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Serum uric acid and risk of multiple sclerosis

J. Massa¹, E. O'Reilly², K.L. Munger³, G.N. DeLorenze⁴, and A. Ascherio⁵

¹ Department of Epidemiology, Harvard School of Public Health, Boston, MA, USA

^{2,3} Department of Nutrition, Harvard School of Public Health, Boston, MA, USA

⁴ Kaiser Permanente Division of Research, Oakland, CA, USA

⁵ Departments of Epidemiology and Nutrition, Harvard School of Public Health; Channing Laboratory, Department of Medicine, Brigham and Women's Hospital and Harvard Medical School, Boston, MA, USA

Individuals with multiple sclerosis (MS) have lower serum uric acid (UA) when compared to controls.^{1–4} Because of evidence implicating oxidative stress in MS pathogenesis,⁵ it has been postulated that high levels of urate, a potent antioxidant,⁶ could reduce risk or favorably influence MS progression.⁵ This hypothesis is supported by observations that experimental allergic encephalomyelitis, an animal model of MS, can be prevented or attenuated by increasing serum UA.^{7,8} However, it is unclear whether the decreased levels of UA in MS cases precede the disease onset or are a consequence of the disease itself. We therefore conducted a prospective study to determine whether serum urate levels predict MS risk.

Methods

This study has been approved by the Institutional Review Boards of the Harvard School of Public Health, Boston, MA, and the Kaiser Foundation Research Institute, Oakland, CA.

Study population

The study population comprised a subset of participants in the Nurses Health Study (NHS) and Nurses Health Study II (NHS II) cohorts, and of members of the Kaiser Permanente Northern California (KPNC) health plan.

NHS and NHS II—The NHS was initiated in 1976, when 121,700 female registered nurses in the United States completed a mailed questionnaire about lifestyle factors and medical history. The NHS II began in 1989, when 116,671 female registered nurses completed a similar questionnaire. Every two years participants of both studies are mailed follow-up questionnaires in order to update information on possible risk factors for disease as well as identify newly diagnosed illnesses.

Participants from both cohorts were asked to provide blood samples to be analyzed for biomarkers of chronic disease. Between 1989 and 1990, 32,826 participants from the NHS provided blood samples; and blood was collected between 1996 and 1999 from 29,613 women in NHSII. Each participant used a collection kit and sent the blood sample by overnight delivery to our lab. 97% of the blood samples were delivered within 26 hours of being drawn.⁹ Blood samples were centrifuged and the components were aliquotted into cryotubes and stored in liquid nitrogen freezers until they were sent to the laboratory for analysis.

Case and control blood samples were analyzed at the Clinical & Epidemiologic Research Laboratory at Boston Children's Hospital. Lab technicians were blinded to the case status of the samples. The concentration of uric acid was determined using a colorimetric enzyme assay

on the Hitachi 917 analyzer (Roche Diagnostics - Indianapolis, IN). The coefficient of variation, determined from blind quality control samples included with the study samples, was 17.2%.¹⁰

Cases of MS were identified through self-report on a follow-up questionnaire and confirmed through medical records obtained from the participants' neurologists. A detailed description of the process by which MS cases are identified has been previously described.⁹ Overall, 31 MS cases were included. Each of the 31 MS cases was matched with one control on year of birth and cohort (NHS or NHSII). Five control serum samples had missing UA values because the quantity of serum was insufficient to do the assay, so that analyses included 31 cases and 26 controls. In 18 of the women with MS, blood was collected before the onset of the first symptoms of MS (median: 1.9 years before onset).⁹ In the remaining 13, blood was collected after the onset of neurological symptoms, but before a diagnosis of MS was established.

KPNC—The KPNC cases and matched controls were selected from the cohort of men and women who participated in a multiphasic examination between 1965 and 1974. During that period new members to the health plan were asked to provide a blood sample and answer a questionnaire on health history and medical risk behaviors. 100,000 KPNC members provided blood samples which were measured for serum UA. Concentrations of serum UA were measured by an AutoAnalyzer from July 1964 to March 1969, by an AutoChemist from April 1969 to January 1972, and by an SMA 12/60 analyzer thereafter.¹¹ Because UA was measured at baseline soon after the blood samples were collected, there is no concern related to the storage of these blood samples.

