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HOW WALKING INDUCES A DECREASE IN URINARY SODIUM EXCRETION IN CHILDREN WITH MESANGIAL CELL PROLIFERATIONS

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ABSTRACT

Glomerular mesangial cell proliferations are common, but not specific, findings in biopsied kidney specimens. Using 246 subjects aged 6 to 18 years with chance proteinuria and/or hematuria and biopsy-proved chronic glomerulonephritis, we clinically studied the roles of proliferated mesangial cells in the regulation of urinary sodium excretion, especially in relation to a positional change from supine to upright, standing, and walking.

Significantly greater decreases in urinary sodium excretion were found to be induced by standing and walking for patients with severe mesangial cell proliferations than in patients with minimal mesangial change. These results suggest that the proliferated mesangial cells decrease urinary sodium excretion in response to standing and walking, which may aid in estimating renal glomerular lesions in patients with chance proteinuria and/or hematuria without using any invasive methods.

INTRODUCTION

There have been many patients found with chance proteinuria and/or hematuria by routine screening tests for renal disease in school children. These screening studies have demonstrated an incidence of 0.05 to 2.0[1,2] per cent.[3] Great increases in the rate of patients with diseases has led to a therapeutic dilemma, since conventional renal function tests of the patients are close to normal.

In recent years, glomerular mesangial cells have been considered to be the predominate regulator of contraction of the glomerular capillary surface area stimulated by angiotensin II, and thereby, of filtration coefficient. A positional change from supine to

upright is known to promote the production of plasma renin-angiotensin-aldosterone.[7~11]

This clinical study was designed to examine whether the proliferated mesangial cells have any effects on urinary sodium excretion upon a postural change from supine to upright, standing, and walking.

METHODS

SUBJECTS

Chance proteinuria and/or hematuria

219 patients aged 6 to 18 (90 males and 129 females) with normal renal functions and found to have chance proteinuria and/or hematuria were classified into four large groups (I, II, III, IV) according to urinary findings as follows;

Group I (n=89, 30 males and 59 females); negative urinary protein with urinary red blood cell (RBC) counts in the centrifuged sediments of less than 20/high-power (×400) field (HPF) on microscope.

Group II (n=63, 25 males and 38 females); negative urinary protein with urinary RBC counts of more than 20/HPF.

Group III (n=36, 21 males and 15 females); positive urinary protein with urinary RBC counts of less 20/HPF.

Group IV (n=31, 14 males and 17 females); positive urinary protein with urinary RBC counts of more than 20/HPF.

The patients in each of group were further subdivided according to urinary RBC counts of more or less than 5/HPF, which is considered generally as the normal limit. In group I, the patients with urinary RBC counts of less than 5/HPF were further subdivided into subgroup I′. In group II, the patients with a urinary RBC too high be counted (designated as numerous in the figures) were subdivided into subgroup II′. In group III, the patients with urinary RBC counts of less than 5/HPF were subdivided into subgroup III′. In group IV, the patients with a high urinary RBC in the sediments were subdivided into subgroup IV′. Thus, subgroup I′ is considered to be an almost normal urine group, while subgroup IV′ is the most severe urine group.

Biopsy-proved chronic glomerulonephritis (GN)

27 patients aged 6 to 18 (14 males and 13 females) with chronic GN and biopsy-proved renal glomerular lesions other than the above subjects were included in this study. These 27 patients had urinary findings ranging from those in groups II to IV described above. Their BUN, serum creatinine, and CCr were all within normal range. These renal glomerular lesions were classified according to the degree of mesangial cell proliferations, based on a table by one of our colleagues who had not been informed of patients' data concerning urinary findings and urinary sodium excretions.

The classifications of the patients were as follows;

1 minimal change GN (n=5, 4 males and 1 females)

2-A-a mild proliferative GN (n=14, 8 males and 6 females)

2-A-b moderate proliferative GN (n=3, 1 male and 2 females)
4 membranoproliferative GN (MPGN, n=5, 1 male and 4 females)

Study protocol (Fig. 1)

The protocol of the study is shown in figure 1. Hereafter, the following abbreviations are used;

FVM-urine: first-voided morning urine, indicating the urine taken after sustained recumbency (sleep) at home.

OPC-urine: urine voided at the outpatient clinic (OPC) of our hospital, indicating the urine taken after orthostatic loading; standing, walking, sitting, or routine daily activities in an upright position, which are all done when "visiting a hospital".

