25-Dihydroxyvitamin D3 Alleviates Diabetic Vascular Endothelial Injury by Inhibiting Proline Isomerase-1 Mediated Mitochondrial Oxidative Stress and Inflammation

Abstract: The incidence of diabetes is increasing, and diabetes has become one of the difficult problems in the medical field. The hyperglycemic environment in diabetic patients can lead to the damage of VEC(vascular endothelial cells) and various abnormal reactions of vascular physiological functions, which can cause diabetic vascular complications and damage the body of diabetic patients. High glucose can up regulate the protein expression of Pin1 (proline isomerase-1) and damage the function of vascular endothelium. Faced with this situation, this paper studied the inhibition of Pin1 by 25-dihydroxyvitamin D3. Studies have shown that 25-dihydroxyvitamin D3 can significantly alleviate the damage of vascular endothelial cells (VEC), and can inhibit the expression level of Pin1 and the phosphorylation level of p66Shc (an oxidative adaptor protein). It can reduce ROS (Reactive Oxygen Species) level and CASP3 (caspase-3) level in vascular endothelial cells. 25-dihydroxyvitamin D3 can inhibit the expression level of Pin1 protein by 9.02% by activating VDR (vitamin D receptor).

Keywords: 25-Dihydroxyvitamin D3; Proline Isomerase-1; Drug Delivery; Diabetic Vascular Endothelial Injury

1. Introduction

As a chronic metabolic disease, diabetes mellitus patients' high glucose environment would cause vascular endothelial damage, which would lead to a series of vascular complications, seriously affecting the life safety and well-being of diabetic patients. In recent years, some studies believe that the lack of VitD (vitaminD) is also one of the factors that cause diabetes. VitD can play a role in relieving vascular endothelial damage in diabetes, but there are few studies related to it. Therefore, this paper studied the topic that 25-dihydroxyvitamin D3 alleviates diabetic vascular endothelial damage by inhibiting mitochondrial oxidative stress and inflammation mediated by proline isomerase-1, so as to alleviate diabetic vascular endothelial damage and ensure the life safety and well-being of diabetic patients.

The study on vascular endothelium and injury of diabetes is beneficial to reduce the incidence of diabetes and to the treatment of diabetic patients. Many scholars have studied this. Zhang, Jinglong believed that endothelial cell inflammation plays a key role in the pathogenesis of type 2 diabetic vascular disease. He discussed the role and molecular mechanism of the activation of NLRP3 (NOD like receptor thermal protein domain associated protein 3) inflammatory bodies induced by low diplococcemia in diabetic vascular endothelial dysfunction [1]. Knapp, Maura elaborated the symptoms of diabetic cardiomyopathy, the potential mechanism of diabetic cardiomyopathy and the mechanism of endothelial dysfunction of diabetic cardiomyopathy. He believed that diabetic vascular endothelial dysfunction may be one of the main causes of diabetic cardiomyopathy [2]. Liu, Yi believed that vascular endothelial injury was the beginning of huge and microvascular system dysfunction in diabetic patients, which would increase the risk of diabetic vascular complications [3]. Jamwal, Shalini discussed the role of vascular endothelium in transporting nutrients and drugs, regulating angiogenesis, increasing vascular tension and permeability, and concluded that diabetes can lead to vascular endothelial dysfunction and damage the integrity of vascular endothelium through analysis [4]. Dong, Yunzhou believed that vascular endothelial injury was a sign of atherosclerosis in diabetic patients [5].

Petrie, John R discussed diabetes and hypertension as complications, focusing on the relationship between endothelial dysfunction, vascular inflammation, oxidative stress and the two diseases, and put forward some views on the current treatment of diabetes and cardiovascular complications [6]. The above scholars have explored the diabetic vascular endothelium and its damage.

Many scholars have studied 25-dihydroxyvitamin D3. Wu, Meifang explored the protective effect of 25-dihydroxyvitamin D3 on human blood vessels and related protective mechanisms. By measuring the relevant indicators of vascular endothelial cells, the conclusion is that 25-dihydroxyvitamin D can reduce the oxidative damage of vascular endothelium induced by high glucose by up regulating Nrf2 (nuclear factor erythroid 2 related factor 2) antioxidant pathway in a VDR (vitamin D receptor) dependent manner [7]. Kim, Il Young used logistic regression analysis to study the correlation between 25-dihydroxyvitamin D and aortic valve calcification, mitral valve calcification and baseline variables in patients with chronic kidney disease [8]. Zeng, Xiaoyun believed that 25-dihydroxyvitamin D3 can interact with VDR, reduce glucose level in blood, and participate in gene regulation in various biological processes [9]. Faridvand, Yousef believed that 25-dihydroxyvitamin D3 could activate the human body's Aperin system and inhibit the production of inflammatory mediators caused by lipopolysaccharide [10]. Tian, Li-Qiang explored the role of 25-dihydroxyvitamin D3 in preventing diabetes, and proved through mouse experiments that 25-dihydroxyvitamin D3 can effectively improve inflammation in blood vessels of type 2 diabetic mice, thereby reducing the incidence of biological diabetes [11]. Jia, Tingting believed that advanced glycosylation end products were potential markers of inflammation level in diabetic patients with periimplant inflammation. Through experimental studies, they proved that 25-dihydroxyvitamin D3 could effectively improve the damage of osteoblastic differentiation caused by advanced glycosylation end products. 25-dihydroxyvitamin D3 can be used as an auxiliary treatment for poor bone integration in patients with type 2 diabetes [12]. To sum up, many scholars have participated in the research on 25-dihydroxyvitamin D3.

