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Reduced level of plasma antioxidant uric acid in schizophrenia

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Abstract

There is evidence of dysregulation of the antioxidant defense system in schizophrenia. The purpose of the present study was to examine whether uric acid, a potent antioxidant, is reduced in the plasma of patients with schizophrenia. To this end, a within-subject, repeated measures, on-off-on haloperidol treatment design was utilized. Male schizophrenic patients with either a haloperidol treatment ($n = 47$) or a drug-free condition ($n = 35$) had significantly lower levels of plasma uric acid than the age- and sex-matched normal control subjects ($n = 34$). Following haloperidol withdrawal, plasma uric acid levels were further reduced in schizophrenic patients ($P = 0.018$; paired t-test, $n = 35$). However, no relationship was found between uric acid levels and the length of the drug-free period (< 5 or > 5 weeks) or days drug free. In addition, the plasma levels of uric acid in patient groups were significantly and inversely correlated with psychosis. There was a trend for lower uric acid levels in relapsed patients relative to clinically stable patients. Smoking, which can modify plasma antioxidant capacity, was not found to have prominent effects on uric acid levels. The present finding of a significant decrease of a selective antioxidant provides additional support to the hypothesis that oxidative stress in schizophrenia may be due to a defect in the antioxidant defense system.

Keywords: Haloperidol; Antioxidant defense system; Plasma uric acid; Psychosis

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1. Introduction

Uric acid is mainly synthesized from adenine- and guanine-based purines. Because uricase (urate oxidase) is lacking in man, uric acid appears to be an end-product of the purine pathway. It is thus traditionally considered as a metabolically inert and waste compound without any physiological significance. However, uric acid can be oxidized following a non-enzymatic degradation and has thus proved to be a selective antioxidant, capable especially of reaction with hydroxyl radicals and hypochlorous acid (Becker, 1993). Uric acid may be found in all tissue compartments with the exception of the lipid phase. In plasma, uric acid, albumin and ascorbic acid, account for more than 85% of total antioxidant capacity (Wayner et al., 1987; Miller et al., 1993). Thus, measuring levels of specific antioxidant molecules, such as plasma uric acid can yield valuable information and low levels of such antioxidants may provide suggestive evidence of oxidative stress.

A role for free radicals in the pathophysiology of schizophrenia has been proposed by several investigators (Cadet and Lohr, 1987; Lohr et al., 1990; Reddy and Yao, 1996). Free radical-mediated pathological processes may account for previous findings of RBC membrane defects in schizophrenia (Yao and van Kammen, 1994, 1996; Yao et al., 1994,b, 1996a,b) including findings of membrane pathology in brains of schizophrenic patients (Horrobin et al., 1991; Pettegrew et al., 1993). Membrane dysfunction can be secondary to free radical-mediated pathology (Reddy and Yao, 1996) and may contribute to specific aspects of schizophrenic symptomatology and complications of its treatment. Specifically, free radical-mediated abnormalities may contribute to the development of a number of clinically significant consequences, including prominent negative symptoms, tardive dyskinesia, neurological 'soft' signs, and Parkinsonian symptoms. Our previous results showing altered membrane dynamics and antioxidant enzyme activities in schizophrenia as well as findings from other investigators, are consistent with the notion of free radical-mediated neurotoxicity in schizophrenia.

The neurotoxicity can be a consequence of increased free radical production and/or an inadequate antioxidant defense system (AODS). The AODS is a complex system that is dynamically linked to free radical production, i.e. increases in free radical burden result in up-regulation of enzymatic and non-enzymatic scavenging activity. Although we and others have previously shown that the critical antioxidant enzyme activities (Abdalla et al., 1986; Reddy et al., 1991; Wang, 1992; Vaiva et al., 1994) and other indices of lipid peroxidation (Kovaleva et al., 1989; Prilipko, 1992; Phillips et al., 1993; Mahadik et al., 1995) are altered in plasma, red blood cells, and cerebrospinal fluid of schizophrenic patients, it is not known whether antioxidant capacity is similarly altered. If so, it would provide further evidence that there exists a substantial risk of free-radical mediated neuronal damage.

