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Journal of Affective Disorders 71 (2002) 205–209

JOURNAL OF AFFECTIVE DISORDERS

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Brief report

# Reduced production of interferon-gamma but not interleukin-10 in bipolar mania and subsequent remission

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Received 12 October 2000; accepted 4 June 2001

**Abstract**

*Background:* Activation of inflammatory response system (IRS) is suggested by increased levels of plasma soluble interleukin-2 receptor (sIL-2R) in patients with bipolar mania. The reasons for changes in stimulated interferon-gamma (IFN-γ) and interleukin-10 (IL-10) production in bipolar mania along with subsequent remission remain unclear. *Methods*: We measured phytohemagglutinin (PHA)-stimulated IFN-γ and IL-10 production in 20 physically healthy inpatients aged between 18 and 45 years with bipolar mania (DSM-IV) using Young Mania Rating Scale (YMRS) scores $\geq 26$ and in subsequent remission (YMRS $\leq 12$), as well as in 15 age- and sex-matched healthy normal controls. *Results:* The mean values of IFN-γ production in patients in acute mania and in subsequent remission were significantly lower than those of healthy controls ($P = 0.0004$, $P = 0.0005$, respectively). There was no significant difference in IL-10 production between bipolar patients in acute mania as well as in subsequent remission and healthy controls. In acute mania, the mean values of IFN-γ and IL-10 production in medicated patients ($n = 13$) did not differ from those of drug-free patients ($n = 7$). Other clinical variables had no effect on IFN-γ and IL-10 production. *Limitation:* The uncontrolled medication, small sample size of the bipolar individuals, and some immune re-measurements prior to full remission periods, limit generalization from the data in this study. *Conclusion*: Reduced production of IFN-γ without alternation of IL-10 in bipolar mania and subsequent remission suggest that the immune modulation may vary in patients with different major psychiatric disorders.


*Keywords:* Immune modulation; Interferon-γ; Interleukin-10; Bipolar disorder in Taiwan; Mania

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0165-0327/02/$ – see front matter 
PII: S0165-0327(01)00369-X

## 1. Introduction

Patients with bipolar disorder have been reported to have altered immune functions (Kronfol and House, 1988a) including leukocytosis (Kronfol et al., 1988b), and higher prevalence of thyroid autoantibodies (Lazarus et al., 1986) as well as immune-related diseases (Tsai et al., 1997b). But reports on the assessment of cytokine regulation in patients with bipolar disorders are limited. Rapaport (1994) found no evidence of immune system activation in euthymic patients with bipolar disorder. However, Maes et al. (1995a) reported that plasma concentrations of plasma soluble interleukin-2 receptor (sIL-2R) and sIL-6R were significantly increased in manic patients as compared with normal controls, and suggested activation of inflammatory response system in manic patients. Tsai et al. (1999) found that lymphocyte proliferation to phytohemagglutinin (PHA) and plasma sIL-2R levels, but not sIL-6R, of bipolar patients, were significantly higher in acute mania than in consequent remission. Thus, it is suggested that cell-mediated immunity activation in bipolar mania may be through a specifically state-dependent immune response. Furthermore, a significant positive correlation between improvement of manic severity and reduction in plasma sIL-2R levels in acute mania and subsequent remission is also demonstrated (Tsai et al., 2001).

Interferon-gamma is associated with T-helper cell type I (TH-1). IL-10, a negative immunoregulatory cytokine, is associated with T-helper cell type II (TH-2) or humoral immune activation. The secretion of interferon-γ (IFN-γ) is increased in patients with major depression (Maes et al., 1994) and decreased in patients with different subtypes of schizophrenia (Wilke et al., 1996). Rothermundt et al. (1996) reported a lower production of stimulated IL-10 and IFN-γ in acute schizophrenic patients. Unlike major depressive disorder and schizophrenia, the reasons for stimulated IFN-γ and IL-10 production in bipolar patients remain unclear. The present study examines IFN-γ and IL-10 production in bipolar patients in acute mania and in subsequent remission.

