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Association of serum uric acid level with ischemic stroke, stroke subtypes and clinical outcome

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Abstract

Background and Objective: Uric acid is a neuroprotective agent. However, its relationship with ischaemic stroke remains controversial. We analyzed the association between serum uric acid and ischemic stroke and clinical outcome. *Methods:* The study subject consisted of 550 ischemic stroke patients from the Nizam's Institute of Health Sciences, Hyderabad, India with 550 matched healthy controls. Serum uric acid levels were estimated, and follow-up interviews conducted with patients. *Results:* There was a significant association of elevated levels of serum uric acid with stroke and its subtypes except lacunar stroke. Patients with high serum uric acid levels had a significant increased risk of poor outcome.
Conclusion: Serum uric acid level is associated with ischemic stroke, and is an independent prognostic factor of poor outcome.

INTRODUCTION

Despite abundant epidemiological evidence, the role of uric acid (UA) as an independent marker of cardiovascular risk has been controversial for over 50 years. The Framingham Heart Study was the first one to evaluate an association between elevated serum UA levels and cardiovascular disease outcome in general population. The study concluded that the association with cardiovascular disease merely reflects the link between serum UA levels and other risk factors including hypertension, kidney disease, elevated lipoprotein levels and use of diuretics.[1,2] On the contrary several other epidemiological studies have shown elevated UA levels to predict increased risk of cardiovascular events, hypertension, congestive heart failure and Type II diabetes mellitus.[3-6]

The administration of UA has been reported to be neuroprotective in rats after transient brain ischemia and may reinforce the benefits provided by alteplase.[7,8] Increased UA levels were found to be associated with increased risk of ischemic as well as hemorrhagic strokes.[9] Higher UA levels in stroke patients have also been described in association with improved or worse outcome.[10-12] The discrepancies in these results could be on account of variable timing of UA, the inconsistent definition of outcome and variable use of therapeutics in these studies.[13] Therefore, the aim of the present study was to assess the serum UA levels in the early phase of ischemic stroke and to determine the relation between serum UA levels and clinical outcome. Furthermore serum UA levels have not been evaluated in ischemic stroke subtypes. Therefore, we determined serum UA levels for stroke subtypes classified according to the trial of ORG 10172 in Acute Stroke Treatment (TOAST) classification.

METHODS

Subjects

Five hundred and fifty ischemic stroke patients (males:females = 376:174), presenting with new stroke evaluated in the stroke clinic of Nizam's Institute of Health Sciences, Hyderabad (Andhra Pradesh, India) between July 2009 and June 2012 were included in the study. The study was approved by the ethical committee of the study hospital. All patients were examined by a qualified neurologist, and ischemic stroke was differentiated by computer tomography (CT) scan and magnetic resonance imaging (MRI). All the patients underwent CT scan as well as MRI. Patients with major cardiac, renal, hepatic, endocrinological disorders, skeletal disorders,

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cancerous diseases, cardiac diseases, and recent infections were excluded from this study. As a control group 550 healthy individuals matched for sex and age were recruited from the same demographic area. The controls were the blood donors from the study hospital and staff members of Institute of Genetics and Hospital for Genetic Hospital. The controls had no clinical evidence of any cerebrovascular disease.

According to Joint National Committee VI–VII, hypertension was defined as a systolic blood pressure >140 mmHg and/or a diastolic blood pressure >90 mmHg based on the average of two blood pressure measurements, or a patient's self-reported history of hypertension or antihypertensive use, supported by documents. Diabetes was diagnosed if fasting plasma glucose was >110mg/100ml or patient was on anti-diabetic medications. Smokers were defined as those reporting daily smoking. Ex-smokers and occasional smokers were classified as non-smokers. Since patients were found to drink alcohol in different forms and many were reluctant to admit the exact amount of consumption, we defined alcohol usage as consumption of at least one alcoholic drink in a week.[14] High-density lipoprotein (HDL) and triglyceride levels were estimated by commercially available kits supplied by ERBA Mannheim (Germany) using an ERBA auto analyzer. Information on demographic features was collected using a structured questionnaire. The ischemic stroke was classified according to Trial of Org 10172 in Acute Stroke Treatment (TOAST) classification.

