Original Article

Histomorphological and Immunohistochemical Study on the Possible Effect of Vitamin E on Aortic Wall Remodeling in Streptozotocin (STZ)-Induced Diabetic Rats

Joseph Amin Aziz

Department of Anatomy and Embryology, Faculty of Medicine, Zagazig University, Egypt

ABSTRACT

Introduction: The number of diabetics around the world will reach three hundred million by 2025. Vascular dysfunction is a common finding in both diabetes and glucose intolerance. The repercussions of hyperglycemia on the endothelium and smooth muscle cells contribute to the high risk of cardiovascular disease in type 1 and type 2 diabetes as well.

Aim of the Work: The purpose of this study was to investigate the possible immunomodulatory effect of vitamin E on the remodeling of aortic wall in STZ-induced diabetic rats histologically, immunohistochemically, and morphometrically.

Materials and Methods: Thirty adult male albino rats were equally divided into 3 groups: control, five rats received a single intraperitoneal injection of 0.1 ml.0.1 M citrate buffer (pH 4.4), and 5 rats administered 2 ml of corn oil by oral gavage.; diabetes group (injected intraperitoneally with 65 mg/kg of streptozotocin in 0.1 ml citrate buffer), diabetic+ vitamin E group given Vitamin E (300 mg/kg) for 10 weeks after induction of diabetes. Sections of aortic specimens were stained with H&E, Masson trichrome, and orcein and processed for immunological evaluation with anti-eNOS and anti-α-SMA.

Results: Vitamin E mended the histomorphological changes induced by diabetes; it significantly decreased tunica media thickness, collagen, and smooth muscle cells. also improved elastin destruction and enhanced the expression of eNOS in the endothelial cells and suppressed α-SMA immunoreactivity in the wall of the aorta in diabetic rats.

Conclusion: Vitamin ameliorated the histological remodeling of the aortic wall in diabetic rats that was confirmed histologically and immunohistochemically through the regulation of e NOS and α -SMA expression.

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Key Words: Aorta, diabetes, eNOS, rat, vitamin E.

Corresponding Author: Joseph Amin Aziz, MD, Department of Anatomy and Embryology, Faculty of Medicine, Zagazig University, Egypt, **Tel.**: +20 12 2752 7558, **E-mail:** Joseph.aziz@su.edu.eg; Jaaziz@Zu.edu.eg

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INTRODUCTION

Vascular dysfunction is a major risk in those with diabetes or glucose intolerance. The impact of hyperglycemia on the endothelium and smooth muscle cells contributes to the development of cardiovascular disease in type 1 and type 2 diabetes as well[1-3]. Vascular inflammation, fibrosis, endothelial dysfunction, and oxidative stress take part in cardiovascular disease, the leading cause of death among diabetic patients[4,5]. Impaired endothelial-dependent relaxation is pivotal to the development of vascular complications[6]. Endothelial cells are not only physical barriers. They are, however, metabolically active cells that secret autocrine, endocrine, and paracrine factors[7]. The lining endothelium maintains vascular integrity by restricting vascular smooth muscle cells (VSMC) proliferation, preventing coagulation, and inhibiting platelet aggregation[8]. Understanding how diabetes causes endothelial dysfunction can aid in the development of new methods to control and prevent diabetic vascular disease[3].

Vascular smooth muscles are highly specialized cells that proliferate slowly in healthy blood vessels whereas, during atherosclerosis, VSMCs undergo phenotypic changes[9]. The Actin Alpha 2 (ACTA2) gene encodes the synthesis of α-SMA, typically expressed in smooth muscle cells (SMCs) of the aortic wall. Alpha-smooth muscle actin is the most abundant protein in differentiated SMCs, comprising up to 40% of total cellular protein. Abnormal expression of α-SMA signals several pathways that together lead to hyperplasia of VSMCs observed in many vascular disorders[9-11].

Hyperglycemia, the whole metabolic disorder in diabetes, damages the cellular components of the blood vessel wall contributing to the high risk of cardiovascular disorders. Nevertheless, strict glycemic control alone has not proven successful in the protection against diabetic cardiovascular morbidity[7,12]. Indeed, a comprehensive approach would be more useful to reduce vascular complications[13]. Diabetes-induced oxygen radicals, particularly superoxide anions, impair endothelial nitric acid production and hence play a key role in the pathogenesis of cardiovascular disease. Therefore, a potent antioxidant is needed to reduce endothelial dysfunction. Interest has been increased in the powerful antioxidant properties and consequently the potential use of vitamin E in protection against diabetic vascular complications[14].

Alpha-Tocopherol is one of eight natural forms of fat-soluble compounds known collectively as vitamin E (vit E). Other forms include β-, γ-, and δ-tocopherol; α-, β-,

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γ-, and δ-tocotrienol. Tocopherols and tocotrienols are naturally found in some foodstuffs like seeds and vegetable oils[15-17]. Vitamin E is a nonenzymatic antioxidant that stabilizes the cell membrane and prevents DNA damage through the prevention of lipid peroxidation and the elimination of reactive oxygen species (ROS). Tocopherols share in gene transcription, modulates inflammatory and immunological responses, and organizes enzyme and molecule activity[18]. Vitamin E can be a valuable supplement to boost antioxidant system and control diabetic complications. Diabetic patients who were supplemented with vitamin E had delayed onset and a slower progression of the complications[19,20].

Thus, the present study aimed at investigating the possible immunomodulatory effect of vitamin E on the remodeling of aortic wall in STZ- induced diabetic rats.

MATERIALS AND METHODS

Drugs

1. Streptozotocin (STZ): obtained from Sigma chemical Co. at Cairo, Egypt.
2. DL-α-tocopherol acetate (Vitamin E 400mg, PHARCO Pharmaceuticals, Alexandria, Egypt) purchased from local pharmacy.

