to *Malassezia*, in cutaneous cell populations. The interactions of *Malassezia* with melanocytes, dermal fibroblasts, keratinocytes and natural killer cells have been studied in some detail and have yielded insight into several diseases. The findings of these studies, described in the following paragraphs, are summarized in Fig. 1.

The interaction of *Malassezia* with normal human dermal fibroblasts *in vitro* has recently been reported (Baroni *et al*., 2001a). *Malassezia* was found to adhere to fibroblasts within 4 h and was internalized into the fibroblast cytoplasm within 24 h, resulting in fibroblast membrane damage. Uptake appeared to be an active process, dependent on F-actin (a cytoskeletal component, involved in cell motility). Although these are interesting findings, *Malassezia* spp. are unlikely to come into contact with dermal fibroblasts *in vivo* and so the clinical relevance of these results is hard to interpret.

Of greater relevance are several studies that have examined the interaction of *Malassezia* with keratinocytes, an interaction that will obviously occur *in vivo*, both in normal and in diseased skin. Keratinocytes are the major cell type in the epidermis, having both a structural role and an involvement in immunological defence. Keratinocytes produce a range of cytokines, some constitutively and others after induction by, for example, microorganisms or skin damage, including IL1α, IL6, IL8 and TNFα. Watanabe *et al*. (2001) were the first group to examine the effects of coculturing *Malassezia* with keratinocytes. Yeast cells of *M. furfur, M. pachydermatis, M. slooffiae* and *M. sympodialis* were cultured, at a ratio of 1 : 1, with normal human keratinocytes for 1–24 h. The supernatants from the cocultures were then assayed for IL1β, IL6, IL8, TNFα and monocyte chemotactic protein 1 (MCP1) at each time-point. For all the species, MCP1 levels were low or undetectable, and for *M. furfur* all the cytokines measured were also low or undetectable. *Malassezia pachydermatis* induced higher levels of cytokines than either *M. slooffiae* or *M. sympodialis*, and levels of IL6 were higher than those of the other cytokines at the same time-points. Culture supernatants from the cultures of *Malassezia* did not induce cytokine production by keratinocytes; therefore, cell–cell contact is required, and cytokine production is not caused by a soluble factor. Watanabe *et al*. (2001) suggested that the higher levels of cytokine induction by *M. pachydermatis* may explain the greater severity of disease associated with this species as seen in animals. Another study, using different isolates from Watanabe *et al*. (2001) cocultured *M. furfur* with the HaCat keratinocyte cell line and also examined cytokine production, as well as yeast uptake (Baroni *et al*., (2001b). The results of this study contradict those of Watanabe *et al*. (2001) finding that *M. furfur* downregulated IL1α production and upregulated IL10 and TGFβ1, leading to inhibition of IL6 and TNFα. *Malassezia* were taken up into the keratinocytes, but there was only limited killing of the organism. It is also interesting that Watanabe *et al*. (2001) found that the species *M. furfur* had minimal effects on cytokine production, whereas Baroni *et al*. found it to have a significant immunomodulatory capacity. Baroni *et al*. postulated that the suppression of IL1α, IL6 and TNFα might allow *Malassezia* to survive within host cells without causing an inflammatory response. Suppression of IL1β, IL6 and TNFα has previously been reported when *Malassezia* was cocultured with peripheral blood mononuclear cells (PBMNC), and that the suppression was IL10-dependent (Kesavan, 1998). A further recent study has examined cytokine and human β defensin (HBD) induction in keratinocytes by *Malassezia* (Donnarumma *et al*., 2004). *Malassezia furfur* was cultured with normal human keratinocytes at a ratio of 30 : 1 for 24, 48 and 72 h. After 48 h of coculture, HDB2, TGFβ1 and IL10 gene expression was induced, but HBD1 gene expression was not affected. The induction of TGFβ1 and IL10, which are anti-inflammatory immunosuppressive cytokines, was suggested as a way in which *Malassezia* could modulate the immune response against it and allow it to live within keratinocytes without stimulating an inflammatory response. This correlates with the situation seen in normal healthy skin and also with the limited inflammation seen in PV, despite the large fungal burden seen in the lesions (Wroblewski *et al*., 2005). The production of IL10 encourages a Th_2-type response with associated IgE production – a feature seen in many patients with AEDS. The differing findings on the effects of *Malassezia*-induced cytokine production by keratinocytes are hard to reconcile; however, it has been suggested that cytokine production may differ in response to different species or even strains (Kesavan, 1998), and this may go some way to explaining the results. It is known that the immunosuppressive effects of *Malassezia* on PBMNC can be reversed by removal of the lipid-rich capsular-like layer around the organism, and it will be interesting to determine if this is also the case with keratinocytes.

