Retinopathy and Visual Acuity in People with Diabetes Mellitus and Hard-to-Heal Diabetic Foot Ulceration

Sellman, Anders

2019

Document Version:
Publisher's PDF, also known as Version of record

Retinopathy and Visual Acuity in People with Diabetes Mellitus and Hard-to-Heal Diabetic Foot Ulceration

ANDERS SELLMAN
FACULTY OF MEDICINE | LUND UNIVERSITY

Retinopathy and Visual Acuity in People with Diabetes Mellitus and Hard-to-Heal Diabetic Foot-Ulcers.

Retinopathy and Visual Acuity in People with Diabetes Mellitus and Hard-to-Heal Diabetic Foot Ulceration

Anders Sellman

LUND
UNIVERSITY

DOCTORAL DISSERTATION
by due permission of the Faculty of Medicine, Lund University, Sweden.
To be defended at Föreläsningssal 4, Blocket, Universitetssjukhuset, Lund.

Date 2019-04-12 at 09.00

Faculty opponent
Associate Professor Stig Attvall, Department of Medicine, Sahlgrenska Akademien, Gothenburg, Sweden.

Organization LUND UNIVERSITY	Document name: Doctoral Dissertation
	Date of issue 2019- 04-12
Author(s) Anders Sellman MD	Sponsoring organization Nil

Title and subtitle Retinopathy and Visual Acuity in People with Diabetes Mellitus and Hard-to.Heal Diabetic Foot Ulceration.

Abstract

The prevalence of diabetes mellitus and its complications are increasing worldwide. Two of the complications:foot ulceration and diabetic retinopathy are especially feared, the former because of the risk of amputation and possible death, the latter because of the risk of.becoming blind The life-time risk of developing a foot-ulcer is estimated to be up to 25%. The world wide prevalence of any diabetic retinopathy is 34,6 %, .6.7 % for proliferative retinopathy, 6.8 % for macular edema and for vision-threatening retinopathy 10.2 %. During the first twenty years of disease retinopathy develops in most patients with Type 1 diabetes and in over 60% in Type 2 . Any degree of retinopathy augments all-case mortality more than 2.3 fold inType 1 diabetes and more than 2.4 in Type 2. Foot ulcers increase mortality by 2.5 compared to diabetic patients without ulcerations. Also lowered visual acuity increases mortality.

In the feet neuropathy, somatic and autonomous, and periferal vascular disease lead to altered blood flow and ischemia. Repetitive trauma then greatly increase the risk of ulceration Some of the ulcers are hard to heal and are therapeutic challenges where Hyperbaric oxygen therapy as an adjunct have been used. Its effects on diabetic eyes are incompletely known.

In the **first paper** we described the rationale of hyperbaric oxygen therapy and designed a study with so few weak points as possible. People with chronic diabetic foot ulcers were to be treated in an hyperbaric chamber with high pressure oxygen (2.5 ATA) or placebo Treatment is given in a double-blinded, randomised, prospektive way. Follow up time of ulcers and eye is two years.

In paper two we measured transcutaneous oxygen of the feet to compare these values with the thickness of the retina, as increased retinal thickness is a sign of retinal vasculopathy. Low O_2 values of the feet covariated with thicker retina..

Several studies have shown improved healing of chronic diabetic ulcers by hyperbaric oxygen treatment. **In paper three** we report on a prospektive, randomised and double-blinded study, where people with chronic ulcers were treated with hyperbaric oxygen or placebo (hyperbaric air) at 2.5 ATA for 40 sessions and then were followed for two years with fundus photography and visual acuity measuring. We found the high pressure oxygen treatment ophtalmological neutral except for a transient drop of acuity after 3 months in the oxygen but not placebo group.

In the **fourth paper** we compared two matched groups, one with chronic diabetic foot ulcers, one without. The. ulcer group had more severe retinopathy, more proliferative changes, more macular edema and lower visual acuity. The group differences also remained in insulin treated people and in people with kidney disease.

In **paper five** we followed the groups above for five and ten years for retinopathy, visual acuity and mortality. DR increased with diabetes duration and was more frequent and severe in the ulcer group. In the ulcer group visual acuity went from 10% severe visual impairment to 15.6% after 5 years and to 15.4% after ten years.This seems to depend on high mortality in the PDR group. Corresponding values in the non-ulcer group were 0.6%, 0.8% and 4.8%. Presence of any DR increased 10-year mortality 1.91 times in the ulcer group and 1.70 times in the control group.

In **paper six** we studied 5-year mortality in people with and without foot ulceration and visual acuity <6/12 (logMAR <0.3).The mortality was significantly higher in people with impaired vision, but worst in the ulcer group. Significant predictors of mortality were: DFU(RR 3.6), male sex (RR 2.34), Visual impairment (RR 1.77), age (RR 1.48) and low eGFR (RR 1.42).

Key words: Hyperbaric oxygen therapy, diabetic retinopathy, diabetic foot ulcers, mortality, visual acuity

Classification system and/or index terms (if any)

Supplementary bibliographical information		Language: English
ISSN and key title 1652-8220		ISBN 978-91-7619-760-8
Recipient's notes	Number of pages 97	Price
	Security classification	

Signature  Date 2019-03-11

Retinopathy and Visual Acuity in People with Diabetes Mellitus and Hard-to-Heal Diabetic Foot Ulceration

Anders Sellman

LUND
UNIVERSITY

Faculty of Medicine
Department of Endocrinology

ISBN 978-91-7619-760-8
ISSN 1652-8220

Printed in Sweden by Media-Tryck, Lund University
Lund 2019

Table of Contents

List of Abbreviations

AGE	Advanced Glycation Endproducts
AMD	Age-related Macular Degeneration
ADP	Adenosine DiPhosphate
ATA	Atmospheres Absolute
ATP	Adenosine TriPhosphate
BRB	Blood Retina Barrier
CI	Confidence Interval
CuZnSOD	Copper-Zinc Superoxide Dismutase
DAG	Diacyl Glycerol
DCCT	Diabetes Control and Complications Trial
DCI	Decompression Illness
DCS	Decompression Sickness
DFU	Diabetic Foot Ulcer
DIRECT	Diabetic Retinopathy Candesartan Study
DME	Diabetic Macular Edema
DR	Diabetic Retinopathy
DRS	Diabetic Retinopathy Study Research Group
ERG	ElectroRetinoGraphy
ET-1	Endothelin-1
ETDRS	Early Treatment Diabetic Retinopathy Study Group
GF-beta	Fibroblast Growth Factor beta
Hb	Hemoglobin
HbA1c	Glycated Hemoglobin
HIF-1	Hypoxia Inducible Factor
IL-1	Interleukin-1
IDDM	Insulin Dependent Diabetes Mellitus
K^+	Potassium Ion

mfERG	multifocal ElectroRetinoGraphy
MnSOD	Manganese Super Oxide Dismutase
NAD	Nicotine Amide Dinucleotide
NIDDM	Non-Insulin Dependent Diabetes Mellitus
NO	Nitric Oxide
NOS	Nitric Oxide Synthetase
NPDR	Non-Proliferative Diabetic Retinopathy
PDR	Proliferative Diabetic Retinopathy
PKC	Protein Kinase C
RAGE	Receptor for AGE
RAS	Renin Angiotensin System
ROS	Reactive Oxygen Species
SOD	Superoxide Dismutase
T1DM	Type 1 Diabetes Mellitus
T2DM	Type 2 Diabetes Mellitus
UKPDS	United Kingdom Prospective Diabetes Study
VEGF	Vascular Endothelial Growth Factor
WESDR	Wisconsin Epidemiological Study of Diabetic Retinopathy

List of papers

This thesis is based on the following papers.

I. Löndahl M, Katzman P, Nilsson A, Hammarlund C, *Sellman A*, Wykman A, Hugo-Persson M, Apelqvist J; A prospective study: hyperbaric oxygen therapy in diabetics with chronic foot ulcers. J Wound Care. 2006:**10**;457-9

II. *Sellman A*, Katzman P, Andreasson S, Löndahl M. Long-term Effects of Hyperbaric Oxygen Therapy on Visual Acuity and Retinopathy in Patients with Chronic Diabetic Foot Ulcers – Outcome of a Randomized Double-blind Placebo-controlled Study. Submitted

III. *Sellman A*, Löndahl M, Andreasson S, Katzman P. Transcutaneous Oximetry but not Arterial Toe Blood Pressure or Ankle-Brachial Index is related to Macular Thickness in Patients with Chronic Diabetic Foot Ulcers. J Expr Integ. Med. 2013;3(2):81-85

IV. *Sellman A*, Katzman P, Andreasson S, Löndahl M. Presence of chronic diabetic foot ulcers is associated with more frequent and more advanced retinopathy. Diabet Med. 2018 Oct;35(10):1364-1370. doi: 10.1111/dme.13682. Epub 2018 Aug 2. PMID: 29791040

V. *Sellman A*, Katzman P, Andreasson S, Löndahl M. Retinopathy and Visual Acuity in People with Type 2 Diabetes with and without Hard- to-heal Diabetic Foot Ulcer– a Ten Year Follow-up. In manuscript

VI. *Sellman A*, Katzman P, Andreasson S, Löndahl M. Visual impairment predicts 5-year mortality in people with Type 2 Diabetes with and without Hard-to-heal Diabetic Foot Ulcer. Submitted

1. Introduction

Diabetes mellitus has pursued mankind since more than 3000 years. Ancient physicians-writers as Sushruta, Aretaeus, Chung Ching, Chen Chuan, Avicenna and Maimonides described diabetic symptoms, inclusive the sweet taste of urine and some other long-time symptoms. Willis in 1674 reported in his *Pharmaceutice Rationalis* the sweet taste of urine in some patients. In 1776 M. Dobson found the sweet taste depending on the presence of a reducing substance, resulting in a sugar-like taste. The reducing substance was shown to be sugar by E.M. Chevreul in 1815 (1).

Mering and Minkowski demonstrated the role of pancreas in diabetes in 1889 and in 1921 insulin was discovered by Banting, Best, MacLeod and Collip and then entered clinical use in 1922-23 (1).

Because of high mortality by other diseases long-time diabetic complications were not identified for a long time. With increased and improved knowledge, long-time complications were unveiled. This was true only for patients of older age because diabetes of acute or subacute type (now known as Type 1 diabetes) had a very short life expectancy due to emaciation and ketoacidosis.

In 1850 the invention of the ophthalmoscope enabled physicians for the first time to visualise the retina and in 1856 von Jäger described diabetic changes in the fundus (2). Already in the late 18th century it became more and more clear that diabetes plays a role in many cases of ulcerations and gangrene of the lower extremity. Following the clinical introduction of insulin in 1923 the life expectancy of the Type 1 dramatically improved and also people with Type 2 diabetes had improved chances of longer life. With a longer life span the risk of developing long-term complications increased.

Because of the risk of blindness diabetic eye-complications were among the most dreadful. Complications involving the lower extremities with ulcerations, life-threatening infections and impending amputations also affect the ability to perform normal life actions. Other complications as renal insufficiency, increased risks of cardiovascular and cerebrovascular events and untimely death got more and more attention.

Despite improved treatment modalities, diabetes related complications are still associated with increased risk of disabilities and death. The significant morbidity also has shown to have significant financial and psychological effects, such as inability to work, procure food and engage in social events.

1.1 Diagnosis of Diabetes mellitus.

Diabetes mellitus (DM) is a metabolic disease with elevated blood sugar levels. For the diagnosis following criteria are used.

Table 1.1
Diagnosis criteria for diabetes mellitus.

	Healthy	Impaired fasting value	Diabetes
Fasting glucose mmol/l	< 6.1	6.3- 6.9	≥7.0
OGTT mmol/l capillary	< 8.9	8.9- 12.1	≥ 12.2
OGTT mmol/l, venous	< 7.8	7.8- 11.0	≥ 11.1
HbA1c mmol/mol	< 48		≥ 48

OGTT= oral glucose tolerance test. Diagnostic criteria from ADA 2019.

1.2 Classification of Diabetes mellitus

Several types of DM are known and currently divided as follows:

- Type 1. A much too low or lack of insulin production because of immune-mediated destruction of the pancreatic islets´ beta-cells. A subgroup is LADA. (Latent Autoimmune Diabetes in Adults). Anti-bodies as GAD, IA and IAA are present in the blood-plasma in about 80% of the diseased, When no antibodies are detected one speaks of Idiopathic Type 1 diabetes.
- Type 2. Insulin production and other insulin modulated effects are maintained but slowly decreasing and insulin resistance prevents an optimal metabolic use of glucose. This is the most common type and affecting mainly middle-aged and older people. Type 2 is often related to overweight, hypertension and high blood-lipids, the metabolic syndrome. A sedentary lifestyle and obesity seem to play an important role. Typical is a very insidious course, where long-time complications may be the first signs of disease.
- Gestational diabetes, a mostly spontaneously resolving type during pregnancy but with increased risk of getting a Type 2 later in life.

- Diabetes due to other causes: including monogenetic diabetes such as MODY, (Mature Onset of Diabetes of the Young), iatrogenic induced diabetes, other endocrine diseases as Cushing´s syndrome, acromegaly, pheochromocytoma, glucagonoma, hyperthyroidism. Diseases of the pancreas, genetic syndromes as the syndromes of Down, Klinefelter, Turner and Prader-Willis.

One can certainly speak of a global epidemic of diabetes, with present estimates of 425 million globally, which may rise to 629 million in 2045 (3). In the Swedish population of 10,120,242 (Dec 2017) 4.2 % have a diagnosis of diabetes, 44,962 Type 1 and 379,481 Type 2. However, many cases are not overt and thus not diagnosed (4).

The incidence of Type 1 is 30/100 000/year. Type 2 diabetes incidence is about 300/100 000/year (5). Genetic inheritance is weak in Type 1 but stronger in Type 2. Risk-genes are common in Type 1 (HLA-DR 3,4 / HLA- DQ 2,8). People with Type 1 diabetes are prone to other auto-immune diseases as thyroid diseases, coeliciasis, and Rheumatoid arthritis (6). In type 2 there are heredity traits and several identified genes associated to the disease but all considered low risk at this time.

Type 1 diabetes typically debuts within weeks or months with obvious symptoms as weight loss, thirst and frequent urination and prostration. Severe derangement of electrolytes and acid-base balance may be present as a consequence of the pathological glucose metabolism. The condition is lethal if not treated with insulin.

The onset of Type 2 is much more insidious, with the disease possibly existing 5 to10 years before diagnosis. Cardiovascular or cerebrovascular problems are often the first signs, but, additional symptoms include neuropathy, vision problems, renal and foot-problems.

1.3 Microvascular disease

The underlying processes of vascular complications in diabetes are very complex and still not fully understood. Macro- and microvascular lesions develop and neuropathy seems to be a very early occurring lesion. Atherosclerosis develop in the larger vessels, starting with insulin resistance and endothelial dysfunction. Microscopically the basement membrane is thickened which is thought to depend on up-regulated fibronectin, collagen and laminin, leading to changes in factors mediating the growth, survival and function of pericytes and endothelial cells. Pericyte loss occurs (7, 8). The pericyte loss results in detoriation of the regulation of blo d flow. The increased blood viscosity, the reduced deformability of red cells, increased platelet aggregation and leukocyte aggregation in diabetes lead to

decreased blood flow at capillary level and even to thrombosis of the smallest vessels of the retinal circulation (9). Soon following the debut of the disease, blood flow and vascular permeability are increased in small vessels and shear-stresses develop. Hyperperfusion leads to capillary damage, then to capillary closure and retinal ischemia(10).

Ischemia activates Hypoxia Induced Factor 1 (HIF-1ά) which mobilises Vascular Endothelial Growth Factor, VEGF. VEGF is a family of molecules, VEGF A-F and Placenta Growth Factor (PlGF) with isoforms. VEGF binds to receptors VEGFR 1 or 2. VEGF-A is "responsible" for most ocular neovascularisation and breakdown of the blood-retina barrier (11, 12). The role of PlGF in humans is unclear. The important vasodilator NO becomes downregulated and the vasoconstrictor endothelin-1 gets enhanced (13).

1.3.1 Hyperglycaemic metabolic changes

Alternative glucose metabolic pathways are activated by the hyperglycaemia and have far reaching consequences.

- Increased hexose amine- pathway activates genes that lead to endothelial dysfunction. However, the mechanism is not known in detail (14).
- Increased polyol pathway flux where glucose is increasingly converted to sorbitol. Its oxygenation leads to increasing methylglyoxal (an AGE precursor) and diacylglycerol (DAG) (14).
- Advanced glycation end products (AGE) formation. AGE damages target cells by modifying their proteins which alter their functions. The end result is production of ROS, increased expression of cytokines and growth factors, including VEGF (15).
- Activation of PKC. Hyperglycaemia elevates synthesis of diacylglycerol (DAG) which activate PKC´s many biochemical signalling pathways. Especially the PKC-β iso-form drives the expression of VEGF and downregulation of the important vasodilator NO (16, 17).
- Poly (ADP-ribose) polymerase (PARP) activation. The involved molecular mechanisms contribute to endothelial dysfunction in diabetic (18).
- Activation of the renin-angiotensin-aldosterone system (RAAS). Local accumulation of glucose activates juxtaglomerular cells to release prorenin and renin. Angiotensin Converting Enzyme (ACE) has been reported to affect capillary perfusion and vascular structure with upregulation of VEGF (19).

It is now well-known that inflammation and leucostasis are factors in capillary occlusion and consecutive retinal hypoxia, which in its turn drives VEGF expression. (20, 21)

The above mentioned pathways all produce superoxides by way of the mitochondrial electron transport chain and it is possible that this overexpression of superoxides is a common denominator of diabetic retinopathy (DR) formation and in combination with inflammation and leucostasis play the main roles in the genesis of DR (9, 16, 22-24).

Chronic inflammation appears as a consequence of continuous oxidative stress, with inflammation upregulating transcription factors, cytokines and chemokines.