The purposes of the present study are: (1) to assess whether plasma levels of the antioxidant uric acid are altered in schizophrenic patients, using a within-subject, repeated measures, on-off-on haloperidol treatment design; (2) if so, to further test whether altered plasma uric acid is associated with specific clinical characteristics in patients; and (3) to assess antipsychotic treatment effects, if any, on the plasma antioxidant uric acid.

2. Methods

2.1. Patients

Patients ($n = 47$) were recruited from the male veteran outpatient population of the Highland Drive VA Pittsburgh Healthcare System. Clinically stable patients who met both DSM-IIIR criteria and Research Diagnostic Criteria (RDC) for schizophrenia were hospitalized after they had signed informed consent forms. The diagnosis of schizophrenia was made using the Structured Clinical Interview for DSM-IIIR (Spitzer et al., 1989) and a DSM-IIIR checklist. Table 1 presents means, standard deviations, and ranges of demographic variables in the schizophrenic patients. All patients adhered to a low-monoamine, alcohol-free, and caffeine-restricted diet. A stan-

Table 1
Demographic variables from healthy volunteers and schizophrenic patients

Demographic variables	Normal volunteers			Schizophrenic patients		
	Mean	S.D.	Range	Mean	S.D.	Range
Age (years)	35	10	18–55	39	7	25–49
Body mass index[a]	26	4	19–36	29	5	20–42
Age of onset (years)	–	–	–	25	5	16–33
Illness duration (years)	–	–	–	15	8	1–27
Haloperidol dose (mg/day)	–	–	–	10	4	4–20
Days drug-free	–	–	–	40	19	17–73

[a] Body mass index: weight (kg)/height (m^2)

dard diet consisting of 100 g protein, 295 g of carbohydrates and 95 g of fat (ratio of unsaturated fatty acids to saturated fatty acids was 2:1) providing a daily energy of 2400 kcal was given to all patients. Current smoking status was ascertained by interview.

Each patient was treated with antipsychotic drugs for ≥ 3 months before the blood draw. If patients were not already on haloperidol, their medication was converted to an equipotent dose (5–20 mg/day) of haloperidol for at least 3 months before the first blood drawing. To assure medication compliance, plasma haloperidol levels were measured by high-performance liquid chromatography (HPLC). No other medications were used during the last 2 weeks of haloperidol treatment.

After patients were stabilized on oral haloperidol, it was then replaced overnight by a placebo in identical looking capsules for a period of up to 3 months. The second and third blood samples were collected from patients who had been free of all psychotropic medications for a period of 3–10 weeks. Patients meeting relapse criteria were restarted on haloperidol treatment. A final blood sample was collected from those patients who had returned to haloperidol treatment for 1 month. Of these 47 patients, 35 patients had completed both haloperidol treatment and drug-free phases. All blood samples were taken in the morning after overnight fasting.

Smoking status of each patient was determined using a questionnaire at the time of each blood draw. Twenty-six of 35 patients are smokers who have > 20 cigarettes per day except for four patients who have < 20 cigarettes per day. Positive smoking was further verified by the presence of plasma cotinine levels > 80 ng/ml. In addition, smoking was prohibited after 23:00 h, before the next day's blood draw.

Relapse criteria consisted of a mean increase of at least three points on the Bunney–Hamburg psychosis score (Bunney and Hamburg, 1963) for three days compared with the mean of the daily psychosis ratings of the last week on haloperidol treatment (van Kammen et al., 1989).