## 2. Methods

Participating subjects were acute inpatients aged between 18 and 45 years. All subjects met DSM-IV (American Psychiatric Association, 1994) criteria for bipolar I disorder, and were manic with Young Mania Rating Scale (YMRS; Young et al., 1978) scores $\geq 26$. They were diagnosed and rated by two senior psychiatrists with a validated semi-structural schedule that has been used elsewhere (Tsai et al., 1997a, 1999). The diagnosis was made by directly interviewing the patients and reviewing available medical records. Patients having mixed episode or other comorbid Axis I psychiatric disorders (e.g. substance abuse) were excluded. All subjects were free from symptoms of infection and immune diseases and had not taken any drug known to affect the immune system for at least 2 weeks before being enrolled in the study. After having obtained written informed consent, the patients and controls provided blood samples between 08:30 and 09:30 h following an overnight fast. The blood samples of those subjects were also collected in subsequent remission state with YMRS scores $\leq 12$ and being free from any sign of depression. During the index hospitalization, all patient subjects still received mood stabilizers, and typical antipsychotic drugs (haloperidol or chlorpromazine) as indicated clinically.

Peripheral blood mononuclear cells (PBMC) of patients and controls were isolated from heparinized peripheral blood with Ficoll-Paque density gradient. For the introduction of IL-10 and IFN-γ, PBMCs ($2 \times 10^6$ cells) were cultured with phytohemagglutinin (PHA) (10 μg/ml) and were incubated in a 37°C, 5% $CO_2$ humidified incubator for 48 h. After incubation, the cells were harvest by centrifugation. The supernatants were kept frozen at $-70$°C till further analysis. The levels of cytokine production were measured in duplicate using commercial ELISA reagents. Determination of cytokine concentration in the supernatants was performed using an IFN-γ kit (BioSource International, Camarilla, CA, USA) and an IL-10 kit (Endogen, Cambridge, MA, USA). The sensitivity of IFN-γ assay was 1000–15.6 pg/ml and that of the IL-10 assay was 600–15.36 pg/ml. The intra- and interassay coefficients of variation for IFN-γ and IL-10 were lower than 5.9 and 6.1%, and 10 and 10%, respectively.

IFN-γ and IL-10 levels were compared between mania and subsequent remission in bipolar individuals by Wilcoxon's non-parametric test or paired *t*-test, as applicable. Mann–Whitney test (Wilcoxon

signed rank test) and *t*-test were used for comparing bipolar and healthy individuals. Among bipolar patients in mania and consequent remission, Spearman's correlations were used to examine the relationship of continuous variables of the clinical features to IFN-γ and IL-10 production, respectively.

Difference were considered statistically significant if a *P*-value was equal to or less than 0.05.

## 3. Results

The participating subjects included 20 (eight male and 12 female) manic patients with a mean age of 31.7±9.9 (S.D.) years, and 15 (seven male and eight female) healthy controls with mean age of 24.3±3.2 (S.D.) years. The mean YMRS score of acute mania at the time of blood withdrawal for study was 35.2±4.7 (S.D.) points (range = 26–40). There were seven patients who were free of any psychotropic drugs for at least 2 weeks, while13 patients (65%) were treated with mood stabilizers and other psychotropic agents including lithium ($n = 8$), carbamazepine ($n = 2$) and valproate ($n = 3$). While follow-up blood samples of patients were collected, 14 patients were treated with lithium, two with carbamazepine, one with valproate, and three with valproate and lithium. The mean score of YMRS in the remission period was 3.5±3.8 points (range = 0–12), which indicates clinically significant improvement in manic symptoms. The mean duration between blood samplings during acute mania and in remission was 43.2±21.2 (S.D.) days.

As shown in Table 1, there is no significant difference in the production of IFN-γ and IL-10 in bipolar patients in acute mania and in remission. However, the production of IFN-γ in bipolar patients both in acute mania and in subsequent remission was significantly lower than in healthy controls. There was no significant difference in IL-10 production between bipolar disorder and healthy controls.

The medication effects on PHA-stimulated IFN-γ and IL-10 production in bipolar patients were examined. The 13 medicated patients had non-significantly higher mean value±S.D. of IFN-γ production (1345.0±2144.0 pg/ml) than seven drug-free patients (698.8±411.2 pg/ml). The mean value±S.D. of IL-10 among medicated patients (464.1±549.8 pg/ml) is comparable to that of drug-free patients (457.3±412.6 pg/ml). We also examined the possible influences of clinical variables on the changes in cytokine production, including current age, age of onset, number of episodes and length of illness. Neither PHA-stimulated IFN-γ nor IL-10 production in acute mania and in subsequent remission showed correlation to any aforementioned clinical variable.

## 4. Discussion

To the best of our knowledge, this study is the first report on IFN-γ and IL-10 production in bipolar patients. One major finding of this study is the reduced production of IFN-γ in bipolar patients as compared to normal controls. This difference in IFN-γ production is not associated with current age, age of onset, number of episodes, or length of illness in patients with bipolar disorder. The reduced IFN-γ production in both acute mania and subsequent remission suggests a trait marker in bipolar disorder.

