Brief report

Monocytic parameters in patients with dysthymia versus major depression

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Abstract

Background: The studies on monocytic function during depression are controversial. A better knowledge of affective disorders may improve the differential diagnosis of depression subtypes. Our goals are to examine if there are differences in monocytic function in patients with major depression and dysthymia.

Method: Twenty-two depressed patients (12 dysthymia and 10 major depression) and 15 healthy controls participated in the study. We analyzed monocyte count, monocyte subsets (CD14+, CD16+, and HLA class-II+), respiratory burst activity, phagocytic index and the interleukin (IL)-1, IL-6 and tumor necrosis factor (TNFα) production. Results: Depressed patients showed elevated IL-1β (P < 0.05) and IL-6 (P < 0.01), elevated monocytic respiratory burst activity (P < 0.01); and reduced surface molecule expression HLA class-II and phagocytosis (P < 0.01) compared with controls. We found no differences in any monocytic parameters between dysthymia and major depression. Limitations: The small sample size and the short wash-out reduce the reliability of the results. Conclusions: Major depression and dysthymia show similar signs of both monocytic activation and suppression. These alterations may be due to the depressive syndrome and not to the characteristics of depression subtypes studied.

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1. Introduction

Over the past few decades, tremendous strides have been made in the understanding of the basic psychoneuroimmunology of affective disorders (Irwin, 2001). Unfortunately, although the data available gives a robust confirmation of the existence of immune system alterations, the results remain controversial and even contradictory. This could be due in part to heterogeneous patient samples investigated in many published studies.

Initial research focussed on global cell counts and on the different subsets of immune cells in depression, with few consistent results (Maes et al., 1992; McAdams and Leonard, 1993). To our knowledge, there are no studies that compare these enumerative measures between major depression and dysthymia.

More recently, several authors have written about the role of the cytokines in the pathophysiology of depression (Maes, 2001). The production of IL-1β upon mitogen stimulation was reported to be increased (Owen et al., 2001), not altered (Brambilla...
and Maggioni, 1998), or decreased (Weizman et al., 1994) in patients with major depression. In patients with dysthymia some studies found elevated IL-1\(\beta\) compared with controls (Anisman et al., 1999b). Most authors have found elevated IL-6 production in major depression (Sluzewska et al., 1996; Natelson et al., 1999). Only one study shows a TNF\(\alpha\) elevation in major depression (Lanquillon et al., 2000). Our group reported possible differences in cytokine patterns between major depression and dysthymia (Schlatter et al., 2001).

Finally, the phagocytic capacity and surface molecule expression (HLA class-II, CD14 and CD16), have shown also some controversial results (McAdams and Leonard, 1993; Cervera-Enguix and Rodríguez-Rosado, 1994; Landmann et al., 1997). Regarding monocytic respiratory burst activity, we have not found any study in depressed patients. To our knowledge, there are no studies available that compare these monocyte parameters between major depression and dysthymia.

The goals of this study are (i) to evaluate monocytes functional and poblational parameters in depressed patients. For this purpose we will include the study of the respiratory burst activity, a novel parameter in immunology research during depression, as well as other parameters previously used; and (ii) to examine whether these alterations differ between two subtypes of depression (dysthymia and major depression).

2. Method

We recruited 37 subjects (age range: 18–65 years) for this study: 15 healthy controls (M/F: 7/8; mean age±S.D. 35.3±8.7 years), and 22 depressed patients (M/F: 8/14; 43.8±12.0 years). Twelve patients had dysthymia (DSM-IV: 300.4) and 10 had major depression (DSM-IV: 296.2) (see Table 1). All patients had a HAM-D score ≥ 7.

We excluded subjects who suffered from any medical disorder that required drug treatment that could affect immune function including psychotropic drugs. Pregnant or lactating women were also excluded.

After the informed consent was obtained, the patients underwent a full physical examination and a general laboratory exam. The symptom severity was measured by the Hamilton Depression Rating Scale (HAM-D, 21 item) (Hamilton, 1967), Hamilton Anxiety Rating Scale (HAM-A) (Hamilton, 1959) and the Clinical Global Impression Scale (CGI) (Guy, 1976). We also used the Newcastle Scale to evaluate the endogeneity of depression.