This leads to changes of the microvascular beds, impaired functions of peripheral nerves and the autonomic nervous system, as well as diabetic Peripheral Arterial Disease (PAD). Further this develops a pro-thrombotic state, vascular endothelial dysfunction and abnormal lipid profiles, affecting the vessels of the lower extremities with risks of occlusion and ischemia (25, 26). Lesions in other vascular beds leads to increased risks of cardiovascular and cerebrovascular events, impaired renal function and retinopathy.

Duration of disease and HbA_{1c} values determine severity and time to retinal complications. Even in a prediabetic state with normal fasting glucose but impaired oral glucose tolerance tests one can find signs of retinopathy with changes in the electroretinogram (ERG) and changes in colour vision and contrast sensitivity (26-28). Other early alterations include altered retinal blood flow with deteriorated autoregulation with arteriolar constriction, decreased blood flow and increased oxygen in the venous capillaries.

In the feet elevated local blood flow and high oxygen saturation despite cellular hypoxia have been found. It may be that the nutritional capillaries are overloaded and that blood bypasses normal circulation via AV-shunts, resulting in ischemia in the nutritional capillaries(29). In 2010 Sangiorgi et al published results from electron microscopy of corrosion casts of diabetic toes. The researchers found deranged microvascular architecture, AV-shunting and signs of vascular microthrombic processes (30). Gabbay et al hypothesise that the findings of elevated blood flow and high oxygen saturation in both artery and vein indicate that the blood flow is redirected by shunts thus not being available for the ischemic tissue (31). Bek found that O_2 saturation in booth arteries and veins increased with increasing numbers of microaneurysms and haemorrhages in the retinal veins is a possible marker of early steps in the chain of causality of formation of DR (32, 33).

In this dissertation Diabetic Retinopaty (DR) and hard-to-heal foot-ulcers (DFU) and their interrelationship will be discussed. It is known that the existence of DR and foot ulceration are related but their exact and close relationship is not fully elucidated.

2. Background

2.1 Diabetic retinopathy

2.1.1 Introduction

Embryologically the retina is a part of the central nervous system and can be regarded as a vascularised neuronal structure (34).

Before the advent of the ophthalmoscope in 1851 it was not possible to inspect the fundus of the eye. After that date many of the serious retinal diseases were recognized and described over a short time period. In 1856 von Jäger described retinal changes in diabetic people, further described by Hirschberg in 1890-1891(2). In 1877 Mackenzie and Nettleship described microaneurysms. The aneurysms got renewed attention in 1943 as early signs of DR through the works of Ballantyne and Loewenstein (35).

The visible signs of diabetic retinopathy are micro aneurysms, hemorrhages, hard and soft exudates formation of new vessels, vitreous hemorrhages and complications such as retinal detachment and retinal edema. Prior to the development of these visible signs there are changes in blood flow and loss of autoregulatory mechanisms which play a role in the pathophysiology. In eyes without visible retinopathy arteriolar constriction with decreased blood flow has been described however later in the disease process the arterioles begin to dilate and the retinal blood flow increases, causing shear stresses in the capillaries (10).

Deterioration of color vision and contrast sensitivity together with electrophysical changes indicate that the neuroretina is affected early (36-38).

Usually DR does not have much symptoms until a hemorrhage or edema appear, a thickening of the macula up to 250-300 μ is compatible with normal visual acuity (39).

The retina´s demand for oxygen is higher than that of any other tissue in the body with the retina fed by two separate systems of vessels. The outer third relies on choroidal circulation and the inner two thirds on retinal circulation. The central artery goes through the optic nerve, enters the inner eye and is divided into small arteries, arterioles and then capillaries partly in the ganglion- and nerve fibre layer,

partly in the inner nuclear layer. The capillary wall consists of endothelial cells, pericytes and the basement membrane. Tight junctions exist between the capillary endothelial cells constituting the inner blood-retina barrier. These junctions limit the passage of larger molecules through the capillary wall. Pinocytosis transfer metabolites through the capillaries to and from the retina.

The blood is returned to the circulation from the venules to smaller veins which then merge into the central retinal vein. The muscle-cells and the pericytes in the vessel-walls allow for changing the diameter of the vessel according to the pressure differential across the lumen.

2.1.2 Pathological findings

Microaneurysms are seen as small red dots and have a diameter between 10 to 100 microns, meaning that small aneurysms of less than 20-30 µm diameter cannot be seen with the ophthalmoscope. However with modern techniques, especially fluorescein-angiography they can be visualized. Their structure have been clarified especially by the works of Ashton, Ballantyne and Lowenstein (35). The basement membrane becomes thickened and pericytes and endothelial slowly disappear leaving the affected vessel non-functional with ischemic areas. The disappearance of pericytes weakens the vessel-wall and there will be a fusiform or unilateral bulging forming the aneurysm. Blood flow may continue but the aneurysm may also be obliterated. The wall is weak and may break resulting in haemorrhage. Aneurysms are principally found in the borders of ischemic retinal areas.

Two types of exudates exist, hard and soft. Hard exudates are signs of leakage because of damaged tight junctions. The exudates consist of lipo-proteins, are yellow-white in colour and sharply demarcated, coalescing exudates in the macular area may severely impair visual acuity and leave scars. Presence, of even small exudates, in the macular area bring changes in the multifocal ERG with prolonged so-called implicit time (40).

Soft or cotton-wool exudates have woolly borders, are grey-white and sometimes difficult to see. They represent areas with stasis of axoplasmic flow and mostly disappear within a few months.

The formation of new vessels, referred to as proliferative retinopathy (PDR), is a very grave sign, with high risk of developing blindness or severely impaired vision if left untreated. In particular new vessels on or in the immediate vicinity of the optic disc give rise to serious complications. They are *de novo* vessels with fragile walls and no blood-retina barrier function. Bleedings from them are common and cause contracting cicatrisation of adjacent vitreous with risks of consecutive tractional retinal detachment. The cause is supposed to be an ischemic retina with expression of HIF-1ά and subsequent production of growth factors with an imbalance between

angio- and antiangiogenic substances (41). A further complication is vascularisation of the iris with its great risks of developing hemorrhagic glaucoma.

Retinal oedema occurs because of leakage from the capillaries, if in the periphery there will be no symptoms but central oedema may cause deterioration of visual acuity. Macular oedema and exudates can appear at all stages of retinopathy, with low visual acuity in diabetes most often caused by central oedema. Photo examination and biomicroscopy are effective in discovering oedema but Ocular Coherence Tomography (OCT) is now the method of choice.

As an aid in the diagnosis and treatment of oedema the conception of Clinically Significant Macular Edema, CSME, was created. The oedema is diagnosed by contact lens examination, photographic methods and according to ETDRS defined as:

- Thickening of the retina within 500 μm of the fovea, or
- Hard exudates within 500 μm of the fovea with adjacent retinal thickening, or
- Thickening of the retina of at least 1-disc area in size that extends to within 1-disc diameter of the fovea (42).

With OCT technique Diabetic Macular Edema, DME, is diagnosed as follows:

- An area of two disc areas or more of thickening of which any part is within one disc diameter of the fovea.

If the criteria above are fulfilled treatment is indicated.

Two barriers prevent leakage of plasma, the retinal pigment epithelium and its tight junctions and the tight junctions between the retinal capillary endothelial cells. This blood-retina-barrier normally prevents leakage of plasma from the vessels into the retina. Early in the disease however, leakage into the vitreous of plasma may be diagnosed. By VEGF-action the proteins Zona Occludens, Occludin, Claudin and more, gradually lose their functions, compromising the intercellular junctions allowing a net leakage of fluid into the retina. The leakage is determined by the factors of Starling´s law (43-45).

Dilation of arterioles, as happens in DR, increases the hydrostatic pressure (Poiseuille´s law) as well as increasing the leakage. The same happens if the oncotic pressure in the interstitium increases, by the presence of above all albumin, in ocular hypotony, or when the capillary-oncotic pressure is lowered by hypoalbuminemia. Co-existing hypertension aggravates the condition. Increased levels of VEGF are strongly correlated to breakdown of the BRB and oedema (41).

It has also been shown that patients with diabetes of long duration and retinopathy of various degree have altered electrical activity in the rods and probably also in the post receptor components (46). However, such findings have also been found in

patients with newly diagnosed Type 2 without visible retinopathy so are not exclusive to those with advanced disease and present retinopathy.

2.1.3 Classification of diabetic retinopathy

It is important to diagnose DR early, follow the retinopathy and estimate the risks of developing vision-threatening lesions and treat them as appropriate as possible. Several systems of classification of the retinopathy have been used, for example Hirschberg´s (1890), Ballantyne and Michaelson`s (1947), Scott´s (1951), Alaerts and Slosse´s (1957) and more.

Currently in use are the Diabetic Retinopathy Disease Severity Scale, the EURODIAB scale and the modified WESDR (47, 48). For diagnosis ophthalmoscopy, photographs, digital or analogue, are used. Photos are taken in the seven standard ETDRS fields of 30°, the original ETDRS method, or two to more 45-50° photos. Ultra-wide photos with 200° coverage, allowing diagnosis of very peripheral lesions have come in use. Today digital cameras are used and the photos are read on a monitor, often at reading centres. Automatic reading systems are under development. A difficulty is that oedema without exudates may be difficult to diagnose because of the need for taking stereo-pictures, contact lens examination or using OCT technique.

Table 2.1
Definitions of grading levels (49, 50) (according to the modified Arlie House definitions.

Level 10	No retinopathy
Level 21	Microaneurysms only, or blot- hemorrhages or cotton wool exudates in the absence of microaneurysms
Level 31	Mild NPDR, characterized mainly by hard exudates and/or mild retinal retinal hemorrhages, less than those in Standard Photo 2A
Level 41	Moderate NPDR, characterized by intra-retinal hemorrhages, cotton wool exudates and IRMAs, level 51 not met.
Level 51	Severe NPDR characterized by one of the following: severe retinal hemorrhages; moderate IRMAs equaling or exceeding those of SP 2A and 8 A respectively, definite venous beading.
Level 60	Fibrous proliferations only.
Level 61 Level 65	Scars of scatter photocoagulation, presumably directed at new vessels but no evidence of levels 60 or 65. Moderate PDR characterised by definite new vessels on the disc, , graded less than SP 10A or moderate new vessels elsewhere or vitreous or preretinal hemorrhages, but level 70 not met
Level 70	PDR with high risk characteristics
Level 80 Grading not possible, because of severe diabetic changes	

IRMA= intraretinal microvascular abnormity. NPDR= Non-proliferative Diabetic Retinopathy. PDR= Proliferative Diabetic Retinopathy. SP= standard photo.

Table 2.2.
Grading levels according to EURODIAB IDDM (REF Aldington et al and compared to the WESDR scale.

Level	Eurodiab	WESDR
No retinopathy	Level 0	Levels 10-15
Mild, non proliferative	Level 1	Levels 20-31
Moderate/severe nonproliferative	Levels 2-3	Levels 41-55
Proliferative (or photocoagulated)	Levels 4-5	Levels 60- 70
Cannot be graded	Unassessable	Level 80

Table 2.3.
Diabetic retinopathy severity levels and associated clinical findings.

Proposed Disease Severity Level	Findings observable on dilated optalmoscopy
No apparent retinopathy	No abnormalities
Mild nonproliferative diabetic retinopathy	Microaneurysms only
Moderate nonproliferative diabetic retinopathy	In between mild and severe
Severe nonproliferative diabetic retinopathy	Any of the following: more than 20 intraretinal hemorrhages in each of 4 quadrants, definite venous beading in 2+ quadrants, prominent intraretinal microvascular abnormalities in 1+ quadrant, No signs of proliferative retinopathy
Proliferative diabetic retinopathy	One or more of the following: neovascularisation, vitreous/preretinal hemorrhage.

Diabetic macula oedema is usually classified as being present or not. If present grading in mild, moderate or severe according to the following is done.

- Mild - Some retinal thickening or hard exudates in posterior pole, distant from centre of macula
- Moderate - Retinal thickening or hard exudates approaching the centre of the macula but not involving it.
- Severe - Retinal thickening or hard exudates involving the centre of the macula.

In summary, the existence of internationally accepted grading systems have greatly facilitated comparing prevalence, incidence and treatment results. They play an important role in screening and controlling systems and in determining when to treat.

2.1.4 Prevalence and incidence of diabetic retinopathy

Diabetic retinopathy (DR) is a very common complication of diabetes in both Type 1 and Type 2 diabetes. It is one of the leading causes of vision impairment and blindness and a leading cause of vision loss in people between 20 and 74 years of age. After twenty years of disease almost all people with T1DM have developed DR; in T2DM about 60% (51).

Incidence and prevalence vary in different studies. This depends on ethnic differences, different patient cohorts, hospital vs general practise, how people follow screening and control programmes, disease advances, therapy and when the study was started but also on inclusion criteria and definitions.

Prevalence of any DR in T2DM was 50% in the WESDR study. Eight years later The Beaver Dam Study showed 35%. The National Diabetic Retinopathy Screening Service for Wales found any DR and sight threatening DR in T1DM 56.o% and 11.2% respectively; in T2DM it was 30.3% respectively 2.9%. In the UK in the general population PDR was 1-8% vs 20% in the hospital database, CSME had a prevalence of about 6% in the general population respectively18%.in the hospital population Any DR was found in 80% of participants in this study (52-58).

In a Swedish cohort of Younger Onset of Diabetes (<30Years) 64% had any DR, with severe or PDR in 25% and CSME in12%. People with Older Onset (≥30 years) showed any DR 36%. Severe or PDR was found in 12% respectively 9% (59).

In a cohort of 2964 newly diagnosed non-insulin dependent in the UKPDS, 39% of men and 35% women showed DR. 8% of men and 4 of women had “marked” retinopathy with cotton wool spots or intraretinally microvascular abnormities (60).

2.1.5 Therapy of diabetic retinopathy

An effective glycaemic control is the most effective way to prevent or postpone the appearance of DR. It has been shown that intensive glucose and blood pressure control reduces risk for developing DR as well as slowing progression and reducing the risk of vision loss(DCCT ,UKPDS, WESDR).

In the 1930s pituitary ablation in diabetic dogs showed that the course of diabetic retinopathy was altered.to the better. Consequently the method was used in humans. In 1969 a pooled report claimed that 75% of 708 patients had a stabilized retinopathy 6 months post-operatively (61). However, long-term results showed that after 5 years approximately one third of the patients exhibited progression. Serious draw-backs were post-operative complications and the need for life-long hormone substitution (61).

Some reports of interferon treatment appeared in 1980s, but further testing was abandoned.

Experimental photocoagulation was reported in 1867, 1882, 1893 and in a human eye in 1927. In 1949 Meyer-Schwickerath reported on therapeutic photocoagulation as well as in 1951, 1951, 1967 and 1969. The technical development went from sunlight to xenon lamps to several types of lasers (62-64).

The Diabetic Retinopathy Study (DRS) showed that pan-retinal photo-treatment reduces the risk of severe vision loss by at least 50% in eyes with proliferative or severe retinopathy (64). On the other hand the ETDRS study showed that treatment of mild, moderate or severe retinopathy did not change the visual outcome (64).

At present proliferative retinopathy is treated with pan-retinal photo-coagulation (scatter treatment), with 1200-3000 burns with a spot size of 200-500 µm, the burns are placed from the vessel arcades out to the periphery. Treatment of very severe pre-proliferative retinopathy might be considered.

Macular oedema can be laser treated with smaller (50-100 µm) and weaker spots in the oedematous area, either in a grid pattern or directly on leaking spots. It is then important to avoid the foveal avascular zone. Treatment decreased the risk of visual acuity loss by 50%, but after 1 year 40% had persistent oedema (65). As VEGF and inflammation plays important roles in the genesis of macular oedema and proliferative retinopathy anti-VEGF (66) and anti-inflammatory substances are now used in treatment as they seem to have superior effects compared to laser treatment (67).

Intraocular long-acting steroids improves oedema but have increased risks of glaucoma and cataract formation. However they are important adjuncts in treating Anti-VEGF non-responders (68).

Drawbacks of pan-retinal laser treatment are deteriorated night-vision and worsened visual field as the treatment is destructive. Another effect sometimes seen is enlarging photo scars, “creeping”, that may affect the macula. The treatment may also be quite painful.

In severe cases with retinal detachment, vitreous bands, organized haemorrhages or traction of the central macula vitrectomy is used most often combined with endolaser treatment (69). Current studies are focusing on pharmacological methods to attain a posterior vitreous detachment and thus diminish traction of the macula and preventing the vitreous to become a scaffold for neovascularisation.

Trials with drugs blocking the pathological metabolic pathways are logical steps but have as yet had only modest success (70).

Anti-VEGF substances are rather effective but about 25% of patients react inadequately or are non-responders. Anti-inflammatory drugs as subtenonal steroids can then be used, or better yet, slow release intraocular steroids and intraocular NSAIDs. New experimental methods include certain antibiotics, immuno-suppressants, oxidative stress reducers, angiopoietin, gene therapy and more (70).

In summary, the introduction of effective laser, anti-VEGF and anti-inflammatory treatment have completely changed the earlier gloomy prognosis of diabetic retinopathy but much research, trials and development remain to do. There is

however rather good hope of bettering the future of visual functions in people with diabetes.

2.1.6 Summary

DR is a leading cause of visual impairment and blindness despite advances in treatment. Occurrence of DR is very common in people with diabetes. After ten years the prevalence in Type 1 DM is 60%, after 15 years 80%. In type 2 where the actual duration is very difficult to determine and found prevalences vary; Klein et al found a prevalence of 67 % after ten years and 10% had PDR, while Yanko et al found 23% after 11-13 years, and after 16 years or more increasing to 60% (71, 72).

DR has two existing forms: one with microaneurysms, bleedings vascular abnormalities and in the end vascular proliferations; in the other form vascular leakage leading to retinal oedema and visual impairment. The two forms most often exist. Despite the differences between Type 1 and Type 2 diabetes there is no visible difference between the ophthalmologic changes in the two forms of diabetes.