Unlike the increased IL-10 production in major depression (Seidel et al., 1995) and the decreased IL-10 production in schizophrenia (Rothermundt et al., 1996), our data showed that neither the presence

Table 1
Interferon-γ (IFN-γ) and interleukin-10 (IL-10) levels (pg/ml) in patients with bipolar disorder and healthy controls

| | Bipolar patients ($n = 20$) | | Healthy controls ($n = 15$) | Statistics (*P*-values) | | |
|---|---|---|---|---|---|---|
| | Acute mania (A) (mean±S.D.) | Remission (B) (mean±S.D.) | (C) (mean±S.D.) | A vs. B | A vs. C | B vs. C |
| IFN-γ | 1118.8±1748.3 | 1126.8±1689.2 | 3527.3±2057.7 | NS[a] | 0.0004 | 0.0005 |
| IL-10 | 461.7±494.6 | 415.9±383.8 | 379.6±273.5 | NS[a] | NS[a] | NS[a] |

[a] Non-significantly different.

of manic symptoms nor psychotropic medication influenced IL-10 production in bipolar patients. Therefore, the present finding provides additional evidence that immune modulation may vary in different major psychiatric disorders and is in agreement with the findings of our early articles (Tsai et al., 1997b).

IL-2 stimulates the production of IFN-γ, which is a proinflammatory cytokine produced by TH-1 cells (like IL-2) and natural killer (NK) cells. The most potent suppressor of IFN-γ production, IL-10, is an anti-inflammatory cytokine produced by variable cells including T-lymphocytes, B-lymphocytes, and monocytes (Rothermundt et al., 1996). The levels of IFN-γ and IL-10 are important to determine the pro- or anti-inflammatory capacity of culture supernatants (Katsikis et al., 1995). Reduced IFN-γ production without alteration of IL-10 production in acute mania along with subsequent remission suggests a negative immunoregulatory status in bipolar patients. Furthermore, this finding shows that the immunomodulatory mechanism in bipolar individuals is partly like that of antidepressants having a suppressive effect on IFN-γ and a stimulatory effect on IL-10 (Maes et al., 1999; Kubera et al., 2000).

In the present study, psychopharmacological treatment and other clinical variables are not found to have an effect on IFN-γ or IL-10 production. However, the effects of mood stabilizers and typical antipsychotic drugs on plasma cytokines (e.g. IL-2, IL-6) along with their soluble receptors and acute phase protein of bipolar patients might be present (Maes et al., 1995b, 1997; Sluzewska et al., 1997; Rapaport et al., 1999; Tsai et al., 2001). The findings of our study imply that the normalization of immunity may be achieved through other mechanisms, such as IL-2 and IL-6 suppression (Maes et al., 1995b; Rapaport et al., 1999).

Uncontrolled medication and small sample size are major limitations of this study. Furthermore, the duration of remission in our sample is relatively short, because it is generally accepted that symptomatic recovery in mood disorders means symptomatic remission for at least 8 continuous weeks (Frank et al., 1991). In conclusion, immune modulation via production of IL-10 and IFN-γ in bipolar disorder may differ from other psychotic disorders and healthy controls. However, large sample size of medicated or drug-free manic patients and immune measurements during full remission of the index episode are warranted in further studies.

## Acknowledgements

The work was supported by the grants NSC 88-2314-B-038-116 and NSC 89-2314-B-038-077 from the National Science Council, Taiwan. The authors thank Dr Chian-Jue Kuo for statistical assistance and Dr Kun-Po Chen for data collection.

## References

American Psychiatric Association, 1994. Diagnostic and Statistical Manual of Mental Disorders, 4th Edition. American Psychiatric Press, Washington, DC.

Frank, E., Prien, R.F., Jarrett, R.B. et al., 1991. Conceptualization and rationale for consensus definitions of terms in major depressive disorder: remission, recovery, relapse, and recurrence. Arch. Gen. Psychiatry 48, 851–855.

Katsikis, P.D., Cohen, S.B., Londei, M., Feldmann, M., 1995. Are CD4 + Th 1 cells pro-inflammatory or anti-inflammatory? The ratio of IL-10 to IFN-gamma or IL-2 determines their function. Int. Immunol. 7, 1287–1294.

Kronfol, Z., House, J.D., 1988a. Immune function in mania. Biol. Psychiatry 24, 341–343.

