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Full Length Research Paper

The effects of combined vitamin C and E in streptozotocin-induced diabetic rat kidney

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Diabetic nephropathy is a serious complication of diabetes mellitus. Oxidative stress has been suggested to play a key role in the pathogenesis of diabetic nephropathy. Vitamins C and E play important roles in the antioxidant defense system. It is likely that both vitamins act in a synergistic manner, with vitamin E primarily being oxidized to the tocopheroxyl radical and then reduced back to tocopherol by vitamin C. The purpose of this study was to determine the effects of supplementation of vitamins C and E (VCE) on diabetic rat kidney. Adult female Wistar rats were used in the study. The animals were divided into three groups. Group I: the control group; Group II: diabetic group, streptozotocin (40 mg/kg) was administered group; Group III: diabetes+VCE group, received a diet containing a combination of ascorbic acid and di- -tocopheryl acetate per kg of feed. Rats were killed on day 21 and renal tissues were taken and fixed in 2.5% glutaraldehyde solution for electron microscopic examination. When compared with the control group, congestion of the glomerular capillaries, increased mesangial cells and distinct mesangium, shortened podocyte processes and disappearance of filtration slit pore of diabetic rat kidney were observed. In the group treated with VCE, glomerular changes were less distinct than the diabetic group. Lengths of the pedicles were similar to the control group. In conclusion, VCE reduced the changes in the glomerular structures due to diabetes.

Key words: Diabetes, kidney, vitamin C, vitamin E, ultrastructure.

INTRODUCTION

Diabetic nephropathy is one of the leading causes of chronic renal failure in the Western European countries, USA and Japan. Renal injury is observed in some 35% of patients with Type I and Type II diabetes. Renal disorders observed in Type I and Type II diabetes are similar. Long-term glycemia, genetic factors, race, sex and hypertension have been implicated in the development of diabetic nephropathy (Monhart, 2008; Rychlik, 2008). Diabetic nephropathy presents itself with ischemic nephropathy, nodular glomerulosclerosis and renal failure. Clinically, 30 - 300 mg/day or 20 - 200 µg/min microalbuminuria indicates diabetic nephropathy (Ritz, 2006). Diabetes is an important etiopathological factor in oxidative stress (Punithavatki et al., 2008). As a result of lipid and protein oxidation, the levels of superoxide dismutase (SOD), glutathione peroxidase (GSH-Px) and catalase (CAT) increase in kidneys (Prakasam et al., 2005; Je et al., 2001; Yıldırım and Büyükbingöl, 2003). Various studies have reported protective effects of antioxidants such as melatonin (Ökten et al., 2006), ginkgobiloba (Welt et al., 2007), Circumin (Murugan and Pari, 2006), Groundnut oil (Ramesh et al., 2006), taurin (Wang et al., 2008), herbal medications (Yokozawa et al., 2008), soybean oil (Sena et al., 2008), naringin, a flavo-noid glycoside which gives the bitter taste of grapefruit juice (Punithavatki et al., 2008), vitamin E (Minamiyama et al., 2008; Hamdy et al., 2008; Ruperez et al., 2008) and Vitamin C (Ruperez et al., 2008; Wu et al., 2007; Fadugpin et al., 2007) against oxidative damage of diabetes.

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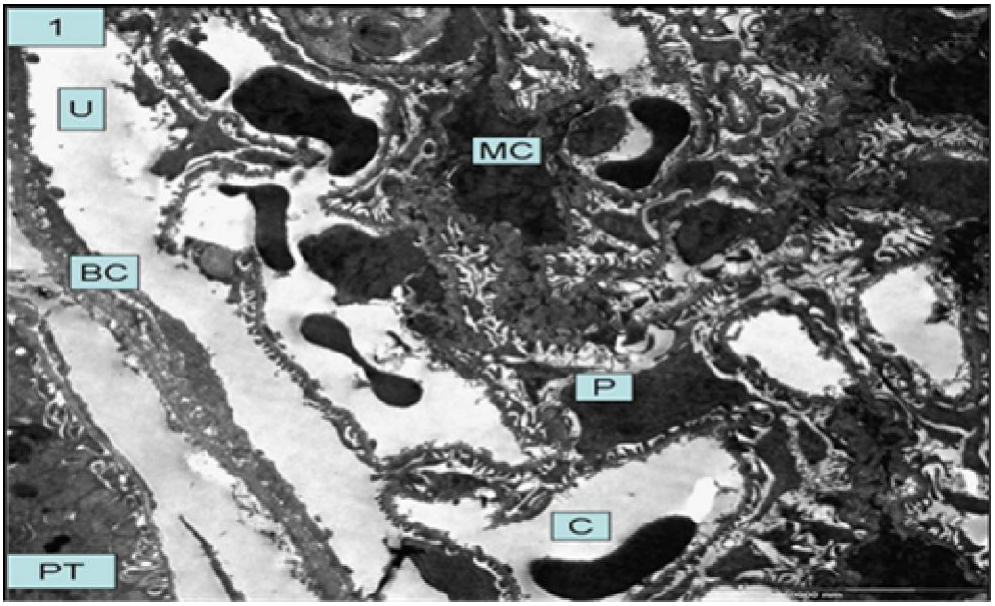

Figure 1. A glomerulus from the control group. Proximal tubule (PT), Bowman's capsule (BC), capillaries (C), podocyte (P), urinary space (U) and mesangial cells (MC).