Animals and experimental design

All experimental procedures were approved and performed in accordance with the guidelines of the Institutional Animal Care and Uses Committee of the Zagazig University (ZU-IACUC). The study was conducted on thirty male albino rats, with a body weight ranging from 220 to 250 g. The animals were kept in clear sided, metal cages at controlled temperature (20-25°c), on a 12-hr light–dark cycle, and with free access to food and water. Animals were randomly equally grouped as the following: Control group: (a)five rats received a single intraperitoneal injection of 0.1 ml.0.1 M citrate buffer (pH 4.4) and (b) 5 rats administered 2 ml of corn oil by oral gavage. Diabetes group: treated with streptozotocin (65 mg/kg body weight in 0.1 ml.0.1 M citrate buffer, pH 4.5) in the intraperitoneal cavity after fasting overnight. Tail vein fasting blood glucose (FBG) levels were tested one week after the streptozotocin injection using GlucoDr (All Medicus Co. Ltd, Republic of Korea). Rats with glucose levels ≥ 200 mg/dl were considered diabetic[21]. Diabetes + Vit E group: after injection with streptozotocin (65 mg/kg body weight) in intraperitoneal cavity and confirmation of diabetes induction, Vitamin E (300 mg/kg[22] was given in corn oil by oral gavage[15] for 10 weeks[23]. After 11 weeks of diabetes induction, all rats were anesthetized by intraperitoneal injection of thiopental 50 mg/kg. Blood samples were taken directly from the heart to measure blood sugar. A thoracic incision was done, and the proximal part (1-2 cm) of the aorta was dissected and fixed in 4% neutral formaldehyde solution after removal of surrounding fat.

Histological study

Formalin fixed aortic specimens were dehydrated with graded alcohol and embedded in paraffin blocks. Cross sections (4μm thick) were then deparaffinized and cleared in xylene, rehydrated with decreasing concentrations of alcohol, and stained with H&E, Masson's trichrome and orcein by conventional methods[24].

Immunohistochemical study for the expression of eNOS and α-SMA

Anti α-SMA rabbit monoclonal antibody (Abcam, EPR5368) and anti-eNOS rabbit polyclonal antibody (Abcam, ab5589) were used to assess smooth muscle and endothelial cells, respectively. Aortic sections (4-5μm) were hydrated after dissolving of paraffin wax. Endogenous peroxidase activity was blocked using 3% hydrogen peroxide. Sections were heated in a citrate buffer (pH 6.0) in a water bath at 95°C for 30 minutes to retrieve the antigens. After antigen retrieval, sections were incubated with primary antibodies at dilution 1:100; at 4°C overnight. Colon and placenta were used for control positive for α-SMA and e NOS respectively. The secondary antibody (biotinylated horse anti-mouse IgG) was added for 30 minutes followed by the avidin-biotin peroxidase complex. The slides were counterstained with Mayer’s hematoxylin and the reaction was visualized with diaminobenzidine solution. The slides were observed under a light microscope.

Morphometry study

Stained slides were photographed using LeicaICC50W light electric microscope at the Image Analysis Unit of Anatomy and Embryology Department, Faculty of Medicine, Zagazig University. All morphological studies were done at magnification 40X using ImageJ (FIJI) software. The thickness of tunica media was measured at 12, 3, 6, 9 o’clock per each slide using H&E-stained sections. The number of SMCs nuclei was calculated in 2500 μm2 square grid in 5 non overlapping fields in each slide. For collagen deposition, the area percentage of greenish discoloration was calculated under Masson’s trichrome in 5 non-overlapping fields in each slide[25]. Quantitative analysis for e NOS and α-SMA immunolocalization was done by measuring the optical density (OD) of eNOS-positive endothelial cells and the area percentage of brown filaments regarding α-SMA.

Statistical analysis

Statistical analyses of data were carried out using GraphPad Prism version 8.0.2 (GraphPad Software, Inc., San Diego, CA). Data were expressed as the mean ± SEM. One-way analysis of variance (ANOVA) followed by the Tukey's multiple-comparison posttest was used to compare between the studied groups. Statistical significance was considered at $P\ value < 0.05$.

RESULTS

Animal deaths

There was not any animal death in any of the studied groups.

Effect of vit E on serological results

The blood sugar level at the time of scarification is shown in figure1; the blood sugar was significantly elevated in the STZ- treated rats (367.6 ± 20.97 mg/dl) versus the control ones (87.89 ± 8.46 mg/dl). There was a non-significant improvement of hyperglycemia in diabetes + vit E group (335.8 ± 14.75 mg/dl) (Figure 1).

Histological and morphometrical results

Under H&E staining, normal aortic histology was observed in both group a and group b of the control group where the aortic wall consisted of 3 clearly demarcated tunics: intima, media, and adventitia. Tunica intima included a single endothelial layer resting on internal elastic lamina, while the media layer included regularly arranged elastic lamellae; smooth muscles and ECM in the interlamellar spaces (Figure 2a). Media in control rats had an average thickness of 91.83 ± 6.78 µm (Figure 2d). Diabetes induced grave destruction in the wall of diabetic rats' aorta (Figure 2b). The media was markedly thickened (175.8 ± 6.87), and the number of SMCs was significantly increased in comparison with control group ($P < 0.0001$) (Figures 2d,c); foam cells were frequently seen with extra deposition of ECM. Moreover, the endothelial lining was hypertrophied (Figure 4b). In diabetes + vit E group, the histological appearance of the aortic wall unmistakably enhanced (Figure 2c), the media thickness (131.1 ± 5.3 µm) and smooth muscle cells (10.88 ± 1.39) declined significantly ($P < 0.0001$) (Figures 2d,c).

In addition, in Masson's trichrome stained sections, the area percentage of collagen was significantly increased in diabetes group when compared with the control group (32.7 ± 0.8 versus 13.5 ± 0.5, $P < 0.0001$). Collagen deposition significantly diminished in in diabetes + vit E group (20,6 ± 1.0, $P < 0.0001$) (Figure 3). At the same time, in orcein stained slides, elastic laminae were disrupted and irregularly arranged in STZ-injected rats. However, the area percentage of elastic fibers were not significantly affected (Figure 4). In diabetic rats supplemented with vit E, the elastic fibers became more regular and showed less disruption than were in diabetic rats (Figure 4c).