In order to alleviate the vascular endothelial injury in diabetes mellitus and ensure the life safety of diabetic patients, this paper carried out an experimental study on 25-dihydroxyvitamin D3 to alleviate the vascular endothelial injury in diabetes mellitus. In this paper, the effects of 25-dihydroxyvitamin D3 on the inhibition of Pin1 expression, the effects of 25-dihydroxyvitamin D3 on p66Shc phosphorylation, and the effects of 25-dihydroxyvitamin D3 on ROS (reactive oxygen species) and CASP3 levels in diabetic vascular endothelial cells were investigated. Finally, the relevant conclusions are drawn.

2. Overview of Diabetes Related Factors and Mechanism of Vascular Endothelial Injury

(1) Brief description of diabetes related factors

Diabetes is a disease caused by factors such as genetics, immunity, diet, and lifestyle habits.

1) Etiology of diabetes

Genetic factors are one of the important influencing factors of diabetes, and diabetes has obvious genetic susceptibility. This means that if a family member suffers from diabetes, other family members have a higher risk of diabetes than the normal population. Immune factors are also factors that affect the onset of diabetes. The decline in the activity of human immune cells and defects in the immune system would increase the risk of diabetes. Dietary factors are closely related to the generation of diabetes. Excessive intake of high sugar, high oil and high fat foods would lead to obesity and reduce the sensitivity of the human body to insulin. Bad living habits such as overeating, refusing to exercise and staying up late would affect the normal absorption of sugar by the body and induce diabetes, as shown in Figure 1.

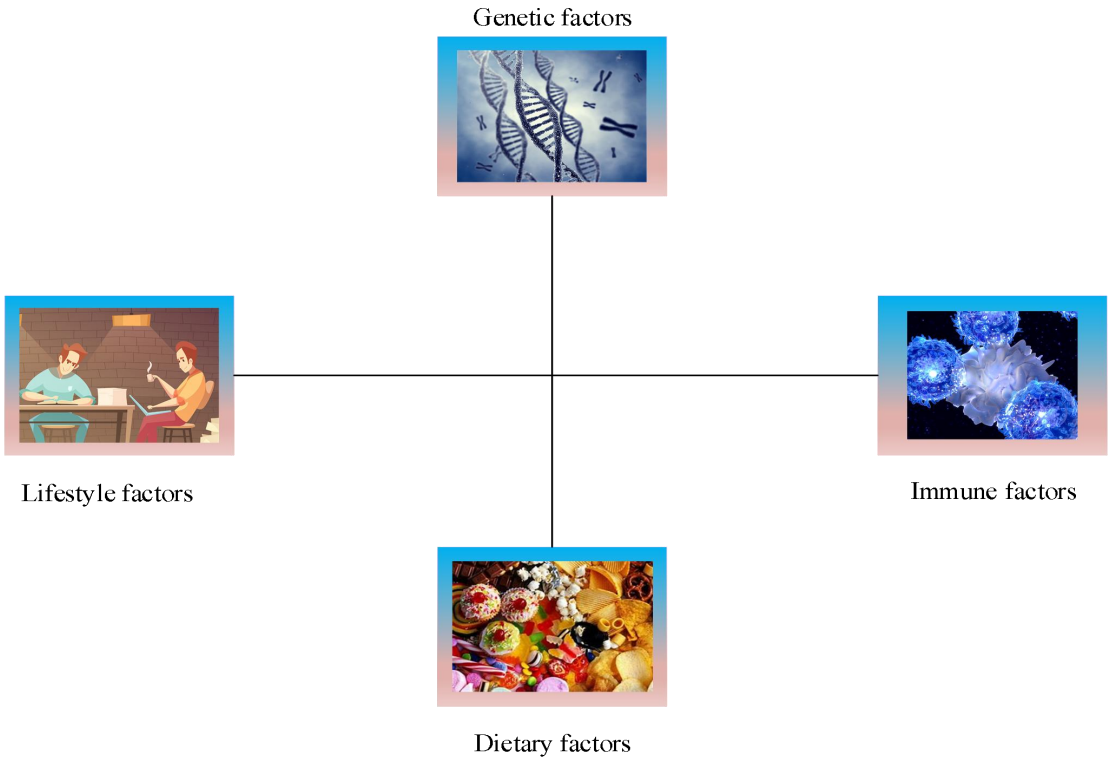

Figure 1: Causes of diabetes

2) Symptoms

The symptoms of polydipsia, polyuria, polydipsia, and emaciation are common in type 1 diabetic patients. Fatigue and weakness, obesity before the onset of diabetes, this symptom is more common in patients with type 2 diabetes.

