Role of Angiogenesis as a Factor Modulating the Course of Cardiovascular Complications in Diabetic Rats

AKEF KHOWEILED, M.D.; HANY EL-SEBAEE, M.D.; SAMAH EL-ATTAR, M.D. and MOHAMED MANSOUR, M.D.
The Department of Physiology, Faculty of Medicine, Cairo University

Abstract

Background: Angiogenesis is an adaptive response that occurs in hypoxic and ischemic states as a compensatory mechanism and is modified by "on" switches as regular exercise and nitric oxide administration, and "off" switches as angiostatin. The normal healthy body maintains a perfect balance of angiogenesis modulators. Some authors believe that angiogenesis is beneficial for the myocardial performance during the course of Type II Diabetes mellitus; through providing more adequate blood supply to the cardiac myocyte. Others believe that angiogenesis may lead to micro-aneurismal formation with subsequent rupture and bleeding.

Aim of the Work: To study the effects of angiogenesis occurring as an adaptive response in diabetes mellitus, on cardiac functions and whether its stimulation by exercise or NO donor is beneficial or harmful on the cardiovascular performance and to study the effect of its inhibition on some cardiac function parameters.

Material and Methods: Fifty male white Albino rats were divided into 5 groups Group 1: (control group) normal rats fed on standard chow, Group 2: (diabetic group) diabetes was induced by fructose enriched chow, Group 3: (diabetic + angiostatin) angiostatin was given in a dose 5 µg/kg each week for 4 weeks starting after 2 weeks from the start of the experiment, Group 4: (diabetic + exercise) exercise [1 hour of daily swimming divided into 4 periods of 15 minutes each, with a 5 minutes rest between each period] and Group 5: (diabetic + NO) L-Arginine was given at a dose 100mg/kg/day.

Results: The results showed a significant impairment in the cardiac performance in all the diabetic groups compared to the control group, and a significant increase in VEGF blood level and gene expression under the effect of exercise and nitric oxide supplementation, which was more marked under the effect of exercise, and a significant decrease in the VEGF blood level and gene expression under the effect angiostatin as compared to the control group. Correlation analysis revealed strong negative correlation between the insulin resistance and the cardiac performance parameters and positive correlation between the angiogenic markers [VEGF blood level and gene expression] and the cardiac performance parameters.

Conclusion: It can be concluded that stimulation of angiogenesis has a beneficial effects on the cardiovascular function in cases of DM, and that exercise exerts additional beneficial effect through improving insulin sensitivity.

Key Words: Angiogenesis – Diabetes mellitus – Cardiac function – exercise – Nitric oxide – Angiostatin.

Introduction

ANGIOGENESIS is a natural process in the body that involves the growth of new blood vessels. It occurs naturally at a certain rate, however, this rate may increase or decrease in certain pathological conditions [1].

Angiogenesis is an adaptive response that occurs in hypoxic and ischemic states as a compensatory mechanism and is modified by several factors including regular exercise and nitric oxide administration. The main "On" switches are known as angiogenesis-stimulating growth factors. The most important of them is vascular endothelial growth factor [VEGF] [2]. The main "off" switches are known as angiogenesis inhibitors. The most important of them are angiostatin, interferons and TGF-Beta [Transforming growth factor Beta]. The normal healthy body maintains a perfect balance of angiogenesis modulators [3].

Some authors believe that angiogenesis is beneficial for the myocardial performance during the course of Type II Diabetes mellitus; through providing more adequate blood supply to the cardiac myocyte, allowing better contractility and cardiovascular performance [4].

Others believe that angiogenesis may lead to micro-aneurismal formation with subsequent rupture and bleeding. This micro hemorrhage may organize into fibrous tissue surrounding the cardiac
myocyte decreasing the contractile functions and impairing cardiovascular performance [5].

