Article

Effects of Soy Protein Isolate Feeding on Severe Kidney Damage in DOCA Salt-Treated Obese Zucker Rats

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ABSTRACT: This study assessed the effects of soy protein isolate (SPI) on severe kidney damage in deoxycorticosterone acetate (DOCA) salt-treated obese Zucker rats. These rats underwent heminephrectomy and were fed either casein or SPI diet for 12 weeks. From weeks 8 to 10 of the experiment, kidney damage was induced by biweekly injection of 25 mg/kg DOCA and administration of 0.5% NaCl (w/v) ad libitum. Urinary protein and N-acetyl-β-D-glucosaminidase excretions of SPI rats were much lower than those of casein rats at weeks 1 ($p < 0.01$) and 2 ($p < 0.05$) after DOCA treatment. Abnormal mineral excretions induced by DOCA treatment in casein rats were hardly detected in SPI rats. Severe atrophy of tubular epithelium and some flattened/detached renal tubules were also observed in casein rats, but not in SPI rats. These results indicate that consecutive treatment of SPI protects against renal dysfunction, particularly tubulointerstitial nephritis, in DOCA salt-treated obese Zucker rats.

KEYWORDS: soy protein isolate, renal dysfunction, obese Zucker rat, deoxycorticosterone acetate, casein, metabolic syndrome

INTRODUCTION

The physiological importance of soy protein isolate (SPI) as a source of protein in the body has been established. The U.S. Food and Drug Administration approved a health claim showing that 25 g of SPI intake reduces the risk of heart failure.[1] Such a reduction induced by SPI is caused by its effects on decreases in serum cholesterol and triglyceride levels.[2] It is suggested that the hypocholesterolemic effect of SPI is mainly due to the high bile acid-binding capacity of the polypeptides contained in SPI and the inhibition of reabsorption of bile acid.[3−5] Moreover, authors have documented that the cholesterol-lowering effect of SPI is related to the up-regulation of 63 genes and the down-regulation of 57 genes in the liver that regulate lipid metabolism, antisuperoxide activities, and energy metabolism, although the administration of casein did not result in any significant changes in such genes as measured by a DNA microarray technique.[6] From this perspective, SPI causes increases in the genes that regulate de novo cholesterol synthesis and steroid catabolism and decreases in the genes that regulate fatty acid synthesis in lipid metabolism. Nagasawa et al. also reported that the administration of SPI, but not casein, induces decreases in plasma glucose and plasma free fatty acid levels and increases in plasma adiponectin levels in KK[Ay] mice.[7] In rats, the administration of SPI decreased the appearance of PAI-1 in the adipose tissue and increased plasma adiponectin levels.[8] In addition, the appearance of SREBP-1c in the liver was inhibited by the administration of SPI.[8] These results indicate that the administration of SPI not only causes physical effects such as inhibition of reabsorption of bile acid but also regulates lipid and glucose levels in the blood through direct actions on adipose tissue, the liver, and so on.

It has also been proposed that obesity and diabetes mellitus induce renal dysfunction, such as chronic kidney disease and severe renal damage.[9,10] Therefore, proper treatment and prevention of renal dysfunction induced by metabolic syndrome must be established as soon as possible. As suggested, SPI results in improved lipid and glucose metabolism; therefore, treatment can be focused on the preventative effects of SPI on renal dysfunction induced by metabolic syndrome.

In this paper, we evaluated the effects of consecutive feeding of SPI on severe renal dysfunction induced by obesity and diabetes mellitus by treatment with deoxycorticosterone acetate (DOCA) salt in Zucker (fa/fa) rats,[11] which display abnormal lipid metabolism and hyperinsulinemia due to the loss of leptin receptors, resulting in mild renal dysfunction.

