Various Candida and Torulopsis species differ in their ability to induce the production of C3, factor B and granulocyte-macrophage colony-stimulating factor (GM-CSF) in human monocyte cultures

A. K. M. HØGÅSEN, T. G. ABRAHAMSEN and P. GAUSTAD*

Department of Pediatric Research and *Kapt. W. Wilhelmsen og Frues Bacteriological Institute, Rikshospitalet, N-0027 Oslo, Norway

Summary. The incidence of infections with Candida albicans and also with non-albicans yeast species is increasing rapidly, particularly in immunocompromised patients. Eight Candida and Torulopsis species were compared for their ability to stimulate production of complement components C3 and factor B by monocytes. In addition, the release of granulocyte-macrophage colony-stimulating factor (GM-CSF) was determined, because this cytokine affects monocyte complement production. The highest ranked pathogenic yeasts, i.e., C. albicans, C. tropicalis and C. parapsilosis, were the most effective inducers of C3, factor B and GM-CSF production. C. krusei and T. glabrata showed intermediate activity, whereas C. kefyr, C. guilliermondii and T. candida had only a moderate stimulatory effect on C3 production and did not affect either factor B or GM-CSF release. The stimulated cytokine and complement production in response to the yeasts was highly variable in monocytes from different donors, but there was a consistent inverse relationship between C3 and GM-CSF concentrations in the monocyte supernates. This is in agreement with the previously described suppressive effect of GM-CSF on yeast-induced C3, but not factor B production. The monocyte responses elicited by a specific yeast species may be linked to its pathogenicity, and may also explain the predilection of some yeasts for particular underlying diseases.

Introduction

Candidosis is the most widespread systemic fungal disease in man, and severe infections caused by Candida species are common in immunocompromised patients. C. albicans accounts for the majority of such serious infections, but non-albicans Candida and Torulopsis species also frequently cause disease in oncological, surgical, HIV-infected and neonatal intensive care patients.

Many studies have characterised and compared the virulence factors of various fungi, but knowledge of the host response towards different yeast species is scarce. Monocytes are important effector cells in the host defence against fungal infections. They engulf and kill the yeast, and they produce various inflammatory mediators such as cytokines, complement components, prostaglandins and oxygen radicals. Fungi activate the complement system through factor B, which results in the formation of opsonins that may bind to the surface of pathogens and enhance phagocytosis. In a previous study, C. albicans was shown to stimulate the production of complement components and granulocyte-macrophage colony-stimulating factor (GM-CSF) in human monocyte cultures, and GM-CSF modulated monocyte C3 and factor B production. Because the monocyte response towards other yeast species may differ from that found for C. albicans, the purpose of the present study was to examine C3, factor B and GM-CSF production when human monocytes were exposed to eight Candida or Torulopsis species.

Materials and methods

Isolation and culture of human monocytes

Monocytes were isolated from buffy coats of healthy, human volunteers as described previously. Briefly, mononuclear cells were obtained according to
the method of Bøyum, adjusted to $4 \times 10^6$ cells/ml in X-VIVO 10 serum-free medium (Whittaker Bioproducts, Walkersville, MD, USA) and seeded on cell culture plates. Viability determined by trypan blue (Sigma) exclusion was always > 98%. After allowing adherence for 4 h at $37^\circ C$ in air+$CO_2$ 5%, non-adherent cells were removed by vigorously washing twice in warm Hank's Balanced Salts Solution (HBSS) without calcium and magnesium (Whittaker Bioproducts); the culture medium was replaced and heat-inactivated yeasts were added. The adherent cell population consisted of > 85% monocytes, determined by differential counts (Diff-Quik, Merz + Dade, Dudingen, Germany) and non-specific esterase staining ($\alpha$-naphthyl acetate kit, Sigma). Contaminating cells were mainly lymphocytes. After cell culture for 5 days, supernates were harvested, centrifuged to remove cell debris, transferred to cryotubes (Micronic, the method of Bøyum, adjusted to 4 x 10^6 cells/ml in X-VIVO 10 serum-free medium (Whittaker Bioproducts, Walkersville, MD, USA) and seeded on cell culture plates. Viability determined by trypan blue (Sigma) exclusion was always > 98%. After allowing adherence for 4 h at 37°C in air + CO_2 5%, non-adherent cells were removed by vigorously washing twice in warm Hank’s Balanced Salts Solution (HBSS) without calcium and magnesium (Whittaker Bioproducts); the culture medium was replaced and heat-inactivated yeasts were added. The adherent cell population consisted of > 85% monocytes, determined by differential counts (Diff-Quik, Merz + Dade, Dudingen, Germany) and non-specific esterase staining ($\alpha$-naphthyl acetate kit, Sigma). Contaminating cells were mainly lymphocytes. After cell culture for 5 days, supernates were harvested, centrifuged to remove cell debris, transferred to cryotubes (Micronic, Lelystad, The Netherlands), and stored at -70°C. More than 90% of the cultured cells were viable after 5 days as judged by trypan blue exclusion. All cell culture experiments were performed in duplicate wells and the results were averaged. Recombinant human GM-CSF was generously provided by Schering-Plough Research, Bloomfield, NJ, USA.