2.2 Diabetic foot ulcers

2.2.1 Introduction.

In the early writings about “honey-urine” sick persons there were remarks about foot and leg ulcers. Lack of communication. resulted in this knowledge only occurring in isolated areas. In the 18th century however scientific meetings and circulation of scientific reports began to take place. In 1888 the London surgeon T.D. Pryce described a case of a diabetic patient, a hawker. with infected plantar ulcers with diminished cutaneous sensibility, and absence of the knee-jerk-reflex. After the patient´s death in diabetic coma Pryce found nerve degeneration and diseased arteries. He concluded that:”Ulcers are not uncommonly associated with locomotor ataxia, with which disease occurs comparatively frequently”; ” - - -does occasionally produce peripheral neuritis”; and finally concluded:”- - -it is possible- nay, even probable- that diabetes may sometimes play an active part in the causation of perforating ulcer.”

Pryce thus described a clinical picture of infected ulcers, peripheral vascular disease, peripheral nerve degeneration, and that, as a hawker, his patient walked a lot probably had had many slight traumas to his feet (73).

Now, 130 years later, we know that diabetes certainly is a causative factor in the genesis of foot ulcers.

In 1975 Walsh et al reported on a group of patients with diabetes, foot ulcers, advanced retinopathy and a peculiar psychological indifference to their situation, despite a grave prognosis (74). One year later Steel et al reported on four young NIDDM patients with short histories of diabetes but with recurrent foot ulceration and severe PDR, three with over-weight and three with hypertension (75). In 1990 Glynn et al pointed at the need of looking for further complications, particularly retinopathy, in diabetic patients presenting with foot ulcers (76).

Life-time risk of developing a diabetic foot ulcer is up to 25% and 3-12 % of all people with diabetes have a history of foot ulceration (77, 78). In amputation cases about 80% of diabetic people have a history of foot ulceration. The rate of recurrence is as high as 50% in some reports (79). Thus, foot ulcers are a leading cause of morbidity and mortality in diabetic people, also severely affecting their quality of life.

When an ulcer is established the risk of infection is high, with risk of gangrene, cellulitis, sepsis, amputation and death. In fact the 5 year mortality is 50-60 % (80).

Important factors in the pathophysiology is neuropathy and peripheral vascular disease with ischemia defined as absence of pulses, low ankle-brachial index and low transcutaneous oxygen pressure. In the UK neuropathy alone was found in 46%, ischemia alone in 12 %, neuroischemia in 30 % and neither in 12 % Walters et al in the UK and Nyamu et al in Kenya found that the greatest ulcer proportion was neuropathic with prevalence of neuropathy varying between 25% and 35% (81-83).

Distal sensorimotor neuropathy causes insensitivity to traumas and temperature, and to small-muscle dysfunction which leads to deformities of the foot.

Autonomic neuropathy leads to reduced sweating, with the skin becoming prone to cracking and to increased blood flow. The increased blood flow does not however improve the oxygenation, probably because of shunting capillaries (29, 30).

The pathoanatomical findings of Sangiorgi of deranged vascular beds in diabetic toes with signs of extensive micro occlusions of capillaries and the existence of arterio-venous shunts in some ways revive the theory of “small vessel disease”

In people with neuropathy the annual incidence of ulceration was 5 times higher than in people without neuropathy (84, 85).

2.2.2 Classification of diabetic foot ulceration

Several methods of classification of ulcers exsist: Wagner´s method, University of Texas system, SINBAD and more (86-88).

Table 2.4
Wagner´s classification of foot ulceration.

Grade 0	Pre-ulcerative lesion
Grade 1	Partial-thickness wound, up to but not through the dermis.
Grade 2	Full thickness wound extending to tendons or deeper subcutaneous tissue but without bone involvement or osteomyelitis
Grade 3	Full thickness wound extending to and involving bone
Grade 4	Localised gangrene
Grade 5	Gangrene of the whole foot

Table 2.5.
University of Texas classification system.

Stage	**0**	**1**	**2**	**3**
A	Pre or postulcerative completely epithelialised	Superficial wound, not involving ten-don or bone	Wound penetrating to tendon, cap-sule or bone	Wound penetrating to bone or joint
B	Infected	Infected	Infected	Infected
C	Ischemic	Ischemic	Ischemic	Ischemic
D	Infected and ischemic	Infected and ischemic	Infected and ischemic	Infected and ischemic

Table 2.6.
The SINBAD system for classifying foot ulcers.

Site and side				
Ischemic?	Yes/No	If yes, is the ischemia critical?*		Yes/No
Neuropathy?	Yes/ No	If Yes Has the patient got Charcot?		Yes/ No
Bacterial infection	Yes/ No	Is there osteo-myelitis		Yes/ No
	0	1	2	3
Area	Skin intact	< 1 cm^2	1-3 cm^2	>3 cm^2
Depth*	Skin intact	Superficial	Deep	Bone

*Categories as defined in UT, S(AD) SAD and Pedis.

2.2.3 Therapy of Foot ulcers

Of primary importance is identifying the foot at risk, then stopping iterative trauma to the foot by better shoes. If an ulcer is present: off-loading and immobilisation,

surgical debridement if necessary and antibiotics after identifying the offending organisms.

If the ulcer is recalcitrant to heal it is important to assess the possibility of endovascular procedures or by-pass operation. As a final procedure minor or major amputations are performed and they may have to be repeated.

The therapy is best carried out in a specialised foot-clinic. Despite this some ulcers are, however, difficult to treat and they heal very slowly if at all. If healing time is three months or more and vascular procedures are not possible they can be characterised as" hard-to heal" or" chronic", other means must then be used, such as topical growth factors, bioengineered biological coverings, vacuum treatment, vascular surgical intervention if possible and Hyperbaric Oxygen Treatment (HBO).

The HBO treatment modality improves oxygenation of ischemic tissue, by increasing the oxygen concentration in plasma thereby increasing the diffusion distances about three times. This stimulates angiogenesis, reduces oedema and inflammation, augments the formation of granulation tissue, improves leukocyte function and inhibits anaerobic bacteria. Most investigations have shown positive effects on ulcer healing with HBO, but it remains somewhat controversial in regard to efficacy (89).

2.2.4 Summary

Diabetic foot ulcers are caused by peripheral neuropathy, sensorimotor and autonomic, and/or macro- and microvascular disease. Ulcers are a leading cause of amputations in diabetic persons and are a serious threat to life itself as well as Quality of Life. Several ulcer classification systems exist. Chronic ulcers demand treatment at a specialised foot clinic and even then the prognosis may be guarded. Of the used therapeutic modalities HBO treatments have shown promising results.

2.3 Hyperbaric Oxygen Therapy

2.3.1 Introduction

Hyperbaric oxygen therapy (HBO) has been used to treat many different diseases and conditions. Approved use of HBO is currently treatment of ischemic ulcers, serious infections with anaerobic bacteria, carbon-monoxide poisoning, radiation injuries and decompression sickness (DCS). Many other diagnoses have been treated, but reports (mostly positive) are anecdotal (90).

In a healthy individual the necessary oxygen, is delivered to the tissues by the blood-vessels, the haemoglobin-bound oxygen being delivered by diffusion to the tissues. If, however, the blood-circulation is damaged and not enough oxygen is delivered the tissue becomes ischemic. To treat the ischemia it is necessary either to diminish the need for oxygen or increase the amount of diffusible oxygen. The haemoglobin-molecules of the erythrocyte are saturated to 97% and more oxygen cannot be bound to it. At normal atmospheric pressure (760 mm Hg =1 ATA)) about 3 ml oxygen is dissolved in one litre plasma. If breathing 100% oxygen about 15 ml oxygen is dissolved. If, however, the pressure is raised to 2.5 ATA and 100% oxygen is used 60 ml oxygen is dissolved and the diffusion distance is augmented three times. This is enough to sustain life in absence of red blood corpuscles

To systemically treat a person with HBO one has to use a hyperbaric chamber, mono- or multiplace, where one can regulate the pressure and monitor the person. The person is placed in the chamber, the pressure is gradually increased to allow for equalisation of pressure in the middle-ear, noted as the compression phase. After 5 minutes a pressure of for example 2,5 ATA (15 m sea-water depth) is reached. When the planned pressure-plateau is reached the person begins to breath 100% oxygen, either by a facemask or an open mask. A treatment session is commonly 60-90 minutes, sometimes interrupted by pauses to breath standard air. Over a period of 5 minutes the pressure is then gradually diminished to 1 ATA, noted as the decompression phase. The person is observed for some time and can then be sent home.

The number of sessions, and the “depth” depends on the treatment plan. Treatments with 100% oxygen are not in point of principle allowed to reach higher pressures than 2.5 ATA because of the risk of oxygen toxicity.

If the decompression is to fast there is risk of decompression sickness (DCS/DCI) which is a serious complication.

2.3.2 Compression and decompression. Gas laws.

The effects of HBO are based on the gas laws and physical, physiological and biochemical effects of high pressure and high pressure oxygen.

The principal gases of the atmosphere are Nitrogen 78% volume %, Oxygen 20.9%, Carbon dioxide 0.04% and trace amounts of inert gases. The total atmospheric pressure is 760 mm Hg at sea-level, this equals 1 absolute atmosphere (1 ATA). According to Dalton´s law the partial pressure of oxygen then is 160 mm Hg, that of nitrogen 600 mmHg, the proportion of the gases are unaltered at different pressures.

The solubility of a gas in a liquid (or tissue) is proportional to its pressure and the coefficient of solubility of the liquid or tissue in question (Henry´s law).

Boyle´s law states that the gas volume is inversely proportional to its pressure. That means that in compression open airways will have the same volume as before but if there is a closed space as the middle-ear, a shut-off sinus or tooth with enclosed air it will diminish in volume and thus hurt or become damaged, due to the "squeezing". An eye prosthesis with enclosed air may implode. Middle-ear barotraumas occurs during the compression phase because of failure of equalising the middle ear and ambient pressure by means of the Eustachian tube.

During treatment session, nitrogen will be dissolved in the blood and especially in fatty tissues (Henry´s law). Time, pressure levels and solubility coefficient determine the extent. During decompression the dissolved nitrogen-gas must be evacuated through the lungs. If decompression is too fast gas-bubbles will form in the tissues and blood because of super-saturation and cause gas-embolism, decompression sickness (DCS). Stationary bubbles can cause pain and mechanical injuries. Rapidly expanding air in the lungs can rupture the alveoli, a very dangerous complication.

Depending on the time spent at a certain pressure one has to monitor the return to atmospheric pressure, To diminish the risks of DCS, decompression tables have been constructed, for example the US Navy tables, the Blackpool tables (UK) and similar tables. The same gas-laws and the same complications are valid both in diving, caisson work and in the hyperbaric chamber. Despite following decompression-tables, DCS is not uncommon in diving and especially in caisson-work. Inter- and intra-individual susceptibility are important factors. In an survey of caisson workers, 4% of the workforce had 50% of the DCS events (91).

The existence of a patent foramen ovale or arteriovenous shunting in the lungs, provides a pathway for getting air into the arterial circulation, with the lungs otherwise acting to filter out bubbles .Persistent foramen ovale is rather common, occuring in 5% in healthy individuals (92). An autopsy report from the Mayo Clinic

claims 27% (93). The foramen allows right-left shunting as does arterio-venous shunting in the lungs

Even when the tables are followed microscopic bubbles are formed while at the same time one finds extracellular vesicles (EV) (94)(95). They are classified in tree classes: exosomes, microparticles and apoptotic bodies. EVs can be released from various tissues and cells and are now considered an intercellular communication and transport system. Their patterns differ in healthy and diseased persons (96, 97).

Table 2.7.
Comparison of Pressure Units.

ATA	mm Hg	kPa	Msw	Atm
1	760	101.3	0	0
1.5	1140	152	5.16	0.5
2	1520	202.6	10.32	1
2.5	2532	253,2	15.48	1.5
3	2280	303,9	20.64	2
4	3040	405.2	30.97	3

ATA= atmospheres absolute; mm Hg= millimeters of mercury; msw= meters of seawater; atm= atmospheres.
Adopted from Textbook of Hyperbaric medicine 5th ed./K.K. Jain

Table 2.8.
Partial pressures of oxygen at ATA 1-4.

ATA	Partial pressure of O_2 in air (ATA)	Partial pressure of O_2 In air (mmHg)
1	0.21	159.7
1.5	0.31	239.4
2	0.42	319.2
2.5	0.53	394.0
3	0.63	478.8
4	0.84	638.4

Adopted from Textbook of Hyperbaric Medicine. 5th ed. K.K. Jain.

Table 2.9.
Signs and symptoms of decompression sickness

Type 1 DCS	Limb and joint pains, itching, skin rash.	Alerting signs
Type 2 DCS	Neurological: (cerebral, spinal, vestibular)	Serious signs
Type 2 DCS	Pulmonary: (dyspnea, hyperventilation, chest pain, acute respiratory distress.)	Serious signs
Type 2 DCS	Cardiac: tachycardia, arrhythmias.	Serious signs

After Jain REF Textbook of Hyperbaric Medicine, 5th ed

There are five issues of hyperbaric treatment:

- compression injuries (barotrauma)
- decompression illness (DCS/DCI))
- toxicity of oxygen and nitrogen.at higher pressures,
- breakdown of the hyperbaric chamber with fast loss of pressure and
- fire in the chamber.

In addition to the oxidative stress in diving or presence in a hyperbaric situation the formation of bubbles damages the endothelium and neutrophils, with the neutrophil destruction inducing an inflammatory cascade further aggravating the Repeated treatment or diving sessions seem to protect tissues by increasing sirtuins. Sirtuins are important in regulating antioxidant and redox signalling and have become known as "anti-aging molecules" (98). Pre-conditioning HBO has been used as an adjunct before coronary by-pass surgery. In the treatment of DCS the patient is recompressed, then slowly returning to normal pressure, according to special decompression tables. Often oxygen is used as an adjunct to diminish lesions.

2.3.3 The road to understand decompression sickness.

The air pump was invented in 1650. Using the air pump Robert Boyle compressed and then decompressed a snake and observed a gas bubble in its eye, the first scientific observation of DCS. (1670). Salvaging companies had used diving bells since at least the early decades of the 17th century. The diver stood on a step with his head and trunk in the bell. When the bell was lowered into the water the water rose in the bell and the air in it was compressed but there was still air-volume enough so the diver could breathe, see and use tools. Notable Swedish achievements were the salvaging of the guns of the ships Sancta Sophia 1658, Vasa and Cronan 1663-65 and 1682-1686 by the von Treileben company. In Francesco Negri´s *Viaggio settentrionale in otte lettere* (1700) the diving process at Vasa is described but without mention of possible decompression complications. According to Negri´s report the diver reported that he could be submerged for 15 to at most 30 minutes. The diving depth was 20-30 m (3-4 ATA at these depths). According to modern decompression tables this barely allowed a direct ascent to the surface without decompression stops.

In 1734 the Swedish engineer and scientist Mårten Triewald published "Konsten att lefwa under Watn,," (The Art of living underwater…) where astonishing modern thoughts of pressure physiology and gases are expressed. He had earlier started a salvaging company using a diving bell influenced by Edmond Halley (99).

In 1837 Siebe´s diving suit and air pump was introduced, the equipment allowing extended stays underwater, which greatly increased the risk of decompression illness as did work in high pressure caissons used in bridge constructions and in pressurised mining work. Instances of decompression illness and death became common. In 1841 Pol and Watelle, found gas bubbles when doing autopsy of fatalities in mine-workers and also observed that recompression had relieved the symptoms in survivors. A report of this and of recompression was sent to l´Àcademie des Sciences and published in 1854.

Paul Bert showed in 1878 that the bubbles were formed of nitrogen, and Haldane (1907) found empirically that a decompression to half the original pressure did not result in bubbles. Haldane constructed tables for safer ascent from different depths and exposition time (working time) These tables are the foundations of today´s decompression tables. In 1937 Behnke and Shaw introduced oxygen as an adjuvant in the treatment of DCS Bull (100). However the process of DCS is complex including oxidative stress reactions and formation of inflammatory components.

In summary, clearly treatment in hyperbaric chambers demands a careful selection of patients if possible, scrupulous adherence to decompression tables, careful monitoring of the patients and first class maintenance of the chambers and to take possible fire hazards into consideration.

2.3.4 Toxicity of nitrogen and oxygen.

2.3.4.1.Nitrogen.

Nitrogen is an inert gas at normal pressures. However gases may have other properties at high pressure than at normal pressure. Nitrogen´s solubility in fatty tissues is five times more than in muscular and connective tissues. It´s elimination in the lungs is about five times slower than that of oxygen and carbon dioxide. When eliminating the soluble nitrogen the decompression must be gradual, or the dissolved gas will get supersaturated and gas bubbles may form, probably emanating from hydrophobic spots in the walls of vessels. In repeated decompression it seems as if these spots diminish in number and an adaption to less bubble formation occurs. Also the sirtuins, SIRT 1-7, have a protective effect in repeated HBO treatments (98).

At higher pressure, 4 ATA and more (partial-pressure of N_2>3.2 ATA), the nitrogen contents of the air begin to have an inebriating effect, which gets worse the higher the pressure gets (Behnke). This effect has caused many accidents and deaths in underwater work and has led to the introduction of other gas mixtures to use in deep water/ high pressure work (101).

2.2.3.2.Oxygen.

Contrary to nitrogen oxygen has many biological effects. Discovered independently by Priestley and Scheele in 1774 respectively 1776, its properties were investigated by Lavoisier and Séguin and others in the 1780ies. Experimenting on guinea pigs and dogs Lavoisier found that prolonged exposure of 100 % oxygen at increased pressure killed the animals (102). It also became evident that low oxygen pressure at altitude, as seen in balloon flights to heights more than 5000-6000 m necessitated breathing extra oxygen. Travellers´experiences of mountain sickness in high mountains as the Alps, inner Asia, the Andes and the Himalayas, underlined this issue. It was thus determined that too little or too much oxygen was dangerous and even lethal.