The interaction of *Malassezia* with dendritic cells has been the subject of several publications since 2000, and is now probably the best characterized of the yeast–host cell interactions. Most of the work described below and summarized in Fig. 2 has concentrated on the role of the interaction in lesions of AEDS. Dendritic cells, specifically Langerhans cells, are present in the epidermis as immature cells efficient at antigen uptake, but with limited antigen presentation ability. After maturation this situation is reversed, and they become very efficient at antigen presentation but have little ability for further antigen uptake. They are also able to present antigen to naive T cells and are involved in directing the Th cell response. Thus, they are likely to be central in the initiation of any immunological response to *Malassezia*. Buentke *et al*. (2000) derived CD1a+ dendritic cells from peripheral blood and determined their ability to take up

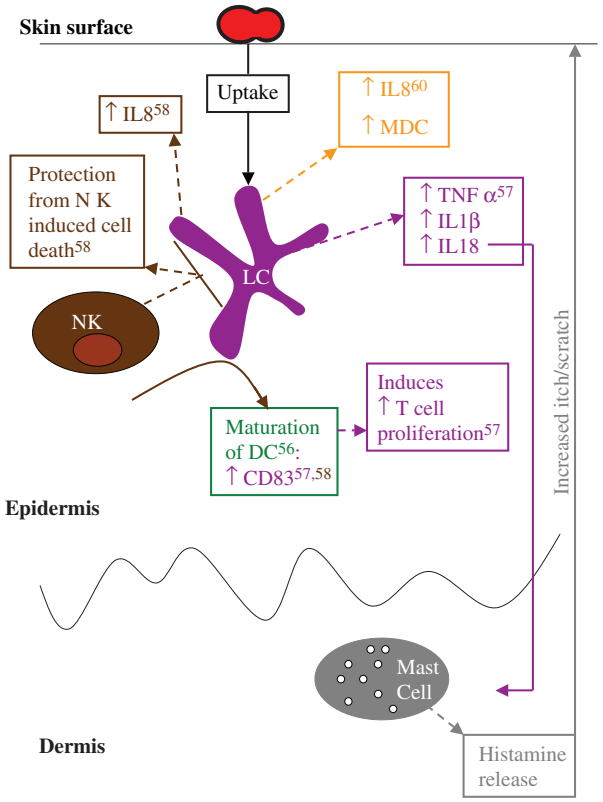

Fig. 2. A schematic illustrating the potential effects of *Malassezia* on Langerhans cells (LC), natural killer cells (NK) and mast cells in atopic dermatitis. Uptake of *Malassezia* by dendritic cells induces their maturation, increased ability to present antigen and leads to an increase in cytokine production and histamine release exacerbating the itch/scratch cycle. Dashed lines indicate the resultant effects after interaction between the cell type and *Malassezia*. Red blobs = *Malassezia* yeasts.