Exercise training improves cardiovascular function and increases vascular transport capacity of skeletal muscle. In human subjects exercise training for as little as 4 weeks has been reported to increase blood flow capacity (BFC) as measured by reactive hyperaemic responses to occlusion of blood flow to the forearm [6]. Furthermore, exercise-induced VEGF expression and angiogenesis as shown by greater density of capillaries and arterioles in skeletal muscles in mice skeletal muscles after exercise training [7,8].

Also, NO supplementation has been recorded to improve the angiogenic response, increase the expression of VEGF, and improve perfusion in skeletal and cardiac muscles [9].

**Aim of the work:**

To study the effects of angiogenesis occurring as an adaptive response in diabetes mellitus, on cardiac functions and whether its stimulation by exercise or NO donor is beneficial or harmful on the cardiovascular performance and to study the effect of its inhibition on some cardiac function parameters.

**Material and Methods**

Fifty male white Albino rats of body weight ranging from 150-200gm were supplied by the animal house unit of Kasr El-Ainy, Faculty of medicine, Cairo University, 2010-2011. The animals had free access to food and water all through the day and were housed with normal light dark cycles and were divided into 5 groups, each group was composed of 10 rats, 4 groups in which type II diabetes mellitus was induced, and one group was designed to be the control group.

Group 1: (control group) normal rats Standard chow contains 75% of its caloric intake as carbohydrates obtained from the animal house.

Group 2: (diabetic group) Induction of Type II diabetes was done by fructose enriched chow contained 60% fructose, 21% proteins, 5% fat and 8% cellulose [10]. Fructose was manufactured by (Panreac Quimica SA , Spain).

Group 3: (diabetic + angiostatin) Type II diabetes was induced by fructose together with an inhibitor of angiogenesis [angiostatin] in a dose 5 g g/kg each week for 4 weeks starting after 2 weeks from the start of the experiment [11].

Group 4: (diabetic + exercise) Type II diabetes was induced by fructose rich feeding with exercise [1 hour of daily swimming divided into 4 periods of 15 minutes each, with a 5 minutes rest between each period] [12] as an inducer of angiogenesis.

Group 5: (diabetic + NO) Type II diabetes was induced by fructose, together with a nitric oxide donor [L-Arginine] at a dose 100mg/kg/day [13] to study the role of nitric oxide as an inducer of angiogenesis. L-Arginine was purchased from Sigma Chemical Co.(St. Louis, Missouri [MO], USA) in the form of powder to be dissolved in the drinking water as a vehicle.

In all the study groups, the following parameters were measured after 6 weeks of experimental period.

- Measurement of CVS functional parameters; Left ventricular developed pressure [LVDP] – Dp/Dt – Heart rate [HR].
- Measurement of serum glucose and insulin levels and calculation of Homa test as an index of insulin sensitivity.
- Measurement of blood VEGF as an indicator of angiogenesis and VEGF gene expression in cardiac tissue.

Blood samples were taken for the measurement of blood glucose, insulin and VEGF level, after which the rats were sacrificed and the cardiac tissue was dissected for the study of physiological parameters and a piece was sent for the biochemical analysis of VEGF gene expression.

**Blood sampling:**

Blood samples were withdrawn from all rats through the retro-orbital route and serum was separated and stored at 70ºC until used. The serum was divided into 3 tubes for further determination of serum levels of glucose, insulin and VEGF.

**Heart perfusion conditions:**

After taking blood samples, all animals were heparinized with 100 U of heparin (i.p.). Animals were then anesthetized with 10mg/Kg sodium pentobarbitone (i.p.). The hearts were excised and placed in ice-cold Krebs-Henseleit Bicarbonate (KHB) buffer. The aorta was cannulated with an 18-gauge plastic cannula, in a non circulating Langendorff apparatus, with modified Krebs Henseleit solution at a constant flow rate of 12ml/min. The perfusate solution consists of the following (mmol/l): NaCl (116), NaHCO 3 (25), CaCl 2 (205), MgSO 4 (102), KCl (407), KH 2PO 4 (102) and glucose (5.5).
The perfusate was oxygenated with 95% O₂ and 5% CO₂ gas mixture to maintain a PO₂ of >40mmHg. A latex balloon-tipped catheter was inserted into the left ventricle through the mitral annulus and inflated with distilled water (0.15-0.3ml) to set an end diastolic pressure of 2mmHg during the initial equilibration.

