Title: Alexithymia and Interleukin Variations in Somatoform Disorder

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Abstract

Background: Previous studies have demonstrated probable involvement of immune activation and selective immune dysfunction in patients with somatoform disorders (SFD). The aim of the present study was to investigate if SFD are associated with changes in the normal serum levels of important interleukins and further, to establish if these changes are related to the presence and severity of alexithymia (reduced ability to identify and express emotions) in the patients with SFD.

Methods: Twenty-four patients who met ICD-10 diagnostic criteria for SFD completed the psychological questionnaire to assess alexithymia (TAS-scale), symptom reporting (SCL-90-R) and diagnostic for SFD (SOMS-scale). Serum concentrations of soluble interleukin 2 receptor alpha (sIL-2 Rα), IL-4, IL-6, IL-10, IL-12 were determined in patients with SFD and in nine healthy subjects.

Results: Patients with SFD exhibited elevated levels of alexithymia compared to population norms, with particularly high scores on the “Difficulty identifying feelings” sub-scale of the TAS-26. Furthermore, serum levels of IL-6 ($p<0.001$) and IL-10 ($p=0.047$) were significantly increased in patients with SFD in comparison to healthy controls. Additionally a negative correlation was observed between the level of alexithymia (total TAS-score) and the serum levels of sIL-2 Rα ($r=-0.538$). Taken together these results suggest that SFD, with clinically significant alexithymia, are associated with a reduction in TH1 mediated immune function and an increase in the activation of the TH2 immune function, indicated by the augmented serum levels of the interleukins (6 and 10).

Conclusions: These data suggest that alexithymia is associated with chronic activation of the immune system and may be critically involved in the pathophysiology of bodily symptoms in SFD.

Key words: Somatoform disorders - Immunology -Interleukin - Alexithymia - IL-6 - IL-10 - sIL-2 Rα

Abbreviations: **SFD =** Somatoform Disorders; **TAS-26 =** 26-Item Toronto Alexithymia Scale; **SCL-90-R =** Symptom Checklist-90 Revised; **IL =** Interleukin; **SOMS =** Screening for Somatoform Symptoms scale; **sIL-2 Rα =** Soluble interleukin-2 receptor α.

1. Introduction

In 1980 the DSM-III introduced the category of somatoform disorders (SFD) as a provisional grouping of psychiatric conditions characterized by often multiple and variable somatic symptoms – commonly seen in general medical practice and primary care – but defying medical explanation (Ustun and Sartorius, 1995). It is clear from their high lifetime prevalence (4-5%) in the general population (Swartz et al., 1986; Escobar et al., 1987, 1998), their high comorbidity with other conditions, their debilitating effect on patients, and their association with high levels of drug use, that SFD are of great clinical and economic importance (Barsky et al., 2005).

Previous studies (e.g. Bach & Bach, 1995; Bankier et al., 2001) have reported that, relative to healthy participants, patients with SFD exhibit elevated levels of alexithymia. Alexithymia is a concept that was developed by Sifneos (1973) and means literally “absence of words for emotion”. Alexithymia is characterised by an inability to describe and identify feelings, by an absence of fantasies, and by the tendency to utilise an externally focused analytical cognitive style. The concept of alexithymia has been examined in a variety of different medical, psychosomatic and psychiatric disorders (Naatanen et al., 1999; Sifneos, 2000; Wise et al., 2000). The findings of these studies support the conceptualisation of alexithymia and the clinical impression of an association between somatization and alexithymia (Bankier et al., 2001; Lundh & Simonsson-Sarnecki, 2001).

Previous research (e.g. Singh & Fudenberg, 1986) has proposed an important relationship between the central nervous system (CNS), the endocrine system, and the immune system, that mediates neuroendocrine regulation of immune function. Furthermore, a number of studies (e.g. Dantzer et al., 1998; Pollmächer et al., 2002; Dantzer, 2005) have identified a key role for cytokines such as Interleukin-1 (IL-1), IL-6 and tumour necrosis factor (TNF-α) in the interaction between the immune and central nervous systems. For example, administration of cytokines (rIL-2 & IL-6) has been shown to induce a number of psychiatric symptoms; most notably depressed mood, apathy, fatigue (Walker et al., 1997; Späth-Schwalbe et al., 1998). Moreover, Kelly-Welch et al. (2005) reported that the deregulation of anti-inflammatory cytokines such as IL-4 may lead to a disturbance of the type1/type2 cytokine balance. A major consequence of this cytokine imbalance is an increase in TH2 response, with a resultant increase IgE antibody synthesis (a type of immunoglobulin usually released during an allergic reaction) leading to an inflammatory response in the body.

As noted earlier, SFD are associated with bodily symptoms (most notably pain) which cannot be adequately explained by organic causes. With this in mind, it is notable that previous studies (e.g. Schaefer & Stein, 1999) have reported that certain cytokines can stimulate inflammatory pain. It would seem plausible that the inflammatory response of the immune system (mediated by cytokines) might underlie the illness symptoms reported by patients with SFD. However, contrary to this notion, Rief et al. (2001) reported that patients with somatization syndrome actually exhibited changes in immune function that are associated with a moderate reduction in the inflammatory response, such as reduced levels of proinflammatory cytokines such as IL-6, and lowered CD8 (lowered T-lymphocytic activity), and yet on the other hand increased interleukin-1 receptor-antagonist (IL-1RA) with monocytic activation. This apparent inconsistency is worthy of further study.