Immunohistochemical and morphometric results of eNOS and α-SMA

The highest OD of eNOS positive endothelial cells was in the lining of the control aorta (0.173 ± 0.007) followed by diabetes + vit E group (0.155 ± 0.009), meanwhile, the least OD was in the diabetic rats (0.115 ± 0.008). The difference was significant between control and diabetic groups ($P< 0.0001$) and between diabetic and diabetes + vit E groups ($P< 0.0046$). Moreover, in diabetic rats, eNOS was expressed in the VSMC (Figure 5). Conversely, the most abundant expression of α-SMA was in the media of diabetic rats. The expression was the most intense near the adventitia in addition to obvious expression in endothelial cells (Figure 6b). The area % of α-SMA expression in diabetes group was 11.74 ± 0.59 which was significantly higher than the control group (3.85 ± 1.48, $P< 0.0001$). In diabetes + vit E group, the area % of α-SMA localization was significantly less than in diabetes group ($P< 0.001$) but still significantly higher than control ($P = 0.0122$) (Figure 6).

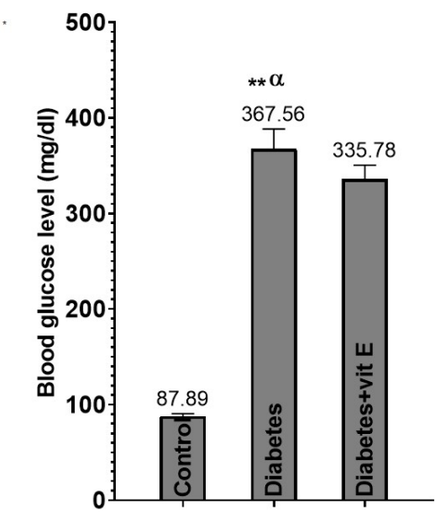

Fig. 1: Showing a histogram representing the mean blood glucose levels in the different studied groups at time of scarification. Glucose level is significantly elevated in diabetes group compared to control group; It decreased insignificantly in diabetes + vit E group compared to diabetes group. **significant at level $p<0.0001$. α versus control.

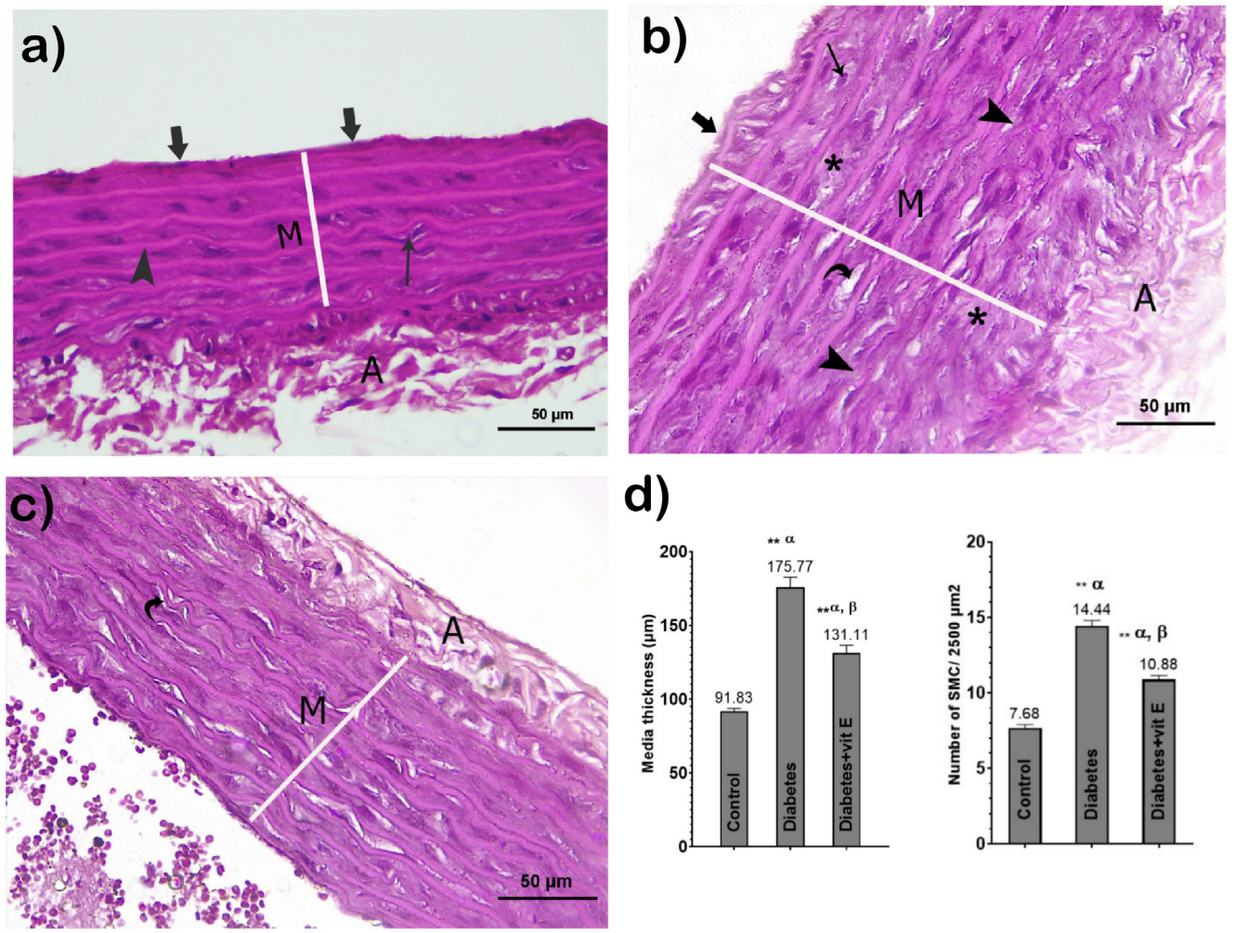

Fig. 2: Photomicrographs showing cross sections of aorta (a-c), a histogram of media thickness (d), and a histogram of smooth muscles cells number (e). In control group (a) normal structure of aorta: thick arrow: tunica intima; M: tunica media; white line: media thickness; arrowhead: elastic lamella; thin arrow: smooth muscle cells; A: adventitia. In diabetes group (b), the media width (white line) was markedly accentuated, and the nuclei of SMCs (thin arrows) were apparently increased. Notice foam cells (curved arrow), deposition of extracellular matrix (asterisks), and destruction of elastic lamella (arrowhead). In diabetes + vit E group, there is less medial thickening, less frequent smooth muscles and foam cells, the elastic lamellae are wavy and more regular (H&E X40). The histograms (d) and (e) represent mean thickness of tunica media in µm and the number of smooth muscles nuclei / 2500 µm^2, respectively. **significant at level p <0.0001. α versus control. β versus Diabetes.