The levels of Vitamin C and E in plasma and renal tissues is significantly reduced in diabetic patients (Wu et al., 2007; Kashiba et al., 2002; Peerapatdit et al., 2006). A decrease in Vitamin C causes hyperlipidemia and hypertension (Wu et al., 2007; Chen et al., 2005). Some studies showed that certain fruits and vegetables (Villegas et al., 2008) and that Vitamins C and E (Harding et al., 2008) are important to prevent or alleviate the complications of diabetes mellitus. have complication-reducing effects. Vitamins C and E not only reduce the risk of thromboembolism in patients with diabetes-related hypertension (Haidara et al., 2004) but also exert favorable effects on wound healing (Musalmah et al., 2005). Vitamins C and E have been shown to prevent the teratogenic effects in diabetic rats and autoimmunity of cells in babies (Uusitalo et al., 2008; Cederberq et al., 2005). Vitamins C and E can be used as antioxidants separately or in combination. Both vitamins act synergistically (Naziroglu et al., 2004; Kutlu et al., 2005). Majority of the studies demonstrated the antioxidant effects of Vitamins C and E while electron microscopic studies are scarce. In the present study, the effects of Vitamin C, a hydrophilic antioxidant and Vitamin E, a lypophilic antioxidant, on structural changes in renal tissue were investigated by feeding the experimental diabetes-induced rats with a combination of these vitamins.

MATERIALS AND METHODS

Fifteen adult female Wistar rats weighing 200 - 220 g were obtained from Experimental Research Center of Firat University Faculty of Medicine (FÜTDAM). The rats were housed at 22 - 24°C and were exposed to alternate cycles of 12 h light and darkness. All animal care and handling procedures conformed to the Guidelines set by the Association for Assessment and Accreditation of Laboratory Animal Care and approval was obtained from the Local Ethics Committee for Animal Studies. The animals were divided into three groups:

Group I: Control group (n = 5). Control rats were given intraperitoneal citrate buffer only (0.1 M, pH = 4.5).
Group II: Diabetic group (n = 5). Streptozotocin (STZ, Serva GmbH, Heidelberg, Germany) was administered intraperitoneally at a dose of 40 mg/kg body weight dissolved in citrate buffer (Naziroglu et al., 2004).
Group III: VCE group (n = 5) . Rats were fed with VCE (Vitamin C and E) supplemented diet for 15 days prior to induction of diabetes. The VCE-supplemented diet contained a combination of ascorbic acid (1 g) and di- -tocopheryl acetate (600 mg) per kg of feed.

VCE supplemented and unsupplemented food compositions were homogenized using a mixer and pellets were prepared in laboratory by heating below 45°C for 2 days. The VCE supplemented diet contained a combination of 1 g vitamin C (ascorbic acid, F. Hoffman La Roche, Istanbul, Turkey) and 600 mg vitamin E (di- -tocopheryl acetate, F. Hoffman La Roche, Istanbul, Turkey) per kg of feed (Naziroglu et al., 2004).

At the end of the experiment, renal tissues were taken under Rompun (5 mg/kg) and Ketamin (60 mg/kg) anesthesia. Renal tissues were fixed in 2.5% glutaraldehyde in 0.1 M sodium phosphate buffer and postfixed with 2% osmium tetraoxide in sodium phosphate buffer. Dehydration was accomplished by gradual ethanol series and tissues were embedded in epoxy resin. Ultrathin sections were stained with Uranly acetate and lead citrate. Sections were then viewed and photographed with a Zeiss 9EM.

RESULTS

Thin sections of the renal proximal tubules and glomeruli of the control group looked normal. Podocytes and cytoplasmic extensions, infiltration slits were evenly