Oxygen, or more correctly dioxygen O_2 is chemically active, the more so in singlet form and as ozone (O_3). Oxygen is necessary for life, except for anaerobic bacteria. These organisms use less effective metabolic pathways for their energy production than aerobic organisms and oxygen may be toxic or lethal for them.

In aerobic organisms the oxygen transported into the cell is used in the mitochondria (about 80%) and in the subcellular organelles (20%). Electrons from various substances combine with oxygen, releasing energy which is used for the pumping of H+ ions from the inside of the mitochondria. When the H+ ions diffuse back energy is formed which is used in phosphorylating adenosine diphosphate (ADP) to adenosine triphosphate (ATP). One glucose molecule gives theoretically 38 ATP, due to procedural losses about 30 ATP are left.

In land living vertebrates atmospheric oxygen is taken up in the lungs. In the lungs the oxygen in the alveoli is transferred by diffusion to the haemoglobin in the erythrocytes through the alveolar epithelial cells. The oxygen rich arterial blood circulates and delivers its haemoglobin bound oxygen to the tissues at the same time receiving carbon dioxide. It follows that decreased number of alveoli, impaired diffusion through the alveolar walls or insufficient air circulation in the lungs will lead to lack of oxygen and excess of carbon dioxide as will a lack of haemoglobin.

In animal experiments Lavoisier and Séguin had found that oxygen at high pressures could be lethal. Later Paul Bert experimented on animals and on himself and found neurological symptoms and injuries after treatment with hyperbaric oxygen. However the symptoms disappeared if breathing air at normal pressure, the so-called Paul Bert effect. Prolonged exposition of high pressure oxygen would, however, cause permanent neurological injuries or death as well as death of the retinal cells, the John Bean effect (103).

In 1899 Lorrain-Smith reported on lung-complications when using normobaric 100% oxygen for 24 hours or more as well as at shorter exposition times but higher pressure, the so-called Lorrain-Smith effect. The cause seems to be increased

thromboxane synthesis in the lungs causing alveolar damage with oedema, haemorrhages and inflammatory reaction. If the exposition is stopped there will be no sequelae but if the exposition is continued it will lead to fibrosis and emphysema.

In normal cellular metabolism ROS are produced. In a hyperoxic state the production of short-lived ROS is markedly increased; they oxidise sulhydryl (SH) groups of enzymes, interact with DNA and promote lipoperoxidation of cellular membranes, oxidation of glutathione and of pyridine nucleotides, resulting in impaired energy production. Lengthy oxygen exposition initiates increasing production of the vasodilator NO. The hyperoxic vasoconstriction in the brain is thus counteracted which allows excessive oxygen to reach the brain. Neurotransmitters are downregulated, for example GABA. In 1927 Tinel showed that cerebral arteries contracted when, oxygen was breathed, it was confirmed 1930 by Wolf and Lennox. It was also found that retinal vessels reacted in the same way. Later it was found that this action was reduced in persons with diabetes.

Acute oxygen poisoning may start as facial twitching, headache, dizziness and constriction of the visual field, followed by convulsions and unconsciousness. The visual field constriction is a common sign, and it has been proposed that it depends on vessel constriction in the peripheral retina, more probable it is due to a direct toxic effect on the retinal cells. It has been shown in young, healthy marine divers that slight toxicity effects may appear at low oxygen pressures when breathing 100% oxygen, where the pressure can be as low as 1.3 ATA. In most cases the symptoms will disappear when the victim breathes room air at the same pressure. Low serum glucose increases the tendency to toxicity.

There are reports of changes of the ocular refraction. in HBO treatment (104-106). The numbers of patients affected are however low with treatment sessions varying from about 28 hours to 40 hours totally. In one report the exhibition time varied from 150 to 850 hours totally. In the latter study cataract formation was seen in 7/ 25 (28%) in earlier clear lenses (104).

In four prospective studies Evanger studied 89 people who underwent standardised HBO treatment and concluded that a myopic shift developed in the majority, that the shift was temporary, and that the refraction returned to baseline after ten weeks. Changes in the refractive index of the crystalline lens seemed to be the mechanism behind the refractive handling (107-109).

Oxygen is necessary for life, however long-term exposures of high concentrations and/or pressure above 3.0 ATA may be dangerous, risking neurological and pulmonary transient lesions if treatment or exposition is interrupted but permanent if exposition is long. HBO increases the formation of ROS, normally this is counteracted by increased scavenger formation (super oxide dismutases).

3. Aims of the study

The aims of this thesis were to

- Evaluate long-term effects of hyperbaric oxygen treatment on visual acuity and retinopathy in people with hard-to-heal diabetic foot ulcers.

- Evaluate a plausible association between vascular impairment in the feet, defined as $TcpO_2$ levels at the dorsum of the feet, and macular thickness.

- Compare retinopathy and visual acuity between people with Type 2 diabetes and chronic foot ulceration and people with Type 2 diabetes but without a history of diabetic foot ulcer.

4. Study population and Methods

4.1 Patients.

4.1.1 Project 1

The **H**yperbaric **O**xygen Therapy in **D**iabetic Patients with Chronic **F**oot **U**lcer study (HODFU) is a prospective, placebo-controlled, randomized and double blinded study primarily evaluating the effects of HBO on ulcer healing in people with diabetes and chronic foot ulceration. The rational and patient enrolment criteria is published in Paper 1 (110).

Study participants were mainly recruited from the Diabetic Foot Clinic at Helsingborg Hospital, and to a minor degree from the Diabetic Foot Clinics at Skane University Hospital and Ängelholm Hospital. All participants had at least one full-thickness, persisting ulcer below the ankle with a duration of at least 3 months in spite of treatment at a multidisciplinary diabetes foot clinic for a period of at least 2 months. Inclusion and exclusion criteria are listed in Table 4.1.

Table 4.1
Inclusion and exclusion criteria for participation in the HODFU study.

Inclusion criteria	Exclusion criteria
Age over 18 years Known diagnosis of diabetes mellitus at time of inclusion Diabetic foot ulcer with a duration of at least three months Treatment at a diabetic foot clinic for at leat two months Need for or if needed possibility of vascular surgical intervention riled out by avascular surgeon Written informed consent	Contraindications for HBO • Severe obstructibe pulomoary disease • Untreated thyreotoxicosis • Untreated pneumothorax • Women in fertile age without conceptive prevention • Ongoing treatment with cisplatin, doxorubicin or disulfiram Vascular surgical intervention in the lower limbs within the last two months C-reactive proteon > 30 mg/L Known malignancy Misuse of alcohol or other drugs Investigators opinion Participation in another clinical trial

As the HODFU EYE study was launched later than the main study, the first 30 HODFU participants was not included in the EYE study. As shown in the Study

Flow Chart in Figure 4.1 two of the 64 consented people were excluded due to severe pre-existing eye disease and twelve were excluded as they received less than the prespecified 36 hyperbaric treatment sessions required for inclusion in the per-protocol analyses. The decision to use the per-protocol and not the intention-to-treat population in primary analyses, is based on the study´s principally safety outcome measures.

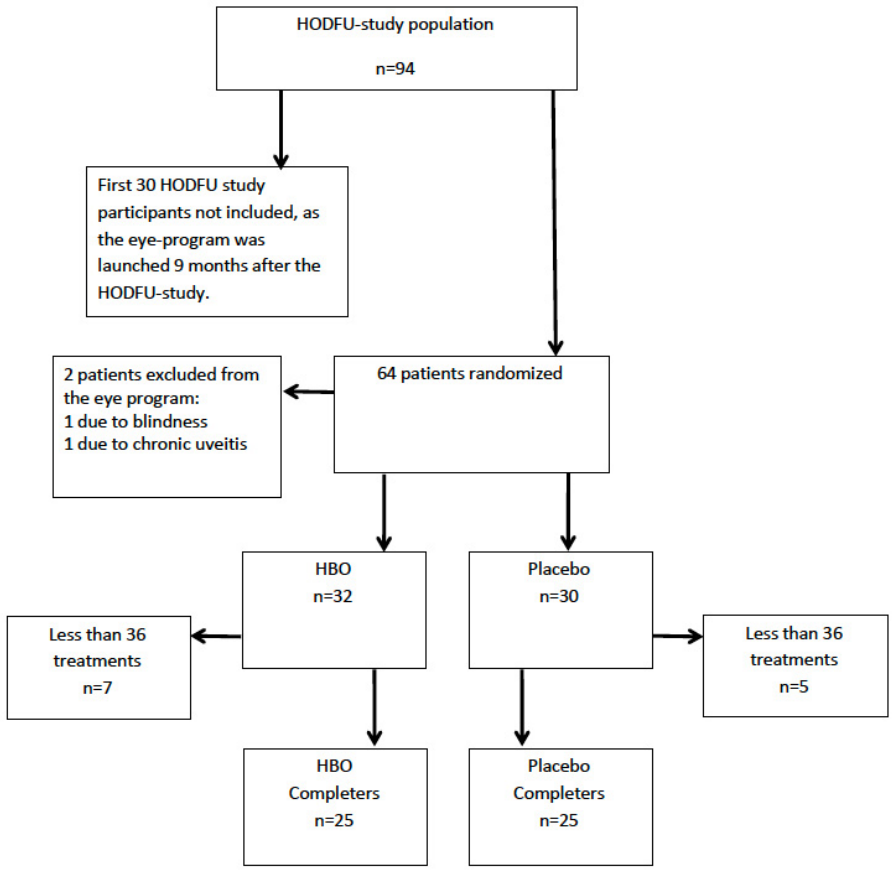

Figure 4.1
Study flow chart of the HODFU EYE study.

Altogether 50 participants, as in most diabetic foot studies mainly men, with a median age of 68 years and a diabetes duration of 20 years were included in the per-protocol analysis. Medical histories included stroke in one fifth of the cases, coronary heart disease in one third and lower limb vascular surgery in almost two thirds. Baseline characteristics were, as shown in Table 4.2, similar between groups. Complete data sets are given in Paper 2 (111).

Table 4.2
Baseline characteristics of the 50 study participants included in the HODFU EYE study per-protocol analysis.

	HBO	Placebo	
Number of participants	25	25	
Number of evaluable eyes	49	48	
Gender (male/female) (%)	84 / 16	76 / 24	n.s
Age (years)	64 (54-73)	70 (65-77)	n.s
Diabetes duration (years)	21 (13-36)	19 (9-39)	n.s
Type of diabetes (1/2) (%)	28 / 72	28 / 72	n.s
Smoker ever (%)	68	64	n.s
Medical history			
Hypertension (%)	88	88	,n.s
Dyslipidemia (%)	92	100	n.s
Coronary heart disease (%)	28	40	n.s
Atrial fibrillation (%)	20	32	n.s
Stroke (%)	20	20	n.s
Lower limb vascular surgery (%)	60	64	n.s
Above ankle amputation (%)	20	8	n.s
On dialysis/Renal replacement (%)	4	4	n.s
HbA_{1c} (mmol/mol)	65 (56-78)	60 (46-75)	n.s

4.1.2 Project 2

In Project 2 twenty consecutive participants in the HODFU EYE study (Project 1) underwent OCT examination. The participants, of which one quarter had type 1 diabetes and three quarters had type 2, were 73 years old and had a median diabetes duration of 22 years. Insulin was prescribed to 90 % of the study participants, ACE-inhibitors or angiotensin receptor blockers to 90 % and statins to 85 % (112).

Table 4.3
Participants´ baseline characteristics in Project 2.

Number of participants	20
Number of evaluable eyes	40
Gender (male/female) (%)	80 / 20
Age (years)	73 (45-85)
Type of diabetes (1/2) (%)	25 / 75
Smoker ever (%)	75
Medical history	
Hypertension (%)	80
Dyslipidemia (%)	85
Coronary heart disease (%)	40
Atrial fibrillation (%)	25
Stroke (%)	20
Lower limb vascular surgery (%)	70
Above ankle amputation (%)	15
On dialysis/Renal replacement (%)	15
HbA1c (mmol/mol)	55 (44-104)

4.1.3 Project 3

Ninety people with Type 2 diabetes and at least one full-thickness diabetic foot ulcer below the ankle of more than 3 months' duration were consecutively recruited between 2003 and 2007 from a Diabetic Foot Clinic that applied international treatment guidelines. Findings from eye examinations in these 90 people were compared with those from 180 people with Type 2 diabetes but without a history of chronic DFUs (control group) from the Eye Register at the Department of Ophthalmology in Helsingborg. Controls were matched for sex, age and diabetes duration, and were required to have had a routine eye examination within 18 months of their matched comparison (113, 114).

In accordance with local recommendations, all people with non-healing diabetic foot ulcers should be referred to the diabetic foot clinic in the catchment area. None of the people in our control group had been referred to the diabetic foot clinic, according to the patients' registry.

Patient characteristics are given in Table 4.4. The mean diabetes duration at time of study inclusion was just above 15 years. Renal function was worse in the diabetic foot ulcer group.

Table 4.4.
Baseline characteristics.

	Diabetic foot ulcer group	Control group	p-value
Participants (n)	90	180	
Gender, men (%)	84	84	n.s
Age (years)	69.1±10.1	69.4±9.6	n.s
Diabetes duration	15.9±10.9	16.4±8.5	n.s
Hypertension (%)	73	73	n.s
HbA_{1c} (mmol/mol)	60±14	58±11	n.s
Diabetes treatment			0.002
Liftestyle intervention only (%)	3	8	
Oral antiglycemic treatment but not insulin (%)	18	36	
Insulin included in glucoslowering treatment (%)	79	56	
eGFR (ml/min/1.73m^2	56±27	65±18	0.001
eGFR subgroup (%)			<0.001
≥60 ml/min/1.73m^2 (%)	41	64	
30-59 ml/min/1.73m^2 (%)	38	32	
< 30 ml/min/1.73m^2 (%)	21	4	
Median (IQR) visual acuity best eye (ETDRS letters)	50 (42.75;55)	55 (52;55)	0.001
Median (IQR) visual acuity worst eye (ETDRS letters)	40 (23;50)	52 (45;55)	0.001

4.2 Examination methods

4.2.1 Visual acuity

Visual acuity (VA) was tested with the Early Treatment of Diabetic Retinopathy Study (ETDRS) chart at 4 m after correction to best visual acuity (REF ETDRS chart). Reading 55 letters on the chart corresponds to VA 1.0 = 6/6 = 20/20 and a logarithm of minimal visual angle (logMAR) of = 0.00. The number of read letters was recorded for both eyes and are presented as best and worst eye visual acuity. A difference of more than 10 letters (2 lines in the examination chart) was considered as clinically significant.

VA for the control group in Project 3 was tested with Snellen charts. The Snellen values were converted to the corresponding number of read letters of the ETDRS chart (REF Kaiser PK. Prospective evaluation of visual acuity assessment : a comparison of Snellen versus ETDRS charts in clinical practice (114).

The World Health Organization criteria for blindness and visual impairment was used. Thus, moderate visual impairment was defined as a best-corrected visual acuity of less than LogMar 0.48 (Snellen 20/60) in the better eye, and equal to or better than LogMar 1.0 (Snellen 20/200) and severe visual impairment as best-corrected visual acuity of less than LogMar 1.0 (Snellen 20/200) and equal to or better than LogMar 1.3 (Snellen 20/400) in the better eye. Severe visual impairment according to WHO states legal blindness.

4.2.2 Fundus photos

Digital fundus photos of the seven standard fields in colour and black/white each covering 50^{o} were taken in pharmacological mydriasis by a certified, registered nurse using Topcon TRC-IX, -IA or -EX fundus cameras (Topcon, Tokyo, Japan) (115, 116). All photos were examined and graded blindly by a senior ophthalmologist (Anders Sellman), using the Topcon Image-net system (Topcon, Tokyo, Japan), which allows enlargement, contrast adjusting, colour enhancement, stereo viewing and measuring. A stereo viewer from Berezin Stereo Photography Products, Mission Viejo, CA, USA was used. If the examiner was in doubt two other experienced consultants in ophthalmology were consulted and a majority decision was taken.

4.2.3 Classification of diabetic retinopathy

Retinopathy was classified per the alternative classification of the Wisconsin Epidemiologic Study of Diabetic Retinopathy (WESDR) Table 4.5, where total retinopathy levels were derived by giving an eye with higher level a greater weight (47). A participant´s DR level was derived by concatenating the levels of the two eyes. The eye with the higher level got a greater weight according to the scheme in Table 4.7. Levels 1 to 5 are equal to none or slight DRP, 6 and 7 to moderate DRP in one or both eyes, levels 8 and 9 to pre-proliferative lesions in one or both eyes and level 10 or higher to new or old proliferative lesions in one or both eyes.

Table 4.5
Scheme for concatenating DR levels of the two eyes. The retinopathy levels were calculated by giving the eye with higher DR level a higher weight.

WESDR		WESDR		WESDR	
10/10	1	41/41	7	65/65	13
21/10	2	51/<51	8	71/<71	14
21/21	3	51/51	9	71/71	15
31/<31	4	61/<61	10	80/ <80	16
31/31	5	61/61	11	80/80	17
41/<41	6	65/<65	12		

4.2.4 Clinical significant macular edema (CSME)

The ETDRS definition of clinical significant macular oedema was used to determine the presence or non-presence of clinical significant macular oedema (117). The condition was considered present when oedema or hard exudates involved the fovea or was within 500 microns of the fovea, or when one disc area of oedema was present with at least a portion of it within one disc diameter from the macular centre.

4.2.5 Ocular Coherence Tomography (OCT)

In Project 2 Ocular Coherence Tomography (OCT) using a Zeiss-Humphrey 2000, application version A2 was used to measure macular thickness. Using the Fast Macula Protocol 6 consecutive radial scans and equally spaced orientation were centred on the foveola. The mean foveal thickness was defined as the calculated mean thickness of the central 1000 µm diameter of the macula. The minimum thickness of the six radial scans was determined manually and the lowest value was taken as the central macular thickness. The central macular thickness in healthy, non-diabetic eyes is between 140-200 µm when measured with this instrument.