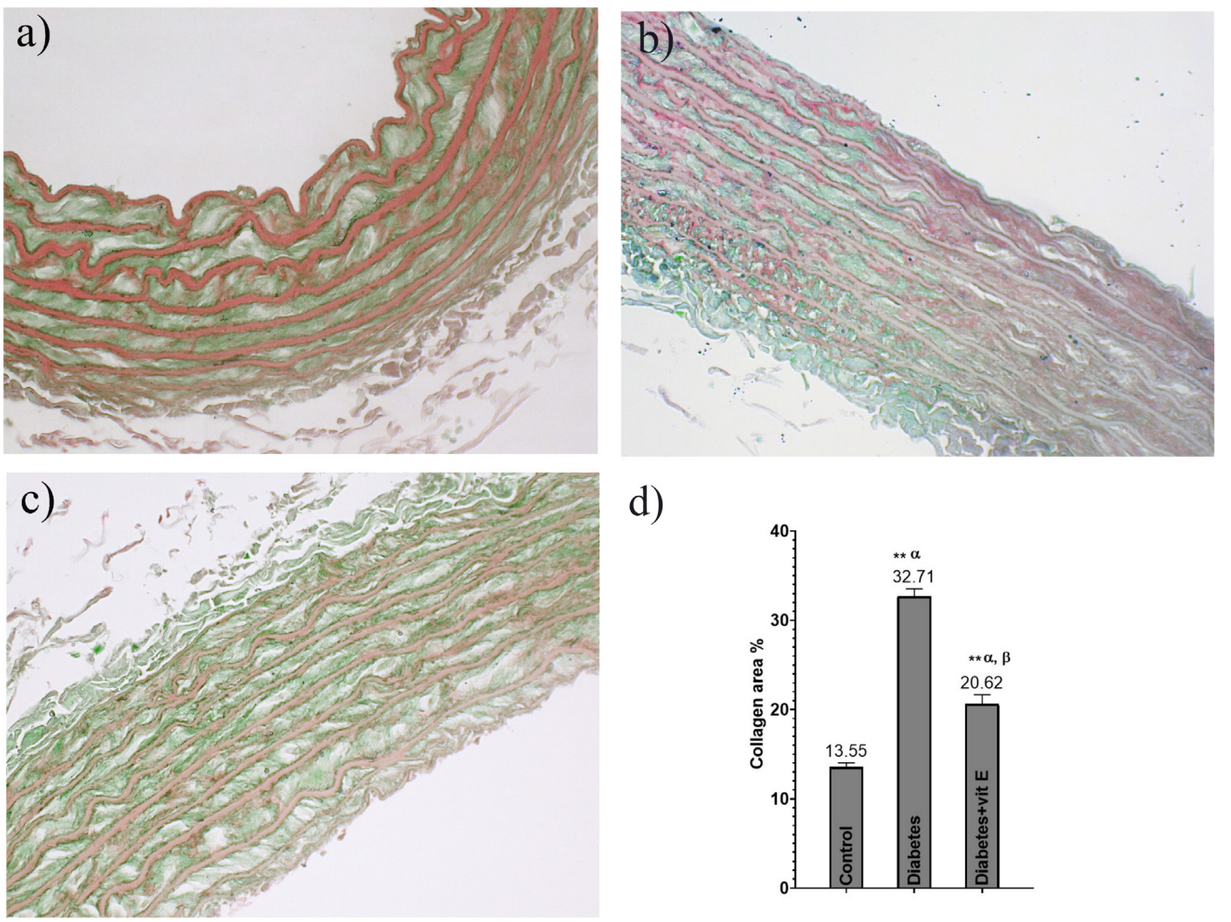

Fig. 3: Photomicrographs showing cross sections of aorta in the 3 groups (a-c) and a histogram of collagen area percentage (d). In control group (a), note the regular distribution of collagen between elastic laminae. In diabetes group (b) there is increased deposition of unevenly distributed collagen fibers both in media and adventitia. In diabetes + vit E group (c) there was less collagen lay down (Masson's trichrome X40). The histogram (d) represents cross sectional area % of collagen in the 3 groups. ** significant at level p <0.0001. α versus control. β versus diabetes.