All 246 subjects were instructed by the physicians not to restrict dietary salts, so the sodium intake was considered to be comparable for statistical analysis using a large number of the subjects in a localized area in Japan.

An individual patient was instructed to empty his/her bladder before going to bed at night prior to visiting the hospital. FVM-urine was obtained between 6:00 am and 8:00 am at home, then he or she visited our hospital, and venous blood and urine samples were taken simultaneously to measure serum sodium (SNa), creatinine (SCr), urinary sodium (UNa), and creatinine (UCr).

Assesment of urinay sodium excretion

Using above obtained values, the following indices were calculated;

$$\text{Fractional sodium excretion rate (FENa)} = \frac{\text{UNa}}{\text{SNa}} / \frac{\text{UCr}}{\text{SCr}} \times 100\%$$

Urinary Na/creatinine ratio (UNa/UCr) = UNa (mEq/L) /UCr (mg/L)

Relation between FENa and UNa/UCr

RESULTS

In the following data, ± values represent the mean ± SD.

Relation between FENa and urinary findings (Fig. 2)

FENa in subgroup I′, the subgroup with almost normal urinary findings, was 1.19± 0.50%, and statistically significant decreases were noted as the urinary findings became more severe in the II′, III′, III, IV, and IV′ groups.

Relation between the ratio of UNa/UCr in OPC-urine to UNa/UCr in FVM-urine and urinary fidings (Fig. 3)

The ratio in subgroup I′ was 1.95±1.54, and statistically significant decreases were noted as the urinary findings become more severe in the III′, III, and IV′ groups.

Relation between the ratio of UNa/UCr in OPC-urine to UNa/UCr in FVM-urine and glomerular mesangial cell proliferations in chronic GN (Fig. 4)

In patients with minimal glomerular lesions (changes), UNa/UCr in OPC-urine was 2.66-fold higher than they were in FVM-urine. These ratios decreased as the glome-

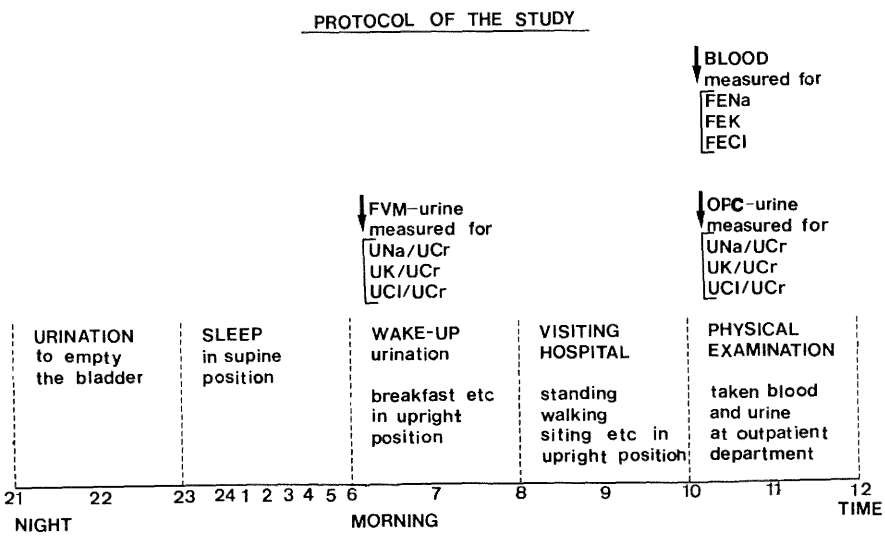

Fig. 1. Protocol of the study
All subjects were instructed to empty the bladder before going to bed. Immediately after wake-up, they took the first-voided morning urine, had breakfast, and visited our hospital. At outpatient clinic they were taken simultaneously urine and blood samples.