2.2. Behavioral ratings

Symptom severity was measured with the Bunney–Hamburg (BH) global rating scale (Bunney and Hamburg, 1963). The BH scale contains, on a 15-point scale, four global items covering psychosis, depression, mania and anxiety. We have been utilizing this scale for the daily or weekly monitoring of the global behavioral status of schizophrenic in- and out-patients. Trained nurses assessed psychosis severity daily, blind to treatment condition. A 3-day mean of BH psychosis ratings (BHPR) was used for analyses. Therapists, also without knowledge of treatment condition, rated the patients weekly with the BH psychosis ratings, the Brief Psychiatric Rating Scale (BPRS, Overall and Gorham, 1962), and the Scale for the Assessment of Negative Symptoms (SANS, Andreasen, 1982). BH psychosis ratings, BPRS, and SANS scores were substantially higher in relapsed than in clinically stable patients during the drug-free condition (Table 2). The rating scale data

Table 2
Behavior rating scale scores of same patients on and off haloperidol

Schizophrenic patients	*n*	Behavior rating scales		
		BHPR	BPRS (total)	SANS (5 items)
HD-treated	35	4.7 ± 1.6 (2–8)	45 ± 12 (29–75)	10 ± 4 (3–17)
Drug-free	35	7.0 ± 2.9 (2–13)	50 ± 15 (26–79)	10 ± 5 (1–17)
Relapsed	16	9.4 ± 1.9 (7–13)	61 ± 13 (36–79)	13 ± 4 (6–17)
Clinically stable	19	5.0 ± 2.0 (2–10)	42 ± 9 (26–64)	8 ± 4 (1–16)

Abbreviations: HD, haloperidol; BHPR, Bunney-Hamburg Psychosis Rating; BPRS, Brief Psychiatric Rating Scale; SANS, Scale for the Assessment of Negative Symptoms.

and weight measurements were taken from the week of the blood draw.

2.3. Normal volunteers

Thirty-four age-matched healthy male volunteers (mean = 35 years, S.D. = 10) were also recruited to participate in the study. They were screened for DSM-III-R Axis I and II diagnoses using the Lifetime version of the Schedule for Affective Disorders and Schizophrenia and the Minnesota Multiphasic Personality Inventory. Prestudy evaluations included a complete medical history, physical examination, and a urine and blood drug screen for alcohol and drug use. All control subjects were on a low-monoamine, alcohol-free, and caffeine-restricted diet for at least 1 week preceding the blood draw. Smoking status of each subject was taken by questionnaire at the time of each blood draw. Only four subjects are smokers with plasma cotinine levels > 80 ng/ml.

2.4. Biochemical assays

2.4.1. Plasma uric acid assay

A kit manufactured by Roche Diagnostics Systems (Cat. No. 47273) enables uric acid concentration in plasma to be measured at 550 nm by the formation of a chromogen. In the presence of O_2 and the enzyme uricase, uric acid is converted to H_2O_2 and allantoin. *N*-ethyl-*N*-(2-hydroxy-3-sulfpropyl)-*m*-toluidine (TOOS) is reacted with 4-aminoantipyrine in the presence of H_2O_2 from the prior reaction and peroxidase to form the red chromogen. The absorbance at 550 nm is directly proportional to the concentration of uric acid in the sample. A Cobas Fara centrifugal analyzer (Roche Diagnostics Systems, Branchburg, NJ) is used for the assay. Exactly 9 μl of platelet-poor-plasma or a standard of known concentration is first diluted with 30 μl of diluent. Then 120 μl of reagent containing 4-aminoantipyrine, TOOS, potassium ferrocyanide, uricase, and peroxidase is added to the diluted sample which starts the reaction. The reaction is followed for 200 s after-which the change in absorbance of the sample, standard, and a reagent blank are used to determine uric acid concentration expressed as mg/dl of platelet-poor-plasma.