Preparation of Candida and Torulopsis species

The yeast species included in the study were C. albicans, C. tropicalis, C. parapsilosis, C. kefyr, C. guilliermondii, T. glabrata and T. candida. They were all clinical isolates from patients with fungaemia, and were stored at -70°C after isolation. One C. albicans isolate was obtained from The National Cancer Institute, Bethesda, MD, USA; all other isolates were from our strain collection. The strains were identified by the germ tube production test and by the 32ID C kit (API-bioMérieux, Montalieu Vercieu, France). The isolates were grown on Sabouraud agar plates at 37°C for 18 h before harvesting the blastoconidia. A culture period of several days was necessary to determine factor B production in monocyte supernates. Therefore, the yeast species were heat-inactivated in boiling water for 1 h before use. Latex beads (particle diameter 3-2 μm, Sigma) were used as a particulate control substrate. Endotoxin contamination of the yeast suspensions was analysed by a Listeria amoebocyte lysate (LAL) chromogen test and was found to be < 40 pg/ml for the dilutions employed.

Determination of complement factor C3 and factor B

Antigenic human C3 and factor B were quantified by previously described double-antibody enzyme-immunoassays (EIAs). Briefly, in the C3 assay a monoclonal anti-C3 antibody (clone 13 F6) kindly provided by Dr K. Högåsen (The National Hospital, Oslo, Norway) was adsorbed to polystyrene wells overnight at a concentration of 2 μg/ml. Then bovine serum albumin (Amtec Diagnostics International Inc., Conroe, TX, USA) 1% was added, followed by cell supernates (diluted 1 in 3), a rabbit anti-human C3c diluted 1 in 3000 (Behring, Marburg, Germany) and a peroxidase-linked donkey anti-rabbit Ig diluted 1 in 1000 (Amersham, Buckinghamshire). Native C3 as well as the activation products C3b, iC3b and C3c are detected in this assay. In the factor B assay, the EIA plates were coated overnight with a monoclonal anti-human Ba at a concentration of 1 μg/ml (Quidel, San Diego, CA, USA), followed by cell supernates (diluted 1 in 1-1), goat anti-human factor B diluted 1 in 4000 (Quidel) and peroxidase-linked mouse anti-goat Ig diluted 1 in 5000 (Jackson Immuno-Research, West Grove, PA, USA). The substrate employed was ABTS [2,2'-azinobis(3-ethylbenz-thiazoline-6-sulphonic acid); Sigma], and E<sub>405</sub>, was determined with an EIA reader. Dilutions of a normal human serum (NHS) pool were used as standards. The C3 concentration in the NHS was 0-91 mg/ml, as measured by nephelometry. One unit of factor B was defined as the amount contained in a 1 in 10<sup>4</sup> dilution of NHS. Cell supernates, NHS and antibodies were diluted in phosphate-buffered saline-Tween 20 (Sigma) to a final Tween 20 concentration of 0.2%. All complement factor determinations were made in triplicate. The minimum detectable amounts of C3 and factor B were 0-4 ng/ml and 1-1 U/ml, respectively. The cell culture medium did not interfere with the EIAs for factor B or C3.

