Polyol pathway: A possible mechanism of diabetes complications in the eye

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In complex diseases such as diabetes mellitus, the causative agents include various serum factors such as glucose, aldose reductase, oxygen-free radicals, advanced glycation end products, protein kinase-C and growth factors. The polyol pathway is a pathway of glucose metabolism and is regarded as an important element in the pathogenesis of refractive changes, cataract formation and diabetic retinopathy in individuals with diabetes mellitus. The focus of this review is on the role of the polyol pathway in the pathogenesis of diabetic complications in the eye. The first enzyme (aldose reductase) in the polyol pathway reduces glucose to sorbitol. The second enzyme (sorbitol dehydrogenase) converts sorbitol to fructose. The accumulation of sorbitol and fructose in the crystalline lens and retina leads to the generation of oxidative stress. Oxidative stress is the imbalance between levels of reactive oxygen species and the antioxidant defence in a biological system, and it results in tissue damage. How hyperglycaemia leads to oxidative stress is not clear but could be through a combination of increased levels of reactive oxygen species and decreased capacity of the cellular antioxidant system. Oxidative stress causes the development of diabetic complications that are seen clinically.

Introduction

Diabetes mellitus is the most common endocrine disorder. It is defined as a group of disorders that exhibit a defective or deficient insulin secretory process, glucose underutilisation and hyperglycaemia.[1,2,3,4,5,6,7,8,9,10,11,12,13,14] All forms of diabetes are characterised by hyperglycaemia (a relative or absolute lack of insulin action), pathway-selective insulin resistance and the development of diabetes-specific pathology in the eye (refractive changes, cataract and retinopathy), peripheral nerve and kidneys (renal glomerulus).[4,8,13] As all forms of diabetes are characterised by chronic hyperglycaemia, it follows that hyperglycaemia is the initiating cause of diabetic tissue damage that we see clinically. However, this pathology can be accelerated by the hereditary characteristics of an individual and other factors such as hypertension and hyperlipidaemia.[8,13] However, it is not clear how chronic hyperglycaemia leads to tissue damage. It is surprising that only some cells are damaged by hyperglycaemia while all cells are exposed to hyperglycaemia. It has been stated that cells not damaged by hyperglycaemia are able to reduce the transport of glucose inside the cell when they are exposed to hyperglycaemia and are consequently able to maintain constant internal glucose concentration.[13] Thus, diabetes selectively damages cells whose glucose transport rate does not decline rapidly as a result of hyperglycaemia, leading to high glucose inside the cell.[13]

The objective of this paper is to review the polyol pathogenetic mechanism of the ocular complications of diabetes. In the eye, diabetes affects both the crystalline lens and retina, resulting in devastating effects on vision.

The polyol pathway

The polyol pathway is based on the enzyme aldose reductase.[4,5,6,7,8,9,10,11,12,13,14] Aldose reductase reduces toxic aldehydes in the cell to inactive alcohols, which is its normalfunction.[10,13] When glucose concentration in the cell becomes too high, aldose reductase also reduces that glucose to sorbitol, using nicotinamide adenine dinucleotide phosphate (NADPH) as a cofactor (Figure 1). NADPH is an essential cofactor for regenerating critical intracellular antioxidants, and is then reduced to glutathione (GSSG). Sorbitol is then oxidised to fructose by the enzyme sorbitol dehydrogenase, which uses nicotinamide adenine dinucleotide (NAD^+) as a cofactor.[4,5,6,7,8,9,10,11,12,13,14] Sorbitol is an alcohol, and is highly hydrophilic.[8,10,13] Therefore it does not diffuse easily through the cell membrane and accumulate intracellularly. Sorbitol is also more difficult to metabolise.[8,10,13] The buildup of sorbitol has damaging effects in cells, with possible osmotic damage. The use of NADP by aldose reductase may result in less cofactor available for glutathione reductase, which