As noted previously, alexithymia is related to an increase in the bodily expression of emotional distress. With this in mind, it is plausible that high levels of alexithymia would be associated with changes in immune function. In line with notion, Todarello et al. (1994, 1997) reported higher levels of alexithymia in patients with cervical intraepithelial neoplasia (precancerous lesions of cervix) compared to patients without such lesions. Furthermore, they reported lower counts of almost all lymphocyte subsets in the alexithymic patients compared to the non-alexithymic patients. Similarly, Dewaraja et al. (1997) reported that healthy men categorised as highly alexithymic (based on their scores on the TAS) exhibited decreased cytotoxic lymphocyte counts (for natural killer subset CD57-CD16+ cells and killer effective T cell CD8+CD11a+ cells) compared to non-alexithymic men. Corcos et al. (2004) examined alexithymia, mood (depression & anxiety) and serum levels of the different cytokines in a group of seventeen healthy women and found a positive correlation between the participants' scores on alexithymia scale (TAS) and their serum levels of IL-4. A similar relationship was observed between the participants' scores on the "difficulty identifying feelings" subscale of the TAS and their observed serum levels of IL-4. A stepwise multiple regression revealed that TAS scores were the only variables that predicted IL-4 levels; as depression, age, and body mass index (BMI) did not enter as significant predictors. It is notable that Corcos et al. (2004) were unable to obtain detectable levels of IL-1 and IL-2 in the majority of their sample, thus they were unable to establish if alexithymia was related to changes in the serum levels of these cytokines.

As there have only been a few studies examining activation of the inflammatory response system (IRS) and changes in immunological variables in SFD (Rief et al., 2001; Dantzer, 2005) it is important to extend this research in order to develop our understanding of the role of the immune response in SFD. Similarly, as there has been relatively little research addressing the effects of alexithymia on the immune function (e.g., Corcos et al., 2004; Guilbaud et al., 2003; Todarello et al., 1994; 1997) it is essential to make advances to this work. Considering that patients with SFD have been shown to exhibit elevated levels of

alexithymia (Bach & Bach, 1995; Bankier et al., 2001) it is plausible that the observed changes in the immune function of patients with SFD might relate to differences in concomitant alexithymia. However, as yet, there has been no study that has examined the relationship between alexithymia and cytokine production in participants with SFD. Therefore, the aim of the present study was to conduct such an investigation.

In the present study, serum levels of key interleukines (IL-6, IL-4 IL-10, IL-12, sIL-2 Rα) were systematically examined in a group of patients with SFD and a group of healthy controls. The presence and severity of alexithymia in patients with SFD was also established using the TAS-26. In line with previous work we expected a higher incidence of alexithymia in SFD compared to the population norms reported in Kupfer et al. (2000; 2001). Second, it was predicted that there would be changes in the serum levels of the key cytokines in the patients with SFD relative to the healthy controls. Furthermore, it was expected that these changes in serum levels would be significantly correlated with the patients' alexithymia (TAS-26) scores.

2. Methods

2.1. Participants

Twenty-four patients (16 females, 8 males) meeting ICD-10 criteria for SFD and nine healthy controls (4 females, 5 males) took part in the present study. Ten of the SFD patients were diagnosed with somatization disorder (F45.0); 13 with somatoform autonomic dysfunction (F45.3); and one with persistent somatoform pain disorder (F45.4). The characteristics of the patients are presented in Table 1. The patients with SFD were recruited from an outpatient clinic of the Department of Medicine (Munich University), where they had been referred for diagnostic interview and counselling in psychiatric and psychosomatic field. Inclusion criteria for the patient group were the presence of SFD diagnosed according to ICD-10 criteria. The diagnosis was established through a clinical interview conducted by a trained psychiatrist (P.G.F.), who performed extensive physical and psychological assessment of the patients;

including checking for the signs and symptoms of SFD as outlined in the diagnostic criteria cited in the ICD-10 (Hiller et al., 1996). A further aid to the diagnosis of SFD was that the participants' fulfil the criteria on the SOMS questionnaire (Rief et al., 1997). Exclusion criteria were presence of other psychiatric disorders (e.g. psychosis, substance abuse disorders, & major affective disorders), medication with benzodiazepines or other psychotropic medication, steroid-hormone intake and contraceptive medication during the past four weeks. The patients were thus in principle drug free in order to minimize pharmacological influence if possible. The presence of an acute or chronic infection or recent history of an infectious disease (e.g. influenza) within the past four weeks was a further exclusion criterion, as was the presence of a serious medical disease (e.g. autoimmune-, neoplasms, cardiac-, pulmonary-, or endocrine diseases). It should be noted that there was a high level of psychiatric comorbidity in the patient sample, as twenty-three of the twenty-four patients exhibited significant symptoms of other psychiatric disorders. The most common comorbid disorder (n = 17) was dysthymia (F34.1). Depressive reaction (F43.2) was the next most common disorder (n = 4). There was also one case of an anxiety disorder (F41.1) and one case of neurasthenia (F48.0). For age and educational level there are no statistically significant differences between patient group with SFD and healthy subjects. The participants (n=9) in the healthy control group were recruited from hospital employees and from the student population at the University of Munich. The nine healthy volunteers were included following a medical examination (medical and psychiatric history) and establishment of normal laboratory parameters (including normal values of including hematological screening, blood chemistry with glucose, total protein, total bilirubin, liver enzymes, electrolytes, creatinine, urea, uric acid, cholesterol, triglycerides, semiquantitative urinalysis, and thyroid hormones, c reactive protein, luteinizing hormone, folliculo-stimulating hormone, prolactin, progesterone, oestrogens, testosterone). In general they were in good general health and there was no evidence of a serious medical disease (like autoimmune-, neoplasms, cardiac-, pulmonary-, or endocrine diseases) or a psychiatric disorder (e.g. depression or anxiety) in any of the participants in the control group. As the controls were screened for psychological

disturbance prior to inclusion in the study it was not considered necessary to conduct the same detailed psychological measurement that was conducted on the patients with SFD. In common with the (SFD) patient group, regular drug intake, the presence of acute or chronic infection and/or a recent history of an infectious disease (e.g. influenza) within the past four weeks were the exclusion criteria. Female subjects were not taking birth control pills, had regular menstrual cycles, but actual menstrual status was not considered. The study protocol was approved by the Ethical Committee of the University Munich in accordance with the declaration of Helsinki. Full written informed consent was obtained from each participant before study inclusion.