MATERIALS AND METHODS

Animals. Twenty-four male obese Zucker (fa/fa) rats were obtained at 5 weeks of age (Charles River Japan, Tokyo, Japan) and were housed individually in stainless steel cages under controlled conditions (temperature, 23 ± 1 °C; humidity, 55 ± 5%; 12:12 h light/dark cycle). Rats were allowed to acclimate to commercial chow (CRF-1; Oriental Yeast Co., Tokyo, Japan) for 1 week prior to entry into this experimental study. Vitamin-free casein (Oriental Yeast Co.) and SPI (Fujipro; Fuji Oil Co., Osaka, Japan) were provided as dietary protein sources. Experimental diets were based on the AIN-93G formula[12] (see Table 1). All animals were treated in accordance with the guidelines established by the Japanese Society of Nutrition and

Table 1. Compositions of Experimental Diets

	groups	
	casein (g)	SPI (g)
casein[a]	22.7	
SPI[b]		23.1
β-cornstarch	36.40	36.95
α-cornstarch	13.2	13.2
sucrose	10.0	10.0
soybean oil[c]	7.0	7.0
cellulose powder	5.0	5.0
mineral mixture[d]	3.5	3.5
vitamin mixture[e]	1.0	1.0
choline bitartrate	0.25	0.25
NaCl[f]	0.1	
NaHCO$_3$[f]	0.8	
total	100.0	100.0

[a]Crude protein/as is 89.1%. Oriental Yeast Co., Tokyo, Japan. [b]Crude protein/as is 86.2%. Fuji Oil Co., Osaka, Japan. [c]Soybean oil contains 0.02% tert-butylhydro-oxyquinone. [d]AIN-93G mixtures, Oriental Yeast Co., Tokyo, Japan. [e]AIN-93 mixtures, Oriental Yeast Co., Tokyo, Japan. [f]To adjust Na content in casein and SPI diets, NaCl and NaHCO$_3$ are added. The percentages of Na content in casein and SPI diet are 0.36 and 0.37%, respectively.

Food Science (Law No. 105 and Notification No. 6 of the Japanese government).

Experimental Procedures. After acclimation to commercial chow for 1 week, the right kidneys of all 6-week-old obese Zucker rats were unilaterally nephrectomized under anesthetic conditions using a gas anesthesia system for small animals (Dainippon Sumitomo Pharma, Tokyo, Japan). After a 10 day recovery period, all nephrectomized rats were randomly divided into two groups of similar average body weights and urinary protein excretion. One group of obese Zucker rats was fed a casein diet (casein rats), and the other group was fed an SPI diet (SPI rats) for 12 weeks. The experimental diets (each containing 20% protein) and water were given ad libitum throughout this study. Food intake was recorded daily, and body weight was measured twice a week. In this experiment, additionally, the mean intake volume of each diet was 26 g/rat/day, and no significant difference of this intake volume between casein and SPI rats was detected. This dosage has

been designed and calculated from our previously reported study.[6] Significant differences of body weight between casein and SPI rats were not seen (data not shown).

DOCA Treatment. From weeks 8 to 10 of the experimental period, severe renal damage was induced in half of the casein and SPI rats by biweekly injection of DOCA (25 mg/kg body weight) suspended in corn oil. During the DOCA treatment period, all rats were given 0.5% NaCl (w/v) ad libitum. After the DOCA treatment period, the rats recovered for 2 weeks. There were no differences in food intake or body weight gain between casein and SPI rats (data not shown).

At week 12, blood was drawn from the abdominal aorta of nonfasted rats into a heparinized syringe under painless conditions. Plasma was separated by centrifugation (3000g, 10 min, 4 °C) and stored at −80 °C until analysis. Kidneys were taken and stored at −80 °C.

Blood Plasma and Urine Analysis. Rats were placed in metabolic cages on the first day of weeks 2, 4, 8, 9, and 10 after either casein or SPI feeding, and urine samples were then collected. Total volumes of urine and food intake were recorded for 24 h on each first day.

Urine samples were centrifuged at 3000g for 10 min at 4 °C, and supernatants were stocked at −80 °C until analysis. Urinary protein excretion was determined according to the CBBG method (Tonein-TP; Otsuka Pharmaceutical Co., Ltd., Tokushima, Japan). The urinary creatinine level was measured by enzymatic methods (7180 autoanalyzer; Hitachi Ltd., Tokyo, Japan). N-Acetyl-β-D-glucosaminidase (NAG) was measured by the colorimetry method (NAG Test Shionogi; Shionogi & Co., Ltd., Osaka, Japan). Blood plasma and urinary P, Na, Cl, and K levels were automatically measured by enzymatic methods (Dri-Chem; Fujifilm Co., Ltd., Tokyo, Japan).