Determination of GM-CSF concentration

GM-CSF was quantified by a GM-CSF EIA kit (QuantiKine, R and D Systems Inc., British Biotechnology Products Ltd, Oxon). The assay was performed according to the manufacturer’s protocol. The minimum detectable amount of GM-CSF was 1-5 pg/ml.

Statistical analysis

The statistical significance of differences between yeast-treated cultures and controls was analysed by a two-tailed Wilcoxon rank sum test.

Results

Effect of Candida and Torulopsis species on production of C3 and factor B by monocytes

C. albicans had been shown to stimulate monocyte biosynthesis of complement component C3 and factor B in a previous study. Therefore, monocytes were examined for a similar response after treatment with other yeasts. Eight different species of heat-killed yeasts (1·5 x 10<sup>6</sup> blastoconidia/well) were added either at the initiation of cell culture (day 0) or 24 h later (day 1). Latex beads were also used as a particulate control substrate. All yeast species stimulated monocyte C3
Table I. Effect of various Candida or Torulopsis species on C3 production by monocytes*

<table>
<thead>
<tr>
<th>Yeast</th>
<th>Day 0</th>
<th>Day 1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>5.8 (0.9-17.0)</td>
<td>5.8 (0.9-17.0)</td>
</tr>
<tr>
<td>C. albicans</td>
<td>12.6 (2.3-100.2)</td>
<td>18.1 (2.3-81.9)</td>
</tr>
<tr>
<td>C. tropicalis</td>
<td>12.5 (3.6-127.2)†</td>
<td>14.8 (3.6-104.0)†</td>
</tr>
<tr>
<td>C. parapsilosis</td>
<td>12.3 (2.1-45.3)†</td>
<td>13.9 (2.4-55.9)†</td>
</tr>
<tr>
<td>C. krusei</td>
<td>14.9 (4.9-85.2)†</td>
<td>11.1 (3.7-48.5)†</td>
</tr>
<tr>
<td>T. glabrata</td>
<td>10.2 (4.0-68.8)†</td>
<td>8.7 (2.6-28.2)†</td>
</tr>
<tr>
<td>C. kefyr</td>
<td>12.2 (3.5-46.9)†</td>
<td>11.4 (3.6-33.2)†</td>
</tr>
<tr>
<td>C. guilliermondii</td>
<td>11.2 (3.5-43.3)†</td>
<td>9.9 (1.8-24.5)†</td>
</tr>
<tr>
<td>T. candida</td>
<td>9.2 (3.1-43.0)†</td>
<td>8.3 (1.6-18.1)†</td>
</tr>
<tr>
<td>Latex beads</td>
<td>6.0 (3.0-20.1)</td>
<td>7.2 (1.7-20.7)</td>
</tr>
</tbody>
</table>

*Yeasts or latex beads were added to monocyte cultures on day 0 or on day 1, and all supernates were harvested on day 5 for determination of accumulated C3 content. This resulted in treatment periods with the yeasts of 120 h and 96 h, respectively. Data are median (range) of at least eight separate experiments. †Significantly increased compared to control (p < 0.01).

production when added at the initiation of cell culture; latex beads were ineffective (table I). However, the accumulated C3 concentration in monocyte cultures treated with C. albicans, C. tropicalis or C. parapsilosis was higher when the yeasts were added on day 1 than when the yeasts were added on day 0, in spite of the shorter treatment period (table I). In contrast, C. guilliermondii and T. candida did not stimulate C3 production significantly when added on day 1 (table I).

Incubation of monocytes before stimulation with the yeasts results in maturation which may affect the secretory capacity of the cells. To elucidate the monocyte secretory response at different stages of cell culture, C. albicans was added daily from day 0 to day 4, and supernates were harvested on sequential days from day 1 to day 5. The addition of C. albicans to freshly isolated monocytes was less effective in stimulating C3 production than treatment at a later stage of cell culture (table II), in accordance with a previous study. There was a successive increase in C3 concentrations during the 5-day culture period when the yeast was added at the initiation of cell culture and when it was added at any later stage (table II). Similar findings were also obtained with other yeast species, as shown in fig. 1a. When C. albicans, C. tropicalis, C. guilliermondii or T. glabrata was added at the initiation of culture, there was a steady increase in C3 concentrations in supernates collected on days 1, 3 and 5.