2.2. Materials and Procedure

Participants fulfilling the inclusion criteria were invited to participate in the study and were assessed using a number of psychological measures. Blood serum levels of the different interleukines were also established for the two participant groups.

2.3. Questionnaires and Measures

The SOMS (Rief, 1997) is a self-rating questionnaire checking for 53 physical symptoms. The questionnaire includes all 33 physical complaints of the DSM-IV somatization disorder symptom list, the symptoms of ICD-10 somatization disorder, and the ICD-10 somatoform autonomic dysfunction symptom list. This questionnaire requires the participants to report if they had experienced any or all of 53 physical symptoms during the past 2 years. They were instructed only to answer "yes" if the symptoms had a significant influence on their subjective well being and if doctors did not find a sufficient explanation for the complaints. Thus, persons with physical illness were not excluded but were instructed only to report physically unexplained symptoms. Item 54 to item 68 of the SOMS cover all inclusion and exclusion criteria (first complaints before age 30 years, symptom duration, acceptance of doctor's explanation that the complaints do not have a physical origin, doctor visits due to the

symptoms, etc.). Adding the number of positively answered symptoms allows computation of the "somatization severity index" (range from 0 to 33 points), which we also used in our study to categorise the SFD patients. The number of somatization symptoms correlated $r = 0.75$ between self-ratings and interview, confirming the high validity of the SOMS.

The 90-item version of the Symptom Checklist-90 Revised (Derogatis, 1994) reveals different aspects of psychopathology; it assesses patients' current symptoms within a specified and optimal point-in-time (i.e., the past 7 days). The SCL-90-R includes three global index scales and nine symptom scales that were based on factor analysis and that include diagnostic-specific and non-specific symptoms. Patients are instructed to rate mental health symptoms that have "bothered" them within the past 7 days on a 5-point scale (0–4) ranging from "Not at All" (i.e., a "0" rating) to "Extremely" (i.e., a "4" rating). The symptom scales include the Somatization, Obsessive-Compulsive, Interpersonal Sensitivity, Depression, Anxiety, Hostility, Phobic Anxiety, Paranoid Ideation, and Psychoticism scales. The SCL-90-R index scales include the Positive Symptom Total, Positive Symptom Distress Index, and Global Severity Index. SCL-90-R index and symptom scale scores are represented as *T*-scores, with a mean of 50 and a standard deviation of 10. Higher *T*-scores reflect greater number and/or severity of patient self-reported symptoms. We defined a "clinically significant" score to be a *T*-score of 63 or higher, based on the recommendations reported in Derogatis (1994).

The TAS-26 was developed to measure alexithymia, a trait that can be characterized as the inability to identify and describe one's own emotions and those of others (Sifneos, 1973). The original version was developed as a standardized self-assessment questionnaire by Taylor et al. (1985;19929. The authorized German version (TAS-26) has subsequently been developed by Kupfer et al., (2000;2001), which consists of 26 items that can be rated on a 5-point Likert scale, was used in the present study to assess the presence and severity of alexithymia in the participants. A three-factor structure has been replicated in clinical and non-clinical groups: This measure includes 26 items that generate scores on three

dimensions: “difficulty identifying feelings”; “difficulty describing feelings” and “externally orientated thinking”. The German version was validated with a representative population sample (n=2,084) and shows adequate internal consistencies ranging between r=.67 and r=.84. The overall TAS-26 scores range from 18 to 90, because only 18 of the 26 items are used to calculate the TAS scores. Scores over raw-value 54 (respectively T-score 61) on the TAS-26 are taken to indicate significant alexithymia and this cut-off point is used to distinguish alexithymic and non-alexithymic individuals.

The Hamilton Rating Scale for Depression (HRSD; Hamilton, 1960) was included in the present study to provide an observer-rated measure of depression severity. This assessment was conducted by a fully trained psychiatrist (P.G.F). The 21-item Beck-Depression-Inventory (BDI; Beck et al., 1961) was utilised in the present study to provide an index of self-rated depression severity.

2.4. Assessment of Serum Levels of Interleukines

Blood was drawn for routine evaluation (hematology, chemistry), and hormones (thyroid hormones, luteinizing hormone, folliculo-stimulating hormone, prolactin, progesterone, oestrogens, testosterone, cortisol), immunophenotyping, and serum cytokine levels. Hematology was measured with an automated hematologic counter (Sysmex, SE 9000). Serum samples were centrifuged and serum was frozen in aliquots at -20°C. Aliquots were defrosted, a standard curve of cytokines was established, and serum levels of the different interleukines (sIL-2 Rα, IL-4, IL-6, IL-10, IL-12) were calculated (pg/ml) by using the quantikine® high sensitivity Enzyme Linked Immunosorbent Assay (ELISA) kit from R&D systems, Wiesbaden, Germany. Means of duplicate samples were used for calculation of the serum levels. The procedure was performed as recommended by the manufacturer. Unspecific laboratory findings in our asymptomatic patients (e.g. lymphopenia) were not assumed to indicate an ongoing infection, because these findings were related to a possible immunopathology in association with SFD. Infectious and inflammatory diseases, which

could interfere with serum cytokine detection, were excluded by C reactive protein and erythrocyte sedimentation rate was within normal limits in all patients. Furthermore, a hormone screen was performed to exclude endocrine diseases as cause of altered serum cytokine levels. There were two considerations when selecting the panel of cytokines investigated in the present study: first we wanted to broadly cover the different immune functions, namely the Th1/Th2 dichotomy and the subsequent T/B cell activation that triggers cellular versus humoral immunity. IL-2 and IL-6 reflect general immune activation, whereas IL-10 and IL-4 are cytokines of the Th2 immunity, which inhibits T effector cells and activates B cell immunity and herewith the humeral antibody response. IL-12 is a member of the Th1 immunity that mediates T cell mediated effector mechanisms. Second, serum levels must be detectable in physically healthy humans. IFNγ and TNFα (Th1), IL-5 and IL-13 (Th2), and IL-1α and IL-2 (general proinflammatory) are not detectable in healthy volunteers. Therefore sIL-2 Rα was chosen instead of IL2, as it has been shown in several investigations that sIL-2 Rα correlates with increased B and T cell activation and immune system activation, e.g. rheumatoid arthritis or systemic lupus erythematosis (Wolf et al., 1988).