Blood Samples and serum uric acid Assay

Blood samples for assessment of serum UA were collected at admission within 24 h after qualifying stroke. Levels of serum UA were determined with a commercially available ELISA kit procured from Diagnostics Biochem Canada. The absorbance of the reaction mixture was measured at 505 nm and the UA level was determined using the formula:

UA (mg/dL) = Abs. of test/Abs. of standard X Concentration of standard

Follow-up

The follow-up telephone interviews were conducted with all the patients at 3, 6, and 12 months after hospital discharge. Poor outcome was defined as either a score of more than two on modified Rankin Scale Score (including death at 3 months) or <5 on an extended Glasgow Outcome Scale (GOS-E) from stroke onset. Outcome was assessed without any knowledge on baseline serum UA levels.

Statistical analysis

The data were expressed as mean±SD value. The difference in demographic features between cases and controls was evaluated by Student's t test. The distribution of serum UA values was not normal. It was positively skewed. Therefore, Mann–Whitney U test (a nonparametric test) was used for comparison of mean values of serum UA in cases and controls. The relationship between serum UA levels with stroke was evaluated by a stepwise multiple logistic regression analysis using SPSS18. Confounding factors included in multiple logistic regression analysis were age, sex, smoking, diabetes, hypertension, alcoholism, total cholesterol, and triglycerides. The relationship of serum UA levels with stroke outcome was also evaluated using multiple logistic regression analysis.

RESULTS

Five hundred and fifty ischemic stroke patients and 550 controls were included in the study. All the patients belonged to a South Indian population from Andhra Pradesh. The clinical characteristics of stroke patients and controls is presented in Table 1.

The serum UA levels were found to be significantly elevated in stroke patients with 54.9% of the patients versus 24.7% of controls had high levels of serum UA (>6mg/dL) (Table 2). Mean serum UA level was significantly high in patients when compared to the controls ($p<0.001$) (Table 3). The significance remained even after adjusting for known risk factors of stoke [adjusted odds ratio=2.93 (95% CI; 1.93-4.876) and $p<0.001$]. The association of serum UA levels with outcome was carried out by multiple logistic regression analysis. High levels of serum UA associated significantly with poor outcome in patients independently of other prognostic factors [adjusted odds ratio=3.86 (95%CI; 1.501-5.317) and $p<0.001$]. The recurrence of stroke and death rate was also high in patients with high levels of serum UA in comparison with patients with low levels of serum UA (Table 4). As for the stroke subtypes, the high level of serum UA were found to be associated significantly with all stroke subtypes except lacunar stroke (Table 5).

Table 1: Clinical characteristics of stroke patients and controls

	Patients (n= 550)	Controls (n= 550)	*p* value
Age	49.3 (17.34)	47.01 (16.78)	
Male : Female	376:174	373:171	
Systolic BP (mmHg)	142 (17.2)	128 (16.2)	<0.001
Diastolic BP (mmHg)	88.4 (20.2)	79 (15.3)	<0.001
Total cholesterol (mg/dl)	197.45 (40.45)	195.36 (47.50)	>0.05
Triglycerides (mg/dl)	178.5 (40.02)	138.68 (43.3)	<0.001
Random Glucose (mg/dl)	128.08 (7.2)	118.73 (23.28)	<0.001
BMI, kg/m2	27.86 ± 2.35	23.2 ± 2.7	<0.001
Hypertension	304 (55.2%)	159 (28.9%)	<0.001
Diabetes	255 (46.3%)	153 (27.8%)	<0.001
Smokers	224 (40.7%)	181 (32.9%)	<0.01
Alcoholics	208 (37.8%)	134 (24.3%)	<0.001
HDL cholesterol (mg/dl)	53.25 (20.23)	59.56 (22.62)	<0.001