4.2.6 $TcpO_2$

$TcpO_2$ values were obtained with a Radiometer TCM2 (Radiometer, Copenhagen, Denmark) in a room temperature of 21-24 ºC. All measurements were performed in supine position after 20 minutes of rest. Study participants were asked to avoid smoking or coffee for at least two hours before investigations. Measuring point was on the dorsum of the foot 2 cm proximal to the base of the third toe. The transducer was heated to 42 ºC and after equilibration values were recorded once every minute for 6 minutes breathing room air, thereafter 6 minutes breathing 100% oxygen. The highest $TcpO_2$ values breathing air or oxygen were used as basal and stimulated $TcpO_2$.

4.2.7 Arterial toe blood pressure

Toe blood-pressure was measured using a small cuff around the first toe or, if not applicable, the second toe. Return of pulsatile blood flow was detected by strain gauge technique or by pulse oximetry. The results from these two methods are comparable according to the laboratory.

4.2.8 Ankle brachial index

Systolic and diastolic arm blood pressure were measured in supine position. Systolic ankle pressure was measured with a Doppler pen over the dorsal pedal and posterior pedal and posterior tibial arteries. The pressures in both legs were measured and the lowest value was used in calculation.

4.2.9 Laboratory analyses

HbA_{1c} measurements were performed using high-performance liquid chromatography according to clinical routines in the central laboratory of the hospital and are reported according to Diabetes Control and Complications Trial and International Federation of Clinical Chemistry standards.

Estimated GFR (eGFR) was calculated from plasma creatinine using the modification of diet in renal disease equation (118).

4.3 Study procedures

4.3.1 Project 1

4.3.1.1 Study design and treatment schedules

The **H**yperbaric **O**xygen Therapy in **D**iabetic Patients with Chronic **F**oot **U**lcer study (HODFU) is a prospective, placebo-controlled, randomized and double blinded study primarily evaluating the effects of HBO on ulcer healing in people with diabetes and chronic foot ulceration. The rational and patient enrolment criteria is published in Paper 1 (110).

The study was performed in an ambulatory setting and the study treatment was given as an adjunct to regular therapy at a multidisciplinary diabetes foot clinic. The treatment sessions were given in a multiplace hyperbaric chamber using face masks, five times a week for eight weeks. Planned number of treatment sessions were 40, and while the treatment period could be extended to ten weeks but the number of treatments was not allowed to exceed 40.

Eligible patients were stratified based upon arterial toe blood pressure (≤ 35 mm Hg vs >35 mm Hg), before being randomly assigned to either of two treatment arms, one with hyperbaric oxygen (100% O_2, 2.5 ATA, 90 minutes/session, 40 treatment sessions) the other with hyperbaric air (air, 2.5 ATA, 90 minutes/session, 40 treatment sessions). Randomization was done in blocks of ten, using sealed, numbered, opaque envelopes.

A study treatment session included a compression time of 5 minutes in air, followed by a treatment period (with oxygen or air) at 2.5 ATA for 85 minutes and then a decompression period for 5 minutes to 1 ATA.

4.3.1.2 Eye examinations

All study participants underwent an ophthalmological examination before the first study treatment and then at three, six, twelve and twenty-four months post Day 1. At each time visual acuity and fundus photographs were taken and visual acuity, retinopathy level and presence of CSME were determined. Numbers of bleedings, micro aneurysms, hard exudates, and edema in the central and horizontal fundus segments of the ETDRS grid were manually counted by a senior ophthalmologist (Anders Sellman). Surfaces of bleedings and exudates were calculated using Topcon Image Net System (Topcon, Tokyo, Japan). Every fifth grading examination was reevaluated to ensure accuracy.

4.3.2 Project 2

OCT examination was performed at the baseline visit in 20 consecutive HODFU EYE study participants. Peripheral circulation was evaluated with ABI, arterial toe blood pressure and $TcpO_2$. Correlations between these baseline parameters and macular thickness were evaluated, as well as macular thickness in participants with and without peripheral ischemia, predefined as $TcpO_2 \leq 55$mmHg, TBP≤60mmHg and ABI≤0.9 or ≥ 1.4.

4.3.3 Project 3

Retinopathy level, visual acuity and presences of CSME were evaluated at inclusion and five and ten years thereafter. Dr Sellman reviewed and classified all fundus photos. Follow-up data was cached from the eye-examination closest to 5 and 10 years after the initial examination. Only examinations within a one-year timespan from these timepoints were included. Baseline characteristics and visual acuity were obtained from patient´s charts. Mortality data was obtained from the Swedish National Death Registry.

4.4 Statistics

Statistical analyses were performed using Statistica version 10.0 software (Statsoft Inc. Tulsa, OK, USA) in Project 2, in the other projects, SPSS 20- 22 (IBM, Chicago, Il, USA) was used.

Normal distribution was tested with Shapiro-Wilks test.

Categorical and ordinal variables are given as percentages and continuous variables as median (quartiles) or mean (SD).

Mann-Whitney U-test and Fisher´s exact test was used to compare outcomes between groups, using the Chi-square test for categorical variables. Wilcoxon´s test was used to compare intra-group differences over time.

Survival data are presented as Kaplan-Meyer curves and the LogRank test was used for comparisons between groups. Multinominal logistic regression analyses have been used to identify predicting factors.

A two sided p-value of <0.05 was considered as statistically significant.

5. Ethical Considerations

Ethical approval was given by the Regional Ethics Committee at the University of Lund. Sweden. The studies were carried out in accordance with the Declaration of Helsinki as revised in year 2000. The patients gave written informed consent to participate in the intervention studies.

The eye examinations, a total of up to 6 per patient in the HODFU-study, took 45-60 minutes, including waiting time for pupil dilation. The flash light of the fundus camera could be experienced as slight to moderate annoying but is harmless. The other patients followed recommended control intervals.

Compression and decompression can be experienced as disagreeable, with difficulties in pressure equalisation. Two patients had to be myringotomized with tube inlaying and then had no troubles. A few patients had mild claustrophobia. However most patient appreciated the care they got and appreciated their new acquaintances in the chamber. As the consequences of diabetic retinopathy may be so grave, we consider the inconveniences as existing but tolerable.

6. Results

6.1 Project 1

The effects of hyperbaric oxygen therapy on retinopathy and visual acuity when used as an adjunctive treatment in people with diabetes and hard to heal foot ulcers were evaluated in a two-year follow-up in this prospective randomized double-blinded placebo-controlled study. The outcomes of those receiving HBO were compared to those receiving study treatment with hyperbaric air (placebo). The predefined per-protocol population (study participants completing more than 35 treatment sessions) are included in this trial, as the outcomes mainly are of safety concerns. The remaining twelve underwent between 1 and 28 (median 15) treatment sessions. Routine clinical follow-ups of these patients did not indicate unexpected detoriation or amelioration of their retinopathy or VA. No signs of decompression sickness or oxygen toxicity were observed (119).

VA was similar between groups (HBO: median 52 letters [quartiles 45-55], Placebo: median 49 letters [quartiles 42-54]) and did not change during the two-year observation period. A trend of transient visual deterioration in the HBO group after three months was noted, this trend had disappeared at the subsequent examinations. No differences in levels of retinopathy between groups during the two-year follow up period were identified (Figure 6.1). Scatter laser therapy was given in three cases in each group. During the two years of follow-up, new clinical significant macular oedema was identified in four eyes in the HBO group and in three eyes in the placebo group. Focal laser therapy was given in four cases in the HBO group and in two cases in placebo group.

Neither could any differences in numbers or areas of bleedings, hard exudates, micro-aneurysms or oedemas be identified, nor between groups or visits (Table 6.1).

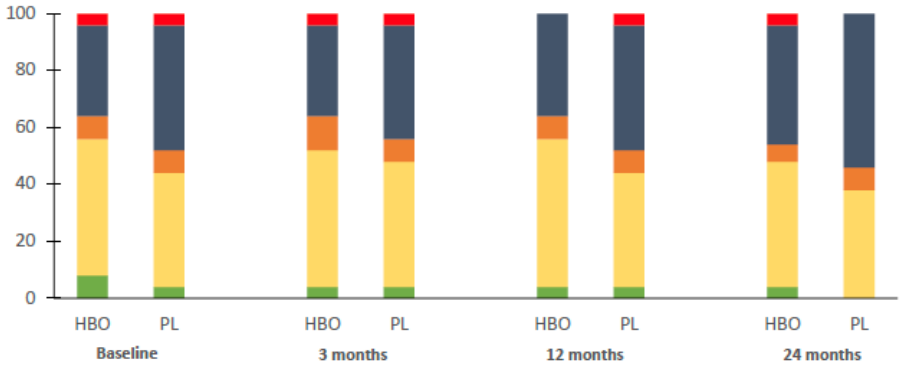

Figure 6.1
Levels of retinopathy at baseline, 3, 12 and 24 months of follow-up in patients treated with hyperbaric oxygen (HBO) and placebo (PL), respectively. no retinopathy, non-proliferative diabetic retinopathy (levels 21-41), pre-proliferative diabetic retinopathy (level 51), laser treated non-active proliferative diabetic retinopathy (level 61) and proliferative active diabetic retinopathy (level 65 or more).

Table 6.1
Number of hard exudates, bleedings and microaneurysms at baseline and during the two year follow up in people receiving 36 to 40 ninety minutes long treatment sessions of hyperbaric oxygen therapy or placebo (100 % air) at 2.5 ATA. Data are given as median, 25 - 75 quartiles and max. No statistical differences could be identified, neither between groups or over time.

	Baseline		**3 months**		**12 months**		**24 months**	
	HBO	**Placebo**	**HBO**	**Placebo**	**HBO**	**Placebo**	**HBO**	**Placebo**
n	25	25	23	23	23	22	20	21
Bleedings, numbers	0.5 (0-3) 13	0.5 (0-3.5) 14	0 (0-2) 19	0 (0-2) 13	1 (0-2) 27	0 (0-2.5) 8	0 (0-1.5) 27	0 (0-3.5) 11
Hard exudates, numbers	0 (0-0.8) 63	0 (0-3.5) 26	0 (0-1.5) 86	0 (0-6) 74	0 (0-1) 48	0 (0-5) 18	0 (0-5) 86	0 (0-4) 54
Micro-aneurysms, numbers	16 (6-28) 74	17 (8-44) 205	15 (6-28) 108	15 (7-26) 149	17 (10-31) 151	17 8-35 170	19 (6-39) 116	21 10-34 126

6.2 Project 2

The aim of the study was to evaluate if macro or micro vascular changes and consequent ischemia in the foot in people with hard to heal foot ulcers was reflected in macular oedema of the retina. Twenty patients (40 eyes) with a median duration of diabetes of 22 years were included. Median ABI was 0.77, median TBP 55 mmHg and median $TcpO_2$ 48 mm Hg. Based on these measurements 13, 13 and 14 patients were considered ischemic. Patients with ischemic $TcpO_2$ levels had significantly higher mean foveal and central foveal thicknesses. Central foveal thickness was 230 µm (159 - 354) vs 204 µm (164 - 283), p = 0.03. Mean foveal thickness 200 µm (140 - 330) vs. 165 µm (130 - 250) p = 0.03. This was not seen using ABI or TBP as circulatory measures.

$TcpO_2$ thus seems to be a clinically relevant measure of peripheral microvascular disease in people with diabetes mellitus, and low values seem to indicate an increased risk of macular oedema.

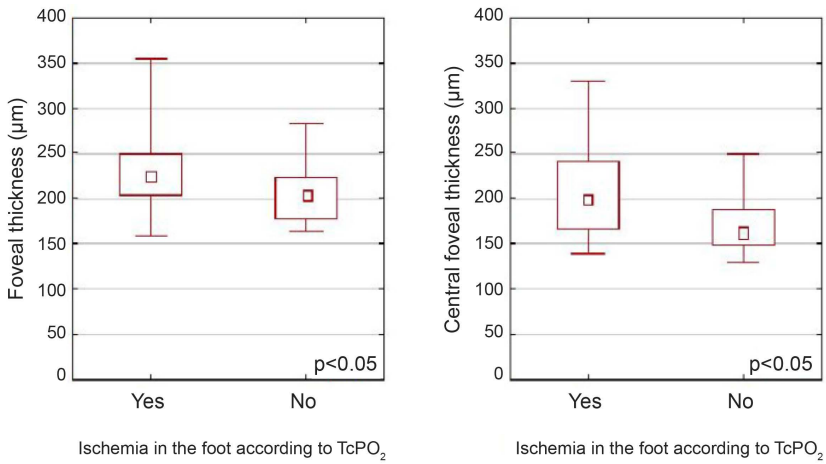

Figure 6.2.
Mean foveal and central foveal thickness in people diabetes and hard-to-heal foot ulcers and peripheral ischemia according to TcpO2 levels. Data are given as median, first and third quartiles and min-max. The predefined cutoff level for ischemia was 55 mmHg. Reprinted from J Exp Integr Med 2013; 3(2):81-85 by permission of GESDAV.

6.3 Project 3.

The aims of this real-life clinical study were to elucidate the frequency and severity of DR in a group of people with Type 2 diabetes and hard-to-heal diabetic foot ulcers, and to compare visual acuity, levels of retinopathy and presence of clinical significant macular oedema in this group with a control group matched for age, sex and Type 2 diabetes duration but without a history of foot ulceration requiring with the health care system.

Altogether, outcomes in 90 people with type 2 diabetes and presence of foot ulceration were compared to those in 180 matched controls. The mean age at inclusion was 69 years, the mean diabetes duration just above 15 years and the mean HbA_{1c} was 59 mmol/mol. Among the foot ulcer patients 79 % were prescribed insulin as compared to 56 % in the control group. Impaired renal function was more frequently present in the diabetic foot group, i.e. 60 vs. 34%. At the time of eye examination the duration of the DFU was 4.9±1.9 months and the mean DFU area was 3.2±1.7 cm^2. Approximately three quarters of the ulcers were classified as Wagner grade 3 or higher.

6.3.1 Retinopathy and visual acuity at time of diabetic foot ulceration (baseline)

Diabetic retinopathy was more severe and more often present in the DFU group (Paper 4). Only 6% of the individuals in the DFU group were without retinopathy as comparted to 34% in the control group, and severe non-proliferative or

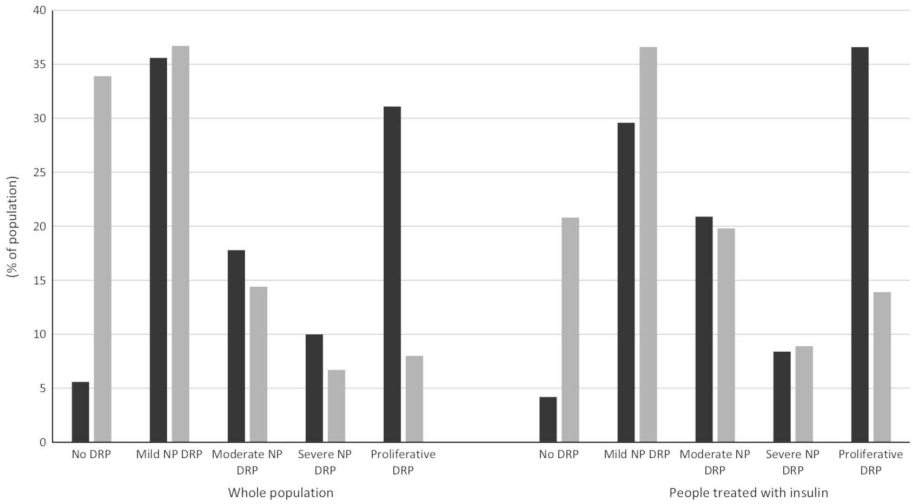

Figure 6.3.
Levels of retinopathy in people with type 2 diabetes and presence of a diabetic foot ulcer (dark bars) or a medical history without a diabetic foot ulcer (light bars). Data are given for the whole as well as the population prescrined insulin. Reprint from paper 4 with permission from Wiley.

proliferative diabetic retinopathy was present in 41% in the DFU population as compared to 15% among controls. The same pattern was seen among insulin users (Figure 6.3). Time to proliferation was not statistically different between groups. In a multinominal logistic regression analysis presence of chronic diabetic foot ulcer and diabetes duration were riskfactors for proliferative retinopathy (Table 6.2).

Table 6.2
Factors predicting proliferative retinopathy. Groups with more severe DR are compared to the group of people with no or mild non-proliferative DR (reference).

Factor	Odds ratio	95%-CI	p-value
Chronic diabetic foot ulcer	5.57	2.04-15.2	<0.001
Diabetes duration (years)	1.16	1.09-1.23	<0.001
Age (decades)	0.92	0.97-0.97	0.002
Hypertension	0.24	0.06-0.96	0.037
Insulin use	4.17	0.83-20.9	0.083
Male sex	2.19	0.65-7-41	0.208
eGFR (3 groups)	0.93	0.4-1.93	0.830
HbA_{1c} (10mmol/mol)	1.02	0.99-1.06	0.168

A history of CSME was more frequent in the DFU group, 40 vs. 16% ($p<0.0001$). Time from diagnosis of diabetes to first visible presence of CSME was not statistically different between the two groups, 13.5±8.7 years in the DFU group and 16.1±4.6 years in the control group ($p=0.17$).

Visual acuity was lower in the DFU group, median (interquartile range) best eye visual acuity was 50 (42.75;55) EDTRS letters in the DFU group and 55 (52;55) in the control group ($p<0.001$). Similarly, the median (interquartile range) worst eye visual acuity was 40 (23;50) vs. 53 (45;55) ETDRS letters ($p<0.001$). Risk factors for poorer visual acuity of the best eye were male sex ($p=0.004$), presence of chronic DFU ($p<0.001$), older age ($p=0.01$) and duration of diabetes ($p=0.024$). Presence of DFU ($p<0.001$), duration of diabetes ($p=0.016$) and older age ($p=0.025$) were risk factors for poorer visual acuity of the worst eye.