Samples of peripheral blood were obtained by vein puncture with EDTA for the monocyte populations study and with natrium heparin for the monocyte function and cell stimulation study. Monocyte count was obtained by the cell counter Coulter-Micro-Dipl II (Coulter Corp., Miami, Florida, USA). The samples were obtained between 8.00 and 9:00 a.m., to minimize the circadian rhythm effect on the comparative studies. Monocyte subsets were determined by flow cytometry using combinations of monoclonal antibodies marked with fluorochromes and directed against leukocyte surface antigens: Anti: CD16 FITC, CD14 PE, HLA-DR PE (Coulter Immonotech. Co., Miami, Florida, USA). As controls, we used antibodies of an irrelevant isotype marked with FITC or PE. The analysis of the data was done in the cytometer.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Control</th>
<th>Major depression</th>
<th>Dysthymia</th>
<th>Total: depression</th>
</tr>
</thead>
<tbody>
<tr>
<td>(N)</td>
<td>15</td>
<td>10</td>
<td>12</td>
<td>22</td>
</tr>
<tr>
<td>Sex</td>
<td>7M/8F</td>
<td>2M/8F</td>
<td>6M/6F</td>
<td>8M/14F</td>
</tr>
<tr>
<td>Age</td>
<td>35.3±8.7</td>
<td>43.8±12.9</td>
<td>43.8±14.7</td>
<td>43.8±12.0</td>
</tr>
<tr>
<td>HAM-D</td>
<td>–</td>
<td>21.3±7.9**</td>
<td>12.8±3.4</td>
<td>16.6±7.6</td>
</tr>
<tr>
<td>HAM-A</td>
<td>–</td>
<td>19.5±9.7</td>
<td>12.4±7.5</td>
<td>15.9±9.2</td>
</tr>
<tr>
<td>Newcastle Scale</td>
<td>–</td>
<td>6.22±2.5***</td>
<td>1.3±1.3</td>
<td>3.6±3.1</td>
</tr>
<tr>
<td>CGI</td>
<td>–</td>
<td>4.9±0.6</td>
<td>4.4±0.8</td>
<td>4.7±0.7</td>
</tr>
</tbody>
</table>

**\(p<0.01\), major depression vs. dysthymia; ***\(p<0.001\), major depression vs. dysthymia. HAM-A (Hamilton Anxiety Rating Scale); HAM-D (Hamilton Depression Rating Scale); CGI (Clinical Global Impression Scale).
EPICS XL (Coulter Corp., Miami, Florida, USA) using XL-2 software (Coulter Corp.). The analysis was done in cell populations selected according to size and cellular complexity (forward scatter and side scatter). Results were expressed in absolute values and in percentages of cell population positive for one or two fluorochromes.

Respiratory burst activity was determined in samples of blood with heparin using the kit BURSTTEST (ORPEGEN, Pharma, Heideberg, Germany). Analysis of data was done in the cytometer FACSCalibur (Becton–Dickinson, New Jersey, USA) using CellQuest software (Becton–Dickinson, New Jersey, USA). Results are expressed as mean fluorescence intensity expressed in the cell population, which was selected according to size and complexity criteria. The assay procedure to evaluate the phagocytic index was similar, but using the PHAGOTEST kit (ORPEGEN, Pharma, Heideberg, Germany), and with FITC marked E. coli.

To measure cytokine production, these are cultured with E. coli lipopolysaccharide (LPS), incubated during 24 h (IL-1β) and with phytohemagglutinin and LPS for 48 h (IL-6 and TNFα). Cytokine quantification was determined with enzyme-linked immunosorbent assay (ELISA) techniques using R&D systems kits (R&D Systems Europe Ltd., Abingdon, Oxon, UK).

We performed comparative analyses of normal variables between all groups with an unpaired two-tailed Student’s t-test. We performed correlations between HAM-D, HAM-A and NS scores with the Pearson’s rank correlation coefficient.