Histology and Image Analysis. Frozen kidneys were embedded in OCT compound. Sections were cut into 2 μm thick sections and stained with hematoxylin and eosin or periodic acid−Schiff. Histological evaluations were performed using light microscopy. Slides were then examined for hyperplasia of the glomerular matrix mesangium, thickness of the glomerular basement membrane, and atrophy and flattening of the tubular epithelium. The extent of each progression was graded as follows: −, normal; +, mild increase; ++, moderate increase; and +++, marked increase.

Statistical Analysis. Data were expressed as the mean ± SEM. Statistical analysis was performed by SPSS software (12.0J; SPSS Inc., Chicago, IL, USA). Data were first analyzed for distribution. The results were then analyzed by the unpaired Student's t test. A level of p < 0.05 was accepted as statistically significant.
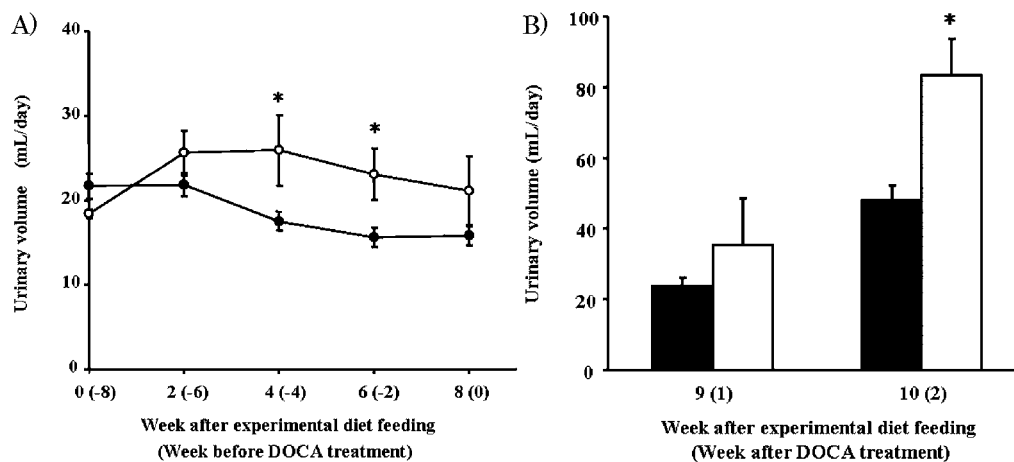