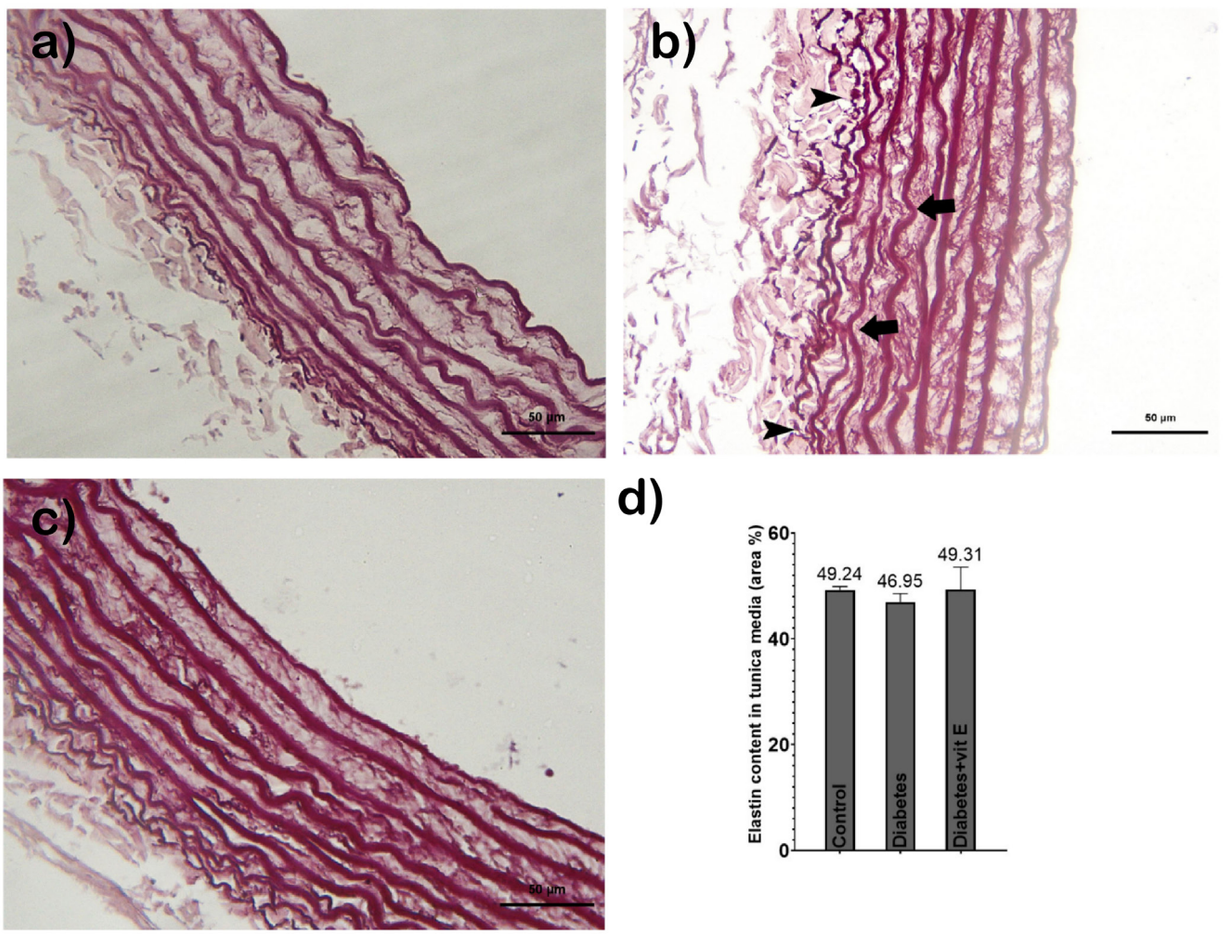

Fig. 4: Photomicrographs showing cross sections of aorta in the 3 groups (a-c) and a histogram of elastin percentage (d). In control group (a), note the regular arrangement of elastic laminae. In diabetes group (b), diabetes induced irregularities (thick arrow), thinning and disruptions of elastic laminae (arrow heads). In diabetes + vit E group (c) there was less disruption with more regularity of elastic lamellae (Orcein X40). The histogram (d) represents cross sectional area % of elastin in media of the 3 groups. There was no significant difference between the studied groups.