3) Inspection method

Blood glucose detection: When there are symptoms of polydipsia, polyuria and polydipsia, the patient can be diagnosed as having diabetes as long as an abnormal blood glucose value is detected once. For asymptomatic patients, abnormal blood glucose values need to be detected twice. The 75g glucose tolerance test was conducted for the suspicious person who was difficult to diagnose whether there was diabetes. Urine glucose detection: Urine glucose detection is an auxiliary method for diabetes detection. Urine ketone body detection: it could check acetone, acetoacetic acid and other intermediate products of fat metabolism in the body. In addition, glucose detection methods include serum insulin and C-peptide level detection, blood lipid detection, immune index detection, urine albumin excretion detection, etc. After the diabetes test, the doctor can judge whether the patient has diabetes according to the test results, and formulate a diagnosis and treatment plan for the diagnosed person in time.

4) Treatment

Diabetes treatment can be started from the following five aspects, as shown in Figure 2. Specifically, it includes knowledge mastery and psychological counseling, blood glucose self detection, drug delivery therapy, exercise therapy and diet therapy.

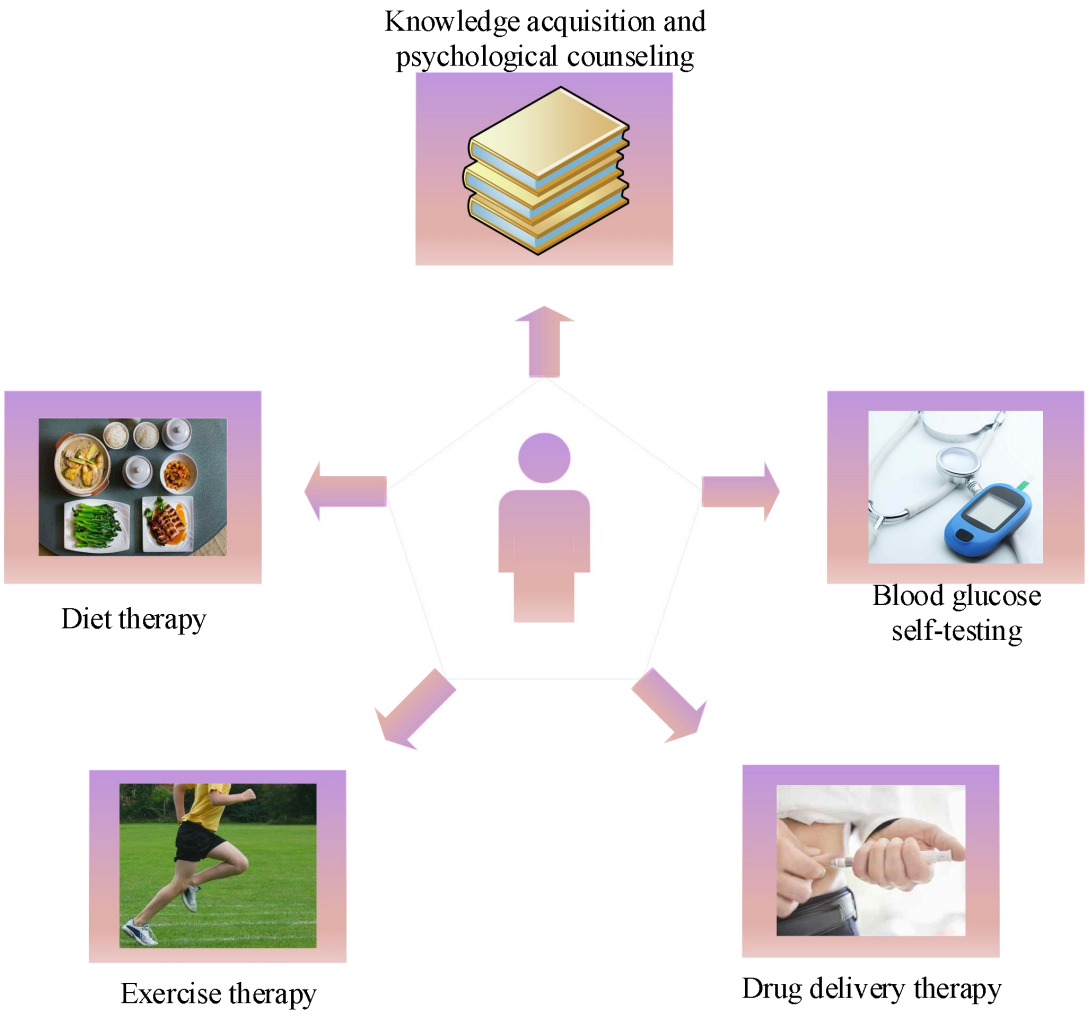

Figure 2: Treatment of diabetes

Knowledge acquisition and psychological counseling: In addition to formulating reasonable and effective treatment plans for diabetics, medical institutions should also teach diabetics to master relevant knowledge about diabetes and help them build confidence in diabetes treatment. Self detection of blood glucose: diabetes is a chronic disease. Diabetic patients should regularly detect their blood glucose before eating in their daily life. If blood glucose is unstable, the frequency of blood glucose self detection should be increased. Drug delivery therapy: drug therapy for diabetes includes oral drug therapy and insulin therapy. The commonly used oral drugs are sulfonylurea drugs, biguanide hypoglycemic drugs, α glucosidase inhibitors, insulin sensitizers, glinide insulin secretagogues, etc. Exercise therapy: The purpose of exercise therapy is to reduce the proportion of body fat, enhance the body's physical strength and immunity, increase the body's sensitivity to insulin, and thus improve the treatment effect. Diet treatment: diet is the basis for controlling and treating diabetes. For diabetics, the intake of sugar, calories, carbohydrates, protein, fat and other substances should be strictly controlled.