KPNC maintained medical records for all members who provided serum samples and electronically stored all summaries of inpatient and outpatient visits for these members. Between the years of 1995 and 1999 the electronic databases which housed these medical records were searched for MS diagnoses and 70 cases were identified. Each case was matched to two controls from the serum database on age at time of blood collection (± 1 year), sex, and date of blood collection. The controls were active members of KPNC at the time the matched cases occurred and had no indication in their records of being diagnosed with MS. More detailed information on the establishment of the electronic databases and the method by which MS cases were identified and matched to controls has been described elsewhere.¹²

There were missing values of UA for 9 of the 70 identified cases and 10 of the 140 matched controls, leaving 61 cases and 130 controls available for analyses. Serum was collected before the date of MS onset in 42 of the 61 MS cases. The cases included 37 females and 5 males. The median number of years between blood collection and MS onset for the 42 cases was 14.5 years with a range of 2 months to 32 years; the median number of years is 14 years for female cases and 15 years for male cases. The years of blood collection for these matched cases and controls span 1964 through 1970.

Statistical analyses

The primary analyses included only the 60 MS cases (18 from NHS/NHS2 and 42 from KPNC) with blood collected before the onset of symptoms. Odds ratios were used to approximate relative risks (RR) estimated by unconditional logistic regression including all controls (matched and unmatched) and adjusting for the matching factors in order to increase power. Analyses were performed within cohort.

In the NHS and NHSII models, the matching factor variables that were controlled for include age (5 year intervals) and cohort (NHS or NHSII); in the KPNC models the variables included age (5 year intervals), gender, and year of serum collection. The multivariable models for both cohorts were additionally adjusted for body mass index (BMI) as a continuous variable, blood

pressure categories (normal, pre-hypertensive, and hypertensive), and smoking status (never, past, and current). A missing indicator variable was used for 4 controls with missing BMI in this model. The multivariable models for KPNC were also adjusted for race using skin color assessed by physician during multiphase check-up (white, black, other). A detailed explanation of using skin color as a proxy for race in this cohort has been previously published.¹³ The relationship between UA and the baseline characteristics were determined by ANOVA. Analyses were also performed adjusting for serum titers of immunoglobulin G antibodies against the Epstein-Barr virus nuclear antigen (EBNA), previously found to be a strong risk factor for MS in these and other cohorts.^{9,12–15}

Secondary analyses were conducted using blood from MS cases which was collected after onset of the disease. There were 13 cases in NHS and NHSII that had blood collected after the first sign or symptom of MS but before a medical diagnosis. In the KPNC cohort, there were 19 cases that provided blood after the date of first symptom or date of possible/probable diagnosis of MS.¹² Unconditional logistic regression analyses were performed using only these prevalent MS cases in each cohort and then combined with the cases whose blood was collected prior to the onset of MS.

Lastly, to better elucidate the relation between serum UA concentration and the onset of MS, we plotted these concentrations according to the time interval between serum collection and date of MS onset using the KPNC cohort. The KPNC cohort was used because it has a longer period of time between serum collection and MS onset (upper range of 32 years) as compared to NHS/NHS II (upper range of 6.5 years). To create this plot, we first removed extraneous variations in serum UA by regressing in a linear model the UA concentrations on age, year of serum collection, BMI, sex, blood pressure, race, smoking, and case status (MS case or control), and then used the residuals of this regression (i.e. the difference between the observed and expected UA concentration) as an adjusted measure of UA concentration. To test the significance of the trend between serum UA levels and years between serum collection and MS onset, we considered the median number of years from each time interval as a continuous variable in a regression model. All statistical tests were two sided and p-values less than 0.05 were considered statistically significant. The statistical software SAS (SAS Institute Inc, Cary, NC) and STATA (StataCorp, College Station, TX) were used for all analyses.

Results

Table 1 presents the baseline characteristics of the study population by tertile of serum UA in the control groups. As expected, higher serum UA concentrations were associated with increasing BMI, gender, and hypertension.¹⁶

In the NHS/NHSII population, risk of MS decreased with increasing UA levels; a 1 mg/dL increase in UA was associated with an RR of 0.55 (95% CI 0.28, 1.10, p-value 0.09). An inverse association of similar magnitude was found when adjusting for BMI, smoking status, and blood pressure categories (RR 0.52, 95% CI 0.22, 1.20, p-value 0.13). In contrast, there was no evidence of a decline in MS risk with increasing serum UA in the KPNC population (RR for 1 mg/dL increase in urate: 1.10, 95% CI 0.78, 1.54, p-value 0.60); results were similar after adjusting for BMI, blood pressure categories, smoking status, and race (RR 1.36, 95% CI 0.87, 2.14, p-value 0.18). These associations remained non-significant after further adjustment for titers of IgG antibodies to EBNA. No significant association was found between tertile of serum UA and MS risk in either population (Table 2).