### Results

The mean ± S.D. HAM-D score in patients with major depression (21.3 ± 7.9) was higher than in patients with dysthymia (12.8 ± 3.4) (*P < 0.01*), but we found no differences in HAM-A scores. The mean of the Newcastle Scale scores were higher in major depression patients (mean ± S.D. 6.22 ± 2.5; *P < 0.001*) than in dysthymia. There were no differences in CGI.

### Table 2
Mean measures and cytokines production (mean ± S.D.) in total depressed patients (major depression and dysthymia) and healthy controls

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Control</th>
<th>Major depression</th>
<th>Dysthymia</th>
<th>Total: depression</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. leukocytes</td>
<td>6933.3 ± 1659.0</td>
<td>6791.3 ± 3585.1</td>
<td>6058.3 ± 1266.6</td>
<td>6391.3 ± 2547.2</td>
</tr>
<tr>
<td>No. monocytes</td>
<td>405.0 ± 75.2</td>
<td>470.3 ± 172.1</td>
<td>436.4 ± 92.0</td>
<td>452.2 ± 134.3</td>
</tr>
<tr>
<td>% Monocytes</td>
<td>6.1 ± 1.5</td>
<td>7.1 ± 2.1</td>
<td>7.1 ± 1.6</td>
<td>7.1 ± 1.7</td>
</tr>
<tr>
<td>No. lymphocytes</td>
<td>2319.1 ± 518.8</td>
<td>2248.4 ± 780.9</td>
<td>2302.0 ± 669.3</td>
<td>2277.6 ± 704.8</td>
</tr>
<tr>
<td>% Lymphocytes</td>
<td>33.9 ± 4.8</td>
<td>34.3 ± 9.7</td>
<td>38.1 ± 8.9</td>
<td>36.4 ± 9.3</td>
</tr>
<tr>
<td>IL-1β</td>
<td>1780.2 ± 394.4</td>
<td>2030.3 ± 966.1</td>
<td>2704.0 ± 1137.4</td>
<td>2435.3 ± 1098.2*</td>
</tr>
<tr>
<td>IL-6</td>
<td>104.6 ± 12.7</td>
<td>125 ± 19.5</td>
<td>121.2 ± 12.4</td>
<td>122.7 ± 15.3**</td>
</tr>
<tr>
<td>TNFα</td>
<td>61.2 ± 1.8</td>
<td>62.8 ± 3.1</td>
<td>61.0 ± 1.9</td>
<td>61.7 ± 2.6</td>
</tr>
</tbody>
</table>

* *P < 0.05, total depression vs. control; **P < 0.01, total depression vs. control.

### Table 3
Monocyte subsets, phagocytic index (PhI) and respiratory-burst activity (RBA) (mean ± S.D.) in total depressed patients (major depression and dysthymia) and healthy controls

<table>
<thead>
<tr>
<th>CD14+</th>
<th>CD16+</th>
<th>HLA class-II+</th>
<th>PhI</th>
<th>RBA mean IF channel</th>
</tr>
</thead>
<tbody>
<tr>
<td>% Cell</td>
<td>Absolute cell</td>
<td>% Cell</td>
<td>Absolute cell</td>
<td>% Cell</td>
</tr>
<tr>
<td>Control</td>
<td>93.6 ± 4.3</td>
<td>383.0 ± 75.5</td>
<td>5.2 ± 3.4</td>
<td>19.4 ± 13.3</td>
</tr>
<tr>
<td>Major depression</td>
<td>91.7 ± 4.3</td>
<td>454.2 ± 166.3</td>
<td>8.7 ± 7.4</td>
<td>41.7 ± 35.8</td>
</tr>
<tr>
<td>Dysthymia</td>
<td>91.6 ± 4.9</td>
<td>399.0 ± 94.3</td>
<td>6.1 ± 4.5</td>
<td>26.1 ± 20.3</td>
</tr>
<tr>
<td>Depressed</td>
<td>91.6 ± 4.5</td>
<td>423.8 ± 130.8</td>
<td>7.3 ± 5.9</td>
<td>33.2 ± 28.6</td>
</tr>
</tbody>
</table>

* *P < 0.05, depressed vs. control; **P < 0.01, depressed vs. control.
scores between the two subgroups of depression (see Table 1).