2.4.2. Plasma total antioxidant status

A kit manufactured by Randox Laboratories (Cat. No. NX 2332) enables the total antioxidant status to be measured in human plasma or serum. The Randox kit utilizes ABTS® (2,2′-azino-di-[3-ethyl-benzthiazoline sulphonate]) which is converted to its cation form when incubated in the presence of a peroxidase (metamyoglobin) and H_2O_2. This cation has a fairly stable blue-green color which can be measured at 600 nm. Antioxidants present in the sample suppress this color formation in the proportion of their concentration. A Cobas Fara centrifugal analyzer (Roche Diagnostics Systems, Branchburg, NJ) is used for the assay. Exactly 4 μl of platelet-poor plasma (PPP) or a standard of known concentration is incubated with 200 μl of a reagent containing metamyoglobin and ABTS® at 37°C for 15 s during which two measurements are taken. Next, 40 μl of H_2O_2 is added and thoroughly mixed starting the reaction. The reaction is followed for

exactly 3 min. The reaction rate of the sample is compared to that of the standard and a blank to determine the concentration of total antioxidants present in the sample, and is expressed as μmol/l or mmol/l of PPP.

2.4.3. Plasma cotinine level

Cotinine is the major metabolite of nicotine. The half-life of cotinine in blood is far longer than that of nicotine (Langone et al., 1973, 1975). Moreover, cotinine levels remain fairly constant in individuals with regular tobacco consumption. Thus, plasma cotinine levels provide us with a better marker than nicotine for smoking status (Langone et al., 1973; Hall et al., 1984; Jarvis et al., 1987). The DRI (Diagnostic Reagents, Inc., Sunnyvale, CA) enzyme immunoassay kit is applied to measure plasma cotinine level. The DRI immunoassay is based on the competition between a cotinine-labeled enzyme glucose-6-phosphate dehydrogenase (G-6-PDH) and the free cotinine in the sample for a fixed number of cotinine-specific antibody binding sites. In the absence of cotinine, the cotinine-labeled G-6-PDH is bound to the antibody and the enzyme activity is inhibited. The G-6-PDH activity is measured spectrophotometrically at 340 nm by converting NAD to NADH. The plasma sample is first centrifuged to remove any interfering debris. In a typical assay, 20 μl of sample is used. The standard curve is obtained from the cotinine calibrators provided by the DRI kit. A normal plasma sample spiked with a known amount of cotinine standard is used as a control for each assay. A Cobas Fara centrifugal analyzer (Roche Diagnostics Systems, Branchburg, NJ) is used to measure the enzymatic reaction rate at 37°C.

2.5. Statistical analyses

Group means ($\pm$S.D.) of healthy volunteers and schizophrenic groups (haloperidol-treated or drug-free) were compared by two-tailed unpaired *t*-tests. Effects of antipsychotic drug withdrawal, length of drug-free period, and duration of haloperidol treatment were evaluated in the same individuals by two-tailed paired *t*-tests. Testing for normality was accomplished by normality plots and Wilks–Shapiro tests. For normally distributed data, means and variances were compared by the use of *t*-tests and *F*-tests, respectively. For non-normally distributed data, the medians and dispersions were compared by the use of the Signed Rank test (paired data), the Rank Sum test (unpaired data, equal dispersions), and the Ansari–Bradley Dispersion test. Pearson correlations and Spearman Rank correlations were, where appropriate, used to determine whether plasma uric acid levels were correlated with demographic features, behavioral ratings, and smoking status.

3. Results

Schizophrenic patients had significantly lower plasma uric acid during both haloperidol-treated (4.58 ± 0.94 mg/dl, $P = 0.024$, $n = 47$) and drug-free conditions (4.29 ± 0.95 mg/dl, $P = 0.001$, $n = 35$) compared to healthy volunteers (5.07 ± 0.96 mg/dl, $n = 34$).

To test whether plasma uric acid is affected by antipsychotic drug withdrawal, the length of the drug-free period, antipsychotic drug treatment, we first compared schizophrenic patients stabilized with haloperidol to the same individuals after haloperidol withdrawal. We also compared patients that were drug-free for less than 5 weeks to the same individuals after they had been drug-free for longer than 5 weeks. Finally, we compared drug-free patients to the same individuals following reintroduction to haloperidol treatment for 1 month. The mean plasma uric acid level of haloperidol-treated patients (4.62 ± 0.99 mg/dl, $n = 35$) was significantly higher ($P = 0.018$) than that of the same individuals following haloperidol withdrawal (4.29 ± 0.95 mg/dl, $n = 35$). No significant differences were found among patients who had undergone the above other clinical phases (Table 3).