2.5. Data analysis

All data were analysed by SPSS for Windows© 11.5. The data were examined for normal distribution using the Shapiro-Wilk Test. Since most parameters were not normally distributed, nonparametric Mann-Whitney U-Tests were performed to establish if any observed differences between the two groups in serum levels of the different interleukines were significant. Mann Whitney U tests and fishers exact test were utilised to establish of the two groups differed significantly in terms of age, education, scores on the SOMS, SCl-90-R and TAS-26. The significance of any observed relationships between the serum levels of the interleukines and participant' alexithymia (indexed by the participants' scores on the TAS-26) were established using Spearman's coefficient tests. In order to elucidate the relationship

between serum levels of the interleukines, participant characteristics, and alexithymia a forward stepwise multiple regression was conducted with serum levels (sIL-2 Rα, IL-4, IL-6, IL-10, IL-12), age and scores on the SCl-90-R entered as predictor variables and participants' TAS-26 score entered as the dependent variable.

3. Results

3.1. Participant characteristics and psychopathology:

Analysis of the participants characteristics revealed that the age of the patients with SFD (Mean=37.1, Standard deviation=10.4) did not differ significantly from the age of the controls (M=32.2, SD=6.5); p = 0.179. 45.8% of the patients and 55.6% of the control have a higher educational level (p=0.123). Analysis of the participants' scores on the SOMS revealed that the patients with SFD exhibited elevated scores (Mean=16.3, SD=10.5), indicating that these patients showed signs of moderate to severe somatization. The HAM-D-score demonstrated with 13.4 (4.2) only moderate depression severity. Analysis of the participants' subjective general psychiatric symptomatology (indexed by the participants' scores on the SCL) revealed that the patients exhibited an elevated General symptomatic-score (Mean= 69.3, SD=12.1). Our patients exceeded the mean value (50) on all scales of SCL-90-R for somatization, obsessive-compulsiveness, social insecurity, depression, anxiety, aggression, phobic anxiety, paranoid ideation, and psychoticism (data not shown).

3.2. Assessment of alexithymia:

Analysis of the participants' alexithymia (TAS-26) scores (presented in Table 2) revealed that the patients with SFD showed a TAS total score (T-value) of 55.7 (SD= 8.2). Further inspection revealed that the patients scored significantly higher on the "Difficulty identifying feelings" subscale (M=63.8, SD=12.1) over the cut-off T-value for TAS-scale 1 (Kupfer et al., 2000, 2001). However, the patients' scores in the subscale "Difficulty describing feelings"

(M=53.5, SD=8.7) and on the “Externally orientated thinking” subscale (M=47.4, SD=12.6) were not in the alexithymic range.

3.3. Interleukines and alexithymia:

Correlational analysis (presented in Table 3) revealed that sIL-2 Rα was significantly negatively related to the patients’ total TAS score; $r_s(24)= -0.54$, $p<0.05$. This analysis also revealed a significant negative correlation between IL-4 serum levels and the patients’ total TAS-26 score; $r_s(24)=-0.49$, $p<0.05$. There were no other significant relationships revealed by this analysis.

In order to further elucidate the nature of these relationships a forward stepwise linear regression was conducted and revealed a significant model with sIL-2 Rα entered as the only significant predictor of the patients’ alexithymia scores ($p = 0.041$) that accounted for 20 % (16%) of the variance in the patients’ alexithymia (TAS-26) scores; $R^2= 0.201$, R^2 adjusted= 0.159; $F(1, 20) =4.8$; $p<0.05$.

3.4. Assessment of interleukines serum levels

Analysis of the serum levels of the different interleukines detected for the patients with SFD and healthy controls (presented in Table 4) revealed that the level of IL-10 was significantly augmented in the patients (M=2.52 pg/ml, SD=2.7) in comparison to control group (M=0.65 pg/ml, SD=1.3); $U(N_1=24, N_2=9)=1.99$, $p<.0.05$. Furthermore, patients with SFD exhibited elevated levels of IL-6 (M=2.61 pg/ml, SD=1.4) relative to the controls (M=1.01pg/m, SD=0.5), $U(N_1=24, N_2=9)=3.61$, $p<0.001$. On the other hand, the two groups did not differ significantly in terms of the serum levels of IL-4, IL-12 and sIL-2 Rα, all tests $p>0.05$. IL-4

levels were not detectable in the majority of probes, in patients only in 7 subjects and in the control group only in 3 subjects were detectable.

3.5. Depression and interleukines:

Inspection of the patients' depression-scores only indicated moderate levels of depression; with 13.4 on the HAM-D (SD=4.2) and 18.8 on the BDI (SD=9.6). Importantly, correlational analyses demonstrated no significant relationships between the patients' depression-scores (HAM-D and BDI) and the observed serum levels of the interleukines (data not shown).

4. Discussion

As expected we found a higher incidence of alexithymia in patients with SFD in comparison to the norms for the German population, regarding the TAS-scale "Difficulty identifying feelings". This finding is in agreement with empirical findings which support a positive association between somatization and alexithymia (Bankier et al., 2001, De Gucht et al., 2003). Consistent with our second hypothesis we found significant changes in the serum levels of the different interleukins in the patients with SFD relative to healthy controls. Specifically, the results revealed augmented IL-6 and IL-10 in the patient group. This pattern is interesting and suggests the immune response in SFD is complex. Interleukin-6 (IL-6) is a pro-inflammatory cytokine that is released in response to trauma or inflammation. Interleukin-10 (IL-10) is a regulatory cytokine that suppresses TH1 response and induces TH2 response in general immune activation (Kidd 2003). The overproduction of IL-6 and IL-10 could lead to an increase of the TH2 response, which in turn is associated with augmented IgE antibody-synthesis contributing to the pathophysiology of allergic disease. This proposition is supported by the unpublished findings of Pedrosa Gil et al. (2006), who reported significantly higher IgE serum levels in patients with SFD in comparison to healthy controls.