Kronfol, Z., Hamdan-Allen, G., Black, D.W., 1988b. Fever and leukocytosis: physical manifestations of bipolar affective disorder? Prog. Neuropsychopharmacol. Biol. Psychiatry 12, 887–891.

Kubera, M., Kenis, G., Bosmans, E., Scharpe, S., Maes, M., 2000. Effects of serotonin and serotonergic agonists and antagonists on the production of interferon-γ and interleukin-10. Neuropsychopharmacology 23, 89–98.

Lazarus, J.H., McGregor, A.M., Ludgate, M., Darke, C., Creagh, F.M., Kingswood, C.J., 1986. Effect of lithium carbonate therapy on thyroid immune status in manic depressive patients: a prospective study. J. Affect. Disord. 11, 155–160.

Maes, M., Scharpe, S., Meltzer, H.Y. et al., 1994. Increased neopterin and interferon-gamma secretion and lower availability of L-tryptophan in major depression: further evidence for an immune response. Psychiatry Res. 54, 143–160.

Maes, M., Meltzer, H., Bosmans, E. et al., 1995a. Increased plasma concentrations of interleukin-6, soluble interleukin-6 receptor, soluble interleukin-2 receptor and transferrin receptor in major depression. J. Affect. Disord. 34, 301–309.

Maes, M., Bosmans, E., Calabrese, J., Smith, R., Meltzer, H.Y., 1995b. Interleukin-2 and interleukin-6 in schizophrenia and mania: effects of neuroleptics and mood stabilizers. J. Psychiatr. Res. 29, 141–152.

Maes, M., Delange, J., Ranjan, R. et al., 1997. Acute phase

proteins in schizophrenia, mania and major depression: modulation by psychotropic drugs. Psychiatry Res. 66, 1–11.

Maes, M., Song, C., Lin, A.H. et al., 1999. Negative immunoregulatory effects of antidepressants: inhibition of interferon-γ and stimulation of interleukin-10 secretion. Neuropsychopharmacology 20, 370–379.

Rapaport, M.H., 1994. Immune parameters in euthymic bipolar patients and normal volunteers. J. Affect. Disord. 32, 149–156.

Rapaport, M.H., Guylai, L., Whybrow, P., 1999. Immune parameter in rapid cycling bipolar patients before and after lithium treatment. J. Psychiatr. Res. 33, 335–340.

Rothermundt, M., Arolt, V., Weitzsch, C., Eckhoff, D., Kirchner, H., 1996. Production of cytokines in acute schizophrenic psychosis. Biol. Psychiatry 40, 1294–1297.

Seidel, A., Arolt, V., Hunstiger, M., Rink, L., Behnisch, A., Kirchner, H., 1995. Cytokine production and serum proteins in depression. Scand. J. Immunol. 41, 534–538.

Sluzewska, A., Sobieska, M., Rybakowski, J.K., 1997. Changes in acute-phase proteins during lithium potentiation of antidepressants in refractory depression. Neuropsychobiology 35, 123–127.

Tsai, S.Y., Chen, C.C., Yeh, E.K., 1997a. Alcohol problems and long-term psychosocial outcome in Chinese patients with bipolar disorder. J. Affect. Disord. 46, 143–150.

Tsai, S.Y., Lee, J.C., Chen, C.C., Chen, J., Chao, W.S., Chen, K.P., 1997b. Co-existing physical illnesses in help-seeking patients of bipolar disorder. Taiwanese J. Psychiatry 11, 249–261.

Tsai, S.Y., Chen, K.P., Yang, Y.Y. et al., 1999. Activation of indices of cell-mediated immunity in bipolar disorder. Biol. Psychiatry 45, 989–994.

Tsai, S.Y.M., Yang, Y.Y., Kuo, C.J., Chen, C.C., Leu, S.J., 2001. Effects of symptomatic severity on elevation of plasma soluble IL-2 receptor in bipolar mania. J. Affect. Disord. 64, 185–193.

Wilke, I., Arolt, V., Rothermundt, M., Weitzsch, C., Hornberg, M., Kirchmer, H., 1996. Investigations of cytokine production in whole blood cultures of paranoid and residual schizophrenic patients. Eur. Arch. Psychiatry Clin. Neurosci. 246, 279–284.

Young, R.C., Biggs, J.T., Ziegler, V.E., Meyer, D.A., 1978. A rating scale for mania: reliability, validity and sensitivity. Br. J. Psychiatry 153, 429–435.