DIETARY RISK FACTORS OF STROKE AND HYPERTENSION IN JAPAN

— Part 2: Validity of Urinalysis for Dietary Salt and Protein Intakes under a Field Condition —

YUKIO YAMORI, M.D., MASAHIRO KIHARA, M.D., JUN FUJIKAWA, M.D.*
YASUNARI SOH, M.D.*, YASUO NARA, Ph.D., MICHIIYA OHTAKA, M.D.**
RYOICHI HORIE, M.D., TOKUGORO TSUNEMATSU, M.D., SHINYA NOTE, M.D.
AND MASAICHI FUKASE, M.D.

Utility of urinalysis as a method for estimating dietary salt and protein intakes was examined in volunteers from farming and fishing villages in Japan and the data was compared to findings in a survey on diets. We found that partial urine samples and even a fasting single spot urine in the a.m. could serve as valid substitutes for 24-hour (hr) urine collections. Further, significant correlations were obtained between levels of salt in the diet estimated by survey interviews and urinary sodium (Na) and between interview-estimated dietary total or animal protein and urinary inorganic sulfate (SO_4). In comparative studies between farming and fishing villages, almost the same findings, higher salt in a farming village and higher animal protein in a fishing village, were obtained in the urinalysis and the survey on diets. These findings clearly indicate that urinalysis, as a method for estimating dietary salt and protein intake in population surveys, has distinct advantages.

IN our studies in spontaneously hypertensive rats (SHR) and stroke-prone SHR, high protein diets were shown to have a preventive effect on hypertension and stroke.¹⁻³ Therefore, the nutritional conditions of low protein intake as well as excessive salt intake seem to be respon-

sible for the high incidence of hypertension and stroke in the Japanese, as had been suspected from epidemiological investigations.⁴⁻⁷ In search of a reliable method for estimating salt and protein intakes, we directed our attention to urinalysis, the utility of which had been shown by other workers.⁸⁻¹¹ We compared our findings with those obtained during interviews regarding diets. In an attempt to ascertain the utility of urinalysis in a field condition, we then evaluated the availability of partial urine samples and further directly compared the data from urinalysis

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Shimane Medical University, Izumo; *Faculty of Medicine, Kyoto University, Kyoto; **Public Health Bureau, Ministry of Health & Welfare, Tokyo, Japan

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Address for correspondence: Yukio Yamori, M.D., Shimane Medical University, 89-1 Enya, Izumo 693, Japan

TABLE I CORRELATION OF EXCRETION RATES (PER KG BODY WEIGHT) IN 24-HOUR URINE WITH CREATININE RATIOS IN VARIOUS PARTIAL DEPOSIT AND SINGLE SAMPLES OF URINE

	<i>n</i>	Na/Cr	K/Cr	UN/Cr	SO ₄ /Cr
Daytime voided urine	83	*** 0.717	*** 0.720	*** 0.531	*** 0.597
Evening voided urine	83	*** 0.559	*** 0.614	*** 0.402	*** 0.421
Overnight voided urine	83	*** 0.419	*** 0.430	*** 0.320	*** 0.457
Single urine sample	81	*** 0.463	*** 0.358	*** 0.330	*** 0.407

*** = statistical significance ($p < 0.001$)

TABLE II CORRELATION BETWEEN VARIOUS URINE SAMPLES AND 24-HOUR URINE SAMPLES

	<i>n</i>	Na/K	SO ₄ /UN
Daytime voided urine	83	*** 0.910	*** 0.914
Evening voided urine	83	*** 0.645	*** 0.738
Overnight voided urine	83	*** 0.797	*** 0.824
Single urine sample	81	*** 0.578	*** 0.363

*** = statistical significance ($p < 0.001$)

Fig.1. Correlation between urinary salt excretion in 24-hr urine and dietary salt intake estimated by dietary survey interview.

to those obtained from the dietary survey interviews.

MATERIALS AND METHODS

Healthy men (51) and women (61) ranging in age from 30 to 50 were randomly selected in two rural areas of Shimane Prefecture. One was Daiwa Village, a farming village in the mountain and the other Fuse Village, a fishing village on the Oki Island. These areas are remote and secluded and their inhabitants are in the lower socioeconomic classes. They almost never dine out, therefore their daily meals are eaten regularly at home.

Twenty-four-hour urine specimens were collected separately as the following 3 preparations: 1) overnight deposit urine: the first voided urine in the a.m., 2) daytime voided urine: urine voided after the first voiding until the evening meal and 3) evening voided urine: urine after the evening meal. Random single spot urines were collected from the subjects in the morning after the overnight urine was voided. All samples were obtained within one day and carefully checked regarding the completeness of the collection. All insufficient samples were discarded.