Figure 1. Changes in urinary volume during the experimental diet feedings (A) before and (B) after DOCA treatment. (A) The numbers in parentheses along the X-axis show weeks before DOCA treatment. Each value shows the mean ± SE of 12 rats. The black circle shows the casein rats. The white circle shows the SPI rats. ∗, significant differences between casein and SPI rats at the same time point, p < 0.05. (B) The numbers in parentheses along the X-axis show weeks after DOCA treatment. Each value shows the mean ± SE of 5−6 rats. The black column shows the casein rats. The white column shows the SPI rats. ∗, significant differences between casein and SPI rats at the same time point; p < 0.05.
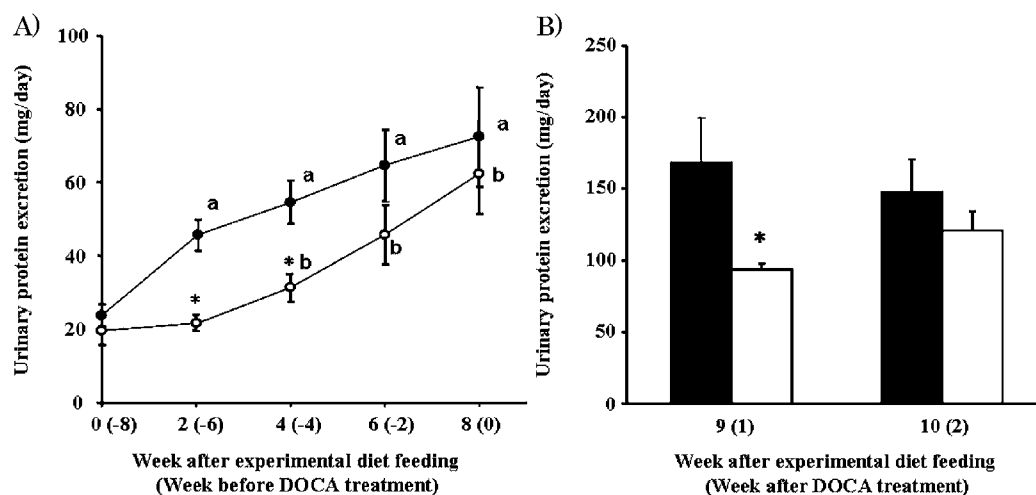
Figure 2. Changes in urinary protein excretion during the experimental diet feedings (A) before and (B) after DOCA treatment. (A) The numbers in parentheses along the X-axis show weeks before DOCA treatment. Each value shows the mean ± SE of 12 rats. The black circle shows the casein rats. The white circle shows the SPI rats. ∗, significant differences between casein and SPI rats at the same time point, $p < 0.05$; (a) Significant differences between the start point (0) and each point after experimental diet feeding in the casein rats, $p < 0.05$; (b) significant differences between the start point (0) and each point after experimental diet feeding in the SPI rats, $p < 0.05$. (B) Each value shows the mean ± SE of 5−6 rats. The black column shows the casein rats. The white column shows the SPI rats. ∗, significant differences between casein and SPI rats at the same time point, $p < 0.05$. (B) The numbers in parentheses along the X-axis show weeks after DOCA treatment. Each value shows the mean ± SE of 5−6 rats. The black column shows the casein rats. The white column shows the SPI rats. ∗, significant differences between casein and SPI rats at the same time point, $p < 0.05$.
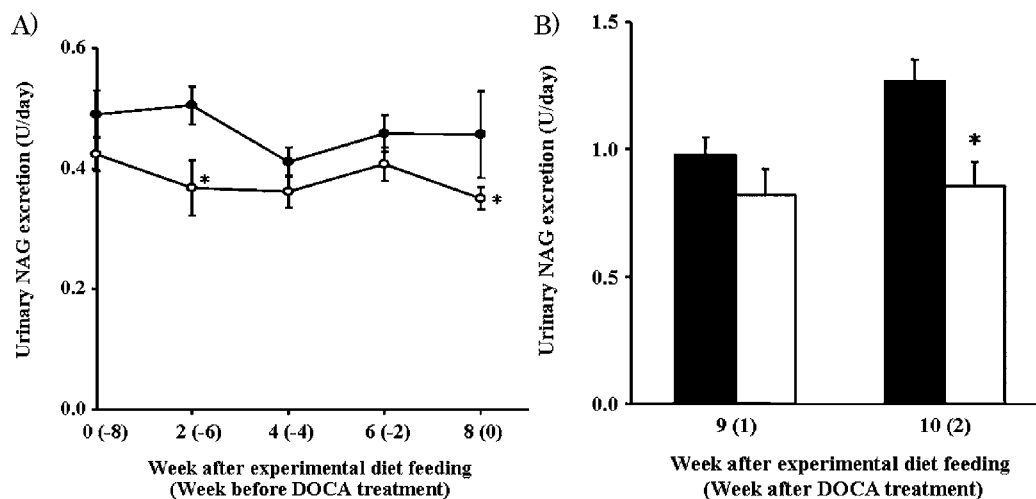

Figure 3. Changes in urinary NAG excretion during the experimental diet feedings (A) before and (B) after DOCA treatment. (A) The numbers in parentheses along the X-axis show weeks before DOCA treatment. Each value shows the mean ± SE of 12 rats. The black circle shows the casein rats. The white circle shows the SPI rats. ∗, significant differences between casein and SPI rats at the same time point, $p < 0.05$. (B) The numbers in parentheses along the X-axis show weeks after DOCA treatment. Each value shows the mean ± SE of 5−6 rats. The black column shows the casein rats. The white column shows the SPI rats. ∗, significant differences between casein and SPI rats at the same time point, $p < 0.05$.