To test whether the plasma uric acid level is influenced by the relapse status, drug-free schizophrenic patients are divided into relapsed and clinically stable groups according to relapse criteria (see methods). The mean levels of plasma uric

Table 3
Effect of antipsychotic drug withdrawal, length of drug-free period, and antipsychotic drug treatment on plasma uric acid levels

Schizophrenic patients	*n*	Uric acid (mg/dl)	*P* (paired *t*-test)[a]
1. Drug withdrawal effect			
On-haloperidol	35	4.62 ± 0.99	**0.018**[b]
Off-haloperidol	35	4.29 ± 0.95	
2. Length of drug-free period			
< 5 weeks	14	4.44 ± 1.04	0.439
> 5 weeks	14	4.60 ± 0.97	
3. Drug treatment effect			
Drug-free	12	4.46 ± 1.01	0.696
Return to haloperidol	12	4.33 ± 1.29	
4. Effect of clinical state			
Relapsed	16	4.03 ± 1.05	0.134
Clinically stable	19	4.52 ± 0.80	

Note: Effect of haloperidol (HD) withdrawal was established by comparing plasma uric acid levels from patients stabilized with HD to those of the same individuals after HD withdrawal. Effect of HD treatment was assessed by comparing plasma uric acid levels from drug-free patients to those of the same individuals returning to the HD treatment after relapse.
[a]With the exception of the comparison between relapsed and clinically stable groups (unpaired).
[b]Statistically significant.

acid are lower in relapsed than in clinically stable groups (Table 3). The difference, however, is not statistically significant.

To test whether plasma individual antioxidant is related to the severity of psychopathology, we compared plasma uric acid of schizophrenic patients with the 3-day mean B-H psychosis ratings, the BPRS total score and the five SANS items. Plasma uric acid was inversely and significantly correlated with the BH psychosis ratings of both haloperidol-treated ($P = 0.044$) and drug-free ($P = 0.015$) schizophrenic patient groups, but not with total BPRS or SANS scores (Table 4).

In addition, relations were examined between plasma uric acid levels and clinically relevant demographic features including body mass index (BMI), age, age of onset of illness, duration of illness as well as length of drug-free period (Table 5). In healthy volunteers, no significant correlations were found between plasma uric acid levels and BMI (or age). In schizophrenic patient groups, however, plasma uric acid levels are significantly correlated with BMI in both haloperidol-treated and drug-free conditions. Age and age at onset of illness were both inversely correlated with plasma uric acid, but only in haloperidol-treated patients. The mean age and S.D. of the schizophrenic patient group (39 ± 7 years) was not significantly different from that of healthy volunteers (35 ± 10 years). Neither duration of illness nor days drug-free were related to plasma uric acid in schizophrenic patients.

Table 4
Correlations between plasma uric acid levels and symptom severity in patients with schizophrenia

Patients	*n*	*r* (correlation coefficient)		
		BHPR	BPRS	SANS
On-haloperidol	35	**−0.3428**[a]	−0.1610	−0.1021
Off-haloperidol	35	**−0.4072**	−0.2149	−0.0194

[a]*r* values in boldfaced type are statistically significant, $P < 0.05$.
Abbreviations: BHPR, 3-day mean Bunney-Hamburg Psychosis Rating; BPRS, Brief Psychiatric Rating Scale (total); SANS, Scale for the Assessment of Negative Symptoms (five items).