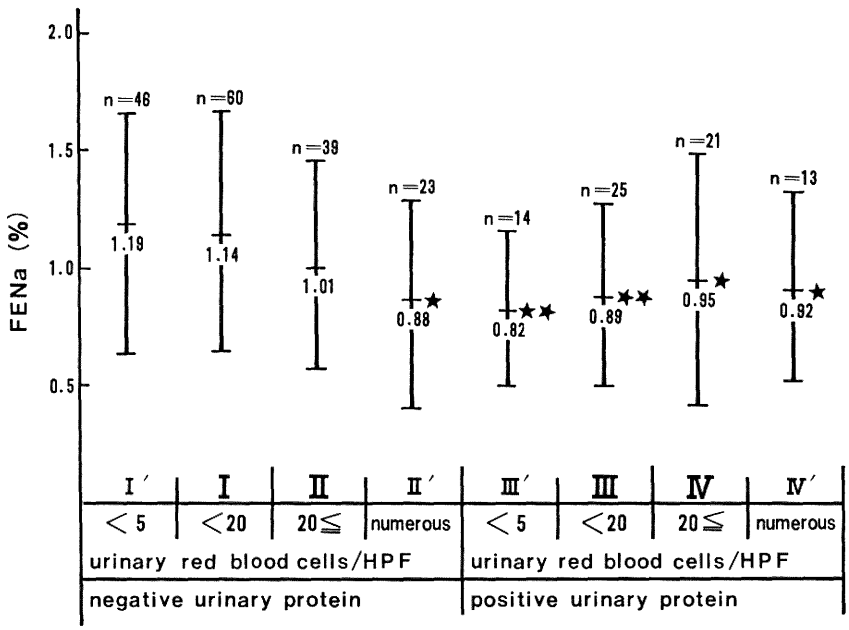

Fig. 2. Relation between FENa measured at outpatient clinic and urinary findings
FENa in subgroup I′ with almost normal urinary findings was 1.19 ± 0.50 % ($m \pm SD$), to which statistically significant decreases were noted as the urinary findings becoming severe in II′, III′, III, IV, and IV′ groups.
(*represents p values: $*0.05 < p < 0.10$, $**0.01 < 6 < 0.05$)

Table 1. Classification of Renal Glomerular Lesions

1	normal or minimal change GN
2-A-a	mild proliferative GN
2-A-b	moderate proliferative GN
2-A-c	severe proliferative GN
2B	proliferative GN with focal crescents
2C	proliferative GN with generalized crescents
3	membranous nephropathy
4	membranoproliferative GN
5	focal GN
6	too advanced to be classified
7	unclassified

mular mesangial proliferations become more severe; 1.44-fold in mild proliferative GN, 1.08-fold in moderate proliferative GN. In MPGN, UNa/UCr in OPC-urine taken after standing and walking were lower than they were in FVM-urine obtained at home. The ratio was inverted to 0.78 as shown is Fig. 4, which limplied the inhibitory effects of standing, walking, or routine daily activities in an upright position on sodium excretion in those patients with proliferated mesangial cells; these were the most conspicuous findings.

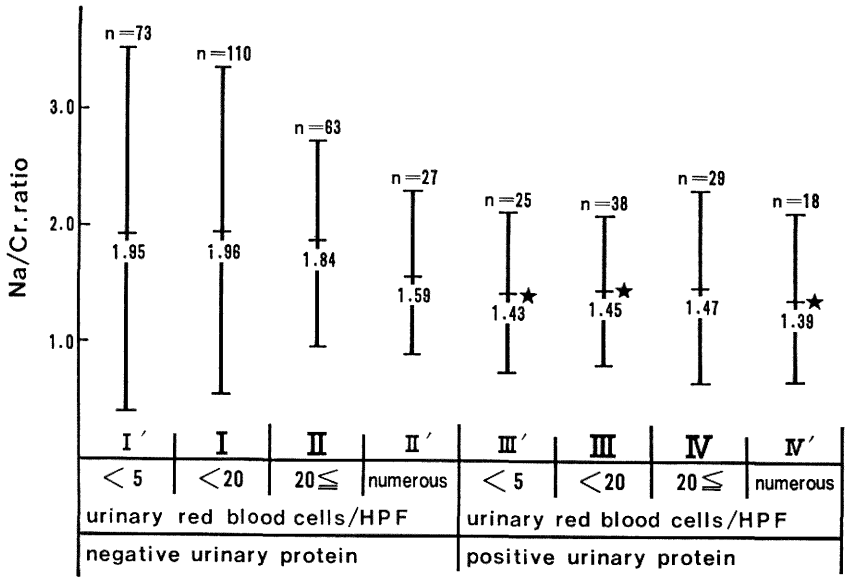

Fig. 3. Relation between the ratio of UNa/UCr in OPC-urine to UNa/UCr in FVM-urine and urinary findings
The ratio in subgroup I′ with almost normal urinary findings was 1.95 ± 1.54 ($m \pm SD$), to which statistically significant decreases were noted as the urinary findings becoming severe in III′, III, and IV′ groups.
(*represents $0.05 < p < 0.10$)