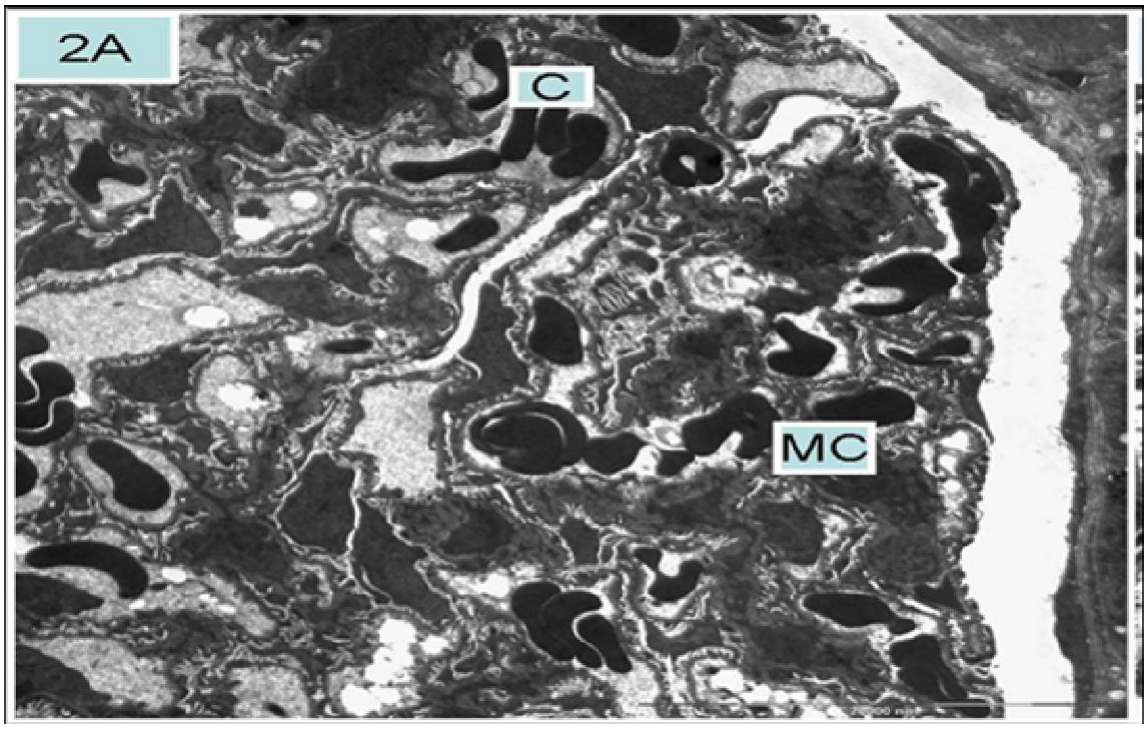

Figure 2a. STZ-induced diabetic rat kidney; Capillary congestion and increase in mesangial cells can be observed.

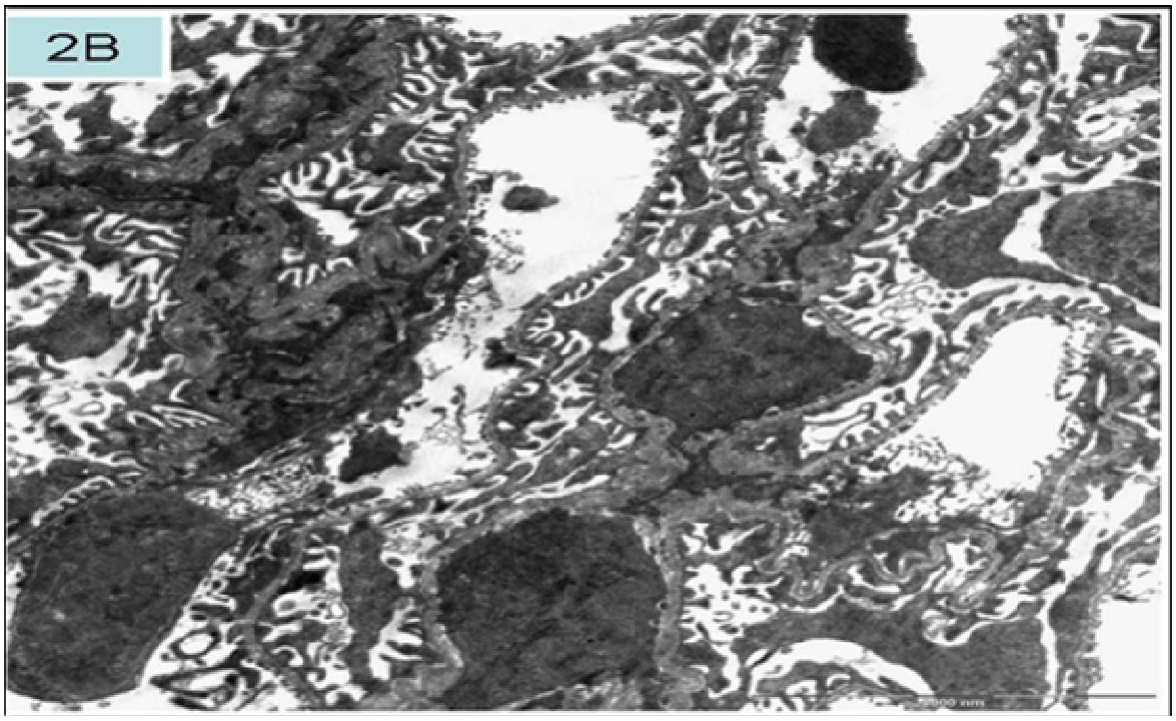

Figure 2b. STZ-induced diabetic rat kidney; Occasional basement membrane thickenings can be observed under higher magnification (arrow).

distributed (Figure 1). Capillary congestion, increase in the number of mesangial cells and shortened podocyte processes were observed in the renal glomeruli secondary to STZ. Basement membrane was thickened in