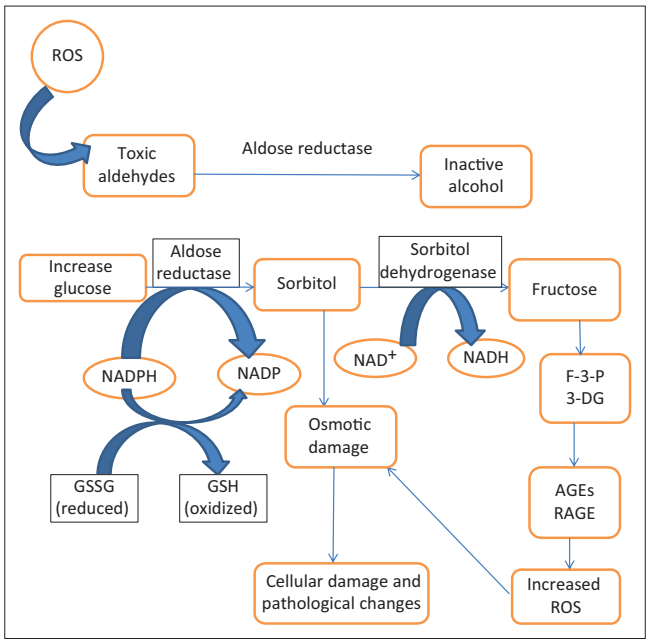

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FIGURE 1: The polyol pathway comprises two enzymes: aldose reductase and sorbitol dehydrogenase. Aldose reductase normally reduces aldehydes generated by reactive oxygen species to inactive alcohol. When glucose concentration in a cell becomes too high, aldose reductase reduces that glucose to sorbitol. Sorbitol dehydrogenase then oxidises sorbitol to fructose. The buildup of sorbitol has damaging effects in cells, including osmotic damage.

is critical for the maintenance of the intracellular pool of reduced glutathione and would otherwise lessen the capacity of cells to respond to oxidative stress (Figure 1).

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Oxidative stress results from the oxidation of sorbitol to fructose because NAD^+ is converted to NADH, which is the substrate for NADH oxidase that generates reactive oxygen species (ROS).[4,6] Oxidative stress is the critical factor in the development of diabetic complications, including diabetic retinopathy.[5] Oxidative stress and the resultant tissue damage are the hallmarks of chronic disease and cell death.

The usage of NAD^+ by sorbitol dehydrogenase leads to an increased NADH/NAD^+ ratio, which is termed '*pseudohypoxia*' and is linked to a number of metabolic and signalling changes that alter cell functions and increased production of ROS within a cell.[4,6] The excess NADH may become a substrate for NADH oxidase, and this would be a mechanism for the generation of intracellular oxidant species. Altering intracellular tonicity would expose cells to oxidative stress (oxidant-derived tissue injury) through decreased antioxidant defences, and generation of oxidant species that would ultimately initiate and multiply several mechanisms of cellular damage. Decreased NADPH/NADP and increased NADH/NAD^+ could potentially account for all of the other biochemical abnormalities seen in diabetic complications,[4,6,8,9,10] In cells where sorbitol dehydrogenase activity is high, this may result in an increased NADH/NAD^+ ratio. A decreased NADH/NADP ratio can increase oxidative stress by decreasing regeneration of cellular antioxidant (reduced glutathione from oxidised glutathione) and by decreasing availability of NADPH, thereby decreasing the activity of catalase, the enzyme responsible for converting ROS and Hydrogen peroxide (H_2O_2) into water. By reducing the amount of reduced glutathione, the polyol pathway increases susceptibility to intracellular oxidative stress.[10] Hyperglycaemia increases the NADH/NAD^+ ratio in complication-prone cell populations. An elevated NADH/NAD^+ ratio could significantly affect the health of the retina.[6]

Produced fructose can become phosphorylated to fructose-3-phosphate which in turn can be broken down to 3-deoxyglucose and 3-deoxyglucosone.[11] These two compounds are strong glycating agents that can result in the production of advanced glycation end products (AGEs).[6,11] AGEs are a heterogeneous group of molecules formed from the non-enzymatic reaction of reducing sugars with free amino groups of proteins, lipids and nucleic acids,[6] and are major pathogenic mediators of almost all diabetic complications. They are found in the retinal vessels of diabetic patients.[6] AGEs increase leukocyte adhesion to retinal microvascular endothelial cells through intracellular ROS generation. Hyperglycaemia inside the cell increases the synthesis of diacylglycerol,[6,13] which is a critical activating cofactor for protein kinase-C (PKC). When PKC is activated by intracellular hyperglycaemia, it alters gene expression and protein function.[13] The factors that promote normal function are decreased, and those that are adverse to normal function are increased, resulting in cell damage.