(2) Vascular endothelial damage in diabetic patients

VEC injury is the initial link of diabetic vascular disease. Effective measures should be taken to timely intervene the high glucose vascular endothelial injury, which is conducive to the prevention and treatment of diabetic vascular complications. In this paper, the mechanism of vascular endothelial damage in diabetes mellitus has been studied from the aspects of hyperglycemia, insulin resistance, lipid metabolism disorder, oxidative stress and inflammatory reaction, as shown in Figure 3.

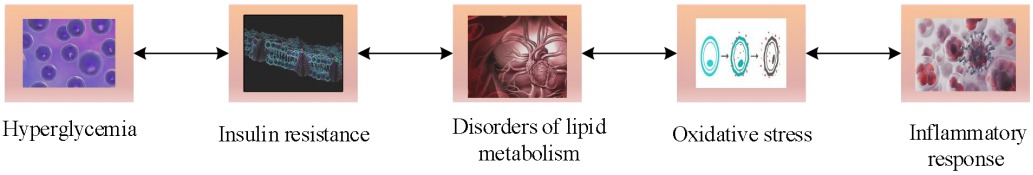

Figure 3: Vascular endothelial damage in diabetic patients

1) Hyperglycemia

Hyperglycemia can significantly affect the damage of vascular endothelial cells. Hyperglycemia can cause mitochondria to produce a large number of superoxide anions, leading to the increase of oxygen free radicals in the body, thus affecting the diastolic function of vascular endothelium. Hyperglycemia would also increase the expression level of diacetylglycerol, reduce the production of NO (nitric oxide), and reduce the production of cyclic guanosine, which would also affect the diastolic function of vascular endothelium. In addition, hyperglycemia can also induce apoptosis of VEC, resulting in the consequences of vascular endothelial dysfunction [13-14].

2) Insulin resistance

The normal level of insulin plays an important role in protecting the function of vascular endothelium. Insulin can protect the diastolic function of vascular endothelium by promoting the production of NO and improving the activity of NO synthase, and can directly interfere with vascular smooth muscle to mediate vasodilation [15]. However, for diabetic patients, long-term stimulation of abnormal levels of insulin would reduce the sensitivity of receptors on adipocyte membrane, inhibit the ability of cells to decompose fat, reduce the synthesis of NO, and cause damage to vascular endothelium. In addition, insulin resistance can also cause leakage and adhesion of vascular endothelium. Vascular endothelium is the target organ of insulin action. Insulin resistance and vascular endothelial damage interact.

3) Lipid metabolism disorder

Lipid metabolism disorder has an important impact on vascular endothelial damage [16]. Diabetic patients are accompanied by TC (high cholesterol), TG (high triglyceride), LDL (low density lipoprotein), HDL (low high density lipoprotein). TCemia would lead to the reduction of NO synthase activity, affect the production and release of NO, and also make NO synthase produce peroxidized free radicals, making NO lose biological activity. TG would affect the diastolic function of vascular endothelial cells in diabetic patients. LDL would increase the production of oxygen and make NO lose its activity, thus leading to dysfunction of vascular endothelial cells.

4) Oxidative stress

Oxidative stress can damage the vascular endothelium of normal people and diabetic patients. Oxidative stress has an important influence on the occurrence of diabetic vascular complications. The early manifestation of vascular injury and microvascular complications caused by oxidative stress is vascular endothelial dysfunction [17-18]. ROS contains O_2 (superoxide anion), H_2O_2 (hydrogen peroxide) and other oxygen-containing chemical reactive substances. The generation and development of ROS in diabetic patients would make NO lose biological activity, thereby affecting the vasodilation function of vascular endothelium.

5) Inflammatory reaction

C-reactive protein (CRP), as an inflammatory marker, has a high level in diabetic patients, especially in diabetic patients with vascular complications. The occurrence of diabetes and insulin

resistance can both improve the CRP level in the human body. CRP can cause dysfunction of vascular endothelium through related channels, resulting in vascular endothelial damage and diabetic vascular complications [19-20].

3. Experimental Method for 25 Dihydroxyvitamin D3 to Alleviate Vascular Endothelial Damage in Diabetes Mellitus

The experimental cultured human vascular endothelial cells were used to simulate the diabetic vascular environment with 33 mmol/L glucose concentration, and the vascular endothelial cells were divided into three groups. The first group of cells is called HG (high glucose) group because the glucose concentration is 33 mmol/L. The second group added 25-dihydroxyvitamin D3 with $10^{-8} \sim 10^{-6}$ mol/L concentration on the basis of 33 mmol/L glucose, which is called VitD3 (vitamin D3) group. The third group increased the concentration of Juglone with 10^{-7} mol/L on the basis of 33 mmol/L glucose, which is called Juglone group. The three groups of cells were placed in their respective environments for 72 hours, and the apoptosis rate of the cells was observed at the end of the time, so as to judge the degree of damage to the vascular endothelium in the three environments, and to judge whether 25-dihydroxyvitamin D3 has any effect on the vascular endothelium damage in diabetes and the extent of the effect.