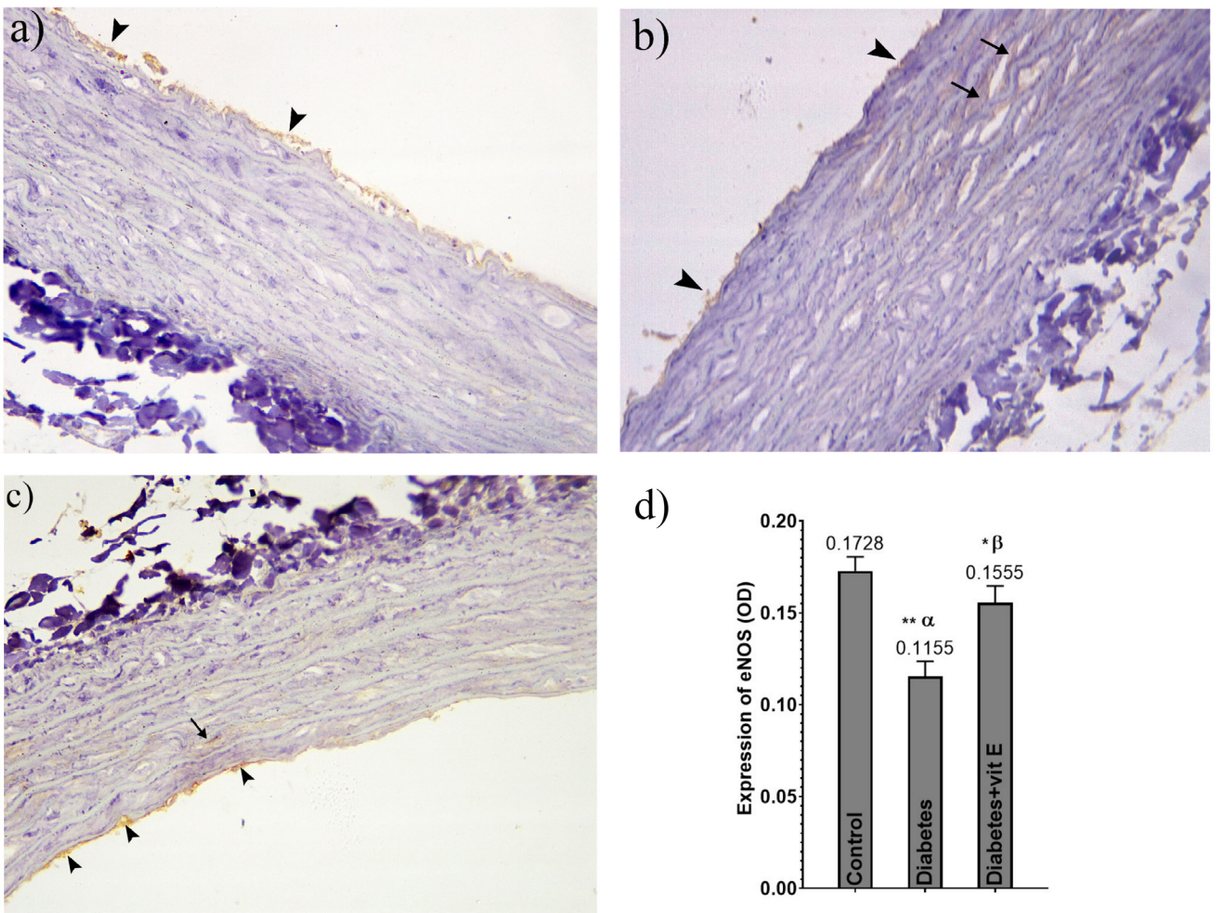

Fig. 5: Photomicrographs showing cross sections of aorta in the 3 groups (a-c) and a histogram of the optical density of eNOS expression in the intimal endothelium. In control group (a) there is strong reaction in the endothelial cells (arrow heads). In diabetes group (b) the expression is weak in tunica intima (arrowhead) but obvious in the smooth cells of the media (arrows). In diabetes + vit E group (c), moderate expression of eNOS can be noticed in the endothelial cells with less expression in smooth muscle cells (arrow) (Anti-eNOS X40). The histogram (d) represents the optical density of anti-eNOS staining of endothelial cells in the 3 groups. * Significant at level p =0.0046, ** significant at level p <0.0001. α versus control. β versus diabetes.

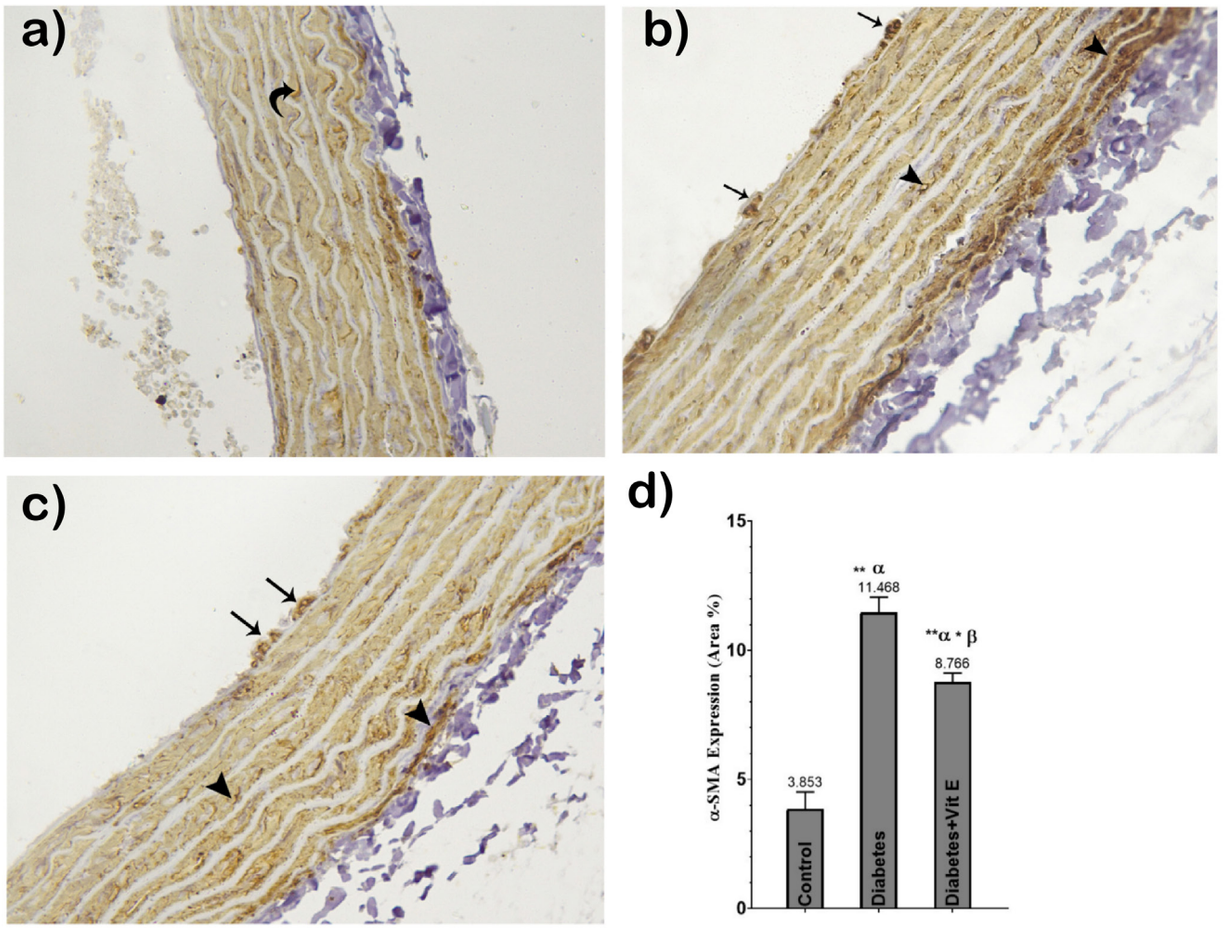

Fig. 6: Photomicrographs showing cross sections of aorta in the 3 groups (a-c) and a histogram of the area percentage of α-SMA expression (d). In control group (a) there is obvious reaction in the smooth muscles. In diabetes group (curved arrow) (b) The intensity of expression is apparently increased especially near the adventitia (arrowheads). Note the strong expression in the endothelial cells (thin arrows). In diabetes + vit E group (c), the immunoexpression of α-SMA was less prominent, when compared with the diabetes group, with few positive endothelial cells (arrows). (Anti- α-SMA X40). The histogram (d) represents the area percentage of α-SMA expression in the 3 groups. * Significant at level p <0.0122, ** significant at level $p \leq 0.0001$. α versus control. β versus diabetes.