The plot of the adjusted mean residuals of serum UA from KPNC MS cases shows increasing serum UA levels as years between serum collection and MS onset increases (p for trend 0.10)

(Figure 1). Fluctuations between the mean serum UA concentrations of MS cases for the time intervals <1 years and 1–5 years may be the result of small case numbers.

Discussion

The results of this study suggest that serum UA is not a strong predictor of MS risk. In analyses including only MS cases with blood collected shortly before onset of MS, there was a trend toward a lower risk of MS among individuals with higher serum UA, but the association was not significant.

An important limitation of our study is that the KPNC is a retrospective cohort, where criteria for eligibility were met at the end of the follow up period, long after the recruitment and collection of blood samples. This study design is vulnerable to selection bias from differential loss to follow up. Specifically, individuals with severe MS could be more likely to be lost to follow-up, either because of death or because of loss of health insurance or change to a different health plan. If, as it has been suggested,^{3,17} high levels of serum UA slow MS progression, individuals with high UA could be overrepresented among the cases, thus creating a spurious positive association between serum UA and MS risk.

Findings from case-control studies conducted on UA and MS, revealed an inverse association between serum UA concentrations and MS risk, but could not explore the temporal nature of this relationship and are thus subject to bias from reverse causality.^{2,3,8} The lack of association in our study between serum UA and MS risk, is consistent with the interpretation that the lower UA levels among individuals of MS are a consequence rather than a cause of the disease. A decrease in serum UA among individuals with MS could be the result of the scavenging activity of UA for peroxynitrite which is produced as a result of the inflammatory process that occurs in MS. It has been hypothesized that high levels of serum UA could be beneficial in patients with MS.¹⁸ Our results do not address this hypothesis, which is currently being evaluated in randomized trials of inosine, a UA precursor.

References

1. Koch M, De Keyser J. Uric acid in multiple sclerosis. *Neurol Res* 2006;28(3):316–9. [PubMed: 16687059]
2. Rentzos M, Nikolaou C, Anagnostouli M, Rombos A, Tsakanikas K, Economou M, Dimitrakopoulos A, Karouli M, Vassilopoulos D. Serum uric acid and multiple sclerosis. *Clin Neurol Neurosurg* 2006;108(6):527–31. [PubMed: 16202511]
3. Toncev G, Milicic B, Toncev S, Samardzic G. Serum uric acid levels in multiple sclerosis patients correlate with activity of disease and blood-brain barrier dysfunction. *Eur J Neurol* 2002;9(3):221–6. [PubMed: 11985629]
4. Knapp CM, Constantinescu CS, Tan JH, McLean R, Cherryman GR, Gottlob I. Serum uric acid levels in optic neuritis. *Mult Scler* 2004;10(3):278–80. [PubMed: 15222691]
5. Gilgun-Sherki Y, Melamed E, Offen D. The role of oxidative stress in the pathogenesis of multiple sclerosis: the need for effective antioxidant therapy. *J Neurol* 2004;251(3):261–8. [PubMed: 15015004]
6. Whiteman M, Halliwell B. Protection against peroxynitrite-dependent tyrosine nitration and alpha 1-antiproteinase inactivation by ascorbic acid. A comparison with other biological antioxidants. *Free Radic Res* 1996;25(3):275–83. [PubMed: 8889493]
7. Hooper DC, Bagasra O, Marini JC, Zborek A, Ohnishi ST, Kean R, Champion JM, Sarker AB, Bobroski L, Farber JL, Akaike T, Maeda H, Koprowski H. Prevention of experimental allergic encephalomyelitis by targeting nitric oxide and peroxynitrite: implications for the treatment of multiple sclerosis. *Proc Natl Acad Sci U S A* 1997;94(6):2528–33. [PubMed: 9122229]