■ RESULTS

Urinary Volume and Proteinuria. In casein rats, no differences in urinary volume were observed during the 8 weeks before DOCA treatment (Figure 1A). In contrast, the urinary volumes of SPI rats were significantly increased compared with those in the initial stage of this experiment (Figure 1A). The urinary volumes between casein and SPI rats showed significant differences at weeks 4 and 6 after feeding of the experimental diet (Figure 1A). The urinary volumes of SPI rats at weeks 1 (35.38 ± 13.17 mL/day) and 2 (83.33 ± 10.25 mL/day) after DOCA administration were much higher than those (18.40 ± 0.48 and 25.90 ± 2.60 mL/day, respectively) during the 8 weeks before DOCA administration (Figure 1). The urinary

volumes of SPI rats were much higher than those of casein rats, and there were significant differences in urinary volume between SPI (83.33 ± 10.25 mL/day) and casein (48.13 ± 4.08 mL/day) rats at week 2 after DOCA treatment ($p < 0.05$) (Figure 1B).

A significant progressive increase in urinary protein excretion was observed in both groups during the 8 weeks before DOCA treatment (Figure 2A). Urinary protein excretion was much lower in SPI rats than in casein rats throughout the period before DOCA treatment (Figure 2A). In particular, significant differences in urinary protein excretion between SPI and casein rats were seen at weeks 2 and 4 after the experimental diet feedings ($p < 0.05$) (Figure 2A). Figure 2B shows the urinary

protein excretion in rats at weeks 1 and 2 after DOCA administration. In casein and SPI rats, the urinary protein excretions at weeks 1 (casein, 168.1 ± 31.5 mg/day; SPI, 93.6 ± 4.5 mg/day) and 2 (casein, 147.5 ± 23.2 mg/day; SPI, 120.9 ± 13.3 mg/day) after DOCA treatment were significantly higher than those (casein, 23.8 ± 3.0 and 72.4 ± 13.5 mg/day, respectively; SPI, 19.7 ± 3.9 and 62.3 ± 10.8 mg/day, respectively) obtained during the non-DOCA treatment period (Figure 2). The urinary protein excretion of SPI rats was much lower than that of casein rats at week 1 after DOCA treatment ($p < 0.01$) (Figure 2B).

Renal Tubular Integrity: Urinary Excretion of NAG and Minerals. Mild increases in urinary NAG excretions were observed in both casein and SPI rats at the beginning of this study (Figure 3A). Continuous urinary NAG excretion was observed in casein rats throughout the experiment. In contrast, urinary NAG excretion in SPI rats gradually decreased compared with that in the initial stage of this experiment (Figure 3A). There were significant differences in urinary NAG excretion between SPI and casein rats at weeks 2 and 8 after both experimental diet feedings ($p < 0.01$) (Figure 3A). Figure 3B shows the urinary NAG excretion in rats at weeks 1 and 2 after DOCA administration. In casein and SPI rats, the urinary NAG excretions at weeks 1 (casein, 0.98 ± 0.07 U/day; SPI, 0.82 ± 0.10 U/day) and 2 (casein, 1.27 ± 0.09 U/day; SPI, 0.85 ± 0.10 U/day) after DOCA treatment were significantly higher than those (casein, 0.41 ± 0.02 and 0.50 ± 0.03 U/day, respectively; SPI, 0.35 ± 0.02 and 0.42 ± 0.03 U/day, respectively) obtained during the non-DOCA treatment period (Figure 3). Urinary NAG excretion was much lower in SPI rats than in casein rats at week 2 after DOCA treatment ($p < 0.05$) (Figure 3B).

Tables 2 and 3 show the changes in mineral excretions in urine and blood plasma, respectively, after DOCA treatment. Before DOCA treatment, urinary P, Na, and K levels in SPI rats were much higher than those in casein rats ($p < 0.05$) (Table 2). In casein rats, urinary P, Na, Cl, and K levels were significantly increased by DOCA administration ($p < 0.05$ and $p < 0.01$) (Table 2). In SPI rats, the increases in urinary P and K excretion induced by DOCA administration were blocked. The ratio of Na/K in SPI rats at week 2 after DOCA administration was significantly higher than that in casein rats ($p < 0.05$). Furthermore, in casein rats, significant differences of Na and Cl levels in blood plasma were not seen compared to those in SPI rats (Table 3). However, P and K levels in the blood plasma of casein rats were significantly lower than those of SPI rats (Table 3).