The finding of augmented IL-6 is inconsistent with a number of previous studies addressing immune response to SFD. Rief et al. (2001), for example, reported significantly lower serum levels of IL-6 in patients with somatization syndrome. However, the findings of the present study are in agreement with the stress-alexithymia hypothesis (Martin & Pihl, 1986), which posits that alexithymia is related to sympathetic overactivity. Furthermore, our findings are consistent with Cohen et al. (1999), who reported that higher levels of reported psychological stress were associated with higher IL-6 lavage concentrations in participants exposed to a viral challenge. However, it is far from conclusive as Peters et al. (1999) demonstrated that the uncontrollability of a stressful event was associated with reduced IL-6 concentrations 15-30 minutes after the stressful event. Furthermore, Weber et al. (2002) observed that relaxation training significantly reduced perceived depression, stress and disturbance in a group of patients with tinnitus. However, this relaxation produced no significant change in IL-6 or IL-10 serum levels. It should be noted that, Maes et al. (1999) found elevated serum interleukin (IL-6) concentration in posttraumatic stress disorder. Similarly, Sluzewska et al. (1996) reported elevated IL-6 concentrations in patients with major depression and in a recent study Tsao et al. (2006) found higher mRNA expressions of IL-1beta, IL-6, Interferon-gamma, and TNF-alpha in a group of depressed patients relative to healthy controls. This is particularly notable as 87.5% of the patient sample in our present study exhibited comorbid depressive symptoms (either dysthymia or depressive reaction), thus it is plausible that the elevated IL-6 levels in the present study were a consequence of concurrent depression in the patients with SFD. However, the lack of a significant correlation, in the present study, between self- or clinician-rated depression severity and serum levels of the interleukins is contrary to this proposal.

In contrast to our hypothesis the observed changes in IL-6 and IL-10 were not significantly related to the participants' alexithymia scores. However, it should be noted that serum levels of IL-4 and soluble IL-2 Rα were negatively related to the patients' alexithymia scores, thus high levels of alexithymia were associated with a reduction in the levels of these interleukins.

Although the function of the soluble IL-2 Rα is unclear, increased levels of the sIL-2 Rα in biological fluids have been reported to correlate with increased immune system activation (Waldmann, 1993; Fernandez-Botran, 1991). In our patients with SFD serum levels of sIL-2 Rα were negatively correlated to alexithymia. A plausible explanation of these results is that patients with more alexithymic features may be suffering from (unnoticed) chronic stress reaction with concomitant activation of TH2 response and an associated reduction in TH1 mediated immunity (indicated by the reduction in sIL-2 Rα). This proposal is consistent with the hypothesis of Guilbaud (2003), who reported that alexithymic individuals may suffer from unnoticed chronic stress with associated changes in endocrine and immune functions, such that the TH1/Th2-balance is shifted towards TH2-dominant-immunity (augmented IgE antibody synthesis) and away from the lower cell-mediated (Th1) immune response. The negative relationship between alexithymia and IL-4 serum levels, observed in the present study, is inconsistent with the findings of Corcos et al. (2004), who reported a positive association, in a group of seventeen healthy females, between serum levels of IL-4 and scores on the TAS-subscale “Difficulty in identifying feelings”. However, in our study IL-4 levels was not detectable in the majority of the patients and therefore the results must be interpreted with care despite the statistical significance. In comparison to Corcos et al. (2004), who only included females, we investigated both genders in SFD in relation to alexithymia and interleukines. The effects are to be considered highly significant without however to regard the gender effect. It is valuable to examine the question of gender in further studies, because most studies in general population samples have found that men show more alexithymia than women do (Parker et al., 2003; Lane et al., 1998), however other studies had observed no association between alexithymia and gender (Loas et al., 2001).

It is plausible that the observed changes in interleukin levels might have contributed to the somatic symptoms of the patients’ disorder. In line with this suggestion, Friedlander et al. (1997) proposed that SFD may result from the alexithymic inability to differentiate and

elaborate affect, which gives rise to physiological arousal and a negative subjective state, which are not regulated by psychological strategies. This decoupling between subjective and physiological arousal when exposed to emotionally negative stimuli may increase alexithymic individuals' risks for stress-related illness (Stone and Nielson, 2001). Furthermore, there is evidence that physical and psychological stressors can provoke transient increases of proinflammatory interleukines (DeRijk et al., 1997; Zhou et al., 1993; Papanicolaou et al., 1998). Finally, increased serum markers of inflammation have been found in obese human (Yoshida et al., 2006, Panagiotakos et al., 2005) although the mechanism of action and the primary event are unclear. With respect to our study, obesity may strengthen an otherwise activated inflammatory process (Oeser et al., 2005). Alternatively, obesity may be the result of a psychological illness and may herewith influence immune responses. As a result, upcoming studies comparing alexithymic patients with healthy controls must include the evaluation of the body mass index and, if possible, the fat distribution pattern as well. Another limitation considering our subject sample is the fact that the control group was not assessed as thoroughly regarding psychology measures as the patient group. However, it should be noted that the participants in the control group were screened prior to participation in the study and no evidence was found of psychological disturbance. Furthermore, the control sample was relatively small, which may have influenced our ability to detect subtle changes in interleukin levels. However, a strength of our study is that our sample of patients with SFD may have been less ill than the psychiatric patients reported in previous studies, as patients with major psychiatric diseases were excluded from our study. Most studies investigating immunological variables have used patients that were being treated with medication at the time of testing. This is important because increases in the circulating levels of TNF-α, IL-6 and other inflammatory cytokines have been reported in response to psychotropic drugs, antipsychotics, antidepressants, mood stabilizers (e.g. Pollmächer et al., 2000; Hinze-Selch et al., 2000; Haack et al., 1999). Therefore, the presence of these drugs makes the interpretation of changes in interleukin levels difficult. Finding patients with SFD who are not currently medicated is difficult, thus a major strength of our study was that none

of our sample (patients and control group) was on medication at the time of testing. Although we have performed a comprehensive analysis of different immunological parameters, it remains difficult to correlate the alterations with the pathophysiology of SFD.