4. Experimental Results of 25 Dihydroxyvitamin D3 in Relieving Vascular Endothelial Damage in Diabetes Mellitus

(1) Apoptosis rate at different levels of 25-dihydroxyvitamin D3

The vascular endothelial cells of HG group and VitD3 group were observed to determine whether 25-dihydroxyvitamin D3 can alleviate the damage of vascular endothelium. The specific results are shown in Table 1.

Table 1: Apoptotic rates at different levels of 25-dihydroxyvitamin D3

	Apoptosis rate
HG	39.85%
VitD3 $\left(10^{-8} mol/L\right)$	37.26%
VitD3 $\left(10^{-7} mol/L\right)$	29.04%
VitD3 $\left(10^{-6} mol/L\right)$	18.27%

The apoptosis rate of vascular endothelial cells in HG group was 39.85%, indicating that hyperglycemia in diabetic patients can indeed damage vascular endothelial cells. In VitD3 group, the apoptosis rate of vascular endothelial cells was 37.26% at $10^{-8} mol/L$ concentration, 29.04% at $10^{-7} mol/L$ concentration, and 18.27% at $10^{-6} mol/L$ concentration. This shows that 25-dihydroxyvitamin D3 has a significant mitigation effect on the damage of VEC, and the apoptosis rate of VEC is low under the concentration of $10^{-6} mol/L$.

(2) Effect of 25-dihydroxyvitamin D3 on apoptosis rate of vascular endothelial cells

TUNEL (TdT mediated dUTP nick end labeling) and flow cytometry were used to observe the apoptosis rate of cells in HG group, VitD3 group and Juglone group. The concentration of $10^{-6} mol/L$ was selected in VitD3 group. The results are shown in Figure 4.

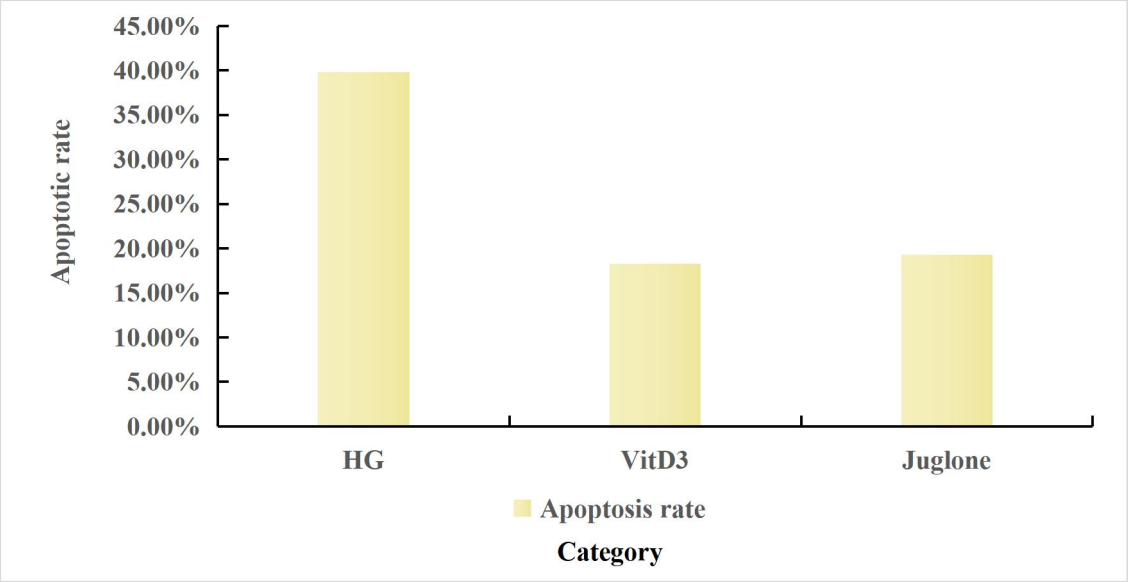

Figure 4. Effect of 25-dihydroxyvitamin D3 on the apoptosis rate of vascular endothelial cells

In view of the injury of vascular endothelium in high glucose environment, both 25-dihydroxyvitamin D3 and walnut quinone can play a mitigating role. The apoptosis rate of VEC in high glucose environment after adding 25-dihydroxyvitamin D3 solvent is 18.27%, and the apoptosis rate of VEC in high glucose environment after adding walnut quinone solvent is 19.26%. Compared with walnut quinone solvent, 25-dihydroxyvitamin D3 has certain advantages in relieving vascular endothelial damage in diabetes mellitus.

(3) Inhibitory effect of 25-dihydroxyvitamin D3 on Pin1 expression at different concentrations

In the experiment, the cultured VEC were placed in a high glucose environment to observe the expression level of Pin1 in endothelial cells under different concentrations of 25-dihydroxyvitamin D3, as shown in Table 2.