Histology and Image Analysis. In the DOCA-treated rats, enlargement of the kidneys was observed compared with the non-DOCA-treated rats (data not shown). In addition, the enlargement of the kidneys induced by DOCA treatment in SPI rats was smaller than that in casein rats (data not shown). Table 4 shows the average scores of hyperplasia of the glomerular matrix mesangium, the thickness of the glomerular basement membrane, and atrophy and flattening of the renal tubules in the DOCA-treated rats. In casein rats, slight hyperplasia of the glomerular matrix mesangium and mild thickening of the glomerular basement membrane were observed. Severe atrophy of the tubular epithelium and some flattened/detached tubules were also observed in casein rats (Figure 4). In contrast, most of these abnormalities were not detected in SPI rats (Table 4 and Figure 4).

Table 2. Changes in Mineral Excretions during the Experimental Diet Feedings before and after DOCA Treatment

		group[a]	
		casein	SPI
Na	before DOCA treatment	2.03 ± 0.09	2.80 ± 0.13 **
(mequiv/day)	1 week after DOCA treatment	4.62 ± 0.39 ##	6.80 ± 1.27 #
	2 weeks after DOCA treatment	5.71 ± 0.77 ##	7.03 ± 1.39 ##
Cl	before DOCA treatment	1.06 ± 0.07	1.35 ± 0.11
(mequiv/day)	1 week after DOCA treatment	3.71 ± 0.34 ##	5.23 ± 0.86 ##
	2 weeks after DOCA treatment	4.87 ± 0.58 ##	6.90 ± 1.12 ##
P	before DOCA treatment	2.43 ± 0.17	3.47 ± 0.33 *
(mg/day)	1 week after DOCA treatment	5.91 ± 0.81 ##	4.57 ± 0.71
	2 weeks after DOCA treatment	5.94 ± 0.66 ##	3.23 ± 0.74
K	before DOCA treatment	1.28 ± 0.07	1.73 ± 0.07 **
(mequiv/day)	1 week after DOCA treatment	2.33 ± 0.21 ##	2.12 ± 0.36
	2 weeks after DOCA treatment	1.63 ± 0.14 #	1.40 ± 0.21
Na/K	before DOCA treatment	1.62 ± 0.15	1.62 ± 0.03
	1 week after DOCA treatment	2.00 ± 0.08	3.36 ± 0.78 #
	2 weeks after DOCA treatment	3.49 ± 0.37 ##	4.95 ± 0.38 * ##

[a]Each value shows the mean ± SE of 6 rats. Significant differences between casein and SPI rats at the same time point: *, $p < 0.05$; **, $p < 0.01$. Significant differences between before DOCA treatment and after DOCA treatment in the same diet rats: #, $p < 0.05$; ##, $p < 0.01$.

Table 3. Changes in Plasma Mineral Levels Just after the Consecutive Feedings of Experimental Diets with DOCA Treatment

	group[a]	
	casein	SPI
Na (mequiv/L)	141.0 ± 0.6	140.0 ± 1.0
Cl (mequiv/L)	92.2 ± 2.0	95.0 ± 1.0
P (mg/day)	5.9 ± 0.1	6.5 ± 0.1 *
K (mequiv/L)	3.2 ± 0.2	4.2 ± 0.2 *
Na/K	45.2 ± 3.2	33.5 ± 2.6

[a]Each value shows the mean ± SE of 6 rats. Significant differences between casein and SPI rats at the same time point: *, $p < 0.05$.

DISCUSSION

In this paper, we showed that severe renal dysfunction, particularly renal tubulointerstitial nephritis, induced by DOCA treatment in obese rats was protected by consecutive treatment with SPI, but not with casein.