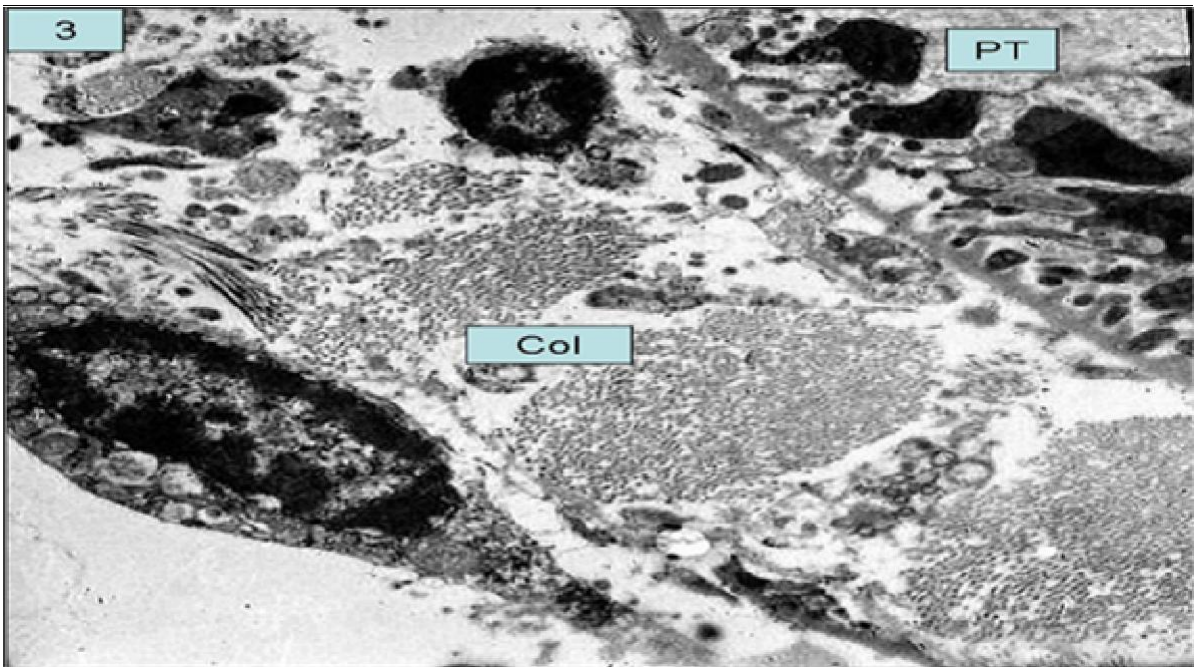

Figure 3. STZ-induced diabetic rat kidney. Irregular basal invaginations in some of the proximal tubules (PT) and peritubular heavy clusters of collagen fibers (Col) can be observed.

Figure 4a. Comparison of podocyte extensions and infiltration slits; Control group.

certain regions (Figures 2a and 2b). The invaginations on the basal regions of the proximal tubules were irregular ad collagen fibers were clustered in the areas between the tubules (Figure 3). Compared to the control group, mesangial accumulation, shortened podocyte processes and obscured filtration slits could clearly be identified in the diabetic group. Obscureness of the infiltration slits were not diffuse (Figures 4a and 4b).

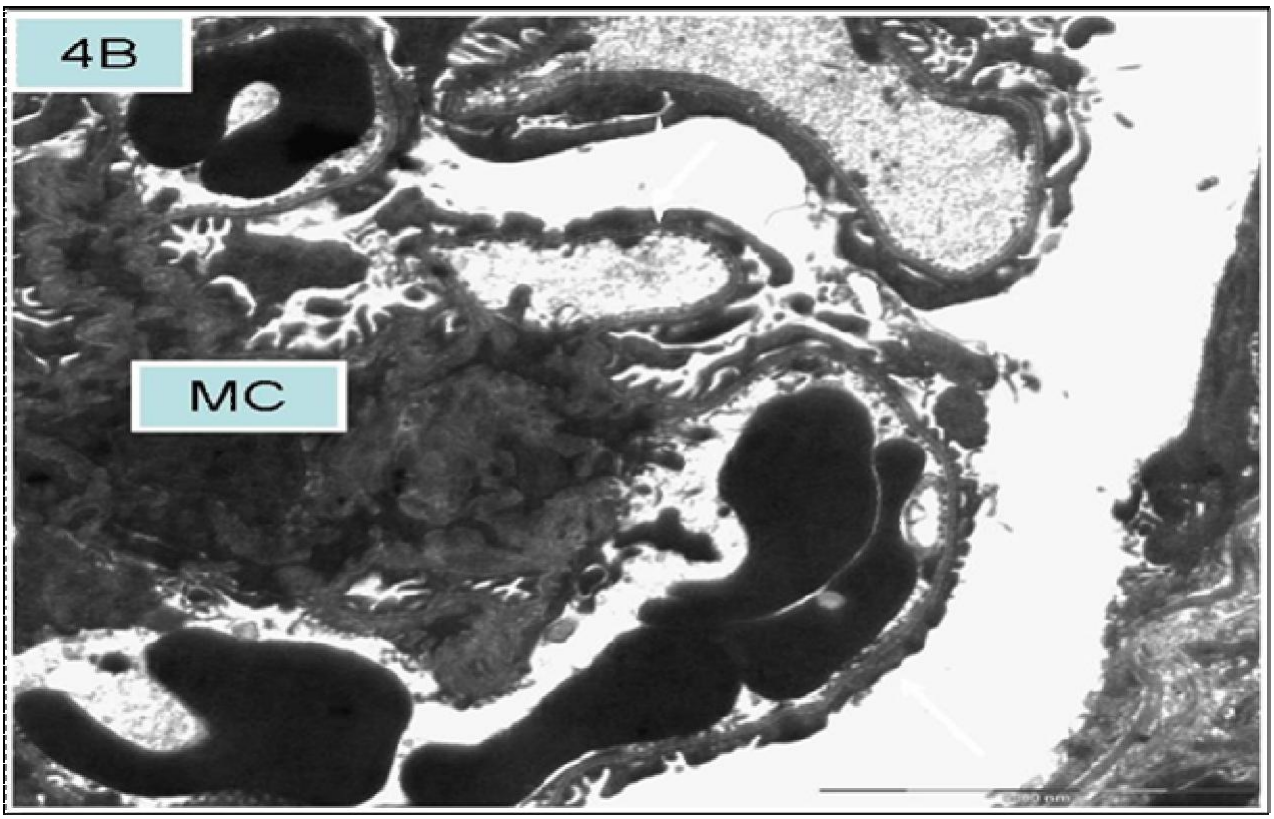

Figure 4b. Comparison of podocyte extensions and infiltration slits; Diabetic rat kidney. Shortened podocyte extensions and obscure infiltration slits (arrow) are marked.