The distal end of the catheter was connected to a polygraph (san-ei, made in Japan) for recording the different haemodynamic parameters, via pressure transducer, according to the experimental design.

Contractile function was assessed by measuring the following parameters:
- Heart rate/min (HR).
- Left ventricular developed pressure (LVDP) which is defined as peak systolic minus end-diastolic pressure.
- The maximum rate of pressure rise (AP/AT).

**Measurement of serum insulin:**
Insulin concentrations were measured by enzyme immunoassay using the rat insulin enzyme linked immunosorbant assay (ELISA) kit (Linco Research, USA).

**Measurement of serum glucose:**
The blood glucose was assayed by the method adopted by [14]. The test materials for this method were supplied as kits by “Diamond Diagnostics, Massachusetts, USA”.

**Homeostasis model assessment of insulin resistance (HOMA-IR):**
HOMA is an indirect method for the assessment of insulin resistance. It depends on relationship between fasting plasma glucose and insulin based on a mathematical model:

\[ \text{HOMA-IR: } \left[ \frac{\text{fasting plasma glucose (mmol/L)}}{\text{fasting plasma insulin (uIU/ml)}} \right] \times 22.5 \]

In cases when HOMA is more than 4.0 this is diagnostic of insulin resistance [16].

**Measurement of VEGF blood level:**
Quantitative sandwich enzyme immunoassay technique. The intensity of the color measured is in proportion to the amount of VEGF bound in the initial step. The sample values are then read off the standard curve.

**Measurement of VEGF gene expression:**
For the detection of VEGF, RNA was extracted, reverse transcribed into cDNA, and amplified by PCR. The E.Z.N.A.® Tissue RNA Kits, supplied by Sigma chemicals Co [Missouri (MO), USA], use the reversible binding properties of HiBind® matrix, a new silica-based material.

The PCR products were semiquantitated using the gel documentation system (Bio Doc Analyze) supplied by Biometra.

**Statistical analysis:**
Data was coded and entered using the statistical package SPSS (version 15). Data was summarized using mean and standard deviation for quantitative variables. Comparisons between groups were done using analysis of variance (ANOVA) and multiple comparisons (post Hoc test) for quantitative variables while non-parametrical (Kruskal-Wallis test) and (Mann-Whitney test) were used for quantitative variables not normally distributed. Correlations were done to test for linear correlations between quantitative variables. p-values <0.05 were considered statistically significant.

**Results**

Table (1) and Fig. (1) show that LVDP is reduced in the diabetic group (Group 2) compared to the control group (Group 1). There is also a significant reduction in the LVDP in the group treated with angiostatin (Group 3), compared to the control group and the diabetic group (Groups 1,2).

The LVDP values are higher in the groups with stimulated angiogenesis, with the highest developed pressure in group 4 (exercise group) which was most close to the control group.

Regarding the contractility, the results showed a significant increase in contractility in the groups 4&5, with a significant increase marked in the exercise group (Group 4) as compared to NO treated group (Group 5) (Table 1, Fig. 2).

The contractility was reduced in group 2 and 3, with a significant reduction in group 3 (the angiostatin group) as compared to diabetic group (Group 2).

Regarding the heart rate, the results show a significant reduction in all groups compared to the control group, signifying diabetes induced cardiomyopathy (Table 1, Fig. 3).

Table (2) and Figs. (4,5,6) displayed that there was a significant increase in the blood glucose level, insulin level and insulin resistance in all the diabetic groups compared to the control group.
However, there was a significant decrease in blood glucose, insulin levels and insulin resistance in group 4 (exercise group) compared to group 2 (diabetic group).