Age, systolic BP, diastolic BP, total cholesterol, high density lipoprotein (HDL) cholesterol, random glucose, and triglycerides are given as mean (SD). Stroke patients have significantly higher levels of systolic and diastolic blood pressure, higher serum triglycerides and random glucose, and a lower level of HDL (p<0.001). *p* values were calculated using Student's t′ unpaired and proportion Z test (SPSS 18)

Table 2: Serum uric acid levels in patients and controls

Study Group	Normal levels (%)	High levels (%)	Total	χ^2	OR	*p* value
Patients n (%)	248 (45.1)	302 (54.9)	550	104.4	3.70 2.869-4.79	<0. 001
Controls n (%)	414 (75.2)	136 (24.7)	550			

For high levels *vs.* normal levels χ^2= 104.4; *p<0.001*; odds ratio=3.70 (2.869-4.79)

Table 3: Mean serum uric acid levels in patients and controls. The difference in mean serum uric acid levels between cases and controls was examined by Mann–Whitney U test (nonparametric test)

Group	Serum uric acid (mean±SD)	*p* value
Patients	6.14±1.68	<0.001
Controls	4.12±2.20	

DISCUSSION

Most epidemiological studies have reported a significant association between elevated serum UA and increased cerebrovascular disease.[15] Although UA is one of the most important antioxidants in plasma/serum and appears to be neuroprotective in animal models, the results from human studies are controversial.[13] Serum UA levels would change

Table 4: Comparison of serum uric acid levels with outcome in stroke patients

	Outcome	Serum uric acid ranges			χ^2	*p* value
		Normal	High	Total		
Performance	Good	142 (51.0)	136 (48.9)	278	126.3	<0.001
	Bad	20 (7.3)	252 (92.6)	272		
Reccurrence	Yes	9 (6.7)	124 (93.2)	133	30.81	<0.001
	No	128 (30.6)	289 (69.3)	417		
Survival Rate	Survival	181 (40.7)	263 (59.2)	444	8.496	0.003
	Death	27 (25.4)	79 (74.5)	106		

Table 5: Distribution of SUA levels in stroke patients classified according to TOAST classification

TOAST Classification	No. of patients	Levels (%)		Odds ratio	*p* value
Large artery atherosclerosis		Low	High		
A. Intracranial large artery	184	67 (36.4)	117 (63.5)	5.31 (3.719-7.599)	<0.001
B. Extracranial large artery	55	9 (16.3)	46 (83.6)	15.5 (7.422-32.61)	<0.001
Lacunar stroke	91	77 (84.6)	14 (15.3)	0.55 (0.303-1.01)	0.051
Cardioembolism	78	38 (48.7)	40 (51.2)	3.20 (1.974-5.201)	<0.001
Other determined etiology	32	6 (18.7)	26 (81.2)	13.19 (5.318-32.72)	<0.001
Undetermined etiology	110	51 (46.3)	59 (53.6)	3.52 (2.31-5.37)	<0.001

noticeably in association with the degree of oxidative stress in acute ischemic stroke.

A meta-analysis of 16 prospective cohort studies including 230,000 patients found that the elevated serum UA level in adults is associated with a modest but statistically significant increased risk of stroke incidence and mortality.[16] In the Apolipoprotein Mortality Risk Study (AMORIS), Holme *et al*.[9] found increased serum UA level to be a risk factor for acute myocardial infarctions, congestive heart failure and stroke. There has been considerable debate whether UA is neuro-protective as an antioxidant or neuro-toxic as a pro-oxidant.[17,18] It is a decade old scientific dispute.[19] The antioxidant properties of urate have long been known. The issue originated in two complementary reports suggesting urate as both a primate evolutionary substitute for ascorbate and as a prooxidant.[18-21] The authors have stated that in addition to being a potent antioxidant, urate mediates radical oxidations and likely, oxidative- stress-related diseases.[18-21] Proctor has suggested that, "The well established association between high urate levels and atherosclerosis could be a protective reaction (antioxidant) or a primary cause (pro-oxidant)".[18] The authors have also suggested that the exact mix of pro-oxidant verses antioxidant properties for uric acid depends on a complex mix of concentration, oxygen availability, electronically active species, other pro- and antioxidant enzymes, transition –series metals and so forth.[18-20] In conditions of extraordinary oxidative stress the balance between the pro- and antioxidant properties of uric acid may shift in favour of tissue protection. This is particularly so because urate scavenges oxidants such as peroxynitrites, where normal background levels are low, except in pathogenic processes.[18] Therefore, it has been suggested that a low level of chronic, sometimes pathogenic oxidative stress may be the price paid for the protective presence of urate when things go bad acutely.