Degeneration of the glomerular endothelia, diffuse or nodular glomerulosclerosis, apoptosis in the podocytes and hyalinization beneath the basal lamina were not observed in this group. Structural changes in the glomeruli of the diabetic rats fed with VCE were milder than the diabetes group. Increase in mesangial cells was not observed. Podocyte processes and filtration slits generally appeared normal (Figure 5).

DISCUSSION

Diabetic nephropathy is the most common cause of chronic renal disease and the foremost indication for dialysis and renal transplantation (Estacio and Schrier, 2001) . Diffuse or nodular glomerulosclerosis, arteriosclerosis, tubulo-interstitial fibrosis and atrophy occur, proteinuria gradually increases and so does the blood pressure (Alsaad et al., 2007) . Glomerulosclerosis develops as a result of injury to the podocytes which plays the key role in glomerular filtration (Lee et al., 2007). Podocyte injury can present itself in two forms: metabolic (biochemical) and hemodynamic (associated with hyperfiltration and hyperperfusion) (Hostetter et al., 2003; Wolf et al., 2003). Glomerular hyperfiltration and hyperperfusion are essential in mesangilisation and changes in the glomerular basal membrane (Wolf et al., 2003; Menini et al., 2007). In diabetic nephropathy, prostaglandin E_2 (PGE_2) synthesis in the glomeruli is significantly increased. This is as a result of an increase in mesangial cells (Iino et al., 2005). -tochopherole inhibits PGE_2 (Peerapatdit et al., 2006; Wu et al., 2007). Oxidative stress and free oxygen radicals, which develop during nephropathy, trigger apoptosis of the tubular epithelial cells and podocytes of the glomeruli (Blauwkamp et al., 2008; Jung et al., 2008; Rüster et al., 2008; Susztak et al., 2006). Various agents used to inhibit apoptosis have been tried in the treatment of nephropathy (Isermann et al., 2007). In the present study was observed an increase in mesangial cell in the glomeruli of diabetic kidneys and mesangial accumulation but not apoptosis of podocytes and tubular epithelial cells.

Antioxidants are frequently used for diabetes and its complications. Plasma Vitamin C and E concentrations are reduced in diabetes (Murugan et al., 2006; Ramesh et al., 2006; Peerapatdit et al., 2006; Wu et al., 2007; Lee et al., 2007). A positive relation has been demonstrated between high plasma vitamin C level and reduction in complications of diabetes (Harding et al., 2008). Vitamin C plays a central role in the antioxidant protective system, protecting all lipids undergoing oxidation and di-minishing the number of apoptotic cells (Sadi et al., 2008;

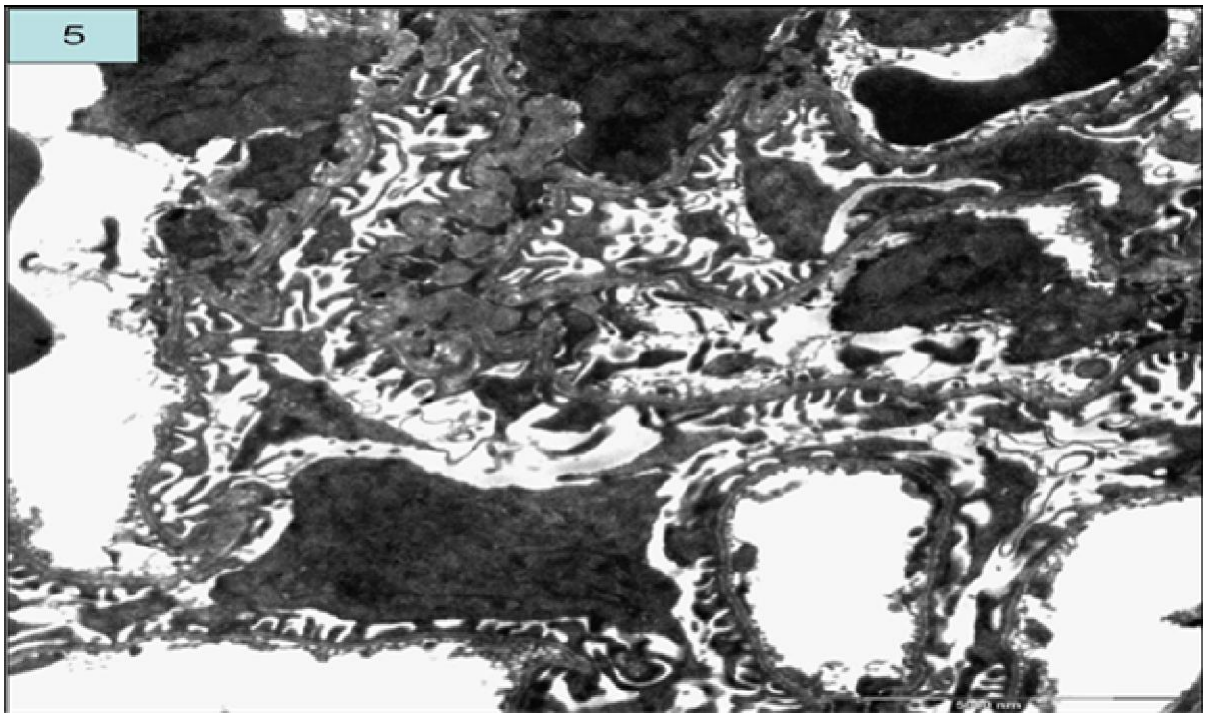

Figure 5. Rats treated with a combination of vitamins C and E had more intact glomeruli in comparison to the diabetic group and increase in mesengial cell was not noted.