In conclusion, in this northern European setting almost all people with Type 2 diabetes and chronic diabetic foot ulcers had diabetic retinopathy. Nearly one-third had proliferative diabetic retinopathy as compared to less than 10% in our matched control group. More advanced diabetic retinopathy was linked to worse visual acuity.

6.3.2 Ten year follow-up of retinopathy and visual acuity

First, this (Paper 5) ten-year follow-up study of the population in paper 4 confirms the high mortality among people with type 2 diabetes and hard-to-heal diabetic foot ulcers. The five year mortality was 60% in DFU group and 21% in the control group, and the ten year mortality was 82% and 47%, respectively (Figure 6.4). Presence of DFU (OR 2.50 (1.73-3.61, $p<0.001$), presence of clinical significant macular oedema (OR 1.81 (1.04-3.12, $p<0.035$), lower eGFR (1.02 per unit (OR 1.01 (1.02, $p=0.006$), older age (1.04 per year (1.02-1.07, $p<0.001$) and higher retinopathy level (OR 1.15 (1.01-1.33, $p=0.045$) were associated with ten-year mortality.

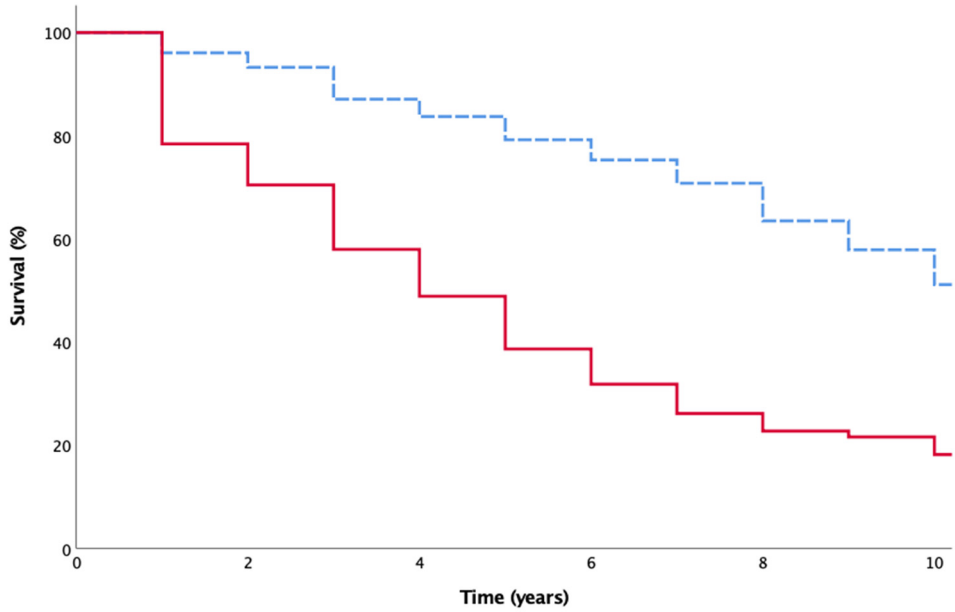

Figure 6.4
Survival in people with type 2 diabetes and presence of a diabetic foot ulcer at baseline (red solid line) and a matched population without a history of diabetic foot ulcer at baseline (blue dotted line), p<0.001.

Progression of retinopathy over the ten year follow-up period is given in Figure 6.5. and as shown, in the DFU group only 6% of the patients were alive and without more than mild diabetes retinopathy ten years after inclusion. Presences of CSME was more frequent and earlier presented in the DFU group (Figure 6.6).

Visual impairment progressed faster in the DFU group. Five years after inclusion best visual acuity had deteriorated with 5 (18;0) letters in the DFU group (p<0.001) and with 0 (5;0) (p<0.01) in the control group, and worst visual acuity with 9 (48;0) and 0 (6;0) letters, respectively. In the DFU group 9.9% of the patients had severe visual impairment at baseline, 15.6% at the five year follow-up and 15.4% at the 10-year follow-up. Each of these prevalences were numerically higher compared to the control group, in which the corresponding prevalences were 0.6% (p=0.0001), 0.8% (p=0.0003) and 4.8% (p=0.14), respectively.

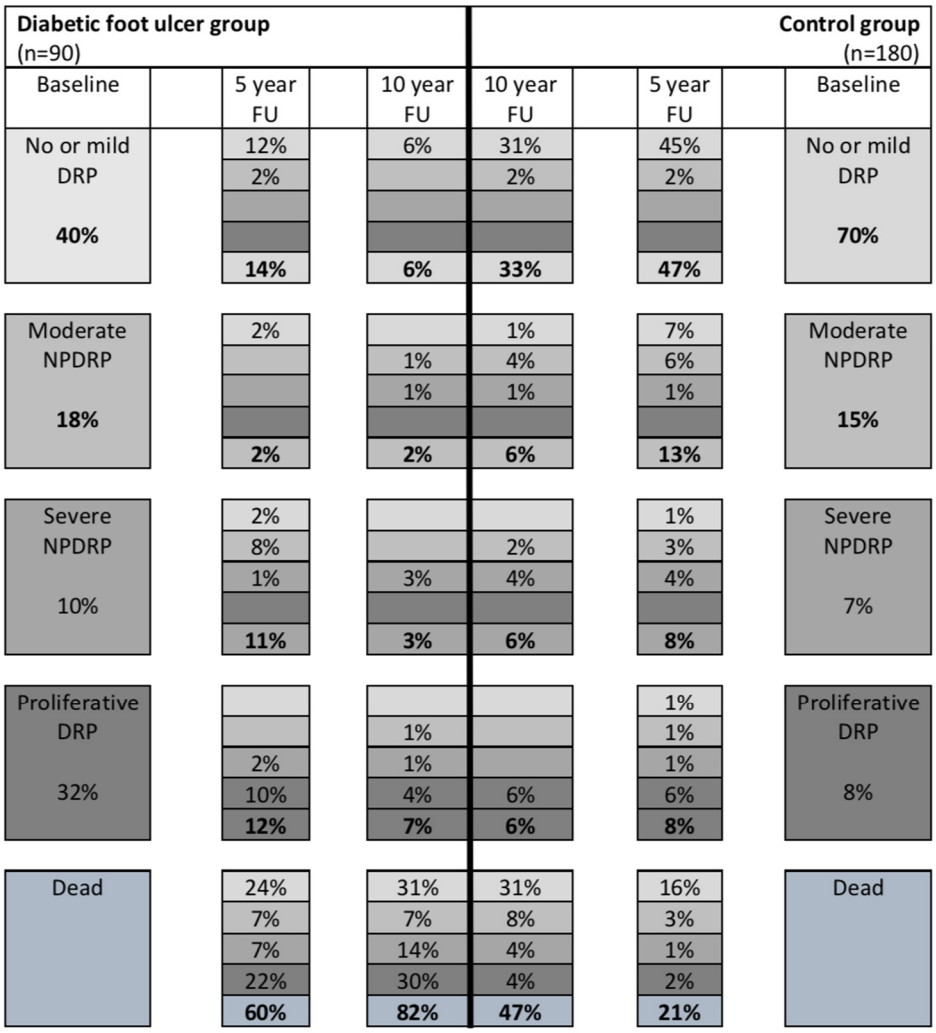

Figure 6.5.
Progression of diabetic retinopathy and mortality in people with and without hard-to-heal diabetic foot ulcers. Percentage of people in each retinopathy category at baseline, 5- and 10 years of follow-up are given, as well as mortality frequencies.

Table 6.3
Five and ten-year follow-up outcomes in people with a DFU at baseline and people without a history of DFU.

	DFU Baseline	Controls Baseline	DFU 5 year FU	Controls 5 year FU	DFU 10 year FU	Controls 10 year FU
No or mild DR	40%	70%	14%	47%	6%	33%
Moderate DR	18%	15%	2%	13%	2%	6%
Severe NPDR	10%	7%	11%	8%	3%	6%
PDR	32%	8%	12%	8%	7%	7%
CSME	40.0%	15.6%				
Dead			60%	21%	82%	47%

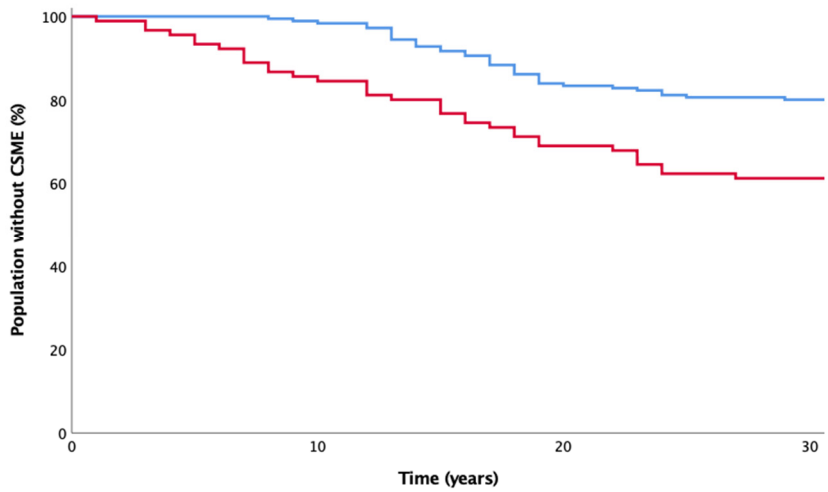

Figure 6.6.
Time to clinical significant macular oedema in people with type 2 diabetes and presences of a hard-to-heal diabetic foot ulcer at baseline (red line) and without a history of diabetic foot ulcer at baseline (blue line), p<0.001.

In conclusion, presences of a hard-to-heal diabetic foot ulcer was accompanied with higher levels of diabetic retinopathy, poorer visual acuity and higher mortality compared to a control group with type 2 diabetes but without DFU over ten years of follow-up.

6.3.3 Moderate visual impairment as a risk factor for mortality in people with hard-to-heal diabetic foot ulcer

Visual impairment has a negative impact on functional independence but also increases the risks of fall, accidents and fractures and could also be a sign of physical frailty, and is associated with increased mortality risk in the general older

population. The aim of the study in Paper 6 was to elucidate if visual acuity is a risk factor for mortality also in people with type 2 diabetes and hard-to-heal foot ulcers.

During the 5-year period, 91 people died, corresponding to a five-year cumulative mortality of 34%. Moderate visual impairment (<6/12) was detected in 32 people (12%), 10 (5.6%) in the control group and 22 (24%) in the DFU group. The group of people with visual impairment were older, had a longer diabetes duration, higher HbA_{1c} and more advanced retinopathy (Table 6.4).

Table 6.4.
Baseline characteristics in people with or without diabetic foot ulcer and with or without moderate visual impairment.

	DFU	Control	p-value	Visual impairment	No visual impairment	p-value
n	90	180		32	238	
Age (years)	69.1±10.1	69.4±9.6	0.83	73.1±10.4	68.9±9.5	0.011
Male sex (%)	84	85	0.96	85	68	0.021
Diabetes duration (years)	15.9±10.9	16.4±8.5	0.89	20.0±11.	15.4±8.9	0.017
HbA1c (mmol/mol) (%)	60±14 (6.8±1.5)	58±11 (6.5±1.2)	0.18	7.16±1.55	6.56±1.88	0.059
No glucose lowering treatment (%)	3	8	n.s	3	7	Ns
Insulin treatment (%)	79	56		84	61	0.006
Blood pressure lowering treatment (%)	73	73	0.67	71	74	Ns
eGRF			<0.0001			0.12
< 45 ml/min/1.73m2 (%)	36	13		31	19	
46-59 ml/min/1.73m2 (%)	23	23		28	21	
≥ 60 ml/min/1.73m2 (%)	41	64		44	59	
Presence of DFU (%)	100	0	na	69	29	<0.001
Presence of CSME (%)	40	16	<0.001	59	19	<0.001
History of proliferative retinopathy (%)	31	8.4	<0.001	56	10	<0.001
Best corrected visual acuity, best eye (ETDRS letters)	50 (42.75;55)	55 (52;55)	<0.001	36 (25;39)	55 (50;55)	<0.001
Best corrected visual acuity, worst eye (ETDRS letters)	40 (23;50)	53 (45:55)	<0.001	17 (6;34)	52 (48;55)	<0.001

The five-year mortality rate was significantly higher among those with visual impairment as compared to those without (69 vs. 30%, p<0.0001). Time to death in people with and without visual impairment is given in Figure 6.7. Presence of a

hard-to-heal DFU was also associated with a higher 5-year mortality (60 vs. 21%, p<0.0001). Time to death considering both visual impairment and presence of DFU is given in Figure 6.8. The difference in 5-year mortality between those with and without visual impairment was statistically significant both among those with a DFU (p=0.049) and those without (p=0.018).

A multinominal regression analysis was performed to adjust for confounding factors. In the final model presence of diabetic foot ulcer (RR 3.6 (2.27-5.75),

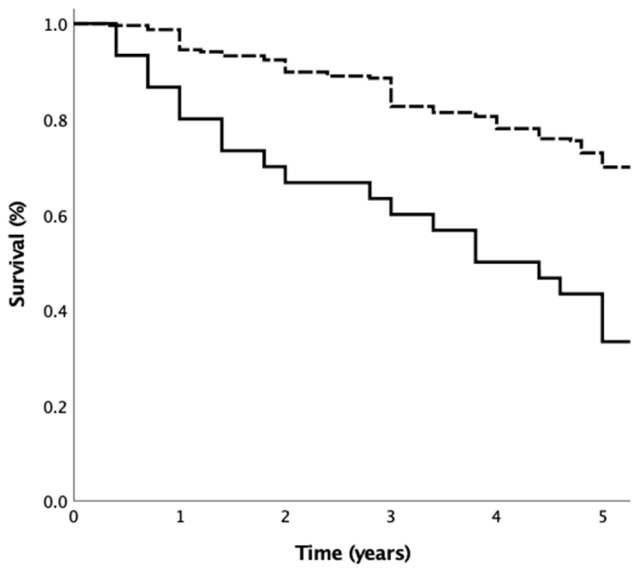

Figure 6.7
Time to mortality in people with (solid line) or without (dotted line) moderate visaual impairment, p<0.001.

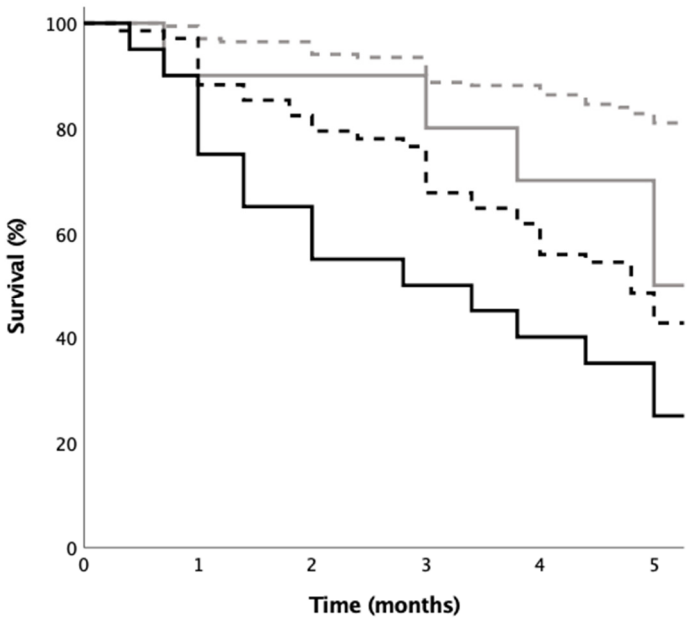

Figure 6.8
Five-year mortality in people with (black lines) or without (gray lines) presence of a hard-to-heal diabetic foot ulcer and with (solid lines) or without (dotted lines) visual impairment defined as visual acuity < 6/12 (LogMar 0.3).

p<0.001), male sex (RR 2.34 (1.16-4.73),p=0.018), moderate visual impairment (RR 1.77 (1.02-3.07), p=0.042), age (decades) (1.48 (1.21-1.79), p<0.001) and eGFR (3 groups) 1.42 (1.09-1.86), p=0.010) were significant predictors of 5-year mortality. Diabetes duration, baseline HbA1c or higher level of retinopathy were not significantly related to 5-year mortality.

In conclusion, impaired visual acuity seems to be associated with increased mortality also in a cardiovascular high risk population such as people with type 2 diabetes and hard-to-heal diabetic foot ulcers.

7. Discussion

7.1 Introduction

Ischemia, neuropathy and infection play important roles in the genesis of foot-ulceration and diabetic retinopathy.

In the retina altered metabolic pathways, inflammation, increased concentration of Reactive Oxygen Species (ROS), “sticky” leukocytes (leukoadhesion), shear stresses of the vessels, impaired blood-retinal-barrier and growth factors as VEGF, all play their roles leading to visible and non-visible changes and altered functions of this vascularized neural tissue. There is also an element of non-visible neurodegeneration, early in the process. Thus, in diabetic retinopathy there is elements of both neuropathy and ischemia along with evidence of inflammatory processes (41).

Distal neuropathy occurs in up to 60 % of patients with long standing diabetes and in more than 80% in people with foot ulcers.

(120, 121) Peripheral Arterial disease (PAD) is present in almost every second patient with DFU. In spite of treatment at specialized foot clinics the ulcerations may become chronic and defy treatment (122).

Accordingly, it appears to be important to consider the relationship between the blood circulation and oxygenation in the diabetic foot and in the diabetic eye.

7.1.1 Examination methods

Simple palpation of the pulses in the feet can give important information of the blood circulation. A somewhat more advanced method is the Ankle-Brachial Index (ABI) where the blood-pressures in the arm and the ankle are compared. An index of ≥ 0.9 to ≤ 1.4 is considered normal There are, however, indications that values between 0.9- 1.0 are borderline values and may be pathological (123). Values above 1.4 indicate sclerosis of the vessels in the lower extremity (Mönkeberg-sclerosis), which is associated with false high values.