To test whether plasma uric acid level is affected by cigarette smoking, plasma uric acid levels of smokers were compared with those of non-smokers in both healthy volunteers and schizophrenic patients (Table 6). No significant differences were found between smokers and non-smokers in patients or controls. In either non-smoking or smoking groups, the mean plasma uric acid levels of schizophrenic patients were lower than in healthy volunteers, although the difference was not statistically significant. Among smokers, however, plasma uric acid levels of drug-free patients (4.28 ± 0.94 mg/dl, $n = 26$) were significantly ($P = 0.046$) lower than levels in these same individuals before haloperidol withdrawal (4.57 ± 0.90 mg/dl, $n = 26$). Such a difference in drug withdrawal was not demonstrated in a lesser number ($n = 9$) of non-smoking patients. Furthermore, there were no significant correlations between plasma uric acid and plasma cotinine levels in either healthy volunteer ($P =$

Table 5
Correlations between plasma uric acid levels and demographic features in healthy volunteers and patients with schizophrenia

Subjects	*n*	*r* (correlation coefficient)				
		BMI[a]	Age	Age of onset	Duration of illness	Days drug free
Healthy volunteers	34	0.0621	0.0378	–	–	–
Schizophrenics						
On-haloperidol	35	**0.4591**[b]	**−0.3752**	**−0.4825**	0.0136	–
Off-haloperidol	35	**0.5330**	−0.2681	−0.1730	−0.1371	0.2344

[a] Body mass index (BMI): weight (kg)/height (m^2)
[b] *r* values in boldfaced type are statistically significant, $P < 0.05$.

0.154) or schizophrenic patient (haloperidol-treated, $P = 0.077$; drug-free, $P = 0.117$) groups.

We also examined whether plasma uric acid was affected by storage time. Samples from healthy volunteers that were stored at −70°C for various time periods up to 7 years and had never been thawed previously, showed no significant trending in the levels of plasma uric acid.

To test whether decreases in individual antioxidant levels reflect similar changes of total antioxidant status in plasma of schizophrenic patients, we correlated plasma uric acid levels with plasma total antioxidant status (data obtained from Yao et al., in press). A significant and positive correlation ($P < 0.005$) was demonstrated in schizophrenic patients but not in healthy volunteers (Fig. 1).

4. Discussion

The is the first study to our knowledge that has examined levels of the selective antioxidant uric acid in schizophrenia. The present findings show that plasma uric acid levels are significantly lower in schizophrenic patients than in normal control subjects, providing further support to the hypothesis that there exist in schizophrenia defects of the antioxidant system (Reddy and Yao, 1996). We also show that haloperidol treatment has significant effects on uric acid levels (Table 3),

Table 6
Effect of cigarette smoking on plasma uric acid levels

Subjects	*n*	Plasma uric acid (mg/dl)	*P* (two tailed *t*-test)		
			Unpaired		Paired
			(NS vs. S)	(SP vs. HV)	(drug free vs. HT)
Healthy volunteers (HV)					
Non-smokers (NS)	29	5.12 ± 0.90	0.798		
Smokers (S)	4	4.90 ± 1.51			
Schizophrenic patients (SP):					
Haloperidol-treated (HT)					
Non-smokers	9	4.74 ± 1.26	0.714	0.428	
Smokers	26	4.57 ± 0.90			
Drug-free					
Non-smokers	9	4.34 ± 1.03	0.865	0.066	0.249
Smokers	26	4.28 ± 0.94			**0.046**[a]

[a] Statistically significant.