The results of the present study shown in the Table (3), Figs. (7,8) displayed a significant increase in VEGF blood level and gene expression under the effect of exercise and nitric oxide supplementation (Groups 4,5), which was more marked under the effect of exercise, and a significant decrease in the VEGF blood level and gene expression under the effect of angiogenesis inhibitor angiostatin as compared to the control group.

Correlation analysis revealed positive correlation between the angiogenic markers (VEGF blood level and gene expression) and the cardiac performance parameters (Figs. 9, 10,11).

Also correlation analysis revealed strong negative correlation between the insulin resistance and the cardiac performance parameters (Figs. 12,13).

Table (1): Measurement of LVDP, dp/dt and heart rate in different groups.

<table>
<thead>
<tr>
<th></th>
<th>LVDP</th>
<th>dp/dt</th>
<th>HR</th>
</tr>
</thead>
<tbody>
<tr>
<td>mmHg</td>
<td>mmHg/sec</td>
<td>Beats/min</td>
<td></td>
</tr>
<tr>
<td>Control group</td>
<td>96.8000±6.44291+</td>
<td>77.0000±6.87992+</td>
<td>192.9000±4.84080+</td>
</tr>
<tr>
<td>Diabetic group</td>
<td>50.9000±8.68524* D 48%</td>
<td>43.1000±8.31932* D 44%</td>
<td>130.9000±5.60653* D 32%</td>
</tr>
<tr>
<td>Diabetic + angiostatin</td>
<td>25.6000±5.6112** D 73%</td>
<td>19.2000±4.15799** D 75%</td>
<td>100.2000±7.58361** D 47%</td>
</tr>
<tr>
<td>diabetic + exercise</td>
<td>90.3000±7.0872** D 6%</td>
<td>68.7000±6.32543** D 11%</td>
<td>179.7000±9.66149** D 7%</td>
</tr>
<tr>
<td>diabetic + NO</td>
<td>69.5000±7.50185** D 28%</td>
<td>54.9000±4.88648** D 29%</td>
<td>151.6000±8.01664** D 23%</td>
</tr>
</tbody>
</table>

*: Significant p as compared to group I (p<0.05).
+: Significant p as compared to group II (p<0.05).
% Change. D: Decrease. I: Increase.

Table (2): Blood glucose, insulin and insulin resistance in different groups.

<table>
<thead>
<tr>
<th></th>
<th>Blood glucose [mmol/dl]</th>
<th>Insulin [uIU/dl]</th>
<th>Insulin resistance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control group</td>
<td>4.6060±0.78262+</td>
<td>10.1180±1.23052+</td>
<td>2.0630±0.39480+</td>
</tr>
<tr>
<td>Diabetic group</td>
<td>11.7600±2.11693*</td>
<td>28.3640±2.25897*</td>
<td>14.8350±3.08873*</td>
</tr>
<tr>
<td>Diabetic + angiostatin</td>
<td>10.4370±1.82809*</td>
<td>31.3110±1.98999*</td>
<td>14.6910±3.50216*</td>
</tr>
<tr>
<td>diabetic + exercise</td>
<td>7.7910±0.94504**</td>
<td>20.9850±1.54174**</td>
<td>7.3010±1.29083*</td>
</tr>
<tr>
<td>diabetic + NO</td>
<td>9.9500±1.69948*</td>
<td>29.2360±2.46791 *</td>
<td>13.0590±3.10622*</td>
</tr>
</tbody>
</table>

*: Significant p as compared to group I (p<0.05).
+: Significant p as compared to group II (p<0.05).
Insulin resistance [Calculated by HOMA, >4 signify IR].

Table (3): Measurement of VEGF blood level [pg/ml] and VEGF gene expression in different groups.