The toe-blood pressure is measured with a cuff around the first toe, levels below 30-50 mm Hg are considered critical ischemic. Both ABI-index and TBP mirror the functions of larger vessels; also toe-blood pressure may be influenced by sclerosis.

Transcutaneous oxygen tension ($TcpO_2$) measurements of the dorsum of the foot are thought to measure both the functions of larger vessels and the capillary network, *i.e.* arterial pressure, local perfusion and local oxygen consumption. A $TcpO_2$ tension below 30 mm Hg is thought of as critical ischemic. A use of all three of the methods can better localize the cause of ischemia and indicate when additional investigation and treatment are needed, for example angiography, Doppler testing, angioplasty or vascular by-pass surgery.

With the help of photography, retinal lesions may be recorded, analyzed and graded. Fluorescein-angiography can show vascular changes, if localized or general ischemia is present, the presence of new vessels and leakage, OCT can quantify leakage and OCT-angiography the finer vascular details.

7.1.2 Association of diabetic eye disease and foot ulceration

In the last 30-40 years there has been an increased interest in the co-existance of retinopathy and other diabetic complications. The early clinical observations of connections between diabetes, DR, kidney disease, cardiac events and more have now been confirmed.

Early observations are Walsh in 1975, Steel in 1976 and Glynn in 1990 who found people with earlier undiagnosed diabetes but with retinopathy and foot ulcers (74-76). In a review (79) of predictive factors for diabetic foot ulcerations, four studies observed that low vision was associated with significantly higher risk for DFU development (124-127). An association between DR and FU was found in six studies (120, 126-131). Two studies reported statistical significance, three reported an association and one reported it as predictive of recurrence. In two studies the presence of retinopathy showed higher prevalence of DFU in one but not in the other. Those patients who had had laser treatment had a higher risk of DFU-development (6, 13)(124, 125).

In a South Indian group of 182 people with risks of DFU (impaired sensibility, foot deformity, arterial disease, ulcers and/or amputation) 67.6% had DR of which 17.9 % was proliferative. There was also more severe DR in patients with greater risk grades of DFU (132).

In a retrospective study of 100 South Korean T2DM patients with ulcers Hwang et al found DR in 90% and PDR in 55%. A non-matched control-group of 2496 people without ulcers had no DR in 95.5 % and DPR in 0.6 %. However, the control group was 10 years younger and their diabetes duration was not recorded (133).

Pearce et al examined 70 studies, from 2001 and forward, also extracted from PubMed (134). They determined that DR has an association with other vascular diabetic complications. Especially DR was a strong risk factor for nephropathy and increased risks of stroke and cardiovascular disease. Proliferative DR was a strong risk factor for peripheral arterial disease with risk of foot and leg ulceration (135) (136).

In five studies amputation was used as a dependent variable. In a study of people with T2DM, Tomita (137) found that the presence of DR significantly increased the risk of developing ulcerations but only when microalbuminuria was present (138). Other authors found that DR *per se* was an independent risk factor for DFU in people with diabetes (139-142). When ABI is used as a diagnostic tool for PAD in T2DM, borderline values between 0.90-0.99 have been found to be independent risk-factors for DR and other micro- and macrovascular complications (123).

The results are varying but indicate that impaired VA and retinopathy are associated with foot ulceration.

On the other hand the presence of hard-to heal DFU or amputation predicts development or existence of DR. In 1975 Walsh found 47 patients with DFU and DR in a group of 6451 patients with newly diagnosed diabetes. Glynn et al identified foot ulcers in 39 previously unknown patients with diabetes of which 13 of 33 had retinopathy (39.4%).

In 1992 Apelqvist et al co-workers found 28.9% severe retinopathy (pre-proliferative, proliferative or previous photocoagulation) in 208 patients with DFU and a toe-blood pressure of ≤ 45 mmHg. In another study of people with DFU they found DR in 54.4% of which 64.4% was severe (137).

Non-healing ulcers are associated with an increased risk of progression from non-proliferative to proliferative DR (Nwanyanwu) (143). In 1999, Hämäläinen et al found 25 amputations in a cohort of 733 individuals with Type 1 or Type 2 DM followed for 7 years. The amputated were compared with the non-amputated, showing DR in 68% vs. 29% and also lower VA in the amputation cohort (144).

Further, in a 19-year follow-up study of 1381 people with Type 2 DM Bruun et al identified an increased risk of DR and impaired VA in patients with a history of amputation (HR 6.42 and 4.92, respectively) (141).

7.2 Hyperbaric oxygen therapy

Even at specialist foot clinics, the treatment of hard-to-heal ulcers can be difficult. Off-loading, debridement, antibiotics are the mainstay with minor and major amputations as a last measure. As the consequences of non-healing can be severe, other methods as HBO may be indicated. The effects of HBO on DR are debatable and more research are needed. The rationale of HBO in DFU is based on increased oxygen diffusion distances under hyperbaric conditions (145).

HBO has been used for some fifty years in the treatment of ulcers. Clinical trials indicate beneficial effects on ulcer healing and amputation frequencies. However, weaknesses in methodology are obvious, as the treatment schedules has varied considerable, as well as inclusion criteria (146). Two studies have not shown improved ulcer healing but their study designs have been questioned.(147, 148)

In Paper 2 we prospectively compared two groups of patients with chronic DFU either treated with HBO or hyperbaric air (placebo) on a randomized, double-blinded basis.

The first study of HBO was reported in 1979 and twenty-eight trials have since been reported: nine retrospective, five prospective but non-randomised, ten randomized and prospective, one prospective, randomized and double-blinded. These studies showed apparent healing with HBO, however several also had methodological problems and the results must be interpreted with caution.

Foot ulcers as well as DR may be associated with ischemic issues. It has been shown that oxygen levels in the retinal venules is high, indicating that the flux from the arterioles is shunted past the capillary bed. Patients with diabetic neuropathy have demonstrated pathological capillary shunting because of malfunctioning of the microvascular regulation in the skin of the foot. The anatomical preparations of amputated toes by Sangiorgi et al (30) have shown widespread occlusions and shunts (29, 31, 149). These findings indicate that blood passes the vascular beds without delivering enough oxygen or nutrients (32). HBO could partially avoid this problem by its increased oxygen diffusion distances (150, 151).

Concerns about HBO have been raised. Firstly, the acute oxygen toxicity and secondly the more chronic effects. The misdirection of the metabolic paths increases the formation of superoxides, and oxygen treatment further increases this formation (23). Oxygen may also be cataractogenous, although most often transient (104, 107). Further, as increased oxygen tension contracts retinal vessels, a plausibly risk of worsening of retinopathy may occur. However, in the treatment of ulcers there are no reports of untoward effects on the retina, but to our knowledge, no systematic study has been done.

7.2.1 Earlier treatment of eye conditions.

HBO has been tried in treating retinal hypoxia of for example central retinal artery occlusion (CRAO) and central retinal vein occlusion (CRVO) but only in small numbers of subjects. Until 2004, a total of 405 patients with CRAO had been treated, of which 261 are claimed to have improved (152). CRVO have also been treated with mixed results.

There are reports of HBO treatment of DR, the earliest by Winstanley who treated 3 cases of PDR, he saw no new fundus changes after cessation of treatment (153). In 1965 Haddad treated 3 cases of PDR with HBO, noting no appreciable complications (154).

Reports on HBO of diabetic macular edema claim improvement. Sindlerova in 1978, reported on 25 patients with diabetic retinopathy who had been treated with at most 20 sessions of 3 hours at 3 ATA with a decompression time of 1 hour. In 88% improvement of the "condition" was claimed (155).

0gura (1988) reported on 11 cases (22 eyes) with diabetic macular edema: 35 treatment sessions of one hour, unclear if at 2 or 3 ATA. Visual acuity improved by 2 lines (= 0.2) in 15 eyes (68%). The acuity tended to decrease after the treatment period. Improvement were also found in static perimetry in 76% (156).

Dumitru in 1993 proposed that HBO could replace laser therapy (157).

Caamaño and de Lara reported on 525 HBO treated persons with DR. HBO was 1 hour at 100% O_2 daily at 2.5 ATA for 7-15 days. Juvenile insulin-dependent patients showed "poor results", in non-insulin users the results were "on the whole encouraging". However, the patients in the cohort were disparate and many seem to have had their diabetes treatment adjusted, as well as their general health problems treated (158).

7.2.2 Eye complications of HBO

There are a few reports on eye-complications in HBO. In 1994 McCartney reported on the presence of vitreous haemorrhages after HBO, Caamano and de Lara noted two cases of vitreous haemorrhage. In 2017 Tran and Smart reported on activated and accelerating PDR after HBOT(159). A non-diabetic woman with ruptured retinal aneurysm was HBO treated after vitrectomy and developed an extensive macular oedema (160). Palmquist reported on cataract formation and Levanger on transiently increased refractive index of the crystalline lens, *Vide supra.* In 24 studies of HBO and chronic foot-ulcers there are no reports of adverse effects in the eye of HBO (161).

7.2.3 The HODFU study

Earlier studies have not followed the diabetic eye-lesions in a standardized way in patients with hard-to-heal ulcers where HBO was used, but the few number of reported untoward effects indicate that the treatment is probably not deleterious but possibly of benefit.

Of the now 64 eligible participants two were excluded, one because of blindness, the other because of chronic uveitis. A total of 50 people fulfilled the pre-determined requirement of at least 36 completed study treatments; 25 were randomized to HBOT, 25 to placebo. Our results during the two years follow-up did not show any differences in VA, retinopathy levels, macular oedema or need for laser therapy between the groups. The number of aneurysms, haemorrhages and exudates did not change significantly. The incidence of PDR and CSME did no differ. There was a transient lowering of the VA in the HBO group after 2 months. We have no explanation to this phenomenon.

It is noteworthy that other researchers have found improvement of macular oedema but we did not. There may be differences in examinations, registrations and definitions and the possibility that 36-40 treatment sessions at 2.5 ATA are not optimal for treating retinal oedema. On the other hand we did a prospective, randomized, double-blinded study with defined inclusion criteria, scheduled treatment and examinations. In our study HBO thus seems to be ophthalmologically neutral, while significantly more ulcers healed in the HBO group (162).

7.3 Association between $TcpO_2$ and macula thickness

In paper 3 we assessed the connection between oxygen tension of the foot as measured with $TcpO_2$ and retinal thickness. Increased retinal thickness is considered as a sign of damaged tight junctions of the retina.

ABI, TBP and TcpO2 were used to assess macro- and micro-vascular function in the feet of people with chronic DFU. ABI and TBP are thought to reflect functions of larger vessels while $TcpO_2$ mirrors the functions of both macro- and micro vessels. Low $TcpO_2$ values then indicate a micro-vasculopathy or a combination of micro- and macro-vasculopathy. $TcpO_2$ is influenced by arterial oxygen supply, local oxygen consumption, capillary density and capillary function or a defective microvascular response. The $TcpO_2$ measurements, also reflect the maximal microvascular response to heightened skin temperature. In both Type 1 and Type 2 DM this important hyperaemic response is impaired. This has been suggested as an explanation for the tendency to develop foot ulceration without large vessel disease

(163, 164). There are some issues in $TcpO_2$ measuring: placement of the probe, skin thickness, oedema of the foot, and use of ointments.

Retinal thickness can be measured using OCT technique. Normal thickness is 140-200µm when using the OCT 2000 instrument. In the presence of retinal oedema, the thickness is increased.

In our study 20 consecutive patients with chronic DFU were included with a median age of 73 years and a median diabetes duration of 22 years. Patients with low $TcpO_2$ had significantly thicker central (230µm range 159- 354µm) and fovea thicknesses (200µm range 140- 300µm) vs. in non-ischemic measurement: central thickness (204µm range 164- 283µm) and foveal thicknesses (200µm range 140- 300µm). These differences were not seen when using TBP or ABI as circulatory measures.

Retinal thickening depends on fluid accumulation from leaking capillaries due to malfunctioning of the blood-retina barrier, a microangiopathy occurring as a result of diabetes. Retinal thickening and low $TcpO_2$ in the absence of low ABI and TBP+ indicate diabetic micro vasculopathy with macular oedema.

The existence of microvascular pathology in both the eye and in the foot indicates a common pathophysiological pathway of these two complications. Huang et al have found a significant negative association between $TcpO_2$ and DR (165). $TcpO_2$ has been shown to predict healing of ulcers but also to independently predict mortality in Type 2 DM (166).

7.4 Diabetic retinopathy and visual impairment in people with hard-to-heal diabetic foot ulcers

The importance of DR as a risk factor for development of foot ulcer and amputation have been reported in several studies (134). There is however a need of a more detailed description of vision and retinal changes in these patients.

7.4.1 Comparison between people with and without chronic foot ulcers

In **paper 4** we compared two groups of people with T2DM. One group consisted of 90 subjects with hard-to-heal DFU, the other a group of 180 patients without DFU. The groups were matched for sex, age, and diabetes duration. The prevalences of diabetic retinal changes and VA at base-line were compared.

Despite a good matching of the two groups, VA, presence of macular oedema and levels of retinopathy differed between the ulcer and control groups. Visual acuity was worse in the DFU group and CSME was present in 40% in the FU group vs

15.6% in the control group (p=<0.000). In the ulcer group 41 % had none or mild DR vs 72.2% in the control group. In the FU group 31.1 % had proliferative DR vs 7.8% in the control group (p=<0.000). When the following sub-groups were analysed a/ insulin treated b/ people with creatinine > 110 c/ people with eGFR < 45 d/ non-insulin users this pattern was repeated. The median of the weighted levels of DR were 6.0 (4.0- 10.0) respectively 3.0 (1.0- 6.0) (p=<0.000) (6.0= moderate DR in one eye, 3.0 = mild DR in two eyes).

Table 7.1
Levels of DR in the FU and control groups WESDR grading.

Levels of DR	FU- group n= 90	Control-group n= 180
10	5.6%	33.3%
21	14.5%	18.4%
31	21.1%	20.6%
41	17.8%	14.4%
51	10.0%	6.7%
61	22.2%	5.0%
65	7.8%	1.7%
≥ 70	2.2%	1.7%
61, 65, ≥ 70	31.1%	7.8%
CSME	40.0%	15.6%

In Hwang´s report on DR in a South Korean cohort of 100 Type 2 patients with FU. 90 (90%) patients had any degree of DR and 55 (55%) had PDR. This compares well with the any degree of DR prevalence in our group of 90 patients: 94.4 %.There is however a significant difference in the number of PDR, 55% vs. ours 31.1%. Hwang´s control group had any DR in 4.5% of which 0.6% was PDR vs. 66.7% and 7.8% in our cohort (133). There are some methodological differences between Hwang`s and our studies. The photographic technique differed and Hwang´s control group was unmatched with 10 year lower age, unknown duration, more hypertension in ulcer group and lower HbA_{1C}, these differences may explain some differences in the results.

In summary, the people in our study with Type 2 DM and chronic foot ulcers had, compared to the control group without foot ulcer, more and more severe DR and lower VA. Also in the subgroup of insulin treated and the subgroup of renal impairment. In fact the prevalence of pre-proliferative and proliferative DR was more than 2.9 timesas high in people with chronic DFU than in people without, while time to PDR and CSME did not differ between groups.

7.4.3 Five and ten year follow up of people with and without chronic DFU

In paper five we followed the ten-year progression of VA and DR. In 1935 Waithe and Beetham found a relation between duration of diabetes and DR.

In a prospective study Pirart studied 4400 diabetic patients. After 25 years about 80% of the patients with poorer glycaemic control had developed DR compared to 40 % with better control (167).

The WESDR study studied patients with younger onset of diabetes (< 30 years) and older onset (≥ 30 years) with and without insulin treatment (53, 168, 169).

In a group of 1780 people with older onset 1370 participated at baseline and were examined after 4 and 10 years. Of the original group 824 used insulin and 956 did not. Grading of retinopathy was performed using a modification of the Airlie House classification scheme (the same as used in our studies). At the baseline examination 674 insulin users and 696 non-insulin users participated. After 4 years 485 insulin users and 502 non-insulin users participated. Of the original groups 167 (24.8%) respectively 173 (24.9%) then had died (47, 53).

The insulin group in the WESDR had more retinopathy, worse VA, more photocoagulation at the 4 year control. However, non-insulin patients were 3 years older than insulin patients and 9 years older at diagnosis and had somewhat lower HbA_{1c} and shorter diabetes duration.

At the 10 year follow-up, 251 of the insulin users and 282 of the non-insulin users still participated, with 219 respectively 194 dead, for a 10 year mortality of 45%. In the insulin group, 35.1% had no DR vs 67% in the non-insulin group. PDR was 9.6% vs 1.8%. There was more improvement and less progression of DR in the non-insulin group than in the insulin group (72).

Table 7.2
Comparison between baseline characteristics.

	Sellman et al 2018 DFU n=90	Sellman et al 2018 DFU Insulin n=60	WESDR 1984 insulin n= 674	WESDR 1984 Non-insulin n= 696	Henricsson 1996 Non-Insulin n= 557	Henricsson 1996 Insulin n=1240
Age_{diagn}	51.7	49.8	50.1	59.3	48.1	55.4
Diabetesduration	16.1	18.7	15.0	8.9	13.1.	6.1
Male	84%	82%			54.5%	58.5%
Age_{exam}	69.0	68.4	65.2	68.0	60.5	61.0
Hypertension	73%	75.2%	-	-	-	-
HbA_{1c}	6.8	6.9	11.9	10.3	7.90	7.04
VA letters Best eye	46.8 = 0.7	46.7 = 0.7	>20/40 in88%	>20/40 =in91.4%	≥ 0.5 94% = ≥ 40 ltrs	≥ 0.5 98% = ≥ 40 ltrs
VA letters Worst eye	37.2 = 0.44	35.8 = 0.42	-	-	-	-
DR any	94.4%	100%	70%	39%	57%	25.9%
PDR	32.2%	12.9%	14.1%	2.9	10%	1.6%
CSME	40.0%	25.7%	-	-	19%	5.0%

Table 7.3
Baseline characteristics in the ETDRS* study and the study by Sellman et al.