Table 2. Inhibition of 25-dihydroxyvitamin D3 on Pin1 expression levels at different concentrations

	Pin1/β-actin
HG	48.35%
VitD3 $\left(10^{-8} mol/L\right)$	46.85%
VitD3 $\left(10^{-7} mol/L\right)$	27.57%
VitD3 $\left(10^{-6} mol/L\right)$	17.39%

Hyperglycemia in diabetic patients can increase the expression level of Pin1, and 25-dihydroxyvitamin D3 can effectively inhibit the expression level of Pin1. When the

25-dihydroxyvitamin D3 solvent was $10^{-8} mol/L$, the expression level of Pin1 decreased slightly. When the 25-dihydroxyvitamin D3 solvent was $10^{-7} mol/L$, the expression level of Pin1 was significantly reduced. When the 25-dihydroxyvitamin D3 solvent was $10^{-6} mol/L$, the expression level of Pin1 was further reduced.

(4) Effect of 25-dihydroxyvitamin D3 on inhibition of p66Shc phosphorylation

The phosphorylation level of p66Shc in vascular endothelial cells of HG group, VitD3 group and Juglone group was observed experimentally, as shown in Table 3.

Table 3. Effect of 25-dihydroxyvitamin D3 on inhibition of p66Shc phosphorylation levels

	p-p66Shc/Total p66Shc
HG	51.59%
VitD3	19.23%
Juglone	21.64%

The level of p66Shc in VEC in HG group was high, indicating that high glucose environment can induce phosphorylation of p66Shc in VEC. Compared with HG group, the level of p66Shc in VEC in VitD3 group and Juglone group was lower, indicating that both 25-dihydroxyvitamin D3 and juglone could inhibit the phosphorylation level of p66Shc, but juglone was not as effective as 25-dihydroxyvitamin D3 in inhibiting the phosphorylation level of p66Shc.

(5) Effect of 25-dihydroxyvitamin D3 on ROS in vascular endothelial cells

Fluorescence microscope and flow cytometry were used to observe ROS generated by endothelial cells in high glucose environment. 25-dihydroxyvitamin D3 was selected as $10^{-6} mol/L$ concentration, and the relevant results were shown in Figure 5.

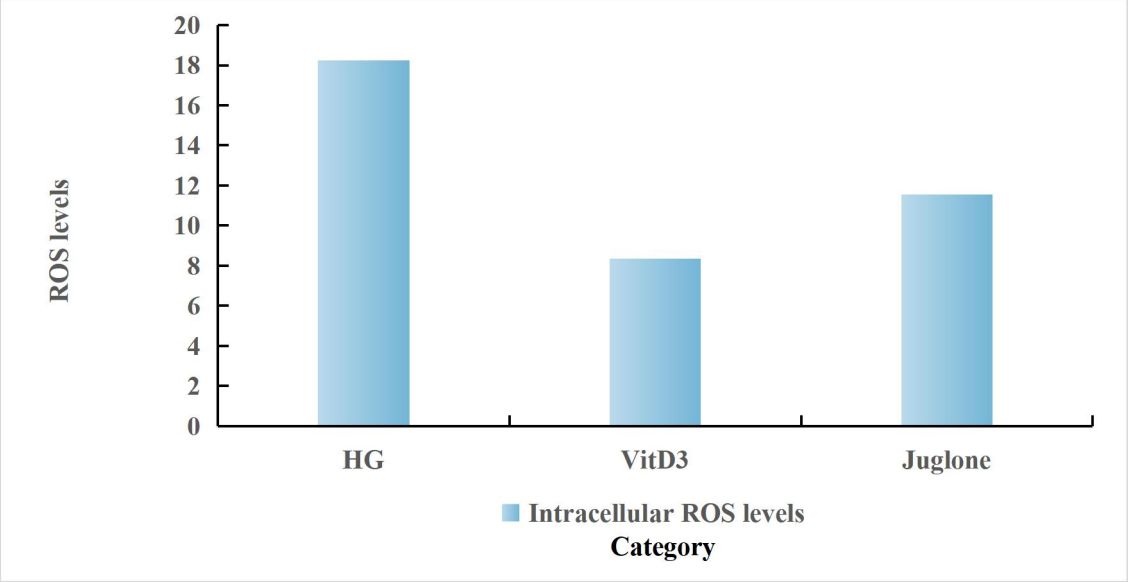

Figure 5. Effect of 25-dihydroxyvitamin D3 on vascular endothelial cell ROS

The ROS level of vascular endothelial cells in HG group was 18.23, indicating that the hyperglycemic environment in diabetic patients can cause the increase of ROS level. The ROS level of cells in VitD3 group was 8.35, and 25-dihydroxyvitamin D3 could play a certain role in reducing the

ROS level of cells. The ROS level of cells in Juglone group was 11.54, indicating that juglone can also reduce the ROS level of cells. The ROS level of cells in VitD3 group was lower than that in Juglone group.

(6) CASP3 level of vascular endothelial cells

CASP3 is a component of the cytotoxic T lymphocyte killing mechanism. The CASP3 level of vascular endothelial cells and the selective concentration $10^{-6} mol/L$ of 25-dihydroxyvitamin D3 in HG group, VitD3 group and Juglone group were observed experimentally. The specific results are shown in Figure 6.