In our present study, we found that the levels of serum UA were significantly elevated in ischemic stroke patients within 24 hours after symptom onset. The results did not change even after adjusting the potential confounders. The strength of the present study was that it was a prospective study with blinding of those who assessed the follow-up events. The study had a large sample size, and the outcome measures were robust. Early measurements of serum UA within 24 hours of stroke onset increased the relevance of our findings. Moreover, we excluded

the patients with renal disorders and gout because of the association of hyperurecemia with these conditions. There are limited studies on the association of serum UA levels with stroke risk and stroke sub types. There is hardly any study from India evaluating the association of serum UA with ischemic stroke. To the best of our knowledge, this is the first study to analyze the association of serum UA values with ischemic stroke, its subtypes, and clinical outcome.

UA is a weak organic acid that is naturally produced in the human body as the end result of purine catabolism from xanthine and hypoxanthine and it has long been considered an anti-oxidant reagent.[22] Because of Its significant capacity as an antioxidant, it behaves as a free radical scavenger, and therefore, may have a protective role in vascular inflammation and dysfunction.[23] The size of the ischemic infarct and consequently the severity of the stroke event have been found to be greater in patients with diminished antioxidant activity. Since the level of free radicals is extremely difficult to measure in the human body, UA can be taken as a potential biomarker of the free radical level, which has been established as an accurate measure of the amount of free radicals generated in the body.

There have been reports suggesting a sudden and drastic increase in the UA level at the onset of an ischemic attack[24], which might be a mechanism to combat the generation of large quantities of free radicals. The idea of increased serum urate contributing to elevated stroke risk suggested by a few studies[25], has not been supported with sufficient evidence. Interestingly, there were a number of studies showing that administration of uric acid at the time of a brain attack or immediately later is associated with reduced degree of brain injury and the infarct size in stroke patients.[8,26] This is consistent with the observation of a better clinical outcome in patients with acute stroke for every milligram per decilitre increase in serum UA.[10]

Although an increased severity of brain damage has been correlated with elevated levels of UA (hyperuricemia) in the body[27], there are no consistent results to prove this. An initial increase in urate levels can act as a predictor of ischemic brain injury, but cannot be considered as a cause of brain damage.[28] In fact, apart from the fact that UA administration in-vivo reduces stroke severity, administering UA to normal individuals did not have an adverse effect[29], indicating that reduced levels of UA might be potentially be linked with an increased stroke severity and vice versa. It has been proposed that serum UA may show both anti- and pro-oxidant properties depending on levels of other antioxidants, levels of oxidative stress and time of interaction with the target tissues, and that the balance between the anti- and pro-oxidant properties shifts in favour of neuroprotection in conditions of extraordinary oxidative stress such as acute ischemic stroke.[18]

Early studies investigating a possible association between serum UA and clinical outcome showed mixed results.[10,11] However, a potential limitation of these studies may be a wider time window for serum UA measurements (e.g up to 48 and 72 hours). The study carried out by Weir *et al*.[11] showed that serum UA level predicts poor outcome after ischemic stroke. Our present study also shows high levels of serum UA (>5mg/dL) associated significantly with poor outcome. These results were confirmed by MLR analysis The recurrence of stroke and death rate was also high in patients with high levels of serum UA in comparison of patients with low levels of serum UA ($p<0.001$ and 0.003 respectively). Our results are also in accordance with the Rotterdam Study and other studies, in which they found a significant association of hyperuricemia with severity of ischemic stroke.[27,30] A recent prospective study carried out by Logallo *et al*.[31] at Haukeland University Hospital, Norway revealed that increased serum UA levels associated with bad outcomes in ischemic stroke patients. Another recent study carried out by Miedema *et al*[32] also confirmed these findings. However, some studies reported that the decreases in serum UA during the first week after onset of stroke correlates with more severe stroke, unfavorable stroke evolution, and poor long-term stroke outcome.[33,34]