The effect on factor B production of adding eight different yeast species either at the initiation of cell culture or after 24 h was examined. As with the C3

Table II. Effect on C3 biosynthesis when C. albicans was added daily from day 0 to day 4 and supernates were harvested on sequential days from day 1 to day 5*

<table>
<thead>
<tr>
<th>Harvest of supernates</th>
<th>Control without C. albicans</th>
<th>Day 0</th>
<th>Day 1</th>
<th>Day 2</th>
<th>Day 3</th>
<th>Day 4</th>
</tr>
</thead>
<tbody>
<tr>
<td>Day 1</td>
<td>1.1 (0.4-2.1)</td>
<td>1.0 (0.5-2.4)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Day 2</td>
<td>3.4 (2.2-5.4)</td>
<td>3.3 (2.1-6.1)</td>
<td>4.9 (2.6-8.9)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Day 3</td>
<td>6.5 (3.0-7.7)</td>
<td>5.9 (2.2-9.6)</td>
<td>8.2 (4.1-12.2)</td>
<td>7.9 (4.8-10.1)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Day 4</td>
<td>6.5 (4.9-11.2)</td>
<td>7.2 (3.2-8.9)</td>
<td>13.1 (5.9-17.8)</td>
<td>10.5 (7.3-22.4)</td>
<td>11.8 (6.1-17.8)</td>
<td></td>
</tr>
<tr>
<td>Day 5</td>
<td>10.3 (7.1-11.5)</td>
<td>8.4 (4.9-12.8)</td>
<td>16.3 (9.4-33.9)</td>
<td>21.1 (12.9-30.1)</td>
<td>20.3 (13.8-29.1)</td>
<td>16.1 (10.1-21.7)</td>
</tr>
</tbody>
</table>

*C. albicans was added to monocyte cultures at the times indicated, resulting in treatment periods varying from 120 h to 24 h. Supernates were harvested after 1, 2, 3, 4 and 5 days for determination of accumulated C3 production. Data are median (range) of four separate experiments.

Fig. 1. Effect of C. albicans (△), C. tropicalis (●), C. guilliermondii (○) and T. glabrata (■) on C3 production (a) and GM-CSF production (b) in human monocyte cultures. Yeasts were added at the initiation of culture, and supernates were collected after 1, 3 and 5 days, respectively. C3 and GM-CSF concentrations were determined as described in Materials and methods. The data presented are corresponding C3 and GM-CSF measurements from one representative experiment (n = 3).
Table III. Effect of various Candida or Torulopsis species on monocyte factor B production*

<table>
<thead>
<tr>
<th>Yeast</th>
<th>Median (range) factor B concentration (U/ml) when yeasts were added on</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Day 0</td>
</tr>
<tr>
<td>Control</td>
<td>51 (1&lt;1-1-13)</td>
</tr>
<tr>
<td>C. albicans</td>
<td>129 (37-29-5)</td>
</tr>
<tr>
<td>C. tropicalis</td>
<td>191 (10-5-34)</td>
</tr>
<tr>
<td>C. parapsilosis</td>
<td>7 (1&lt;1-5-2)</td>
</tr>
<tr>
<td>C. krusei</td>
<td>20 (8-7-38)</td>
</tr>
<tr>
<td>T. glabrata</td>
<td>6 (19-26-8)</td>
</tr>
<tr>
<td>C. kefyr</td>
<td>8 (4-0-16-7)</td>
</tr>
<tr>
<td>C. guilliermondii</td>
<td>5 (1&lt;1-13-1)</td>
</tr>
<tr>
<td>T. candida</td>
<td>3 (1&lt;1-7-5)</td>
</tr>
<tr>
<td>Latex beads</td>
<td>1.5 (1&lt;1-50)</td>
</tr>
</tbody>
</table>