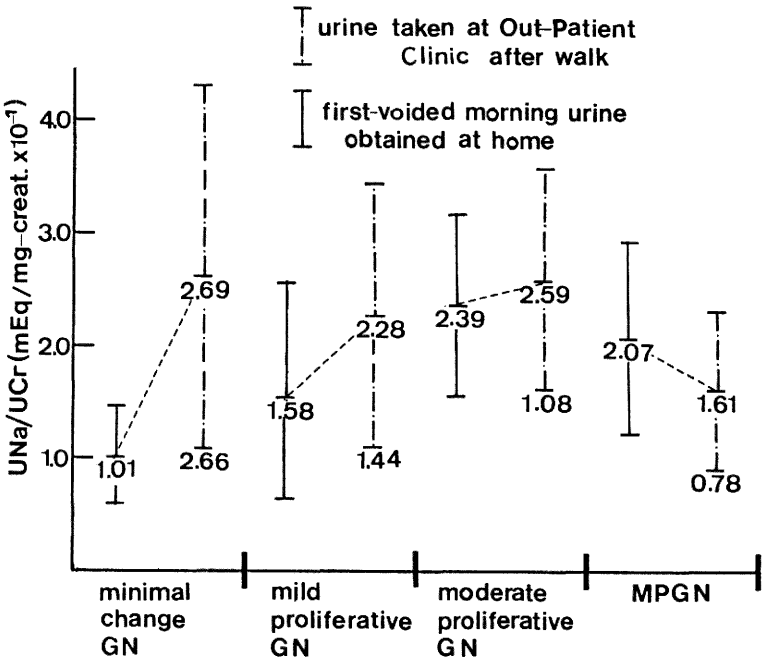

Fig. 4. Relation between the ratio of UNa/UCr in OPC-urine to UNa/UHr in FVM-urine and glomerular mesangial proliferations in chronic GN
In patients with minimal change GN UNa/UCr in OPC-urine was 2.66-fold higher than those in FVM-urine. These ratio were more decreased as the glomerular mesangial proliferations becoming severe, 1.44-fold in mild proliferative GN, 1.08-fold in moderate proliferative GN. These ratio become inversed to 0.78 in MPGN. UNa/UCr in FVM-urine was higher in patients with mild proliferative GN ($0.01<p<0.05$), moderate proliferative GN ($p<0.001$), and MPGN ($0.001<p<0.01$) respectively than in those with minimal change GN.

In contrast to the ratios in OPC-urine, UNa/UCr in FVM-urine increased as the mesangial cell proliferations changed from mild to moderate to severe, and MPGN. The sum total of UNa/UCr in FVM-urine and OPC-urine was 3.70 (1.01+2.69) in minimal change GN and 3.68 (2.07+1.61) in MPGN, respectively; this showed no difference in total daily urinary Na excretion, and implied that increases of urinary Na excretion in supine position compensated for the Na retention in upright position.

Relation between FENa and UNa/UCr (Fig. 5)

A significant correlation was noted between FENa and UNa/UCr ($r=0.92$, $n=19$, $p<0.001$) as shown in Fig. 5.

Discussion

Morphological studies have demonstrated fine structures of smooth muscle fibers in the cytoplasma of mesangial cells.[12,13] Cultures of adult human glomeruli were observed by Bernik[14] using microcinematography, and contractile activities of human glomerul