DISCUSSION

The results of this study showed that vitamin E can play a valuable role in the prophylaxis of diabetic macrovascular complications by targeting endothelial dysfunction and smooth muscle activation. The number of diabetics around the world will reach three hundred million by 2025[26]. Angiopathy complicating diabetes involves large conduit arteries and microvascular arterioles and capillaries[5]. The major pathological feature in diabetic macroangiopathy is thickening and calcification of the arterial wall[23].

The Streptozotocin model of diabetes is suitable for studying the basic mechanisms of diabetic cardiovascular complications and consequently can help to discover new therapies for diabetes sequelae. The most frequently used protocol is intravenous or intraperitoneal single dose of 65 mg/kg STZ[21,27].

Although some studies showed that tocotrienols could be more powerful as antioxidants and have different health-promoting capabilities[14,18], α-tocopherol was chosen in this experiment because alpha-tocopherol is the only form that is known to meet human requirements[28]. Moreover, it is the most used as it is the main form of vitamin E in tissues[16].

The histomorphological alterations in the wall of the aorta in the diabetic rats in this study were in concordance with those in previous literature. Marked thickening, and disorganization of the media in have been described in diabetic animal models[29-31]. Human studies also showed that the media thickness of aorta increased significantly in diabetic patients when compared with the healthy people[22,29,32]. On the other hand, Salum *et al*.[33] and Bahar *et al*.[34] found non-significant difference in the media thickness between diabetic and control rats. According to Cüce *et al*.[35], 60 days after induction of diabetes were not sufficient to damage the wall of aorta.

Consistent with the present work results, Li *et al*.[36] found that proliferated smooth muscles account for most

of the media thickening. Under basic conditions, VSMC proliferate slowly. However, in vascular injury, they undergo abnormal proliferation[37]. In this work, vitamin E supplementation limited the proliferation of smooth muscle. In another study, vitamin E consumption decreased the number of proliferating cell nuclear antigen (PCNA) positive muscle cells in the aorta by inhibition of low-density lipoproteins oxidation in diabetic rats[22].

Smooth muscle cells, the main stromal cell in the wall of vasculature, are of great importance in regulating the functions of the blood vessels.[37,38]. A little is recognized about the way diabetes affects smooth muscles in the vascular wall[3]. High glucose level can activate the protein kinase C and Rho/Rho-kinase signaling pathways and stimulate actin polymerization[2]. Immunohistochemistry is a sensitive method for localizing α-SMA stains in the aortic tissue in human and experimental studies[11]. In healthy arterial wall, α-SMA is highly expressed in the VSMC denoting contractile phenotype[39]. In this study, the aortic wall of control rats showed obvious expression of α-SMA. Although Chen *et al*.[39] mentioned that in pathological conditions the expression of such protein diminishes defining the transformation of smooth muscle cells into more proliferative and migratory phenotype, in the current study and in line with Elbe *et al*.[22], the expression of α-SMA increased significantly in STZ-treated rats. According to Yuan and Wu[11], disruptions in α-SMA may result in SMC hyperplasia via focal adhesion kinase, p53, and platelet-derived growth factor receptor-α signaling pathways ending in aortic disease. Moreover, a-SMA is not a definitive SMC-lineage marker[9]. More recently, diabetes increased αSMA-expressing in perivascular mural cells in the choriocapillaris[40]. Additionally, new expression of α-SMA is noticed in activated fibrogenic cells known as myofibroblasts[41,42]. Myofibroblasts are likely derived from adventitial fibroblasts, SMC to myofibroblast transition, and endothelial to mesenchymal transition[43]. This is supported by the presence of strong anti-α-SMA staining juxta-adventitia and the expression of α-SMA in the endothelial cells. Alpha-smooth muscle actin-positive endothelial cells in adult aortic endothelium can be associated with progression of atherosclerosis[9]. The results of Zhao *et al*.[44] showed that the elevated α-SMA protein level in diabetic rats was markedly attenuated by vitamin E treatment which agrees with the results of the present study.

The Abundancy of foam cells in the media of diabetic rats in this study are supported by the observations of Brahmanaidu *et al*.[45]. Vascular smooth muscles uptake extra amounts of oxidized and non-oxidized lipids in pathological conditions and look much like foam under microscopic examination[38].

Fibrosis is a well-known diabetic vascular complication[5]. Variety of mechanism mediate the possible contribution of diabetes to extracellular matrix production[23]. Myofibroblasts regulate connective tissue remodeling. Activated myofibroblasts play a central role in the fibrotic process through the deposition of excess extracellular matrix[41,42]. Similarly, there was a significant increase in the deposition of collagen between elastic bundles and in the adventitia of aortic wall of the diabetic rats in the current study. However, concomitant treatment with vitamin E amended diabetes-induced fibrosis. Also, vitamin E has significantly reduced liver collagen in a diverse of pathological conditions. The antifibrotic properties of vitamin E were attributed to its ability to reduce lipid peroxidation and to enhance the activity of antioxidant enzymes[46].

Previous studies showed that diabetes induces deformation of elastic laminae and fibers[23,34]. Komolafe *et al*.[47] demonstrated a reduction in the elastic fibers in the tunics of the aorta and pulmonary trunk in the diabetic group. Hyperglycemia may render elastic fibers stiffer and more susceptible to degradation. Besides, diabetic rats had low levels of elastin protein in aortic tissues besides an increase in elastolytic activity[48,49]. Vitamin E normalized the amount of collagen and restored the organization of elastic laminae in the media of the aorta treated with homocysteine and methionine[50]. In line, in this work, vitamin E improved the diabetes-induced disruption and irregularity of elastic lamina.

Regarding the impact of diabetes on the thickness of vascular endothelium, Thent *et al*.[29] found no change in the intimal thickness between euglycemic and hyperglycemic rats. In contrast, the findings of this work and the findings of El-kassaby *et al*.[51], Komolafe, *et al*.[47] and Badalzadeh *et al*.[32] described the presence of hypertrophied aortic intima in diabetic rats.

Vascular endothelium plays a crucial role in modulating the tone and structure of the underlying vascular smooth muscle cells via NO production and synthesis of bioactive substances that keep the vascular homeostasis. Endothelial nitric oxide synthase (eNOS) is of major importance for many of these endothelial functions. Inhibition of eNOS, either directly or indirectly, can lead to the occurrence of vascular complications[3,8,52].