In conclusion, in our study, high serum UA levels were observed to be associated significantly with ischemic stroke, stroke subtypes (except lacunar stroke), and poor outcome including death at 3 months. Considerable progress has been made in identifying and treating modifiable risk factors for stroke. If identified as an etiological agent in the pathogenesis of vascular disease including stroke, hyperurecemia could be targeted therapeutically in the same way as other risk factors like dyslipedemia and blood pressure are routinely treated after stroke. However, more research is needed at scientific and clinical levels before routine treatment of serum urate can be recommended.

DISCLOSURE

Conflict of interest: None

REFERENCES

1. Abbott RD, Brand FN, Kannel WB, Castelli WP. Gout and coronary heart disease: the Framingham Study. *J Clin Epidemiol* 1988; 41(3):237-42.
2. Culleton BF, Larson MG, Kannel WB, Levy D. Serum uric acid and risk for cardiovascular disease and death: the Framingham Heart Study. *Ann Intern Med* 1999; 131(1):7-13.
3. Lehto S, Niskanen L, Ronnemaa T, Laakso M. Serum uric acid is a strong predictor of stroke in patients with non- insulin-dependent diabetes mellitus. *Stroke* 1998; 29(3):635-9.
4. Niskanen LK, Laaksonen DE, Nyyssönen K, *et al.* Uric acid level as a risk factor for cardiovascular and all-cause mortality in middle-aged men: a prospective cohort study. *Arch Intern Med* 2004; 164(14):1546-51.
5. Kawamoto R, Tomita H, Oka Y, *et al.* Association between uric acid and carotid atherosclerosis in elderly persons. *Intern Med* 2005; 44(8):787-93.
6. Gerritsen WB, van Boven WJ, Boss DS, *et al.* Malondialdehyde in plasma, a biomarker of global oxidative stress during mini-CABG compared to on- and off-pump CABG surgery: a pilot study. *Interact Cardiovasc Thorac Surg* 2006; 5(1):27-31.
7. Yu ZF, Bruce-Keller AJ, Goodman Y, Mattson MP. Uric acid protects neurons against excitotoxic and metabolic insults in cell culture, and against focal ischemic brain injury in vivo. *J Neurosci Res* 1998; 53(5):613-25.
8. Romanos E, Planas AM, Amaro S, Chamorro A. Uric acid reduces brain damage animproves the benefits of rt-PA in a rat model of thromboembolic stroke. *J Cereb Blood Flow Metab* 2007; 27(1):14-20.
9. Holme I, Aastveit AH, Hammar N, Jungner I, Walldius G. Uric acid and risk of myocardial infarction, stroke and congestive heart failure in 417,734 men and women in the Apolipoprotein MOrtality RISk study (AMORIS). *J Intern Med* 2009; 266(6):558-70.
10. Chamorro A, Obach V, Cervera A, *et al.* Prognostic significance of uric acid serum concentration in patients with acute ischemic stroke. *Stroke* 2002; 33:1048-52.
11. Weir C, Muir S, Walters M, Lees KR. Serum urate as an independent predictor of poor outcome and future vascular events after acute stroke. *Stroke* 2003; 34:1951-7.
12. Seet RC, Kasiman K, Gruber J, *et al.* Is uric acid protective or deleterious in acute ischemic stroke? A prospective cohort study. *Atherosclerosis* 2010; 209(1):215-9.
13. Amaro S, Urra X, Gómez-Choco M, *et al.* Uric acid levels are relevant in patients with stroke treated with thrombolysis. *Stroke* 2011; 42(1):S28-S32.