8. Hooper DC, Spitsin S, Kean RB, Champion JM, Dickson GM, Chaudhry I, Koprowski H. Uric acid, a natural scavenger of peroxynitrite, in experimental allergic encephalomyelitis and multiple sclerosis. *Proc Natl Acad Sci U S A* 1998;95(2):675–80. [PubMed: 9435251]
9. Ascherio A, Munger KL, Lennette ET, Spiegelman D, Hernan MA, Olek MJ, Hankinson SE, Hunter DJ. Epstein-Barr virus antibodies and risk of multiple sclerosis: a prospective study. *Jama* 2001;286(24):3083–8. [PubMed: 11754673]
10. Kageyama N. A direct colorimetric determination of uric acid in serum and urine with uricase-catalase system. *Clin Chim Acta* 1971;31(2):421–6. [PubMed: 5573684]
11. Friedman GD, Siegelaub AB. Changes after quitting cigarette smoking. *Circulation* 1980;61(4):716–23. [PubMed: 7357713]
12. DeLorenze GN, Munger KL, Lennette ET, Orentreich N, Vogelmann JH, Ascherio A. Epstein-Barr virus and multiple sclerosis: evidence of association from a prospective study with long-term follow-up. *Arch Neurol* 2006;63(6):839–44. [PubMed: 16606758]
13. Munger KL, DeLorenze GN, Levin LI, Rubertone MV, Vogelmann JH, Peck CA, Peeling RW, Orentreich N, Ascherio A. A prospective study of Chlamydia pneumoniae infection and risk of MS in two US cohorts. *Neurology* 2004;62(10):1799–803. [PubMed: 15159481]
14. Vaughan JH, Riise T, Rhodes GH, Nguyen MD, Barrett-Connor E, Nyland H. An Epstein Barr virus-related cross reactive autoimmune response in multiple sclerosis in Norway. *J Neuroimmunol* 1996;69(1–2):95–102. [PubMed: 8823380]
15. Wagner HJ, Munger KL, Ascherio A. Plasma viral load of Epstein-Barr virus and risk of multiple sclerosis. *Eur J Neurol* 2004;11(12):833–4. [PubMed: 15667414]
16. Saag KG, Choi H. Epidemiology, risk factors, and lifestyle modifications for gout. *Arthritis Res Ther* 2006;8 (Suppl 1):S2. [PubMed: 16820041]
17. Spitsin S, Hooper DC, Leist T, Streletz LJ, Mikheeva T, Koprowski H. Inactivation of peroxynitrite in multiple sclerosis patients after oral administration of inosine may suggest possible approaches to therapy of the disease. *Mult Scler* 2001;7(5):313–9. [PubMed: 11724447]
18. Spitsin SV, Scott GS, Kean RB, Mikheeva T, Hooper DC. Protection of myelin basic protein immunized mice from free-radical mediated inflammatory cell invasion of the central nervous system by the natural peroxynitrite scavenger uric acid. *Neurosci Lett* 2000;292(2):137–41. [PubMed: 10998568]

1	Yrs between serum collection and MS onset	<=0	1-5	6-10	11-20	>=21
	Mean Age at Blood Collection (y)	35.4	36.0	31.0	31.8	30.3
	Mean Age at MS Onset (y)	30.3	36.0	35.6	45.4	57.4
2						

Figure 1.
Change in residuals of plasma UA among MS cases by time of serum collection in years in KPNC

Table 1

Baseline Characteristics According to Tertiles of Serum UA Levels

	Tertiles of Serum Uric Acid (mg/dL)		
	1	2	3
NHS and NHS II			
Median Serum Acid Concentration	3.2	4.4	6.2
No. Controls	7	11	8
Age (y)	48.9	51.1	52.9
Cohort (% NHS vs NHSII)	71.4	100.0	75.0
Body mass index (kg/m^2)	23.9	25.4	25.8
Smoking Status (%)			
Never Smoker	28.6	36.4	50.0
Past Smoker	57.1	54.6	12.5
Current Smoker	14.3	9.1	25.0
Missing	0	0	12.5
Blood Pressure			
Normal	85.7	54.6	50.0
Pre-hypertensive	14.3	36.4	37.5
Hypertensive	0	9.1	0
Missing	0	0	12.5
KPNC			
Median Serum Acid Concentration	3.3	4.0	5.4
No. Controls	43	44	43
Age (y)	32.0	32.9	33.6
Gender (% female)	90.7	88.6	58.1
Year of Serum Collection	1967	1966	1965
Race			
White	65.1	79.6	62.8
Black	18.6	13.6	25.6
Other	16.3	6.8	11.6
Body mass index (kg/m^2)	22.8	22.8	24.8
Missing (%)	2.3	6.8	0
Smoking Status (%)			
Never Smoker	44.2	27.3	27.9
Past Smoker	7.0	13.6	4.7
Current Smoker	39.5	43.2	55.8
Missing	9.3	15.9	11.6
Blood Pressure (%)			