The negative impact of diabetes on the expression or eNOS or its activity has been shown by low level of eNOS protein in the vascular endothelium of diabetic heart[53]. eNOS-knockout diabetic mice exhibited distinct features of accelerated retinopathy[54] and progressive diabetic nephropathy[8]. Diabetes also downregulated aortic eNOS expression in rat aorta[55]. Moreover, advanced glycation end-products, associated with hyperglycemia, decreased the expression of eNOS in human coronary artery through oxidative stress[56].

On the other hand, activation of eNOS provided atheroprotection in diabetes-accelerated atherosclerosis[57]. Additionally, the administration of teucrium polium[55] and clinacanthus nutans leaves extract[58] resulted in reversal of endothelial dysfunction in diabetic rat aorta through the upregulation of eNOS.

Supporting the results of this study, the tocotrienol content of tocomin restored endothelium-dependent relaxation in the aorta of diabetic rats by increasing NO bioavailability and improving the expression of eNOS[14]. In this study, diabetes downregulated eNOS expression in the endothelium and induced its expression in the smooth muscles. Vitamin E supplementation increased the expression of eNOS in the endothelium and prevented its expression in the VSMCs. According to Pandolfiet *et al*.[59], cultures of vascular smooth muscle cells from diabetic rats exhibited phenotype modulation that was associated with increased eNOS expression and superoxide anion generation.

In the present study although the effect of vitamin E on hyperglycemia was insignificant, treatment of diabetic rats with vitamin E reversed the histopathological and immunohistochemical alterations induced by diabetes. In agreement, the histological and ultrastructural alterations in addition to the increase in glial fibrillary acidic protein (GFAP)-positive astrocytes in the cerebellar cortex of alloxan-induced diabetic rats were ameliorated with vitamin E treatement without any significant improvement of hyperglycemia[15]. Similarly, in the study of Yonguc *et al*.[60] the blood glcouse didn't differ in both diabetes + vitamin E and diabetes + grape seed extract groups and their neuroprotective effect has been attributed to removing oxidative stress and decreasing the expression of apoptotic genes in the hippocampus. The triple-blind, placebo-controlled clinical trial of Boshtam *et al*.[61] concluded that insulin, glyosylated hemoglobin, or fasting blood sugar in type II diabetic patients were not affected by the daily administration of 200 IU of vitamin E for 27 weeks. Neverthelss, the antioxidant vitamin E is beneficial in other ways in these patients.

CONCLUSION

The results of this study demonstrated the ability of vitamin E to revert diabetes-induced aortic remodeling through the modulation of α-SMA and eNOS expression.

CONFLICT OF INTERESTS

There are no conflicts of interest.

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الملخص العربى

دراسة نسيجية ومناعية كيميائية حول التاثير المحتمل لفيتامين على إعادة تشكيل جدار الأبهر في الجرذان المصابة بداء السكري المستحث بالستربتوزوتوسين

جوزيف امين عزيز

قسم التشريح والأجنة، كلية الطب، جامعة الزقازيق

الخلفية: سيصل عدد مرضى السكرى بحلول عام ٢٠٢٥ الى ثلثمائة مليون. ضعف الأوعية الدموية هو نتيجة شائعة لكل من مرض السكري وعدم تحمل الجلوكوز. تساهم تداعيات إرتفاع السكر في الدم على البطانة وخلايا العضلات الملساء في زيادة معدل الإصابة بأمراض القلب والأوعية الدموية في مرض السكري من النوع الاول والثانى على السواء.

الهدف من البحث: كان الغرض من هذه الدراسة هو التحقق من التاثير المناعى المحتمل لفيتامين (هـ) على إعادة تشكيل جدار الأبهر فى الجرذان المصابة بداء السكرى المستحث بالستربتوزوتوسين من الناحية النسيجية والنسيجية المناعية والقياسات الشكلية.

المواد والطرق: تم تقسيم ثلاثين ذكرا من جرذان الوستار البيضاء بشكل متساو الى ثلاث مجموعات: المجموعة الضابطة التى أعيد تقسيمها بالتساوى الى مجموعتين فرعتين (تلقت احداها حقنة واحده من ٠,١ مل من محلول سيترا ت داخل الصفاق والاخرى ٢ مل من زيت الذرة عن طريق التزقيم الفمو ى). مجموعة داء السكر ى (تم حقنها داخل الصفاق ب ٦٥ مجم لكل كجم من الستربتوزوتوسين فى من ٠,١ مل من محلول سي ترات ومجموعة داء السكرى مع فيتامين هـ أعطيت فيتامين ه) ٣٠٠ مجم لكل كجم لمدة ١٠ اسابيع بعد احداث السكرى). عينات الاورطى تم صباغتها بصبغات الهيماتوكسيلين والايوسين والماسون تراى كروم والاورسين. تم معالجه بعض المقاطع للتقييم المناعى لإنزيم أكسيد حمض الن يتريك البطانى وأكتين ألفا العضلات الملساء.

النتائج: فيتامين هـ أصلح التغيرات النسيجية التي يسببها مرض السكري. قلّل بشكل كبير من سماكة وسائط الغلالة والكولاجين وخلايا العضلات الملساء؛ كما أدى فيتامين هـ أيضًا إلى تحسين تدمير الإيلاستين وتعزيز تعبير إنزيم أكسيد حمض النيتريك البطانى في الخلايا البطانية وقمع النشاط المناعي لأكتين ألفا العضلات الملساء في جدار الشريان الأورطي في الفئران المصابة بداء السكري.

الخلاصة: أدى فيتامين هـ إلى تحسين إعادة التشكيل النسيجي لجدار الأبهر في الفئران المصابة بداء السكري والتي تم تأكيدها من الناحية النسيجية والمناعية الكيميائية من خلال تنظيم تعب ير إنزيم أوكسيد حمض النيتريك البطانى وأكتين ألفا العضلات الملسا ء