14. Munshi A, Shehnaz S, Kaul S, *et al.* Angiotensin converting enzyme insertion/deletion polymorphism and the risk of ischemic stroke in a South Indian population. *J Neurol Sc* 2008; 272(1–2):132-5.
15. Hong JM, Bang OY, Chung CS, *et al.* Influence of recanalization on uric acid patterns in acute ischemic stroke. *Cerebrovasc Dis* 2010; 29(5):431-9.
16. Kim SY, Guevara JP, Kim KM, *et al.* Hyperuricemia and risk of stroke: a systematic review and meta-analysis. *Arthritis Rheum* 2009; 61(7):885-92.
17. Kanellis J, Johnson RJ. Editorial comment-Elevated uric acid and ischemic stroke: accumulating evidence that it is injurious and not neuroprotective. *Stroke* 2003; 34(8):1956-7.
18. Proctor PH. Uric acid: neuroprotective or neurotoxic? *Stroke* 2008; 39(5):e88. author reply e89.
19. Dawson J, Quinn T, Lees K, Walters M. The continued Yin and Yang of uric acid . *Stroke* 2008; 39:e9.
20. Proctor P. Electron transfer factors in psuchosis and dyskinesia. *Physiol Chem phys* 1972; 4:349-60.
21. Proctor PH. Free radicals and Human Disease. In: CRC Handbook of Free Radicals and Antioxidants in Biomedicine. Boca Raton, Fla:CRC Press, Inc ;1989;209-21.
22. Squadrito GL, Cueto R, Splenser AE, *et al.* Reaction of uric acid with peroxynitrite and implications for the mechanism of neuroprotection by uric acid. *Arch Biochem Biophys* 2000; 376:333-7.
23. Waring WS. Uric acid: an important antioxidant in acute ischaemic stroke. *QJM* 2002; 95(10):691-3.
24. Nieto FJ, Iribarren C, Gross MD, Comstock GW, Cutler RG. Uric acid and serum antioxidant capacity: a reaction to atherosclerosis? *Atherosclerosis* 2000; 148(1):131-9.
25. Vannorsdall TD, Jinnah HA, Gordon B, Kraut M, Schretlen DJ. Cerebral ischemia mediates the effect of serum uric acid on cognitive function. *Stroke* 2008; 39:3418-20.
26. Amaro S, Planas AM, Chamorro A. Uric acid administration in patients with acute stroke: a novel approach to neuroprotection. *Expert Rev Neurother* 2008; 2:259-70.
27. Chien KL, Hsu HC, Sung FC, *et al.* Hyperuricemia as a risk factor on cardiovascular events in Taiwan: the Chin-Shan Community Cardiovascular Cohort study. *Atherosclerosis* 2005; 183:147-55.
28. Hozawa A, Folsom AR, Ibrahim H, *et al.* Serum uric acid and risk of ischemic stroke: the ARIC Study. *Atherosclerosis* 2006; 187(2):401-7.
29. Chamorro A. Role of inflammation in stroke and atherothrombosis. *Cerebrovasc Dis* 2004; 17(3):1-5.
30. Bellodi G, Bernini G, Manicardi V, *et al*. Arterial hypertension in relation to life style and other cardiovascular risk factors: epidemiologic study of a population of blood donors. Project AVIS. *Minerva Cardioangiol* 1994; 42:73-84.
31. Logallo N, Naess H, Idicula TT, *et al.* Serum uri acid: neuroprotection in thrombolysis. The Bergen NORSTROKE study. *BMC Neurol* 2011; 11:114.
32. Miedema I, Uyttenboogaart M, Koch M, *et al.* Lack of association between serum uric acid levels and outcome in acute ischemic stroke. *J Neurol Sci* 2012; 319(1-2):51-5.
33. Brouns R, Wauters A, Van De Vijver G, *et al.* Decrease in uric acid in acute ischemic stroke correlates with stroke severity, evolution and outcome. *Clin Chem Lab Med* 2010; 48(3):383-90.
34. Chen YC, Su CT, Wang ST, Lee HD, Lin SY. A preliminary investigation of the association between serum uric acid and impaired renal function. *Chang Gung Med J* 2009; 32(1):66-71.