Afkhami-Ardekani et al., 2007; Al-Shamsi et al., 2006). Furthermore, vitamin C regenerates the oxidized vitamin E (Chen et al., 2005). Vitamin E, on the other hand, acts as a non- enzymatic antioxidant and reduces lipid peroxidation and glutathione (Punithavatki et al., 2008; Minamiyama et al., 2008; Lee et al., 2007). Vitamin E is very effective in glycemic control, lowering the HbA_{1c} levels (Ihara et al., 2000) and preventing the hypertrophic effects of hyperglycemia (Nascimento et al., 2005). However, this is in contrast to the results of some studies, which showed that vitamin E was not beneficial in glycemic control and lipid metabolism in Type II diabetes (Ble-Castillo et al., 2005). In another study, the researchers demonstrated that a combination of vitamins C and E improved the glomerular functions but did not have any effect on the tubular functions (Farvid et al., 2006). When exercise is given to rats with STZ-induced diabetes in addition to Vitamins C and E, it was observed that lipid peroxidation was significantly reduced, glutathione peroxidase (GSH-Px) was increased and reduced glutathione (GSH) level was decreased (Kutlu et al., 2005).

The earliest structural changes in diabetic nephropathy are the increase in mesangial cells and mesangial dilatation. Diffuse thickening of the glomerular basement membrane depends on the severity of the disease. Vitamins C and E reduce the thickness of the basement membrane (Kedziara- Kornatowski et al., 2003; Davila-Esqueda et al., 2005). It has been argued that glomerular changes can occur in diabetic nephropathy without arterial or tubulo-interstitial changes (Fioretto and Mauer, 2007). Vacuolization within the podocytes, myelin figures and blebs were noted (Farvid et al., 2006). In our results, thickening of the glomerular basement membrane was not diffuse but they were in certain regions. Occasional irregularities of the podocyte processes, shortened podocyte processes and obscureness of the infiltration slits were observed.

In conclusion, alterations were not observed in the renal tubules of rats with STZ induced-diabetes while increased mesangial cells, increased capillary permeability, and obscureness of the filtration slits were noted in the glomeruli. Neither apoptosis of the podocytes nor thickening of the basement membrane was observed. These alterations were less pronounced in the diabetic rats treated with VCE. VCE helped alleviation of the renal degeneration by protecting the glomerular structures from oxidative injury. Concomitant administration of Vitamins C and E would be more effective in preventing the complications of diabetes.

REFERENCES

Afkhami-Ardekani M, Shojaodding-Ardekani A (2007). Effect of vitamin C on blood glucose, serum lipids and serum insulin type 2 diabetes patients. Indian J. Med. Res. 126: 471-4.

Alsaad KO, Herzenberg AM (2007). Distinguishing diabetic nephropathy from other causes of glomerulosclerosis: An update. J. Clin. Pathol. 60: 18-26.