No information on the purpose of the study was given to the participants before the examination in order to avoid intentional changes in usual dietary fare.

All samples were put into brown plastic bottles with a small amount of toluene as a preservative and stored at -20°C until measurement. They were analyzed for urinary Na and K by flame photometry (Hitachi 205), urea nitrogen (UN) by urease method, creatinine (Cr) by Jaffe's procedure and for inorganic sulfate (SO₄) by turbidimetric method described by Dodgson.¹²

Detailed interviews as to dietary intake for the past 24 hours were conducted by expert dieticians

Fig.2. Correlation between urinary inorganic sulfate (SO_4) excretion in 24-hr urine and dietary protein intake estimated by dietary survey interview.

Fig.3. Correlation between urinary inorganic sulfate (SO_4) excretion in 24-hr urine and dietary animal protein intake estimated by dietary survey interview.

to estimate dietary electrolytes and protein ingested on the same day when 24-hr urines were collected. The data obtained were compared with data on the urinalysis in terms of Cr ratio of each component, Na ratio to K (Na/K) and SO_4 ratio to UN (SO_4 /UN).

RESULTS

Completely collected urine samples were obtained from 39 men (Daiwa 22, Fuse 17) and 44 women (Daiwa 26, Fuse 18) in both villages, and 37 men (Daiwa 18, Fuse 19) and 54 women (Daiwa 25, Fuse 29) complied with dietary

TABLE III ANALYSIS OF 24-HOUR URINE FROM INHABITANTS OF FARMING AND FISHING VILLAGES

	Farming village (48)	Fishing village (35)
Na (mEq/day)	202 ± 79	198 ± 80
K (mEq/day)	51 ± 20	45 ± 15
Urea nitrogen (mg/day)	9099 ± 3324	8443 ± 2525
Sulfate (mg/day)	1093 ± 367	1311 ± 425*
Creatinine (mg/day)	1231 ± 371	1209 ± 380
Urine volume (ml/day)	1081 ± 440	1285 ± 476*
Body weight (kg)	55.4 ± 9.3	54.5 ± 7.8

Values are shown as mean ± SD.

* = statistical significance from farming village ($p < 0.05$)

TABLE IV SALT AND PROTEIN INTAKE ESTIMATED BY DIETARY SURVEY INTERVIEW IN FARMING AND FISHING VILLAGES

	Farming village men: 18 women: 25	Fishing village men: 19 women: 29
Salt (g/day)	17.0 ± 8.3	14.4 ± 5.8
Total protein (g/day)	77.9 ± 37.8	93.1 ± 29.8*
Animal protein (g/day)	37.1 ± 28.3	51.2 ± 25.0*

Values are shown as mean ± SD.

* = statistical significance from farming village ($p < 0.05$)

interviews.

Correlations between excretion rates of urinary constituents in 24-hr urine with their Cr ratio in various partial urines are shown in Table I. Na/Cr, K/Cr, UN/Cr and SO₄/Cr in the urine samples highly correlated with the 24-hr urine specimens, the highest correlation occurring in the daytime voided urine and the second in the evening voided urine. Although correlation coefficients were rather low in single spot urines, a statistical significance was detected in all urinary constituents. Correlations of Na/K and SO₄/UN between 24-hr urine and various partial urine samples are listed in Table II; they were high in urine samples voided and collected during various times of the day, and relatively low in single spot urines.

Figure 1 shows the correlation between salt intake estimated by dietary survey interviews and urinary salt, and Figs. 2 and 3 show the correla-

tions between interview-estimated total or animal protein intake and urinary SO₄ data from 63 subjects (men 28, women 35) who complied with both urine collections and dietary survey interviews. The correlation coefficients ranged from 0.3 to 0.4 and as such were statistically significant. On the other hand, there was no significant correlation between interview-estimated protein intake and urinary UN.

The data from analysis of 24-hr urine specimens and dietary survey interviews are compared in Tables III and IV. Generally speaking, a similar tendency was noted between urinalysis and dietary survey interviews, that is, Na intake and urinary Na tended to be higher in the farming village while animal protein intake and urinary SO₄, a possible index of animal protein intake was significantly higher in the fishing village.

The importance of urinary Cr for assessing the accuracy of the urine collection has been stressed by Joossens et al.¹³ and according to their standard curves of urinary Cr, all the examinees in our study provided a complete collection of 24-hr voided urine.