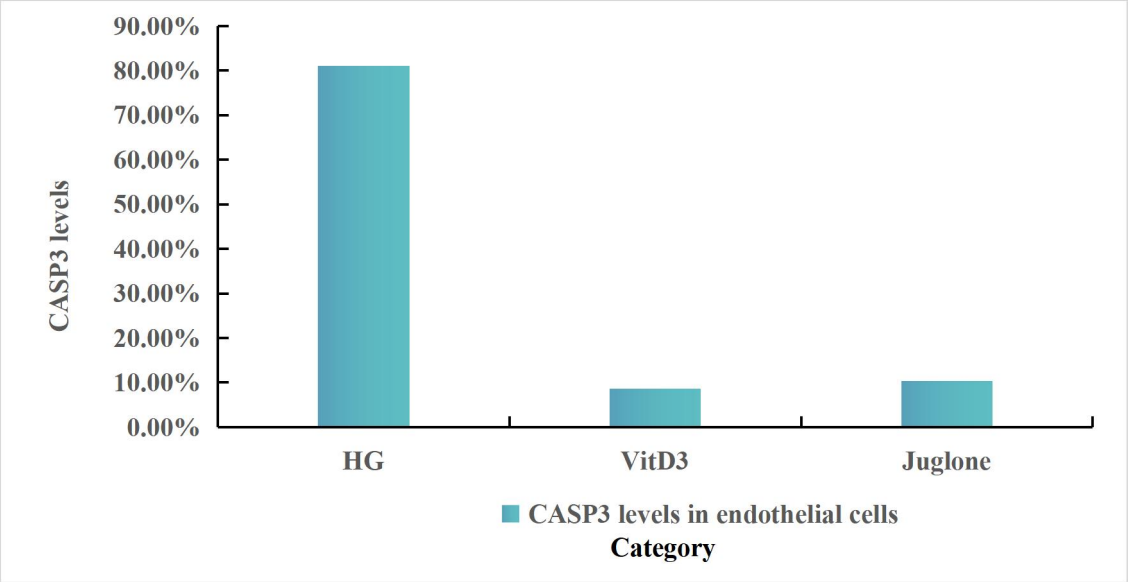

Figure 6. Vascular endothelial cell CASP3 levels

The level of CASP3 of cells in HG group reached 81.06%, and the level of CASP3 of cells was significantly abnormal in hyperglycemia environment, which was not conducive to the normal function of vascular endothelium in diabetic patients. The level of CASP3 in cells in VitD3 group was 8.56%. Compared with HG group, the level of CASP3 in vascular endothelial cells in VitD3 group decreased significantly, indicating that 25-dihydroxyvitamin D3 can effectively inhibit the level of CASP3 in cells. The level of CASP3 in cells in Juglone group was 10.37%. Compared with the HG group, the level of CASP3 in cells in the Juglone group was also significantly reduced. Walnut quinone also had a significant effect in reducing the level of CASP3 in cells, but the effect was not as good as that of 25-dihydroxyvitamin D3.

(7) VDR (vitamin D receptor) mediates the inhibitory effect of 25-dihydroxyvitamin D3 on the expression of Pin1

In the experiment, VDR siRNA (Small interfering RNA) was used to transfect vascular endothelial cells to observe whether 25-dihydroxyvitamin D3 could inhibit the expression level of Pin1 by activating VDR, thereby inhibiting the mitochondrial stress oxidation and inflammation mediated by Pin1, so as to alleviate vascular endothelial damage under hyperglycemic environment. The results are shown in Table 4.

Table 4. VDR mediates the inhibitory effect of 25-dihydroxyvitamin D3 on Pin1 expression levels

	Pin1/ β -actin
HG	39.27%
VitD3 $\left(10^{-6} mol/L\right)$	10.39%
Scrambled siRNA $\left(25nmol/L\right)$	11.45%
VDR siRNA $\left(25nmol/L\right)$	30.25%

The expression level of Pin1 protein was 39.27% in HG group and 30.25% in VDR siRNA group. Compared with HG group, the expression level of Pin1 protein in VDR siRNA group decreased by 9.02%. It shows that 25-dihydroxyvitamin D3 can inhibit the expression level of Pin1 protein, reduce the production of mitochondrial ROS mediated by p66Shc, and alleviate diabetic vascular endothelial damage by activating VDR.

5. Conclusions

In order to alleviate the damage of diabetic vascular endothelium, so as to ensure the life safety of diabetic patients and improve their sense of happiness, this paper first analyzed the causes, symptoms, examination and treatment of diabetes. Secondly, the mechanism of vascular endothelial injury in diabetes mellitus was discussed, including hyperglycemia, oxidative stress, inflammatory reaction, etc. Finally, an experimental study was conducted on 25-dihydroxyvitamin D3 to alleviate vascular endothelial injury in diabetes mellitus. The conclusion is that 25-dihydroxyvitamin D3 can play a significant role in reducing the expression level of Pin1 protein, p66Shc phosphorylation level, ROS level and CASP3 level in diabetic vascular endothelial cells. The study in this paper provided a reference for alleviating vascular endothelial injury and preventing and treating diabetic angiopathy, but there are also shortcomings in this study. This paper failed to further detect the O2 - in mitochondria, mitochondrial DNA damage and other indicators, and did not study the function of 25-dihydroxyvitamin D3 in resisting mitochondrial oxidative stress in detail. It is hoped that the mechanism of 25-dihydroxyvitamin D3 in inhibiting the expression of Pin1 can be further studied in future studies.