whole *M. furfur* cells, various components of the yeast, and recombinant Mala f5 allergen. Immature cells were significantly better at uptake than mature cells, with whole-cell uptake mediated mainly by the mannose receptor and not influenced by IgE. The uptake of the yeast by the dendritic cells induced maturation of the cells and a significant increase in the production of TNFα, IL1β, and IL18, but not IL10 or IL12, after 46 h of coculture (Buentke *et al.*, 2001). The dendritic cells that had been exposed to *Malassezia* induced proliferation of autologous lymphocytes in a dose-dependent way. Dendritic cell maturation may also be influenced by interaction with natural killer (NK) cells. Buentke *et al.* (2002) examined whether this may occur in AEDS by comparing the numbers of NK cells in the normal skin of healthy controls with those in the APT-positive skin of AEDS patients. They found that there were only scanty NK cells in normal skin, but that they were 'numerous' in the APT-positive skin, and that where they occurred in the APT-positive skin they were in close apposition with dendritic cells. Dendritic cells that had been preincubated with *Malassezia* for 46 h were less susceptible to NK-mediated lysis, and this protection was mediated by soluble factors. This protection of dendritic cells against NK-mediated lysis, if it were to occur *in vivo*, would allow the mature dendritic cells to remain in the epidermis, presenting *Malassezia* antigens to T cells and hence contributing to the maintenance of the inflammatory response in AEDS lesions. When dendritic cells preincubated with *Malassezia* were then cocultured with NK cells, production of IL8 increased and, in some cases, IL6 and IFNγ also increased (Buentke *et al.*, 2003). Therefore, the interaction between dendritic cells in the presence of *Malassezia* with NK cells leads to the production of proinflammatory cytokines and will contribute to the maintenance of inflammation in AEDS lesions. Recently, it has been shown that the dendritic cells of patients with AEDS respond differently to *M. sympodialis* than those of healthy individuals (Gabrielsson *et al.*, 2004). *Malassezia sympodialis* was cocultured with the dendritic cells from patients with moderate/severe AEDS or healthy controls, and the gene expression of the dendritic cells analysed. Although there were no genes exclusively upregulated in either patients or controls, there were differences in the magnitude of the upregulation between the two groups. Six genes were upregulated more than fivefold in patients with AEDS: IL8, CD54, CD83, IL1R, BTG1 (B-cell translocation gene 1) and MDC (monocyte-derived chemokine). The authors postulated that, as these molecules are involved in cell adhesion, costimulation and chemoattraction, they may indicate increased cell traffic and stimulation in patients. In turn, this may result in increased potential for cell interactions between dendritic cells presenting *Malassezia* antigens and T cells. Dendritic cells are known to produce numerous cytokines in response to microbial stimulation, and in this environment continued *Malassezia*-driven inflammation will occur in the lesions. Dendritic cells have a differential response to the various phases of growth of *Candida albicans* (d'Ostiani *et al.*, 2000). The yeast phase elicits production of IL12 and priming of Th_1 cells, whilst the hyphal phase inhibits IL12 and Th_1 priming, and induces IL4 production, a Th_2-type cytokine. The studies carried out so far with *Malassezia* have used yeast-phase cells, and it would be of interest to determine the dendritic cell response to the hyphal phase and whether the differential response seen with *C. albicans* also occurs with *Malassezia*.

Another disease with which *Malassezia* has been associated is psoriasis (Rosenberg *et al.*, 1980). Psoriasis is characterized by hyperproliferation of keratinocytes, but the cause is not known. The application of heat-killed *Malassezia* to rabbit skin elicits lesions similar in appearance and histology to those in human psoriasis (Rosenberg *et al.*, 1980), and its role in the disease has been debated for a number of years. A recent study added to the evidence of its

role in the hyperproliferation seen in psoriasis. Using normal human skin and biopsies from psoriatic skin, the effect of *Malassezia* on TGFβ1, HSP70 and integrin expression by keratinocytes was determined (Baroni *et al*., 2004). *Malassezia furfur* upregulated the expression of all three molecules, and this was more pronounced in psoriatic skin than in healthy keratinocytes. If the psoriatic skin was colonized with *Malassezia*, upregulation was greater than for noncolonized skin. Thus, *Malassezia* upregulates the production of various molecules involved in hyperproliferation and cell migration, which may exacerbate psoriatic lesions.

Conclusion and outlook

Our increased understanding of the interactions occurring in normal and diseased skin highlights the ability of *Malassezia* to modulate cutaneous immune responses. In normal skin, most data suggest that *Malassezia* reduces the inflammatory response, enabling it to live as a commensal. In AEDS and psoriasis, the response against it maintains the inflammation associated with the diseases and contributes to the longevity of the lesions. A key area for future research is to determine ways in which the inflammatory cycle can be broken and how the exacerbation of these diseases can be controlled or prevented.

Acknowledgement

I would like to thank Eileen Ingham for helpful discussions on the manuscript.

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