*Yeasts or latex beads were added to monocyte cultures on day 0 or on day 1, and all supernates were harvested on day 5 for determination of accumulated factor B content. This resulted in treatment periods with the yeasts of 120 h and 96 h respectively. Data are median (range) of at least eight separate experiments. †Significantly increased compared to control (p < 0.01).

results, C. albicans, C. tropicalis and C. parapsilosis were the most potent inducers of factor B, followed by C. krusei and T. glabrata (table III). Treatment on day 1 with any of the yeast species was usually not more effective in inducing factor B production than initial treatment (table III), in contrast to what was observed for C3.

To determine the optimal timing for stimulation of monocytes with the various yeasts, a set of experiments was performed in which six different yeast species were added daily from day 0 to day 4, and supernates were harvested on day 5. As already indicated by the previous findings, there was a different stimulatory response pattern for C3 and factor B production (fig. 2). C. albicans, C. tropicalis and C. parapsilosis were most effective C3 inducers when added on day 2 or day 3, whereas C. krusei, T. glabrata and C. guilliermondii were more effective when added at the initiation of culture (fig. 2). In contrast, the stimulatory effect on factor B production was optimal for all yeast species when the yeasts were added on day 0 (fig. 2). The complement-inducing effect of different clinical isolates within the same species was very similar for all species tested (data not shown).

Effect of Candida and Torulopsis species on production of GM-CSF by monocytes

C. albicans was shown previously to induce the release of GM-CSF in human monocyte cultures. Therefore other yeasts were examined for this capacity. Freshly isolated monocytes were treated with heat-killed yeasts (1.5 x 10^6 blastoconidia/well) or latex beads from the initiation of cell culture, and supernates were collected and examined for GM-CSF content. There was a gradual increase in GM-CSF concentration during 5 days of cell culture (fig. 1b). Therefore, in all remaining experiments, GM-CSF concentrations were determined in monocyte supernates harvested on day 5. The yeast species differed remarkably in their capacity to induce GM-CSF production. As shown in fig. 3, C. albicans, C. parapsilosis and C. tropicalis were the most effective inducers of GM-CSF. However, the increase in GM-CSF release in response to these yeasts in monocytes from the different donors was highly

Fig. 2. Effect on C3 (□) and factor B (■) production when (a) C. albicans (b) C. tropicalis, (c) C. parapsilosis (d) C. krusei, (e) T. glabrata or (f) C. guilliermondii were added to monocytes at different stages of cell culture. The yeasts were added to the cell cultures on days 0, 1, 2, 3 and 4, and all supernates were harvested on day 5 for determination of accumulated C3 and factor B production. The data presented are median values of at least five separate experiments. Control values (C) were from monocyte cultures without addition of yeasts.
MONOCYTE RESPONSES ELICITED BY YEASTS

Fig. 3. Effect of eight different Candida or Torulopsis species on the production of GM-CSF in human monocyte cultures. Yeasts or latex beads were added at the initiation of cell culture, and supernates were harvested after 5 days. Data are median (vertical bar) and range (box diagram) of at least eight separate experiments.

Fig. 4. C3 concentrations (■) and corresponding GM-CSF concentrations (—■—) in monocyte supernates harvested after cell culture for 5 days. The results presented are from two different donors, in which (a) C. tropicalis and (b) T. glabrata had been added to the cell cultures on days 0, 1, 2, 3, and 4. Control valves (C) were from monocyte cultures without yeasts.

variable, with no detectable production in some experiments and 16–28-fold increased amounts in other experiments. C. krusei and T. glabrata were much less potent in inducing GM-CSF release, and T. candida, C. kefyr, C. guilliermondii and latex beads were ineffective (fig. 3). Different clinical isolates within the same species had very similar GM-CSF-inducing effect on monocytes from the same donor (data not shown).