Al-Shamsi M, Amin A, Adeqhate E (2006). Effect of vitamin C on liver and kidney functions in normal and diabetic rats. Ann. NY Acad. Sci. 1084: 371-90.
Blauwkamp MN, Yu J, Schin MA, Burke KA, Berry MJ, Carlson BA, Brosius FC, Koenig RJ (2008). Podocyte specific knock out of seleneproteins does not enhance nephropathy in streptozotocin diabetic C57BL/6 mice. BMC Nephrol. 9: 7.
Ble-Castillo JL, Carmona-Diaz E, Mendez JD, Larios-Medina FJ, Medina-Santillan R, Cleva-Villanueva G, Diaz-Zagoya JC (2005). Effect of alpha-tocopherol on the metabolic control and oxidative stress in female type 2 diabetics. Biomed. Pharmacother. 59: 290-5.
Cederberq J, Erikssan UJ (2005). Antioxidative treatment of pregnant diabetic rats diminishes embryonic dysmorphogenesis. Birth Defects Res. A. Clin. Mol. Teratol. 73: 498-505.
Chen L, Jia RH, Qiu CJ, Ding G (2005). Hyperglycemia inhibits the uptake of dehydroascorbate in tubular epithelial cell. Am. J. Nephrol. 25: 459-465.
Davila-Esqueda ME, Vertiz-Hernandez AA, Martinez-Morales F (2005). Comparative analysis of the renoprotective effects of pentoxifylline and vitamin E on streptozotocin-induced diabetes mellitus. Ren. Fail. 27: 195-207.
Estacio RO, Schrier RW (2001). Diabetic nephropathy; pathogenesis, diagnosis and prevention of progression. Adv. Int. Med. 46: 359-408.
Fadupin GT, Akpoqhar AU, Okunade KA (2007). Comparative study of serum ascorbic acid level in people with and without type 2 diabetes in Ibadan Nigeria. Afr. J. Med. Med. Sci. 36: 335-339.
Farvid MS, Jalali M, Siassi F, Hosseini M (2006). Comparison of the effects of vitamins and/or mineral supplementation on glomerular and tubular dysfunction in type 2 diabetes. Diabetes Care 29: 747-8.
Fioretto P, Mauer M (2007). Histopathology of diabetic nephropathy. Semin. Nephrol. 27: 195-207.
Haidara MA, Khloussy H, Ammar H, Aal Kasem LA (2004). Impact of alpha-tocopherol and vitamin C on endothelial markers in rats with streptozotocin-induced diabetes. Med. Sci. Monit. 10: BR41-6.
Hamdy NM, Suwailem SM, El-Mesallamy HO (2008). Influence of vitamin E supplementation on endothelial complications in type 2 diabetes mellitus patients who underwent coronary artery bypass graft. J. Diab. Complications 23:167-73.
Harding AH, Wareham NJ, Bingham SA, Khaw K, Luben R, Welch A, Forouhi NG (2008). Plasma vitamin C level, fruit and vegetable consumption and the risk of new-onset type 2 diabetes mellitus: The European prospective investigation of Cancer - Norfolk prospective study. Arch. Inter. Med. 168: 1485-1486.
Hostetter TH (2003). Hyperfiltration and glomerulosclerosis. Semin. Nephrol. 23:194-9.
Iino K, Iwase M, Sonoki K, Yoshinari M, Iida M (2005). Combination treatment of vitamin C and desferrioxamine suppresses glomerular superoxide and prostaglandin E production in diabetic rats. Diab. Obes. Metab. 7: 106-9.
Ihara Y, Yamada Y, Toyokuni S, Miyowaki K, Ban N, Adachi T, Kurae A, Iwakuru T, Kubota A, Hiai H, Seino Y (2000). Antioxidant alpha-tocopherol ameliorates glycemic control of GK rats, a model of type 2 diabetes. FEBS Lett. 473: 24-6.
Isermann B, Vinnikov IA, Madhusudhan T, Herzog S, Kashif M, Blautzik J (2007). Activated protein C protects against diabetic nephropathy by inhibiting endothelial and podocyte apoptosis. Nat. Med. 13: 1349-58.
Je HD, Shin CY, Park HS, Huh IH, Sohn UD (2001). The comparison of vitamin C and vitamin E on the protein oxidation of diabetic rats. J. Auton. Pharmacol. 21: 231-236.
Jung DS, Li JJ, Kwak SJ, Lee SH, Park J, Song YS, Yoo TH, Han SH, Lee JE, Kim DK, Moon SJ, Kim YS, Han DS, Kang SW (2008). FR167653 inhibits fibronectin expression and apoptosis in diabetic glomeruli and in high-glucose-stimulated mesangial cells. Am. J. Physiol. Renal Physiol. 295: F595-604.
Kashiba M, Oka J, Ichikawa R, Kasahara E, Inayama T (2002). Impaired ascorbic acid metabolism in streptozotocin-induced diabetic rats. Free Radic. Biol. Med. 33: 1221-1230.
Kedziara-Kornatowski K, Szram S, Karnatowski T, Szadujkis -Szadurski L, Kedziora J, Bartosz G (2003). Effect of vitamin E and vitamin C supplementation on antioxidative state and renal glomerular basement membrane thickness in diabetic kidney. Nephron Exp. Nephrol. 95: e134-43.
Kutlu M, Nazıoglu M, Simsek H, Yılmaz T, Kukner AH (2005). Moderate exercise combined with dietary vitamins C and E counteracts oxidative stress in the kidney and lens of streptozotocin-induced diabetic rat. Int. J. Vitam. Nutr. Res. 75: 71-80.
Lee EY, Lee MY, Honq SW, Chunq CH, Honq SY (2007). Blockade of oxidative stress by vitamin C ameliorates albuminuria and renal sclerosis in experimental diabetic rats. Yonsei Med. J. 48: 847-55.
Menini S, Iacobini C, Oddi G, Ricci C, Simonelli P, Fallucca S, Grattarola M, Peqliese F, Pesce C, Peqliese G (2007). Increased glomerular cell (podocyte) apoptosis in rats with streptozotozocin-induced diabetes mellitus: role in the development of diabetic glomerular disease. Diabetologia 50: 2591-9.
Minamiyama Y, Takemura S, Bito Y, Shinkawa H, Tsukioka T, Nakahira A, Suehiro S, Okada S (2008). Supplementation of alpha-tocopherol improves cardiovascular risk factors via the insulin signaling pathway and reduction of mitochondrial reactive oxygen species in type II diabetic rats. Free Radic. Res. 42: 261-271.
Monhart V (2008). Diabetes mellitus, hypertension and kidney. Vnitr. Lek. 54: 499-504.
Murugan P, Pari L (2006). Antioxidant effect of tetrahydrocurcımin in streptozotocin-nicotinamide induced diabetic rats. Life Sci. 79: 1720-1728.
Musalmah M, Nizrana MY, Fairuz AH, Noor Aini AH, Azian AL, Gapor MT, Wan Ngah WZ (2005). Comparative effects of paim vitamin E and alpha-tocopherol on healing and wound tissue antioxidant enzyme levels in diabetic rats. Lipids 40: 575-80.
Nascimento Gomes G, Barbosa FT, Radaeli RF, Cavaral MF, Mello Aires M, Zaladek Gil F (2005). Effect of D-alpha-tocopherol on tubular nephron acidification by rats with induced diabetes mellitus. Braz. J. Med. Biol. Res. 38: 1043-51.
Naziroglu M, Simsek M, Simsek H, Aydilek N, Özcan Z, Atılgan R (2004). The effects of hormone replacement therapy combined with vitamins C and E on antioxidants levels and lipid profiles in postmenopausal women with type 2 diabetes. Clin. Chem. Acta. 344: 63-71.
Ökten F, Özgüner F, Yılmaz HR, Uz E, Dündar B (2006). Melatonin reduces urinary excretion of N-acetyl-beta-D-glucosaminidase, albumin and renal oxidative markers in diabetic rats. Clin. Exp. Pharmacol. Physiol. 33: 95-101.
Peerapatdit T, Patchanans N, Likidlilid A, Poldee S, Sriratanasathavorn C (2006). Plasma lipid peroxidation and antioxidant nutrients in type 2 diabetic patients. J. Med. Assos. Thai. 89 Suppl. 5: 5147-55.
Prakasam A, Sethupathy S, Puqalendi KV (2005). Antiperoxidative and antioxidant effects of casearia esculenta root extract in streptozotocin-induced diabetic rats. Yale J. Biol. Med. 78: 15-23.
Punithavatki VR, Anuthama R, Prince PS (2008). Combined treatment with naringin and vitamin C ameliorates streptozotocin-induced diabetes in male Wistar rats. J. Appl. Toxicol. 28: 806-813.
Ramesh B, Saravanan R, Pugalendi KV (2006). Effect of dietary substitution of Groundnut oil on blood glucose, lipid profile, and redox status in streptozotocin-diabetic rats. Yale J. Biol. Med. 79: 9-17.
Ritz E (2006). Diabetic nephropathy. Saudi J. Kidney Dis. Transpl. 17: 481-490.
Ruperez FJ, Garcia-Martinez D, Baera B, Maeso N, Cifuentes A, Barbas C, Herrera E (2008). Evolution of oxidative stress parameters and response to oral vitamins E and C in streptozotocin-induced diabetic rats. J. Pharm. Pharmacol. 60: 871-878.
Rüster C, Bondeva T, Franke S, Förster M, Wolf G (2008). Advanced glycation end-products induce cell cycle arrest and hypertrophy in podocytes. Nephrol. Dial. Transplant. 23: 2179-91.
Rychlik I (2008). Epidemiology of diabetic nephropathy. Vnitr Lek. 54: 488-493.
Sadi G, Yılmaz O, Güray T (2008). Effect of vitamin C and lipoic acid on streptozotocin-induced diabetes gene expression: mRNA and protein expressions of Cu-Zn SOD and catalase. Mol. Cell Biochem. 309: 109-16.
Sena CM, Proenca T, Nunes E, Santos MS, Seica RM (2008). The effect of soybean oil on glycaemic control in Goto-Kakizaki rats, an animal model of type 2 diabetes. Med. Chem. 4: 293-297.
Susztak K, Raff AC, Schilffer M, Bottinger EP (2006). Glucose-induced reactive oxygen species cause apoptosis of podocytes and podocyte