<table>
<thead>
<tr>
<th></th>
<th>VEGF [pg/ml]</th>
<th>% change</th>
<th>VEGF gene</th>
<th>% change</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control group</td>
<td>22.9300±4.06531*</td>
<td>1.0140±0.23950*</td>
<td>1.18%</td>
<td>1.0800±0.26293*</td>
</tr>
<tr>
<td>Diabetic group</td>
<td>26.6100±5.10696*</td>
<td>1.0800±0.26293*</td>
<td>1.63%</td>
<td>0.1210±0.06790*</td>
</tr>
<tr>
<td>Diabetic + angiostatin</td>
<td>9.7200±1.15451 ++</td>
<td>2.1400±0.36237+</td>
<td>1.41%</td>
<td>1.8090±0.19376+</td>
</tr>
<tr>
<td>diabetic + exercise</td>
<td>36.1800±4.10333 ++</td>
<td>2.1400±0.36237+</td>
<td>1.41%</td>
<td>1.8090±0.19376+</td>
</tr>
<tr>
<td>diabetic + NO</td>
<td>31.7300±3.65211 ++</td>
<td>2.1400±0.36237+</td>
<td>1.41%</td>
<td>1.8090±0.19376+</td>
</tr>
</tbody>
</table>

*: Significant p as compared to group I (p<0.05).
+: Significant p as compared to group II (p<0.05).
D: Decrease. I: Increase.
LVDP (mmHg)

*: Significant \( p < 0.05 \) as compared to group I
+: Significant \( p < 0.05 \) as compared to group II

Fig. (1): LVDP among different groups (mmHg).

dp/dt mmHg/sec

*: Significant \( p < 0.05 \) as compared to group I
+: Significant \( p < 0.05 \) as compared to group II

Fig. (2): \( \text{dp/dt} \) among different groups (mmHg/s).

Insulin (uIU/dl) HR Beats/min

*: Significant \( p < 0.05 \) as compared to group I
+: Significant \( p < 0.05 \) as compared to group II

Fig. (3): Heart rate among different groups (BPM).

Blood glucose (mmol/dl)

*: Significant \( p < 0.05 \) as compared to group I
+: Significant \( p < 0.05 \) as compared to group II

Fig. (4): Blood glucose among different groups (mmol/dl).

Insulin resistance

*: Significant \( p < 0.05 \) as compared to group I
+: Significant \( p < 0.05 \) as compared to group II

Fig. (5): Blood Insulin among different groups (uIU/ml).

Fig. (6): Insulin resistance among different groups.
Role of Angiogenesis as a Factor Modulating the Course of Cardiovascular Disease

Fig. (7): Blood VEGF level among different groups (Pg/ml).

Fig. (8): VEGF Gene expression among different groups.

Fig. (9): Correlation scatter blot between LVDP and VEGF Gene expression. $r = 0.49$.

Fig. (10): Correlation scatter blot between VEGF gene expression and Dp/Dt. $r = 0.47$.

Fig. (11): Correlation scatter blot between VEGF gene expression and Insulin resistance. $r = -0.07$.

Fig. (12): Correlation scatter blot between Insulin resistance and Dp/Dt. $r = -0.72$. 
Insulin resistance

\[
\begin{array}{|c|}
\hline
\text{IR} \\
\text{Linear (IR)} \\
\hline
\end{array}
\]

Strong negative correlation between the degree of insulin resistance and the LVDP.

Conclusion: Insulin resistance depresses cardiac contractility.

Fig. (13): Correlation scatter blot between Insulin resistance and LVDP. \( r = -0.74 \).

**Discussion**

Data of the present work demonstrated that relative to normal control group I, cardiac mechanical performance was significantly reduced in type 2 diabetic hearts (groups II to V). A significant decrease in HR, LVDP and dp/dt was recorded signifying that isolated hearts from type II diabetic rats exhibited a certain degree of diabetic cardiomyopathy and heart failure.

The results of the present study are in agreement with those of several authors who reported similar findings in diabetic rats [17,18]. Indeed, alterations of diastolic and systolic function are widely reported in healthy diabetic subjects and often predict the development of other chronic diabetic complications [19].