Refractive changes in diabetes mellitus

The crystalline lens is covered by a collagenous capsule containing matrix proteins and proteoglycans.[13] The main energy supply of the lens is the glucose that enters the lens via facilitated transport.[13] In uncontrolled diabetes, refractive error may change or fluctuate.[15,16,17,18,19,20,21,22] This refractive change may be the first sign of undiagnosed diabetes. Both acute and chronic refractive changes are associated with diabetes. Duke-Elder[16] reported an association of myopia with hyperglycaemia and hyperopia with hypoglycaemia in diabetic patients. However, other researchers have reported that decreasing plasma glucose levels in diabetic patients cause hyperopic changes in diabetic patients.[17,18,19,20] Some researchers have found both myopic and hyperopic changes in diabetic patients.[20,21,22]

When glucose concentration in the crystalline lens becomes too high, aldose reductase reduces the glucose to sorbitol. Sorbitol then accumulates in the lens. When the body changes from a hyperglycaemic to a hypoglycaemic state, excess glucose in the lens flows out into the aqueous humour

but the sorbitol remains in the lens. The osmotic pressure difference that develops results in the influx of water from the aqueous humour into the lens, causing lenticular swelling with hyperopic refractive change. Even a decrease in the refractive index of the crystalline lens produces a significant transient hyperopic change;[22] this is a possible explanation for the occurrence of refractive change in diabetic patients.

Cataract formation

Cataract formation is another complication of diabetes. The key factor in diabetic cataract formation is the excess conversion of glucose to sorbitol.[23,24,25,26,27,28,29,30] In the lens, sorbitol is produced faster than it is converted to fructose by the enzyme sorbitol dehydrogenase.[24,25] The increased accumulation of sorbitol creates a hyperosmotic effect that results in the infusion of fluid to counteract the osmotic gradient. The osmotic stress or imbalance leads to fibre swelling, liquefaction and eventually cataract formation.[23,24,25,26]

Diabetic retinopathy

Diabetic retinopathy is one of the major complications in patients with diabetes; the retina becomes progressively damaged, leading to vision loss and blindness as a result of long-term accumulated damage to small blood vessels in the retina. The pathogenesis of diabetic retinopathy is complex, involving different cells, different molecules and different factors.[6,7,8,9,10,11] Hyperglycaemia leads to several biochemical disorders that are thought to contribute to diabetic retinopathy by causing damage and dysfunction of retinal capillary endothelial cells.[6,7,8,8,9,10,11,31,32,33,34,35] Aldose reductase is also present in the retina. Because sorbitol is less permeable to the cellular membrane, it accumulates within retinal capillary cells in response to hyperglycaemia and causes hyperosmolality of the cells.[6,8,10] Consequently, hyperosmolality induces an increase in intracellular water and lactate production and a decrease in oxygen uptake (oxidative stress). The fructose produced by the polyol pathway is phosphorylated to fructose-3-phosphate which is then degraded to 3-deoxyglucosone. Both the fructose-3-phosphate and 3-deoxyglucosone are glycating agents that can produce AGEs.[4,6] Generation of AGEs decreases antioxidant defences and initiates several mechanisms of cellular damage.[32,33,34]

Hyperglycaemia damages retinal microvascular cells and causes various changes in retinal tissues (such as enhanced vascular permeability) owing to pericyte loss, followed by microvascular occlusion in the retina.[35] Pericytes are elongated cells of mesodermal origin, wrapping around and along endothelial cells of small vessels. AGEs play a role in the development of microvascular disease in diabetes. Retinal pericytes accumulate AGEs during diabetes, which have a detrimental influence on pericyte survival and function.[4,6] AGEs cause apoptosis of retinal pericytes and induce vascular endothelial growth factor (VEGF).

VEGF is a growth factor that is studied in connection with diabetic retinopathy; it promotes angiogenesis (which causes breakdown of the blood-retinal barrier), stimulates endothelial cell growth and neovascularisation (formation of new blood vessels), and increases vascular permeability in the ischaemic retina.[31,34] The binding of VEGF to membrane tyrosine kinase receptors leads to vascular leakage and breakdown of the blood-retinal barrier. VEGF has been linked with leukocyte adhesion to retinal endothelial cells. An increased intraocular level of VEGF correlates with increased vascular permeability which contributes to haemorrhage, exudates and vascular leakage, leading to non-proliferative diabetic retinopathy. Angiogenesis and vasculogenesis lead to proliferative diabetic retinopathy.[35,36,37,38,39,40,41,42]

In addition to vascular changes, structural and functional damage to nonvascular cells (ganglion cells, glial cells and microglial) contribute to the pathogenesis of diabetic retinopathy.[38] Neurodegeneration of retinal neurons and glial cells can occur even before the development of microaneurysms.