depletion at the onset of diabetic nephropathy. Diabetes 55: 225-233.
Uusitalo L, Nevalainen J, Niinistü S, Alfthan G, Sandvall J (2008). Serum alpha and gamma- tocopherol concentrations and risk of advanced beta cell autoimmunity in children with HLA-conferred susceptibility to type 1 diabetes mellitus. Diabetologia 51: 773-80.
Villegas R, Shu XO, Gao YT, Yang G, Elasy T, Li H, Zhenq W (2008). Vegetable but not fruit consumption reduces the risk of type 2 diabetes in Chinese women. J. Nutr. 138: 574-80.
Wang L, Zhang L, Yu Y, Wang Y, Niu N (2008). The protective effects of taurine against early renal injury in STZ-induced diabetic rats, correlated with inhibition of renal Lox-1 mediated ICAM-1 expression. Ren. Fail. 30: 763-771.
Welt K, Weiss J, Martin R, Hermsdorf T, Drews S, Fitzi G (2007). Ginkgobiloba extract protects rat kidney from diabetic and hypoxic damage. Phytomedicine 14: 196-203.
Wolf G, Butzmann U, Wenzel UO (2003). The renin-angiotensin system and progression of renal disease: from hemodynamics to cell biology. Nephron. Physiol. 93: 3-13.
Wu JH, Ward NC, Indrawan AP, Almeida CA, Hodgson JM, Porudfoot JM, Puddey IB, Croft KD (2007). Effect of -tocopherol and mixed tocopherol supplementation on markers of oxidative stress and inflammation in type 2 diabetes. Clin. Chem. 53: 511-519.
Wu X, Iguchi T, Hiranoj J, Fujita I, Ueda H, Itoh N, Tanaka K, Nakahishi T (2007). Upregulation of sodium-dependent vitamin C transporter 2 expression in adrenals increases norepinephrine production and aggravates hyperlipidemia in mice with streptozotocin-induced diabetes. Biochem. Pharmacol. 74: 1020-1028.
Yıldırım O, Büyükbingöl Z (2003). In-vivo effect of vitamin C with cobalt on oxidative stres in experimental diabetic rat kidney. Diab. Nutr .Metab. 16: 208-213.
Yokozawa T, Yamabe N, Kim HY, Kang KS, Hur JM, Park CH, Tanaka T (2008). Protective effects of Morroniside isolated from Corni Fructus against renal damage in streptozotocin- induced diabetic rats. Biol. Pharm. Bull. 31: 1422-1428.