Alterations of cardiac sympathetic innervation, tone, and responsiveness, coupled with abnormal myocardial blood flow regulation, have emerged as potential contributing factors [20]. Also reduced phosphatidyl-inositol (PI) 3-kinase signalling in the heart and decreased L-type Ca\(^{2+}\) current density in diabetic myocytes as compared to control non diabetic were reported as an aetiology of cardiomyopathy reported in diabetes [21].

Decrease in L.V.D.P. might be due to increased collagen deposition or increased glucose-mediated collagen cross-linking in diabetic mice independent of myocyte pathology as reported by [22].

Also, hyperglycemia is associated with an increase in oxidants. The mechanism whereby hyperglycemia mediates tissue injury through the generation of reactive oxygen species has been elucidated largely through the work of Crook et al. [23].

Recently, both type 1 and type 2 diabetes have been shown to affect angiogenic growth factors and inhibitors in skeletal muscle. Previous studies show that there is marked imbalance involving several pro-angiogenic proteins and anti-angiogenic ones in mouse skeletal and cardiac muscles [24].

The change in balance between stimulators and inhibitors may be one of the reasons for the markedly increased risk for peripheral cardiovascular complications in diabetes. There is a controversy between authors regarding the net effect of angiogenesis, and whether it is beneficial or harmful for the myocardial function [25].

The results of the present study showed that exercise improved insulin sensitivity and decreased the degree on insulin resistance as well as reduces blood glucose level. Also correlation analysis revealed strong negative correlation between the insulin resistance and the cardiac performance parameters.

These results come in agreement with Kirwan et al. [26] and Chibalin et al. [27].

Exercise training leads to increased expression of glucose transporter 4 (GLUT-4) content in skeletal muscle, however, the improvements in insulin sensitivity after exercise training may be related to changes in expression and/or activity of proteins involved in insulin signal transduction in skeletal muscle [27]. These effects include increasing the expression of insulin receptor substrate molecules [IRS] especially IRS-1 and IRS-2, with increasing its phosphorylation and activity [26].

Our work revealed that a significant increase in the angiogenesis in exercise or NO treated groups, while the increase is mostly marked in the exercise group.

These data are consistent with those of previous studies showing that capillary density was increased in the hypertrophic left ventricle by exercise training as shown by Ziada et al., 2005 [28], who showed that exercise increased the myocardial contractility, improved cardiovascular parameters and increased the neovascularisation and capillarization of the cardiac muscles.

Also the results are consistent with Binder et al. [8] and Laughlin et al. [29] who investigated the effect of regular exercise on arteriolar and capillary density and found significant increase in both the arteriolar and capillary density in skeletal and
cardiac muscles after regular exercise, and proved that arteriolar and capillary remodelling is the main mechanism underlying the increase in blood flow post exercise.

As shown from the results of this work that blood VEGF and its gene expression in cardiac muscle were significantly increased in diabetic rats underwent exercise training as compared to the diabetic group. It is believed that the hypoxic conditions, including those induced by exercise are potent stimuli for angiogenesis, together with the shear stress induced by the marked increase in the blood flow during exercise, which contributes to the upregulation of proangiogenic mechanisms and the increased expression of proangiogenic growth factors [7,8].

Guifu et al. [30] reported upregulation of VEGF occurs in rat skeletal muscle following chronic muscular exercise or with a single bout of moderately intense treadmill running. The mechanism involved is that muscle VEGF contents may be mobilized as VEGF increases in the interstitium and released by active muscles.

These results are consistent with those of Guifu et al. [30], Junichi Suzuki et al. [31], and Matsunaga et al. [32].

The present work demonstrated that diabetic cardiomyopathy and heart failure was improved by the induction of angiogenesis and deteriorated by the inhibition of angiogenesis. Correlation analysis revealed strong positive correlation between the VEGF blood level and gene expression and the cardiac performance. This signifies that the angiogenic response is beneficial for the cardiac performance in the cases of diabetes mellitus and helps to improve the myocardial performance.