Effect of GM-CSF on yeast-induced production of C3 and factor B by monocytes

In a previous study, yeast-induced GM-CSF release from monocytes was shown to suppress C3 production by the same cells. Therefore, it should be noted that the three most effective GM-CSF-inducing yeasts (fig. 3) were the same species that had been shown to be more effective stimulators of C3 production when added at a late stage of cell culture than when added to freshly isolated cells (fig. 2). To clarify this possible relationship, corresponding GM-CSF and C3 concentrations in different monocyte supernates were examined. The results from two separate experiments with C. tropicalis, an effective inducer of GM-CSF, and T. glabrata, a poor inducer of GM-CSF, are shown in fig. 4. In experiment 1, a strong GM-CSF response was observed when C. tropicalis was added to freshly isolated monocytes, but with a gradually attenuated response by the cultured cells. The corresponding C3
concentrations were very low. The GM-CSF response to T. glabrata was much less pronounced, and again attenuated in cultured cells. In experiment 2, there was no GM-CSF release in response to either C. tropicalis or T. glabrata, and the corresponding C3 concentrations were remarkably high. As monocyte C3 production probably was unaffected by GM-CSF in this experiment, the stimulatory effect of both species increased with increasing duration of the treatment period. Thus, local GM-CSF concentrations seem to be important in regulating monocyte C3 production.

To further document that GM-CSF may suppress C3 production induced by yeast species other than C. albicans, recombinant GM-CSF was added at the initiation of cell culture together with either C. tropicalis or T. glabrata. As shown in fig. 5a, GM-CSF suppressed constitutive C3 production and nearly abrogated the stimulatory effect of both C. tropicalis and T. glabrata on monocyte C3 production. However, GM-CSF did not affect yeast-induced factor B production (fig. 5b).

**Fig. 5.** Effect of GM-CSF on basal and yeast-induced monocyte production of C3 (a) and factor B (b). C. tropicalis and T. glabrata were added to the monocyte cultures alone or in combination with GM-CSF (100 ng/ml) on day 0, and supernates were collected after cell culture for 5 days. Data are median (vertical bar) and range (box diagram) of four separate experiments.

**Discussion**

In immunocompromised patients, Candida and Torulopsis species are important causes of morbidity and mortality. Host defence mechanisms towards fungal diseases include activation of the complement system, antibody and cytokine responses, engulfment...
and killing by phagocytes, and development of cell-mediated immunity.\textsuperscript{9, 18} However, knowledge of host responses towards specific yeast species is limited.

Eight different yeast species were studied for their ability to stimulate monocytes. The four most frequent fungaemia isolates in Norway in 1991 were \textit{C. albicans} (69\%), \textit{T. glabrata} (14\%), \textit{C. parapsilosis} (9\%) and \textit{C. tropicalis} (3\%) (Per Sandven, personal communication). Therefore, these common species were chosen and compared with four species that cause disease only occasionally in man, namely \textit{C. krusei}, \textit{C. kefyr}, \textit{C. guilliermondii} and \textit{T. candida}. Because all yeasts were killed by heat-inactivation before use, differences in properties such as germination were eliminated.

All eight yeast species stimulated C3 production when added to freshly isolated monocytes, and there was a steady increase in C3 concentrations in the monocyte supernates during a 5-day culture period. However, the optimal time for adding the yeasts differed for the various species. \textit{C. albicans}, \textit{C. tropicalis} and \textit{C. parapsilosis} were better inducers of C3 when added at a late stage of monocyte culture, whereas all the other yeasts were more effective when added at the initiation of culture. In contrast, only \textit{C. albicans}, \textit{C. tropicalis}, \textit{C. parapsilosis}, \textit{C. krusei} and \textit{T. glabrata} stimulated factor B production, and the stimulatory effect was always optimal when the yeasts were added on day 0. These five species, which are the most virulent yeasts,\textsuperscript{19} were also the most effective inducers of GM-CSF. In contrast, \textit{T. candida}, \textit{C. kefyr} and \textit{C. guilliermondii}, which seldom cause disease in man, were ineffective. Thus, the complement- and cytokine-inducing capacity seems to be linked with the pathogenicity of the yeasts.