It is now established that inflammation is crucial in the pathogenesis of diabetic retinopathy because it may cause neovascular damage and ischaemic neovascularisation.[38] Inflammation in the retina leads to increased intraocular blood pressure via endothelial nitric oxide synthase, formation of new weak vessels and their increased permeability owing to VEGF which in turn leads to haemorrhages in the retina, and leukostasis.[31,35,38] Leukostasis is an important event in diabetic retinopathy pathogenesis because it leads to capillary occlusion, ROS-mediated cell death and inflammatory response in the retinal tissue.[33]

Protein kinase-C (PKC) is also a mediator of vascular endothelial growth factor activity. When PKC is activated, it contributes to basement membrane thickening, vascular occlusion, increased permeability and activation of angiogenesis.[13] As ROS-producing H_2O_2 increases, it activates PKC. Therefore, ROS also activate PKC in vascular endothelial cells.

Exposure of retinal cells (pericytes, retinal pigment epithelial cells, neurons, ganglion cells, glial and Muller cells) to increased concentrations of glucose results in reduced viability of cells.[31,35] Pathophysiological changes that can occur during diabetic retinopathy include thickening of the retinal capillary basement membrane, leukocytes adhering to endothelial cells (leukostasis), vascular permeability and breakdown of the blood-retinal barrier.

Stages of diabetic retinopathy

Diabetic retinopathy is classified into non-proliferative diabetic retinopathy and proliferative diabetic retinopathy. It begins as mild, non-proliferative abnormalities and progresses to moderate and severe non-proliferative diabetic retinopathy and then proliferative diabetic retinopathy,

which is characterised by the growth of new blood vessels.[36,37,38,39,40,41,42]

The clinical features of non-proliferative diabetic retinopathy include the appearance of dot and blot haemorrhages (intraretinal blood) and microaneurysms.[36,37,38,39,40,41,42] Differentiation between a microaneurysm and a dot and blot haemorrhage is subjective because it is based on size. To distinguish them by direct ophthalmoscopy may be difficult. Haemorrhage stems from breakdown of the blood-retinal barrier, resulting in exudation of blood and exudates.[41] Hard exudates caused by leakage of proteins and lipids from the damaged arterioles appear as small white or yellow areas.[36,37,38,39,40,41,42] Cottonwool spots (soft exudates) result from micro-infarctions of nerve fibres caused by focal ischaemia after occlusion of terminal retinal arterioles occurs (ischaemia of the retinal nerve-fibre layer). These cottonwool spots appear as white spots on the retina;[36,37,38,39,40,41,42] they are an accumulation of cytoplasmic debris in the retina.[41] Macular oedema occurs when the oedema is located in the macular and is one of the clinical features that best correlates with the degree of vision loss in diabetic patients. It is the result of leakage from microaneurysms.

Ischaemia from vascular damage and disruption of local perfusion results in angiogenesis and neovascularisation, which manifest as proliferative diabetic retinopathy. Proliferative diabetic retinopathy is the late stage of diabetic retinopathy and is characterised by neovascularisation which is a response to continued retinal ischaemia.[36,37,38,39,40,41,42] The new blood vessels formed are fragile and prone to haemorrhage, which impairs vision, ultimately causing blindness.[36,37,38,39,40,41,42] Vision loss in diabetic retinopathy occurs from breakdown of the blood-retinal barrier, resulting in macular oedema, retinal detachment and inner retinal and vitreous haemorrhage.[42]

Conclusion

Diabetic complication is one of the leading causes of blindness and visual impairment. It is influenced by both genetic and independent factors, such as hypertension. Hyperglycaemia is considered the major determinant of diabetic microvascular complications. The polyol pathophysiology of diabetic complications has been briefly explained. The potential sources of tissue damage in diabetes mellitus could be a combination of overproduction of ROS, glucose auto-oxidation and non-enzymatic glycation. A cure for diabetic retinopathy is unlikely at present but the continuing research into diabetic retinopathy will provide better understanding of diabetic complications and will help in the development of novel therapeutic agents for effective treatment.

Competing interests

The author declares that he has no financial or personal relationships which may have inappropriately influenced him in writing this article.

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