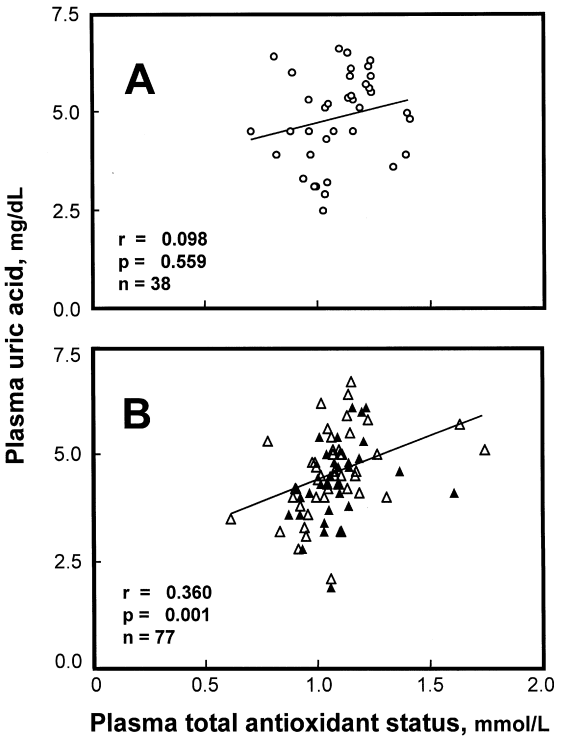

Fig. 1. Correlations between plasma uric acid levels and plasma total antioxidant status in normal volunteers (A) and schizophrenic patients (B). Note: △ = haloperidol-treated; ▲ = drug-free.

although these effects do not account for the absolute decreases in uric acid levels relative to normal control subjects. No patients were on any uricosuric agents that potentially could decrease uric acid levels. If anything, they were at greater risk for increased uric acid levels because of increased body mass index (BMI), which was correlated with plasma uric acid levels in patients (Table 5). Diet, caloric intake, and alcohol are the major factors that can affect both the antioxidant system and the production of free radicals and reactive oxygen-containing species (Papas, 1996). Thus, plasma levels of uric acid may be affected by diet. In the present study, all our patients were hospitalized and maintained on a control balanced diet without alcohol consumption. Thus, it is unlikely that *decreased* uric acid in plasma of schizophrenic patients resulted from a dietary deficiency or alcohol consumption as compared to their healthy volunteer counterparts.

In normal control subjects, plasma uric acid levels were not correlated with age. By contrast, there was an inverse correlation with age in schizophrenic patients, although there was no significant difference in mean age between the two groups. Furthermore, age at onset of illness was inversely correlated with plasma uric acid level. These two relationships were observed only during the haloperidol treatment condition. Any discussion of the implications of these relationships can only be regarded as highly speculative at this time.

4.1. Neuroleptic effects on plasma uric acid levels

The controlled discontinuation of haloperidol in patients allowed the examination of neuroleptic effects on plasma uric acid. The further significant *decrease* of uric acid levels after haloperidol was discontinued suggests: (1) that baseline uric acid levels in untreated schizophrenic patients may be even lower than those observed in this study; and (2) that haloperidol treatment is associated with increased uric acid levels. Haloperidol treatment in animals has been related to decreased lipid peroxidation (Murthy et al., 1989). Interestingly, no effects of length of drug-free period, days drug free, or return to treatment were observed. This may be due to the small number of subjects in these comparisons. Although a clearer picture of treatment effects can be best provided by prospective treatment studies of neuroleptic-naïve patients, the decreased uric acid levels observed in this study are not due to neuroleptic effects, because haloperidol treatment was associated with increased uric acid levels within schizophrenic patients, but still significantly lower than normal control subjects.

4.2. Correlation between plasma uric acid levels and psychosis

The finding of a significant inverse correlation of plasma uric acid to the psychosis scores on the Bunney–Hamburg global rating scale suggests that the alterations in individual antioxidant levels may have pathophysiological significance in schizophrenia. It is unclear why plasma uric acid

levels were not correlated with BPRS ratings. The BPRS assesses symptoms other than psychosis,, such as depression and negative symptom equivalents. Thus, the relations between psychosis and uric acid may be quite specific, reflected in the correlation with BH psychosis ratings and not simply reflect overall psychopathology severity.

These findings add to previous findings suggestive of oxidative stress in schizophrenia. However, it remains to be determined which specific free-radical mediated pathophysiological processes have clinical implications for schizophrenia. A variety of antioxidant defense system alterations, all favoring oxidative stress, have been associated with tardive dyskinesia, negative symptoms, neurological signs, poor premorbid function, and CT scan abnormalities (reviewed in Reddy and Yao, 1996). It is not definitively known whether oxidative stress is of etiological relevance or is secondary to the disease state. Thus, it is possible that the decreased uric acid levels reflect state-related changes associated with worsening of psychosis.