We detected a non-significant ($P = 0.271$) elevation in the monocyte count in depressed patients (see Table 2). There were no correlations between monocyte count and HAM-D and HAM-A scores.

There was a significant increase of IL-1β ($P < 0.05$) and IL-6 ($P < 0.01$) production in depressed patients compared with controls, but there were no differences between major depression and dysthymia (see Table 2). We found no correlation between cytokine production and the severity of depression or anxiety. The production of TNFα following mitogen stimulation was unchanged in patients compared to controls.

The phagocytosis ($P < 0.01$) and the HLA class-II expression ($P < 0.01$) were lower in depressed patients than in controls. The CD14 and CD16 expression was unchanged in patients with major depression compared to dysthymia. We found a significant increase of the respiratory-burst activity in depressed patients compared with controls ($P < 0.05$). None of the above-mentioned parameters showed a significant difference between major depression and dysthymia. We found only a correlation between CD16 expression, absolute ($r = 0.6; P < 0.01$) and relative ($r = 0.7; P < 0.01$), and the HAM-A scores (see Table 3).

### 4. Discussion

Absolute and relative immune cell count did not differ between depressed patients and controls. However, the patient’s sample showed a tendency towards monocytosis as reported by the authors (Maes et al., 1992; Seidel et al., 1996).

The IL-1β, IL-6 and TNFα are proinflammatory cytokines. The increase found in our sample in IL-1β and IL-6 suggests an unspecific inflammatory reaction (Sluzewska et al., 1996). We were not able to reproduce Anisman’s findings (Anisman et al., 1999b) who found a significant correlation between IL-1β production and HAM-D scores.

The phagocytosis appears lower in our depressed patients’ sample. This previously observed fact is consistent with the decrease of HLA class-II expression supported by the alteration of the vimentin filaments (Cervera-Enguix and Rodriguez-Rosado, 1994). Depressed patients had higher respiratory-burst activity than controls, however these changes may also be due to anxiety or to perceived stress.

The monocytic parameters evaluated showed no correlation with HAM-D and HAM-A scores. However, the few statistically significant data available in the literature is contradictory (Maes et al., 1992; Anisman and Merali, 1999a). We only found correlation between CD16+ monocytes ($r = 0.7; P < 0.01$) and HAM-A scores, and also between this parameter and NS ($r = 0.6; P < 0.05$), that suggests its possible use as a biological marker for the autonomic syndrome. To our knowledge, this is the first time that this finding is reported.

It is difficult to evaluate the role of perceived stress. Some studies (Song et al., 1999) describe the increase of monocyte count and their cytokines in stressed subjects. However, we should refer more to an acute or sub-acute stress in major depression and to a chronic stress in dysthymia, even though differences between both of them have not been detected.

When we compared the two subgroups of depressed patients (major depression and dysthymia), differences in monocytic parameters could not be detected. There are few studies that compare both subtypes of depression. Moreover, the diagnostic group of major depression and dysthymia according to DSM-IV criteria is too heterogeneous. It may be interesting to know the pathophysiology of dysthymia (i.e. immune function) in order to differentiate from major depression, specially from melancholic depression (Maes et al., 1995c; Rothermundt et al., 2001).

To our knowledge, although there are some studies comparing minor with major depression (Maes et al., 1992, 1993) there are few authors that compare major depression and dysthymia (Ravindran et al., 1995; Zaharia et al., 2000). Despite the small sample size, our results suggest that the monocytic dysfunction is similar in major depression and dysthymia, and that the extent of immune alteration is not related to depression severity. These results will need to be replicated in order to confirm our findings.

### References


Anisman, H., Ravindran, A.V., Griffiths, J., Merali, Z., 1999. IL-1β