A recent paper suggested that progression of chronic renal disease is much more strongly related to the severity of tubulointerstitial disease than to the aggravation of glomerular

Table 4. Pathological Evaluation on Kidney of DOCA-Treated Zucker (fa/fa) Rat Fed either Casein- or SPI-Containing Diet

	group[a]	
	casein	SPI
hyperplasia of glomerular matrix mesanjiumu	++	+
thickness of glomerular basement membrane	++	−
atrophy of tubular epitheliuma	++	+
flattened tubular epitheliuma	+++	+

[a]The extent of each progression was graded as follows: −, normal; +, mild increase; ++, moderate increase; +++, marked increase.
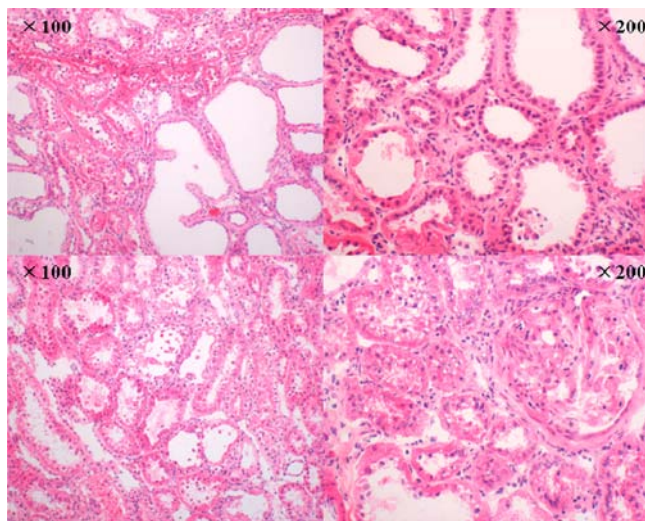

Figure 4. Microscopic analysis of kidney slices from DOCA-treated Zucker (fa/fa) rats fed either a casein- or SPI-containing diet: (upper left) casein-fed DOCA-treated rat (×100); (upper right) SPI-fed DOCA-treated rat (×100); (lower left) casein-fed DOCA-treated rat (×200); (lower right) SPI-fed DOCA-treated rat (×200).

dysfunction.[13] Therefore, the significance of protection of renal tubular function must be considered. It has been documented that SPI, a vegetal protein, protects against severe renal dysfunction much better than does casein, an animal protein, in animal studies.[14−16] A Mexican research group reported that SPI inhibits the production of proteinuria and the development of glomerular diseases in Zucker rats, which concurs with the renal dysfunction caused by lipogenous diabetes.[17] However, most research has targeted glomerular dysfunction, not tubulointerstitial disease. It has been suggested that Zucker rats are the suitable animal model for renal dysfunction caused by lipogenous diabetes, although the blood pressure of Zucker rats is not particularly high.[14] Few studies have evaluated the effects of SPI on the severe renal dysfunction induced by DOCA. It has been well documented that DOCA affects mineralocorticoid receptors and induces retention of the Na ion, causing hypertension and severe renal dysfunction with glomerular damage.[11,19,20] Furthermore, DOCA-treated rats show several types of remarkable kidney damage, including tubular dilatation, atrophy of tubular epithelial cells, and cast formation in the early stages of DOCA treatment.[11,19]

In this paper, we examined the effects of SPI on severe renal dysfunction with tubular integrity using DOCA-treated Zucker rats. The aim of this paper was to evaluate prophylaxis of SPI by administration of specific diets for 8 weeks before DOCA treatment. During the 8 week administration of SPI and casein

diets, the body weights of all animals significantly increased daily. Proteinuria increased linearly with growth, and the progression of renal dysfunction was accelerated. During this period, the SPI-treated rats, but not the casein-treated rats, showed significant inhibition of increases in proteinuria, although both the total volume of feeding and body weights were similar. In casein rats, significant increases in proteinuria induced by DOCA occurred, although these increases were blocked by the feeding of SPI. These results indicate the presence of prophylactic effects of SPI on renal dysfunction. Moreover, the volume of urine in both casein and SPI rats significantly increased after DOCA treatment. The increase in urine volume in SPI rats was much higher than that in casein rats. This increase in SPI rats may have been related to the decrease in proteinuria.