GM-CSF release from monocytes in response to the yeasts varied remarkably between the different donors tested, but was consistently higher in freshly isolated cells than in cultured cells, in agreement with previous work.\textsuperscript{13} Moreover, the yeast-induced GM-CSF release seemed to closely regulate C3 production by the same cells. When GM-CSF production was high, C3 production was low and vice versa. As GM-CSF release was attenuated in cultured cells, this may explain that the three most potent GM-CSF-inducing yeast species were better stimulators of C3 when added at a late stage of cell culture. The suppressive effect of GM-CSF on yeast-induced C3 production was further confirmed in experiments in which recombinant GM-CSF was added together with the yeasts at the initiation of culture. Both \textit{C. tropicalis}- and \textit{T. glabrata}-induced C3 production was suppressed very effectively by exogenous GM-CSF.

The yeast-stimulated release of GM-CSF may be important in host defence against fungal disease because this cytokine augments human neutrophil and monocyte fungicidal activity.\textsuperscript{20-23} In a previous study it was concluded that large granular lymphocytes were the major source of GM-CSF production when mononuclear cells were stimulated with \textit{C. albicans}.\textsuperscript{22} In the monocyte cultures in the present studies, contaminating large granular lymphocytes may also have contributed to the GM-CSF production induced by the yeasts.

Other studies have also reported a diversity of cellular responses against different yeast species. For instance, \textit{C. albicans}, but not \textit{T. glabrata}, stimulated the release of prostaglandins and other arachidonic acid metabolites from endothelial cells.\textsuperscript{24} Brief exposure to \textit{C. albicans} suppressed the adherence ability of human neutrophils, whereas \textit{C. parapsilosis} and \textit{C. guilliermondii} did not affect this important cell function.\textsuperscript{25} Furthermore, \textit{C. parapsilosis} elicited a greater oxidative metabolic response from murine macrophages than \textit{C. albicans}.\textsuperscript{26} The cell wall of \textit{C. albicans} contains polysaccharides such as mannan and \(\beta\)-glucan, components that may be important in initiating an inflammatory response by the host. It was reported recently that alveolar macrophages secrete TNF-\(\alpha\) in response to \textit{C. albicans} mannan.\textsuperscript{27} Interestingly, there was also a detectable response to \textit{C. tropicalis} mannan, although much less pronounced, whereas \textit{S. cerevisiae} mannan was ineffective. Thus, expression of the stimulatory compound is not an all or none feature of the cell wall, but may vary between related yeast species and strains.

Since leucocytes are important effector cells in host defence, leukopenic patients are particularly vulnerable to serious fungal infections. Large clinical studies on candidaemia have shown a predilection of some \textit{Candida} species for certain underlying diseases. Specifically, \textit{C. tropicalis} and \textit{C. krusei} have emerged as the pre-eminent pathogens in leukaemic patients, \textit{C. albicans} fungaemias are evenly distributed between patients with haematological and non-haematological diseases, whereas \textit{C. parapsilosis} and \textit{T. glabrata} are isolated most frequently from patients with solid tumours or non-oncological diseases.\textsuperscript{19, 28} Interestingly, neutropenia preceded fungaemia in only a minority of patients with \textit{T. glabrata} infections.\textsuperscript{17, 19} Moreover, previous studies have documented a poor host response against \textit{T. glabrata}.\textsuperscript{29, 30} This may be in accordance with the relatively poor monocyte-stimulating ability reported in the present study, a property which may increase the risk of infection with this particular species in non-leukopenic patients with normal monocyte and granulocyte responses. However, since the potent inducer of GM-CSF, \textit{C. parapsilosis}, is also frequently isolated from non-leukopenic patients, additional factors may also contribute to this relationship, such as parenteral hyperalimentation and the presence of prosthetic devices.\textsuperscript{2, 19} The low incidence of fungal disease caused by \textit{C. kefyr}, \textit{C. guilliermondii} and \textit{T. candida} may be due to poor invasive properties of these species combined with a rare occurrence in the host's natural flora, factors which may be more important than their monocyte-stimulating capacity.

Further knowledge of host response mechanisms towards different yeast species may lead to a better understanding of the pathogenicity of specific fungi,
including the predilection for infection in patients with certain underlying diseases.

References


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