In summary, our study represents the first investigation of the relationship between alexithymic symptoms in somatoform disorders and changes in the serum levels of important cytokines. Patients with SFD exhibited higher levels of IL-6 and IL-10 than did the controls indicating the presence of an inflammatory response in these patients, plausibly relating to their reported somatic pain. Although the levels of IL-6 and IL-10 were not associated with the patients' alexithymia score, there was evidence of a relationship between alexithymia and an augmented TH-2 inflammatory response (indexed by reduced sIL-2 Rα).

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References:

Bach, M., Bach D., 1995. Predictive value of alexithymia: a prospective study in somatizing patients. Psychother. Psychosom. 64, 43-48.

Bankier, B., Aigner, M., Bach, M., 2001. Alexithymia in DSM-IV Disorder. Comparative Evaluation of Somatoform Disorder, Panic Disorder, Obsessive-Compulsive Disorder, and Depression. Psychosomatics 42, 235-240.

Barsky, A.J., Orav, E., Bates D.W., 2005. Somatization increases medical utilization and costs independent of psychiatric and medical comorbidity. Arch. Gen. Psychiatry 62, 903-910.

Beck, A.T., Ward C.H., Medelson M., Mock F., Erbaugh F., 1961. An inventory for measuring depression. Arch. Gen. Psychiatry 4, 561-571.

Cohen, S., Doyle, W.J., Skoner, D.P., 1999. Psychological stress, cytokine production, and severity of upper respiratory illness. Psychosom. Med. 61, 175–180.

Corcos, M., Guilbaud, O., Paterniti, S., Curt, F., Hjalmarsson, L., Moussa, M., Chambry, J., Loas, G., Chaout, G., Jeammet, P., 2004. Correlation between serum levels of interleukin-4 and alexithymia scores in healthy female subjects:preliminary findings. Psychoneuroendocrinology 29(5), 686-691.

Dantzer, R., Bluthe, R.M., Laye, S., Bret-Dibat, J.L., Parnet, P., Kelley, K.W., 1998. Cytokines and sickness behaviour. Ann. NY. Acad. Sci. 840, 586-590.

Dantzer, R., 2005. Somatization: A psychoneuroimmune perspective. Psychoneuroendocrinology 30, 947-952.

De Gucht, V., Heiser W., 2003. Alexithymia and somatisation: quantitative review of the literature.J Psychosom Res. 54(5), 425-434

DeRijk, R., Michelson, D., Karp, B., Petrides, J., Galliven, E., Deuster, P., Paciotti, G., Gold, P.W., Sternberg, E.M., 1997. Exercise and circadian rhythm-induced variations in plasma cortisol differentially regulate interleukin-1ß (IL-1ß), IL-6, and tumor necrosis factor-α (TNF-α) production in humans: high sensitivity of TNF-α and resistance of IL-6. J. Clin. Endocrinol. Metab. 82, 2182–2192.

Derogatis, L.R., 1994. SCL-90-R. Self-Report Symptom Inventory. In: Collegium Internationale Psychiatriae Scalarum (Ed.), Internationale Skalen für Psychiatrie. Weinheim: Beltz-Verlag.

Dewaraja, R., Tanigawa, T., Araki, S., Nakata, A., Kawamura, N., Ago, Y., Sasaki, Y., 1997. Decreased cytotoxic lymphocyte counts in alexithymia. Psychother. Psychosom. 66(2), 83-86.

Escobar, J.I., Burnam, M.A., Karno, M., Forsythe, A., Golding, J.M., 1987. Somatization in the community. Arch. Gen. Psychiatry 44, 713-718.

Escobar, J.I., Waitzkin, H., Silver, R.C., 1998. Abridged somatization: a study in primary care. Psychosom. Med. 60:466–472.

Fernandez-Botran R., 1991. Soluble cytokine receptors: their role in immunoregulation. FASEB J. 5, 2567-74

Friedlander, L., Lumley, M.A., Farchione, T., Doyal, G., 1997. Testing the alexithymia hypothesis: physiological and subjective responses during relaxation and stress. J. Nerv Ment. Dis. 185(4), 233-139.

Guilbaud, O., Corcos, M., Hjalmarsson, L., Loas, G., Jeammet, P., 2003. Is there a psychoneuroimmunological pathway between alexithymia and immunity? Immune and physiological correlates of alexithymia. Biomed. Pharmacother. 57, 292-295.

Haack, M., Hinze-Selch, D., Fenzel, T., Kraus, T., Kühn, M., Schuld, A., Pollmächer, T., 1999. Plasma Levels of cytokines and soluble cytokine receptors in psychiatric patients upon hospital admission: effects of confounding factors and diagnosis. J. Psychiatric Res. 33, 407-418.

Hamilton, M., 1960. A rating scale for depression. J. Neurol. Neurosurg. Psychiatry 23, 56–62.

Hiller, W., Zaudig, M., Mombour, W., 1996. International diagnostic checklists for ICD-10 and DSM-IV (Handbook), Seattle: Hogrefe & Huber.