These results come in agreement with Riika et al., 2008 [33] who concluded that the angiogenic response is increased in diabetic subjects and is associated with better muscular and myocardial performance.

Moreover, Petra et al., [34] proved that the pharmacologically induced angiogenic response improved the muscular and myocardial performance and improved the ischemic state in diabetic rabbits. The mechanism by which the angiogenic response is augmented in diabetes mellitus involves the up-regulation of the mRNAs of some extracellular proteins, which have proangiogenic properties, which is up-regulated in diabetic muscles and, thus, may induce compensatory protective events.

However, other investigations into expression of VEGF in mice models revealed deleterious effects of stimulation of angiogenesis due to the formation of leaky immature vessels/hemangiomas and subsequent death of the experimental animal [35,36]. Furthermore, transgenic mice over-expressing VEGF revealed lengthy and leaky dermal vessels with evident inflammation [36,37].

Our results show that nitric oxide (NO) is a mediator of angiogenesis and that vascular endothelial growth factor (VEGF) is stimulated by NO.

These results are in consistent with results of Van Der Zee et al., [38] who reported increased VEGF release from cultured human umbilical venous endothelial cells and upregulation by the more expression of nitric oxide synthase (NOS). VEGF also increases the release of NO, starting a vicious circle that leads to augmentation of angiogenesis.

Also, Babaei et al., [39] reported that the release of NO plays a critical role in the angiogenic actions, and angiogenesis is attenuated when NO bioactivity is reduced. The angiogenic response to ischemia is impaired in the eNOS-deficient mice, an effect that cannot be reversed by administration of recombinant VEGF protein or adenovirus-mediated VEGF gene transfer [40].

The results of the present study proves that NO donor (L-Arginine) improved the angiogenic response, reduced the cardiovascular complications and improved the myocardial performance without affecting the insulin sensitivity.

These results come in agreement with Junichi, 2005 [31], Kapila et al. [9], who proved that NO supplementation achieved promising results improving the angiogenic response, increased the expression of VEGF, improves perfusion and functional parameters and the muscular and myocardial contractility in rats.

The mechanisms by which NO promotes angiogenesis involves multiple mechanisms. NO is an endothelial survival factor, inhibiting apoptosis and enhancing endothelial cell proliferation, by increasing the expression of VEGF. NO also enhances endothelial migration by stimulating endothelial cell podokinesis [41]. Finally, the hemodynamic effects of this potent vasodilator may play a role in its angiogenic effects [42].

Another mechanism by Matsunaga et al., [43] is that NO may suppress the production of angiotatin, which is the most potent endogenous antagonist of angiogenesis.
Of relevance to this discussion is the existence of an endogenous antagonist to NO synthase. Asymmetric dimethyl arginine (ADMA) is an arginine analogue that competes with L-arginine for NOS. Plasma ADMA levels are elevated in animals and humans diabetes mellitus. In experimental animal models, endogenous or exogenously elevated plasma ADMA levels are associated with impaired angiogenic response to ischemia, an effect that is reversed by supplemental L-arginine [44].

In this study, it was also found that there is a significant decrease in the VEGF blood level and gene expression under the effect of angiogenesis inhibitor angiostatin.

Our results are in agreement with results of Yamahara et al. [45], who reported that there is a harmful effect for angiostatin treatment in cases of heart failure and this pathological effect is through inhibiting angiogenesis.

From the above results, it can be concluded that there was significant impairment in the cardiac performance in all the diabetic groups compared to the control group. However angiogenesis induction either by exercise or nitric oxide significantly improved the cardiac performance especially in the exercise group. Moreover the inhibition of angiogenesis further depressed the cardiac performance to the lowest values.

From the above discussion it can be concluded that stimulation of angiogenesis has a beneficial effects on the cardiovascular function in cases of DM, and that exercise exerts additional beneficial effect through improving insulin sensitivity.

References


Role of Angiogenesis as a Factor Modulating the Course of Cardiovascular Disease