Patients who relapsed after drug discontinuation had lower uric acid levels relative to patients who remained clinically stable, although this difference did not reach statistical significance. It is possible that lower uric acid levels, which lead to lower antioxidant capacity, increase the risk for worsening psychopathology. Although lipid peroxide levels were not determined in the study sample, it is known that lipid peroxide levels covary with psychosis severity and treatment (Prilipko, 1992). Increased neutrophil superoxide production has been found in schizophrenic patients (Sirota et al., 1997), indicating *peripheral* oxidative stress. Plasma uric acid may be utilized as a free radical scavenger at increased rates in schizophrenia, accounting for the observed decreased levels (Table 3). However, prospective studies in neuroleptic-naïve patients are needed to further clarify the relations between plasma uric acid levels, psychopathology and treatment effects. We have initiated these studies.

4.3. Plasma total antioxidant capacity and individual antioxidants

A major contribution to the total antioxidant capacity comes from antioxidant molecules in plasma. Plasma, however, is not a simple chemical system for combating oxidative stress. In addition to the preventive antioxidants (reducing the rate of new chain reaction) of the iron-scavenging proteins, e.g. transferrin and ceruloplasmin (Stocks et al., 1974), plasma also contains chain-breaking antioxidants that can trap radicals directly and thereby shorten the chain length (Wayner et al., 1987). The relative contribution of each antioxidant in vivo depends not only on its efficacy but also its concentration in biological fluid. It is known that uric acid alone with albumin (sulfhydryl group), ascorbic acid and α-tocopherol are the major source of the chain-breaking antioxidant activity in human plasma (Wayner et al., 1987; Miller et al., 1993). Although individual antioxidants play a specific role in the antioxidative defense system, these antioxidants may act cooperatively in vivo to provide synergistic protection to the organs against oxidative damage. Therefore, it is more meaningful to assess antioxidant status by measuring both the individual levels and the overall antioxidant capacity. The present data demonstrating a decrease in plasma uric acid level in schizophrenic patients support our previous findings of a reduced plasma total antioxidant status (Yao et al., 1998). Furthermore, changes in individual antioxidant levels, i.e. uric acid, are positively and significantly correlated with the status of total antioxidant activities (Fig. 1).

In addition to its chain-breaking antioxidant activity, uric acid may also exhibit preventive antioxidant activity through metal ion chelation (Davies et al., 1986). Lam et al. (1984) demonstrated that uric acid is able to inhibit oxidation of ascorbic acid through cupric ion. Such a protection to stabilize ascorbic acid can be achieved without apparent consumption of the uric acid (Sevanian et al., 1991).

4.4. Effect of cigarette smoking on antioxidant capacity

Cigarette smoking is associated with lower levels of plasma antioxidants of dietary origin (Chow, 1992; Pryor and Stone, 1992; Stegmayr et al., 1993). One of the major compounds in the gas

phase of tobacco smoke is nitric oxide. It has been suggested that nitric oxide reacts with smoke olefins to form carbon-centered radicals (Pryor and Stone, 1992). On the other hand, the tar phase consists of a semiquinone radical that promotes hydrogen peroxide formation. Moreover, tobacco smoke may increase free radical formation by activating neutrophils.

In the present study, no differences in plasma uric acid levels were found between smokers and non-smokers in the normal control group, although a majority of subjects in this group were non-smokers. Similarly, no differences in uric acid levels were found between treated schizophrenic patients who smoked and those who did not. The most marked difference in uric acid levels was observed between schizophrenic smokers while on haloperidol and while drug free, the latter condition being associated with significantly lower uric acid levels. It is possible that smoking cigarettes magnifies the difference in uric acid levels associated with haloperidol discontinuation. Thus, smoking-associated decreases of plasma antioxidants of dietary origin (Chow, 1992; Pryor and Stone, 1992; Stegmayr et al., 1993) may interact with schizophrenia-related decreases in uric acid to result in greater decrements of uric acid. However, it does not appear that cigarette smoking in this group of patients independently explains the significant decreases in plasma uric acid levels in schizophrenic patients on and off haloperidol treatment and that smoking effects are quite modest.

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