Hinze-Selch, D., Schuld, A., Kraus, T., Kühn, M. , Uhr, M. , Haack, M., Pollmächer, T., 2000. Effects of antidepressants on weight and on the plasma levels of leptin, TNF-α and soluble TNF receptors: a longitudinal study in patients treated with amitryptiline or paroxetine. Neuropsychopharmacology 23, 13-19.

Kelly-Welch, A., Hanson, E.M., Keegan, A.D., 2005. Interleukin-4 (IL-4) pathway. Sci. STKE. 293, 9.

Kidd P., 2003. Th1/Th2 balance: the hypothesis, its limitations, and implications for health and disease. Altern Med Rev 8(3), 223-246

Kupfer, J., Brosig, B., Brahler, E., 2000. Testing and validation of the 26-item Toronto Alexithymia Scale in a representative population sample. [in German] Z Psychosom Med Psychother 46, 368.

Kupfer, J., Brosig, B., Brähler, E., 2001. Toronto-Alexithymie-Skala-26, German Version,. Göttingen-Seattle, Hogrefe Verlag.

Lane, RD., Sechrest, L., Riedel, R., 1998. Sociodemographic correlates of alexithymia, Compr. Psychiatry 39, 377–385

Loas, G., Corcos, M., Stephan, P., Pellet, J., Bizouard P. Venisse JL., 2001. Factorial structure of the 20-item Toronto Alexithymia Scale. Confirmatory factorial analyses in nonclinical and clinical samples. J Psychosom Res. 50, 255–261.

Lundh, L.G., Simonsson-Sarnecki, M., 2001. Alexithymia, emotion, and somatic complaints. J. Pers. 69(3), 483-510.

Martin, J.B., Pihl, R.O.,1986. Influence of alexithymic characteristics on physiological and subjective stress responses in normal individuals. Psychother. Psychosom. 45(2), 66-77.

Maes, M., Lin, A., Delmeire, L., Van Gastel, A., Kenis, G., DeJongh, R., Bosmans, E., 1999. Elevated serum interleukin-6 (IL-6) and IL-6 receptor concentration in posttraumatic stress disorder following accidental man-made traumatic events. Biol. Psychiatry 45, 833–839.

Naatanen, P., Rynamen, A., Keltikangas-Jarvinen, L., 1999. The influence of alexithymia characteristics on the self-perception and facial expression of physiological stress state. Psychother. Psychosom. 68, 252-262.

Oeser, A., Chung, C.P., Asanuma, Y., Avalos, I., Stein, C.M., 2005. Obesity is an independent contributor to functional capacity and inflammation in systemic lupus erythematosus. Arthritis Rheum. 52, 3651-3659.

Panagiotakos, D.B., Pitsavos, C., Yannakoulia, M., Chrysohoou, C., Stefanadis, C., 2005. The implication of obesity and central fat on markers of chronic inflammation: The ATTICA study. Atherosclerosis 183, 308-15.

Papanicolaou, D.A., Wilder, R.L., Manolagas, S.C., Chrousos, G.P.,1998. The pathophysiologic roles of interleukin-6 in human disease. Ann. Intern. Med. 128, 127–137.

Parker, JDA., Taylor, GJ., Bagby, R., 2003. The 20-Item Toronto Alexithymia Scale: III Reliability and factorial validity in a community population. J Psychosom Res. 55, 269–275.

Pedrosa Gil, F., Nickel, M., Ridout, N., Schmidmaier, R. Lymphocyte subsets and Immunglobuline differences in Somatoform disorders, in preparation, 2006.

Peters, M.L., Godeart, G.L.R., Ballieux. R.E., Brosschot, J.F., Sweep, F.C.G.J., Swinkels, L.M.J.W., van Vliet, M., Heijnen, C.J., 1999. Immune responses to experimental stress: effects on mental effort and uncontrollability. Psychosom Med. 61, 513–524.

Pollmächer, T., Haack, M., Schuld, A., Kraus, T., Hinze-Selch, D., 2000. Effects of antipsychotic drugs on cytokine networks. J. Psychiatric Res. 34, 369-382.

Pollmächer, T., Haack, M., Schuld, A., Reichenberg, A., Yirmiya, A., 2002. Low Levels of circulating inflammatory cytokines – Do they affect human brain functions. Brain Behav. Immun. 16, 525-532.

Rief, W., Hiller, W., Heuser, J., 1997. SOMS- Das Screening für Somatoforme Störungen Manual zum Fragebogen [SOMS-Screening for somatoform symptoms. Manual for the self-rating scale]. Hans Huber, Bern.

Rief, W., Pilger, F., Ihle, D., Bosmans, E., Egyed, B., Maes, M., 2001. Immunological differences between patients with major depression and somatization syndrome. Psychiatric Res. 105, 165-174.

Rief, W., Barsky, A., 2005. Psychobiological perspectives on somatoform disorders. Psychoneuroendocrinology 30,996-1002.

Sifneos, P.E., 1973. The prevalence of 'alexithymic' characteristics in psychosomatic patients. Psychother Psychosom 22(2), 255-262

Sifneos, P.E., 2000. Alexithymia, clinical issues, politics and crime. Psychothr. Psychosom. 69, 113-116

Singh, V.K., Fudenberg, H.H., 1986. Can blood immunocytes be used to study neuropsychiatric disorders ? J. Clin. Psychiatry 47, 592-595.

Sluzewska, A., Rybakowski, J., Bosmans, E., Sobieska, M., Berghmans, R., Maes, M., Wiktorowicz, K., 1996. Indicators of immune activation in major depression. Psychiatry Res 64(3), 161-167.

Späth-Schwalbe, E., Hansen, K., Schmidt, F., Schreznemeier, H., Marshall, L., Burger, K., Fehm, H.L., Born, J., 1998. Acute effects of recombinant human interleukin-6 on endocrine and central nervous sleep functions in healthy men. J. Clin. Endocrinol. Metabol. 83, 1573-1579.

Stone, L.A., Nielson, K.A., 2001. Intact physiological response to arousal with impaired emotional recognition in alexithymia. Psychother. Psychosom. 70(2), 92-102.