	Tertiles of Serum Uric Acid (mg/dL)		
	1	2	3
Normal	46.5	36.4	30.2
Pre-hypertensive	11.6	18.2	23.3
Hypertensive	16.3	18.2	14.0
Missing	25.6	27.3	32.6

*Values are means unless otherwise indicated. All values (except age) are standardized to the age distribution of the study population
≈P-values based on ANOVA test

Table 2

Relative Risk of Multiple Sclerosis by Tertiles by Serum UA, considering only MS cases whose blood was collected before onset of disease

	T1	T2	T3
NHS and NHSII			
UA (mg/dL) *	3.2	4.4	6.2
No. of Cases/No. of Controls	8/7	6/11	4/8
Age Adjusted RR	1.00	RR (95% CI) 0.55 (0.12–2.55)	RR (95% CI) 0.40 (0.07–2.24)
Multivariable RR [∞]	1.00	p-value 0.44 0.32 (0.05–2.06)	p-value 0.30 0.39 (0.05–3.22)
KPNC			
UA (mg/dL) *	3.3	4.0	5.4
No. of Cases/No. of Controls	11/43	17/44	14/43
Age Adjusted RR [‡]	1.00	RR (95% CI) 1.97 (0.78–5.00)	RR (95% CI) 1.90 (0.71–5.11)
Multivariable RR [≡]	1.00	p-value 0.15 1.48 (0.42–5.22)	p-value 0.20 2.24 (0.62–8.06)

* Median concentration among controls

[∞] RR and 95% CI from unconditional logistic regression, cases and controls matched on age (controlled for in 5 year intervals) and cohort (NHS or NHSII)

[‡] RR and 95% CI from unconditional logistic regression (||) additionally adjusted for BMI, smoking status (never, past, current smoker), and blood pressure (normal, pre-hypertensive, hypertension)

[≡] RR and 95% CI from unconditional logistic regression cases and controls matched on age (controlled for in 5 year intervals), year of serum collection, and gender

[≡] RR and 95% CI from unconditional logistic regression (¥) additionally adjusted for BMI, smoking status (never, past, current smoker), and blood pressure (normal, pre-hypertensive), and race (using skin color: white, black, other)

Table 3

Relative Risk of Multiple Sclerosis by Tertiles by Serum UA, considering MS cases whose blood was collected both before and after disease onset

	T1	T2	T3
NHS and NHSII			
UA (mg/dL) *	3.2	4.4	6.2
No. of Cases/No. of Controls	13/7	11/11	7/8
Age Adjusted RR	1.00	RR (95% CI) 0.52 (0.14–1.87)	RR (95% CI) 0.47 (0.11–2.08)
Multivariable RR [∞]	1.00	0.48 (0.12–1.90)	0.59 (0.12–3.04)
		p-value 0.31	p-value 0.32
KPNC			
UA (mg/dL) *	3.3	4.0	5.4
No. of Cases/No. of Controls	17/43	20/44	24/43
Age Adjusted RR [‡]	1.00	RR (95% CI) 1.34 (0.60–3.00)	RR (95% CI) 1.56 (0.68–3.61)
Multivariable RR [≡]	1.00	1.14 (0.39–3.30)	1.81 (0.59–5.55)
		p-value 0.48	p-value 0.30
		0.82	0.30

* Median concentration among controls

[†] RR and 95% CI from unconditional logistic regression, cases and controls matched on age (controlled for in 5 year intervals) and cohort (NHS or NHSII)

[∞] RR and 95% CI from unconditional logistic regression (!) additionally adjusted for BMI, smoking status (never, past, current smoker), and blood pressure (normal, pre-hypertensive, hypertension)

[‡] RR and 95% CI from unconditional logistic regression cases and controls matched on age (controlled for in 5 year intervals), year of serum collection, and gender

[≡] RR and 95% CI from unconditional logistic regression (¥) additionally adjusted for BMI, smoking status (never, past, current smoker), and blood pressure (normal, pre-hyper tensive), and race (using skin color: white, black, other)