DISCUSSION

Significant correlations were confirmed in a field condition between a 24-hr urine and various partial urine specimens, with regard to Na/Cr, K/Cr, UN/Cr, SO₄/Cr, Na/K and SO₄/UN. In terms of the degree of correlation coefficient daytime voided urine was the best substitute for a 24-hr urine collection, however, the evening and overnight voided urine were practically better than daytime voided urine simply because urinary collection at home is far easier than the collection during daytime when most of these

country people are occupied in works outdoors. Single spot urine is also a method for use in population surveys, particularly in a large population, because it can be readily collected.

In a comparison of the data obtained by urinalysis with those from dietary survey interviews, low but significant relations were observed between dietary Na and urinary Na and between dietary total or animal protein and urinary SO_4 . These results are of interest because interview-estimated dietary salt and protein intakes theoretically do not relate directly with urinary components of 24-hr urine collected on the same day when the dietary intakes were examined by interviews. Such consistency of interview-estimated values with urinary components may be ascribed to the relatively steady dietary customs of these village people who regularly eat 3 meals at home and do not dine out. In this sense these village people are more suitable subjects for epidemiological surveys of hypertension-related diseases than are persons living in urban areas. Similar results were obtained as to the nutritional situations in both villages, by both urinalysis and dietary survey interviews.

Our findings show the utility of urinalysis as a method for estimating nutritional situations, in field conditions.

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REFERENCES

1. YAMORI Y, HORIE R, HANDA H, OHTAKA M, NARA Y, FUKASE M: Pathogenetic approach to the prophylaxis of stroke and atherogenesis in SHR. In Spontaneous Hypertension, DHEW Publication No (NIH) 77-1179, 1977, p 269
2. YAMORI Y, HORIE R, IKEDA K, NARA Y, LOVENBERG W: Prophylactic effect of dietary protein on stroke and its mechanisms. In Prophylactic Approach to Hypertensive Diseases, ed by YAMORI Y, LOVENBERG W, FREIS ED, Raven Press, New York, 1979, p 497
3. YAMORI Y: Environmental influences on the development of hypertensive vascular diseases in SHR and related models, and their relation to human disease. In New Trends in Arterial Hypertension, ed by WORCEL M, BONVALET JP, LANGER SZ, MENARD J, SASSARD J, Elsevier/North-Holland Biomedical Press, Amsterdam, 1981, p 305
4. DAHL LK: Salt and hypertension. *Am J Clin Nutr* 25: 231, 1972
5. SASAKI N: High blood pressure and the salt intake of the Japanese. *Jpn Heart J* 3: 313, 1962
6. MENEELY GR, BATTARBEE HD: High sodium-low potassium environment and hypertension. *Am J Cardiol* 38: 768, 1976
7. KIMURA M, TOSHIMA H, NAKAYAMA Y, MIZUGUCHI T, TAKAYAMA K, YOSHINAGA M, FUKAMI T, TAHIRO H, KATAYAMA F, ABE K, ARIMA T, YOKOTA Y, MINAGAWA E, TANAKA R, AKIYOSHI T, SOEJIMA K, YAMADA T, IKEDA H, NAKAMICHI E, AGETA M, MIIKE T, SHIMADA S, FUKUMOTO T: Population survey on cerebrovascular and cardiovascular diseases. *Jpn Heart J* 13: 118, 1972
8. DAHL LK: Salt intake and salt need. *N Engl J Med* 258: 1152, 1958
9. SIMMONS WK: Urinary urea nitrogen/creatinine ratio as indicator of recent protein intake in field studies. *Am J Clin Nutr* 25: 539, 1972
10. SIMMONS WK: Use of the inorganic sulfate sulfur/creatinine ratio in field studies. *Am J Clin Nutr* 26: 72, 1973
11. DEHAVEN I, SHERWIN R, HENDLER R, FELIG P: Nitrogen and sodium balance and sympathetic-nervous system activity in obese subjects treated with a low-caloric protein or mixed diet. *N Engl J Med* 302: 477, 1980
12. DODGSON KS: Determination of inorganic sulfate in studies on the enzymic and non-enzymic hydrolysis of carbohydrate and other sulphate esters. *Biochem J* 78: 312, 1961
13. JOOSSENS JV, CLAESSENS J, GEBOERS J, CLAES JH: Electrolytes and creatinine in multiple 24-hr urine collections (1970-1974). In Epidemiology of Arterial Blood Pressure, ed by KESTELOVT H, JOOSSENS JV, Martius Nijhoff Medical Division, Hague, 1980