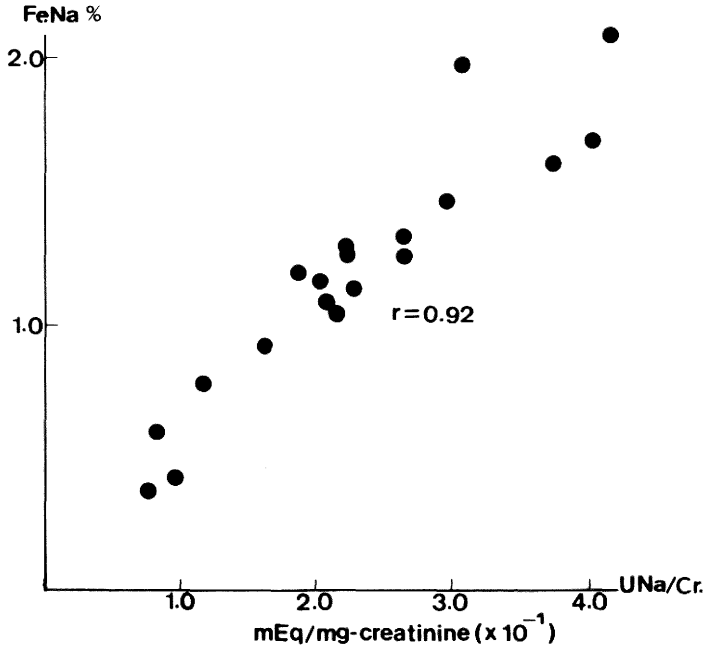

Fig. 5. Correlation between FENa and UNa/UCr
A significant correlation was noted between FENa and UNa/UCr ($r=0.92$, $n=19$, $p<0.001$).

were shown. Becker[15] demonstrated actomyosin in mesangial cells. In rat experiments, Blantz[16] showed the effects of angiotensin II upon the glomerular microcirculation and ultrafiltration coefficient. In Blantz's study,[16] contraction of mesangial cells was induced by stimulation of angiotensin II. Contraction of mesangialc cells by stimulation of arginine vasopressin and angiotensin II was visualized for the first time by Ausiello[5] and considered to be important in the regulation of glomerular filtration.

Martino[17] reported that the amount of muscle fibers similar to those seen in smooth muscle cells were higher in mesangial cells from patients with mesangial prliferative GN than in those from normal subjects. Furtermore, actomyosin normally localized in mesangium was shown to have wider distributions in mesangial cells from patients with glomerulonephritis compared with from normal subjects.[18]

Ultrastructural studies have revealed that glomerular endothelial and visceral epithelial cells also possess contractile elements in the cytoplasma.[19,20] However, both Ausiello[5] and Mahieu[4] found that whereas cultured mesangial cells exhibited a contractile response when exposed to physiologic concentrations of angiotensin II, ADH, or norepinephrinte, no response was elicited in cultured epithelial cells exposed to these same hormones. Therefore, at present, the available evidence is most consistent with view that the mesangial cells are the predominant regulators of glomerular capillary surface area and, thereby, of filtration coefficient.[6]

The influence of posture on kidney functions has been reported in healthy subjects[21,22] and ascribed to changes in renal hemodynamics. In an upright position, hydrostatic pressure of the lower vessels below the heart increases and extravasation of plasma components other than protein occurs,[22] which leads to increases in hematocrit, plasma protein concentrations and decreases in circulating volume.[24] In these processes, rises in plasma renin-angitensin-aldosterone concentrations have been reported.[7~10]

As decribed above, mesangial cells are now considered to be stimulated by angiotensin II to contract and reduce the glomerular capillary surface area for filtration, and an injection of low-dose angiotensin II has been reported to decrease glomerular filtration coefficient.[25] Our study showed that in patients with moderate to severe mesangial proliferative GN, including MPGN, urinary Na excretions were significantly further decreased upon standing and walking than in minimal change GN, and we found silificantly greater decreases in urinary Na excretions induced by walking in severe chance proteinuria and/or hematuria groups may also support the possible contribution of proliferated mesangial cells to the regulation of urinary Na, because a nation-wide study of 172 children with chance proteinuria and/or hematuria conducted in Japan[26] revealed that glomerular lesions, especially mesangial cell proliferation, in biopsied specimens are more frequent and more severe in patients with severe urinary findings than in those with only slight hematuria.

Orita[27] et al reported that in patients with mesangial proliferative GN, changing the position from supine to upine to upright for two hours induced significant greater decreases in creatinine clearance, FENa, and urinary Na excretion than in healthy subjects, followed by increases in tubular Na reabsorption, serum and plasma aldosterone concentration.

In the patients with severe mesangial proliferative GN and MPGN, the transient retention of Na induced by orthostatic positional change was found to be compensated by the increased urinary excretion in supine position during sleep, as reflected in increased urinary Na excretion in FVM-urine. Therefore clinically neither weight gain nor fluid retention occurs in these patients who have normal conventional renal function test results. However, latently inpaired renal functions seem to be reflected in greater decreases of urinary Na excretion in the upright position in these patients than in those with minimal change GN.

It seems that the two consecutive urine specimens, the first taken in the morning and the second taken at outpatient clinic after walking, may aid inestimating presence and severity of glomerular mesangial cell proliferations and latently impaired renal function without using any invasive methods.

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