Casein rats showed slight increases in NAG, a marker of renal tubular damage, before DOCA treatment, although the level of NAG in SPI rats gradually decreased during the period of SPI feeding. The tubular function damage induced by DOCA was improved by the feeding of SPI. In the pathological analysis, abnormalities such as severe applanation of renal tubules were clearly detected in casein rats, but not SPI rats. These results suggest that the feeding of SPI protects against renal tubule damage induced by DOCA and maintains normal tubular function.

Minerals in the body may help to maintain many physiological functions. In particular, increased Na ion in the blood causes hypertension induced by renal dysfunction; thus, control of Na intake is important for nephropathy patients. In this experiment, excretion of Na and K ions in urea in SPI rats was higher than that in casein rats before DOCA treatment, although the Na/K ratio did not show any significant changes between SPI and casein rats. These results suggest that the increased excretion of Na and K ions induced by SPI is related to increased urine volume. After DOCA treatment, the excretion of Na, Cl, K, and P in casein rats was much higher than that before DOCA treatment, although these significant expressions, with the exception of Na and Cl ions, seen in SPI rats could not be detected. The increased excretion of Na and Cl ions in both rats was caused by salt loading, and the severely increased excretion of K and P ions seen in casein rats was caused by dysfunction of mineral reabsorption in the renal tubules. Meanwhile, Na and Cl levels in blood plasma did not show any significant differences between casein and SPI rats, although K and P levels in blood plasma of casein rats were much lower than those of SPI rats, significantly. These decreases were caused by the extraordinary increases of K and P excretions in urine induced by renal dysfunctions. These results indicate that the feeding of SPI protects against damage to tubular function induced by DOCA treatment and maintains the balance of Na/K ions, even with excessive treatment with NaCl solution.

In our previous paper, we measured the apparent absorption rate of SPI and casein in Zucker rats using the same method described in this paper. These results showed that the apparent absorption rate, which was calculated from the ratio of the intake of nitrogen and feces excretion of nitrogen, was between 93 and 94% in both SPI- and casein-treated Zucker rats.[6] Thus, the absorption rate between SPI and casein is not shown as the significant difference. Then the different effects of SPI and casein on DOCA-induced renal dysfunctions could not be explained by the absorption rates of SPI and casein. Because of the different effects of SPI and casein on renal dysfunction

induced by DOCA and lipogenous diabetes, the different contents of vegetal and animal proteins, such as β-conglycinin, glycine, lipophilic proteins associated with phospholipids, composed amino acids, phospholipids, isoflavone, and saponin, may affect these different findings obtained between casein and SPI feeding.[21] In particular, reduced cholesterol caused by the polypeptides in SPI, which comprise amino acids and isoflavones, has been documented.,[3−5,22,23] Furthermore, SPI has a large amount of Arg in its composed amino acids and increases the production of endothelial nitric oxide (NO).[17] These studies suggest that the inhibitory effects of SPI on renal dysfunction are caused by reduced cholesterol levels and increased NO production.[17,18,24]

This paper first documented that the feeding of SPI can inhibit increases in specific markers of tubular function damage in severe renal dysfunction. These effects cannot be fully explained by reduced cholesterol levels and increased NO production induced by SPI. In severe tubular dysfunction, the cell membrane and components of the tubular membrane might be damaged. The specific phospholipids contained in SPI may repair the damage of the tubular membrane because the main components of the cell membrane are phospholipids. By measurement of the primary phospholipid fraction, which was extracted by chloroform and methanol (2:1), the ratios of phospholipid of SPI and casein were 5 and 0.9%, respectively. Thus, the content of phospholipid of SPI is 5 times more than that of casein.

In conclusion, this paper demonstrated that vegetable proteins such as those in SPI can alleviate abnormal renal tubule function and increase proteinuria induced by lipogenous diabetes and DOCA treatment. These mechanisms must be evaluated in further studies. However, our results indicate that the feeding of SPI as opposed to casein leads to the development of complementary medicines to treat the decreased renal function that occurs with aging and metabolic syndrome.

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Notes
The authors declare no competing financial interest.

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