References

[1]Zhang, Jinglong. "A novel mechanism of diabetic vascular endothelial dysfunction: Hypoadiponectinemia-induced NLRP3 inflammasome activation." *Biochimica et Biophysica Acta (BBA)-Molecular Basis of Disease* 1863.6 (2017): 1556-1567.

[2]Knapp, Maura, Xin Tu, and Rongxue Wu. "Vascular endothelial dysfunction, a major mediator in diabetic cardiomyopathy." *Acta Pharmacologica Sinica* 40.1 (2019): 1-8.

[3]Liu, Yi. "Human umbilical cord-derived mesenchymal stem cells not only ameliorate blood glucose but also protect vascular endothelium from diabetic damage through a paracrine mechanism mediated by MAPK/ERK signaling." *Stem cell research & therapy* 13.1 (2022): 1-20.

[4]Jamwal, Shalini, and Saurabh Sharma. "Vascular endothelium dysfunction: a conservative target in metabolic disorders." *Inflammation Research* 67.5 (2018): 391-405.

[5]Dong, Yunzhou. "Role of endoplasmic reticulum stress signalling in diabetic endothelial dysfunction and atherosclerosis." *Diabetes and Vascular Disease Research* 14.1 (2017): 14-23.
[6]Petrie, John R., Tomasz J. Guzik, and Rhian M. Touyz. "Diabetes, hypertension, and cardiovascular disease: clinical insights and vascular mechanisms." *Canadian Journal of Cardiology* 34.5 (2018): 575-584.
[7]Wu, Meifang. "Protective effects of 1, 25-dihydroxyvitamin D3 against high-glucose-induced damage in human umbilical vein endothelial cells involve activation of Nrf2 antioxidant signaling." *Journal of Vascular Research* 58.4 (2021): 267-276.
[8]Kim, Il Young. "1, 25-dihydroxyvitamin D deficiency is independently associated with cardiac valve calcification in patients with chronic kidney disease." *Scientific reports* 12.1 (2022): 1-9.
[9]Zeng, Xiaoyun. "Effects of 1, 25-dihydroxyvitamin D3 on pathological changes in rats with diabetic cardiomyopathy." *Lipids in health and disease* 16.1 (2017): 1-7.
[10]Faridvand, Yousef. "1, 25-Dihydroxyvitamin D3 activates Apelin/APJ system and inhibits the production of adhesion molecules and inflammatory mediators in LPS-activated RAW264. 7 cells." *Pharmacological Reports* 71.5 (2019): 811-817.
[11]Tian, Li-Qiang. "Early 1, 25-Dihydroxyvitamin D3 Supplementation Effectively Lowers the Incidence of Type 2 Diabetes Mellitus via Ameliorating Inflammation In KK-Ay Mice." *Journal of nutritional science and vitaminology* 67.2 (2021): 84-90.
[12]Jia, Tingting. "1α, 25-dihydroxyvitamin D3 promotes osseointegration of titanium implant via downregulating AGEs/RAGE pathway in T2DM." *Endocrine Connections* 7.11 (2018): 1186-1195.
[13]Zhu, Wuzhengl. "Mesenchymal stem cells ameliorate hyperglycemia-induced endothelial injury through modulation of mitophagy." *Cell Death & Disease* 9.8 (2018): 1-17.
[14]Khaddaj Mallat, Rayan. "The vascular endothelium: A regulator of arterial tone and interface for the immune system." *Critical reviews in clinical laboratory sciences* 54.7-8 (2017): 458-470.
[15]Fratantonio, Deborah. "Cyanidin-3-O-glucoside ameliorates palmitate-induced insulin resistance by modulating IRS-1 phosphorylation and release of endothelial derived vasoactive factors." *Biochimica et biophysica acta (BBA)-molecular and cell biology of lipids* 1862.3 (2017): 351-357.
[16]Ghosh, Arijit. "Role of free fatty acids in endothelial dysfunction." *Journal of biomedical science* 24.1 (2017): 1-15.
[17]Li, Shuang. "Exosomes from hyperglycemia-stimulated vascular endothelial cells contain versican that regulate calcification/senescence in vascular smooth muscle cells." *Cell & bioscience* 9.1 (2019): 1-15.
[18]Berezin, Alexander E. "Endothelial progenitor cells dysfunction and impaired tissue reparation: the missed link in diabetes mellitus development." *Diabetes & Metabolic Syndrome: Clinical Research & Reviews* 11.3 (2017): 215-220.
[19]Xu, Suowen. "Endothelial dysfunction in atherosclerotic cardiovascular diseases and beyond: from mechanism to pharmacotherapies." *Pharmacological Reviews* 73.3 (2021): 924-967.
[20]Xing, Ying. "Netrin-1 restores cell injury and impaired angiogenesis in vascular endothelial cells upon high glucose by PI3K/AKT-eNOS." *Journal of molecular endocrinology* 58.4 (2017): 167-177.