Swartz, M., Blazer, D., Georg, L., Landerman, R., 1986. Somatization disorder in a community population. Am. J. Psychiatry 143, 1403-1408.

Taylor, G.J., Ryan, D., Bagby, R.M., 1985. Toward the development of a new self-report alexithymia scale. Psychother Psychosom 44,191.

Taylor, G.J., Bagby, R.M., Parker, J.D.A., 1992. The Revised Toronto Alexithymia Scale: some reliability, validity, and normative data. Psychother Psychosom 57, 34-41

Taylor, G.J., Bagby, R.M., Parker, J.D.A., 1997. Disorders of Affect Regulation: Alexithymia in Medical and Psychiatric Illness. New York: Cambridge: Univ. Press.

Taylor, G.J., Bagby, R.M., 2004. New Trends in Alexithymia Research. Psychother Psychosom. 73, 68-77.

Todarello, O., Casamassima, A., Marinaccio, M., La Pesa, M.W., Caradonna, L., Valentino, L., Marinaccio, L., 1994. Alexithymia, immunity and cervical intraepithelial neoplasia: a pilot study. Psychother. Psychosom. 61(3-4),199-204.

Todarello, O., Casamassima, A., Daniele, S., Marinaccio, M., Fanciullo, F., Valentino, L., Tedesco, N., Wiesel, S., Simone, G., Marinaccio, L., 1997. Alexithymia, immunity and cervical intraepithelial neoplasia: replication.Psychother. Psychosom. 66(4), 208-213.

Tsao, C.W., Lin, Y.S., Chen, C.C., Bai, C.H., Wu, S.R., 2006. Cytokines and serotonin transporter in patients with major depression.
Prog Neuropsychopharmacol Biol Psychiatry. [Epub ahead of print]

Ustun, T.B., Sartorius, N., editors. Mental illness in general health care. Chichester, UK: John Wiley; 1995.

Waldmann, T.A., 1993. The IL-2/IL-2 receptor system: a target for rational immune intervention. Immunol Today 14, 264-70.

Walker, L.G., Walker, M.B., Heys, S.D., Lolley, J., Wesnes, K., Eremin, O., 1997. The psychological and psychiatric effects of rIL-2 therapy: a controlled clinical trial. Psycho-Oncology 6, 290-301.

Wise, T.N., Mann, L.S., Sheridan, M.J., 2000. Relationship between alexithymia, dissociation and personality in psychiatric outpatients. Psychothr. Psychosom. 69,123-127

Wolf, R.E., Brelsford W.G., 1988. Soluble interleukin-2-receptors in systemic lupus erythematosus. Arthritis & Rheumatism 31(6), 729-735.

Yoshida, T., Kaneshi, T., Shimabukuro, T., Sunagawa, M., Ohta, T., 2006. Serum C-reactive protein and its relation to cardiovascular risk factors and adipocytokines in Japanese children. J Clin Endocrinol Metab. 91, 2133-7.

Zhou, D., Kusnecov, A.W., Shurin, M.R., DePaoli, M., Rabin, B.S., 1993. Exposure to physical and psychological stressors elevates plasma interleukin 6: relationship to the activation of hypothalamic-pituitary-adrenal axis. Endocrinology 133, 2523–2530

Table 1: **Demographic and clinical characteristics of patients with SFD (Standard deviations are presented in parentheses)**

Variable	Patients with SFD (n=24)
Age	37.1 (10.4)
Gender (male/female)	8 / 16
Higher education (%)	45.8%
Somatization severity score (SOMS)	16.3 (10.5)
General symptomatic (SCL-90-R)	69.3 (12.1)
HAM-D	13.4 (4.2)
BDI	18.8 (9.6)

<u>Table 2:</u> Mean alexithymia (TAS-26) scores for patients with SFD (Standard deviations are presented in parentheses)

Scores on the TAS-26 (T-values)	**Patients with SFD (n=24)**
	Mean (SD)
Total Score	55.7 (8.2)
***"Difficulty identifying feelings"* subscale**	63.8 (12.1)
***"Difficulty describing feelings"* subscale**	53.5 (8.7)
***"Externally orientated thinking"* subscale**	47.4 (12.6)

Table 3: Relationships between alexithymia (TAS-26 score) and serum levels of the different interleukins in the patients with SFD (n = 24)

	Type of Interleukin				
	sIL-2 Rα	**IL-4**	**IL-6**	**IL-10**	**Il-12**
TAS, *"Difficulty identifying feelings"* subscale	r_s=-0.256	r_s=0.025	r_s=-0.204	r_s=-0.083	r_s=0.014
TAS *"Difficulty describing feelings"* subscale	r_s=-0.306	r_s=-0.071	r_s=0.000	r_s=0.133	r_s=-0.088
TAS *"Externally orientated thinking"* subscale	r_s=-0.027	r_s=-0.248	r_s=0.202	r_s=-0.05	r_s=0.346
TAS Total score	**r_s=-0.538***	**r_s=-0.491***	r_s=0.162	r_s=-0.031	r_s=0.019

* Significance ($p<0.05$) of correlations computed using Spearman's tests

Table 4: Mean serum levels (pg/ml) of the different interleukins for the patients with SFD and healthy controls (Standard deviations are presented in parentheses)

Type of Interleukin	Patients with SFD (N=24)	Healthy Controls (N=9)	P-Value*
sIL-2 Rα	884.1 (206.4)	898.5 (208.4)	p = 0.894
IL-4	0.004 (0.01)	0.32 (0.8)	p = 0.185
IL-6	2.61 (1.4)	1.01 (0.5)	**p = 0.000**
IL-10	2.52 (2.7)	0.65 (1.3)	**P= 0.047**
Il-12	0.27 (0.1)	0.71 (1.4)	p = 0.329

*Significance determined using Mann-Whitey U-Tests with adjusted alpha for multiple comparisons