	Sellman et al Foot ulcers all	Sellman et al Foot ulcers insulin users	ETDRS 2005 N= 1681 Survivors	ETDRS 2005 N= 586 Non-surviors
Age at diagnose (years)	51.7	49.8	54.4	58.8
Duration (years)	16.9	18.7	13.5	14.5
Male sex (%)	84%	82%	52.6%	53.4
Age at examination (years)	69	68.4		
VA best eye (ETDRS letters)	46.4	46.7	≥20/220 48.8% 20/20-20/40 39% <20/40 11.4%	34.6% 44.2% 21.2%
VA worst eye	37.2	45.8	-	-
Hypertension	73%	75%	24.2	302
Creatinin	137	96.9	-	-
HbA1c (%)	6.8	6.9	9.4	9.6

*Early Treatment of Diabetic Retinopathy Study (ETDRS) Report No 27(170).

The cohorts of WESDR, Henricsson and ETDRS have no reported foot-ulcers, but it is probable that about 3% of the participants had ulcers (78, 85). In our group with 100% chronic DFU it is evident that these patients have more and more severe DR than the other cohorts.

In 1996 Henricsson et al in a cohort of 1729 patients considered to have Type 2 DM of which 28% were insulin treated, found a prevalence of any DR in 31%, of which 5% were severe or proliferative and 2.7 % were visually impaired. Diabetes duration was 8.3 ±7.3 years and age 60.6 ±10.7 years.

In another cohort of 2737 patients with a mean age of 61.5 ±11.4 years and a duration of diabetes of 7.1±7.4 years, and HbA_{1c} of 7.3±1.5, 25% were insulin treated, 72 % had no DR, 4% had severe or proliferative DR, 3.2% had impaired vision. After 3.4 years 9.1% had died (250 people) against expected 5.6 % (158 people). After 5 years the all-cause mortality was 21% in people with severe or proliferative DR at baseline (171).

In 2012 Jones et al published a study of incidence and progression of retinopathy during a 17 year follow up-period. The cohort consisted of 20,686 patients without PDR. At baseline 79% had no DR, 18% had non-proliferative DR and 2.9% had pre-proliferative DR. After 5 years 36% of the patients without DR at baseline had non-proliferative DR, 4% pre-proliferative DR, sight threatening maculopathy in 0.6% and PDR in 0.7%.

After 10 years these prevalences were 66%, 16.4%, 1.2% and 1.5%, respectively. 23% of patients with non-proliferative DR at baseline developed pre-proliferative DR, 5.0 % maculopathy and PDR in 6.1%. After ten years the prevalences were 53%, 9.6% and 11% (172).

In our study of people with T2DM and DFU we found higher prevalence of retinopathy, more PDR, more macular edema and lower VA in the ulcer group than in the matched control group. Both in the control group but most in the DFU-group the number of no or mild DR had diminished. From baseline 70% in the control group to 47%, to 33% after 5 and ten years respectively. In the DFU group the corresponding decrease was from 40% to 14% and 6% (113).

Retinopathy and foot-ulcers are signs of severely damaged vascular beds and also of neurological damage. Retinopathy in many cases leads to increased morbidity, impaired vision and legal blindness and is a leading cause of these issues but is also an indicator of increased mortality. Foot ulcers alone lower quality of life and lead to high morbidity and mortality (166).

7.4.4. Mortality

Survival of people with diabetes is lower than in healthy people.(173, 174) WESDR reported in 1989 on survival rates of diabetic patients with older onset of diabetes (≥ 30years of age) with a survival rate of 77.1 % after 4 years if no DR, but 52.9% if PDR (47). Early non-proliferative DR and moderate to severe non-proliferative DR had a survival of 67.8% respectively 66.2%. Patients with impaired vision also had lower survival, VA 20/40-20/63 correlated with 47.4% survival and 20/80-20/160 with 27.5% survival. VA of ≤ 20/200 had a survival of 30.6%. (175).

Henricsson reported a mortality rate of 9.1% vs. the expected of 5.8% after 3.4 years in a cohort of 2737 with diabetes diagnosis above 30 years. (171). A Cox regression analysis showed a mortality risk of 1.8 at moderate NPDR and 2.3 at severe or PDR. After 5 years, survival was 79% in patients with severe or PDR and 97% in the group without DR at baseline.

ETDRS report no 27 (CUSICK) estimates all-cause mortality after 5 years to 18.9% in Type 2 DM. Probability of death with none, mild, moderate DR is about 0.15, in severe NPDR about 0.22 and in PDR 0.3. Also impaired vision is associated with worse outcome with a probability of death 0.3 if VA is worse than 20/40, if VA is 20/20 or better the probability is 0.15 (170).

In a Spanish study of 345 patients with DFU and diabetes (93% Type 2) 60.2 % had DR of which 29.1% had PDR or macular edema. After 5 years, the survival was 60% and after 7 years 45%. Multivariate analysis showed that age, earlier amputation, smoking, cerebrovascular disease, renal dysfunction and ischemic heart disease were significantly related to lower survival (176).

In a prospective survey by Mundet et al 2008 of a cohort of 317 Type 2 DM, 22.9% had DR at baseline of which 8.7% had PDR, after five years 26.1 % had DR, 10.8% PDR and after ten years 29.1% DR , 12.2% PDR. Foot ulcer or amputation in 2.8%, 2.8 % and 2.2%. Five year mortality was 12.3 % but ten year mortality 24.3%. The mean duration of disease was 7.15 years (SD±7.52) (177).

People with diabetes has about twice the mortality of people without diabetes. It is however very striking how the presence of foot ulceration further detoriates survival of diabetes sick people. Presence or non-presence of DR also influences mortality especially PDR. In our patient material of Type2DM with FU 32% had PDR at baseline. After 5 years 12% had PDR, after 10 years 7%. In the group without ulcer 8% had PDR at baseline, 8% after 5 years and 6 % after 10 years, the number seems to be stationary (Sellman et al Paper 5)

No or mild DR was present in 40% in the ulcer group at baseline but in only 14% respectively 6% after 5 and 10 years. In the non-ulcer group presence of no or mild DR was 70% at baseline, decreasing to 47% respectively 33% after 5 and 10 years.

Thus the number of none or mild DR diminished in both groups but relatively more in the ulcer group (113).

One plausible explanation behind the "improvement" of PDR in our study depends on excessive mortality: the sickest died. This seems to be true also in the non-ulcer group, otherwise the number of PDR should have increased. The same phenomenon of mortality in PDR is seen in the WESDR, ESDRT and more.

Insulin treatment also influences mortality, probably because the diabetic disease is more advanced and the hypoglycemic complications.

We may assume that booth the foot ulcer group and the non-ulcer group were well managed as HbA_{1c} did not statistically differed, hypertension was the same but creatinine and eGFR were worse in the ulcer group.

Table 7.4
Five and ten year mortality in people with type 2 diabetes.

	Five year mortality	Ten year mortality
Sellman et al Control	21%	47%
Sellman et al Chronic Foot ulcer	60%	82%
WESDR,4 & 10 year(47, 53)	22.9%	45%
ETDRS (170)	18.9%	
Henricsson	21%*	
Rubio(Foot ulcer)(176)	40%	55%
Mundet et al(177)	12.3%	24.3%

*Severe or PDR at baseline

In the non-ulcer studies mortality is about the same except in the Mundet study with its much lower mortality. The coexistence of chronic foot ulceration totally changes the image with its high mortality. The very high mortality in our study may depend on a rather small number of patients with a skewed selection of very sick patients or that chronic foot ulcers are signs of a very seriously deranged vascular system.

7.4.5 Visual impairment and mortality.

7.4.5.1 Background

Visual impairment is a risk factor for untimely death. This is valid for cataract, glaucoma, age related retinal diseases and diabetic retinopathy (178). The risks of falling, accidents and fractures increase (179, 180). A visual improvement after successful cataract surgery decreased t the 5 year mortality risk, HR 0.73, compared to those with persistent visual impairment (181). Klein 1999 reported on visual impairment as a risk of ischemic heart disease death (175).

Studies to evaluate the association between people with visual impairment, chronic DFU and mortality has, to our knowledge, not been done before. Visual impairment

with a VA of less than 6/12= 20/40 = <logMAR 0.30 is considered to impede modern life activities and to increase mortality. Five year mortality was increased in the Beaver Dam study, after control for age and gender and Kulmala found an association in people aged 75 years (182). Other studies have not found this association but there are differences in age distributions and comorbidities including other eye diseases (183). However visual impairment can also be a sign of generalized frailty and aging which is a predictor of mortality (184, 185).

In the Singapore Malay Eye study Siantar et al investigated visual impairment in 3280 people between 40- 80 years. VA was < logmar 0.30(<20/40) in the best eye. Follow up was 7.24 years (median). Of the cohort 398 (12.2%) died. People with visual impairment had an all-cause mortality hazard of 1.57. Diabetic retinopathy had an all-cause mortality HR of 1.70 (186).

In the Salisbury Eye evaluation project mortality and visual impairment were studied in 2520 people. Worse baseline acuity was associated with higher mortality rate, Hazard rate 1.05, but those who gained ≥ 2 ETDRS lines had a lower adjusted risk of dying (HR =0.47) over two years (178).

7.4.5.2 Mortality in individual with visual impairment with or without chronic foot ulcers

Paper six. In the patients in paper four (90 patients with chronic foot ulcer and 180 matched controls) visual impairment, defined as VA < 20/40; <20/40, logMAR 0.3 was detected in 22 (24%) of the ulcer group and in 10 (5.6%) of the control group. The five-year mortality was significantly higher in the visual impairment group (69 % vs 30%, p <0.0001). Mortality in the DFU group was significantly higher than in the control group (70% vs 30%). The combination DFU and impaired vision had a five year mortality of 77%.

The number of patients in our study is low but the results are in accordance with other researchers. Visual impairment brings still more risk of mortality. The mortality is extremely high in people with both chronic DFU and visual impairment.

8. Conclusions

Diabetes mellitus is a common, multifaceted disease with typically late occurring but very serious complications. The pathophysiology is very complex and not currently fully understood. Neurological and macro- and microvascular lesions develop leading to retinopathy and foot and leg ulcerations.

A number of studies have tried HBO as an adjunct in the treatment of chronic foot ulcers, mostly with positive results. However many of the studies have methodological weaknesses. Therefore we designed a randomised, prospective, double blinded, placebo controlled study with defined inclusion criteria and defined control schedules for two years, the HODFU study.

We have tried to assess the common coexistence of DR and microvascular lesions in the feet comparing skin ischemia in the feet with retinal thickness as a sign of vascular retinal pathology. We found a significant relationship which may speak of a common pathology.

In the HODFU study 62 patients underwent hyperbaric therapy with Hyperbaric Oxygen (2.5 ATA) in 40 sessions. Before starting the study all went through examinations with VA assessment and retina photography. These examinations were repeated after 3, 6, 12 and 24 months, 50 patients fulfilled the study schedule and their data could be statistically calculated. The study did not find any differences of the retinal changes or changes of visual acuity between the two groups.

Two groups, one of 90 people with chronic foot ulcers the other of 180 people without present or earlier ulcers were ophthalmologically compared. The people of the ulcer group had worse VA and more and more severe retinopathy than the control group.

The two groups above were followed for five and ten years. Retinopathy deteriorated in booth groups but more in the ulcer group. The mortality was much higher in the ulcer group.

In the last study we studied mortality in people with impaired vision, considered a risk factor for death. People with ulcers and impaired vision had higher mortality than people without ulcers over a five year perspective.

As a general conclusion we can say:

- that we have found an association between hypoxia in the foot skin and retinal vasculopathy,
- that HBO seems to be ophthalmological neutral,
- that the presence of chronic diabetic foot ulceration indicates more advanced and serious retinal changes,
- that follow up for five and ten years show a striking high mortality in the foot ulcer patients and
- that visual impairment combined with foot ulceration indicate higher mortality risk.

9. Future Perspectives

There are two causes of the world spanning diabetes epidemic: the world population and the incidence of diabetes are increasing. It is hoped that the increase of population may flatten and stabilise. The diabetes incidence however is heavily dependent on ways of living and of food resources, sedentary living and too much food lead to overweight, the leading cause of Type 2 diabetes. A ticking bomb is the developing countries where people go from a life of hard labour and limited access to food to a more “modern” life with more high caloric food and less hard work. In the developed countries the same development can be seen if in a lesser scale.

Morbidity and mortality is high in diabetes. The presence of diabetic retinopathy or a foot ulcer is an alerting sign of the probable presence of other microvascular lesions that demand appropriate and timely action. Screening programmes with retinoscopy or still better photography are important to find retinopathy, but it is just as important to regularly search for ulcers, cardiac signs and kidney disease. The existence of one type of complication also shows there is a risk of other.

No health service can thus be at rest with its accomplishments, it is necessary to develop new affordable therapies, screening systems and bettered health systems. Otherwise diabetes and its complications may overwhelm the health services.

For the nearest future we plan:

To analyze macular edema of our patients in a more extensive way. We will then use existent photos and relevant patients´charts.

To study the changes of the ERG, full-field and multifocal, comparing them with changes of the ECG.

10. Summary in Swedish

Diabetes mellitus, ”sockersjuka”, är en vanlig sjukdom. Den förekommer i huvudsak i två former, typ 1 och typ 2 diabetes. I Sverige finns för närvarande 45 000 personer med typ 1 och 380 000 med typ 2 diabetes. Sjukdomen ökar raskt, antalet individer med diabetes i världen beräknas öka från dagens 415 miljoner till 640 miljoner år 2040.

Behandling vid typ 1 diabetes är alltid insulin, vid typ 2 diabetes kan insulin ingå i den blodsockersänkande behandlingen. Sjukdomen kan så småningom orsaka en mängd komplikationer i blodkärl och nervsystem. Komplikationerna är lika vid båda typerna. Två fruktade komplikationer är fotsår och förändringar i ögonens näthinnor.

Vi utarbetade en forskningsplan för att utröna om syrgas under övertryck förbättrade sårläkningen och om denna behandling påverkade ögonen. Tidigare undersökningar har gett diskutabla resultat.

94 patienter med svårläkta fotsår rekryterades, varav 62 ingick i ögonprojektet. Patienterna delades slumpvis upp till syrgasbehandling eller luftbehandling, som skedde i en tryckkammare vid 2.5 atmosfär absolut tryck. Undersökningar av sår och ögon skedde enligt ett fast schema under två år. Resultatet blev att såren visade bättre läkning vid syrgasbehandling men att ögonförändringarna inte tycktes påverkas.

Vår undersökning av syrgaskoncentrationen i huden på foten hos patienter med kroniska sår visade att lägre syrgaskoncentration hade samband med påverkad näthinna i form av ökad tjocklek. Vi uppfattar detta som diabetesorsakat.

Vi jämförde en grupp av 90 patienter med diabetiska fotsår med en kontrollgrupp på 180 utan sår. Det visade sig att fotsårsgruppen hade avsevärt mer ögonförändringar och också sämre synskärpa än kontrollgruppen. Slutsatsen blev att närvaro av fotsår är ett allvarligt tecken på diabetesskador.

Ovanstående två grupper följdes sedan under 10 år. Vi fann då att ögonförändringarna ökade och blev allvarligare. Samtidigt fann vi att dödligheten var påtagligt hög framför allt i sårgruppen men också att personer med allvarliga ögonförändringar också hade högre dödlighet än de med lindrigare förändringar.

En undergrupp med nedsatt syn (< 0.5) hade hög dödlighet, högst i kombination med fotsår.

11. Acknowledgements

I would like to express my sincere gratitude to the many who have supported me during the years as a PhD student.

My supervisors Ass. Professor Magnus Löndahl, Ass. Professor Per Katzman, Professor Sten Andreasson and. Professor Mona Landin-Olsson. Without their interest, urging, pushing and encouragement this work would have come to nothing.

To Anders Nilsson MD and Christer Hammarlund MD, PhD for introducing and including me into the hyperbaric treatment world.

To the former heads of the Dept of Ophtalmology at Helsingborgs Hospital Kerstin Sjöström MD, the late Eskil Olivius MD, Lena Brenner MD, Division Head Peter Olsson MD PhD for giving me the opportunity to work with the project.

To Marianne Henricsson MD, PhD who showed the way forward.

To Karin Filipsson MD, PhD and Ass. Prof. Monica Lövestam-Adrian for their interest in my work and constructive discussions.

To Katarina Fagher MD, PhD for discussions and much needed help.

To my collegues at the Eye Department Lena Rung MD, Maria Tyrberg MD, PhD, Göran Svensson MD, Annika Rosell MD, Anna Ljungberg MD for cooperation, many laughters and discussions.

To the RNs who did it, Beata Dus, Cathrin Karlsson, Eva Steffert and Agneta Byström.

To Dick Nelson who patiently guided me into the mysteries of statistics.

To the staff of the library at Helsingborgs Hospital and especially to Gitte Jönsson.

To Anna Ljungberg MD, Margareta Hansson MA, Beatha Sellman MA, and Emiko Tamura for linguistic help and to Annika Martinsson and Lilian Hagert for secretarial help.

To all my friends near-by or far away for their interest and for just being there.

A sincere thank you to all the patients who in spite of severe illness, tiredness and sometimes long waiting times assumed a favorable attitude towards these studies.

A very special thank you to my dear family: Christina, Eva, Håkan, Mattias, Liv, and Beatha without your support and common sense this work would not have succeeded.

The study was supported by the Zoega, Gorthon and Segerfalk Foundations and by the Sydvästra Skånes Diabetesförening.

References

1. Duke-Elder S. System of Ophtalmology Volume X. London: Henry Kimpton; 1967. p. 408.
2. Duke-Elder S. System of Ophthalmology Volume X. London: Henry Kimpton; 1967. p. 410.
3. DiabetesAtlas [Internet]. 2017.
4. The Swedish National Diabetesregister [Internet]. 2018.
5. Federation ID. IDF DiabetesAtlas 8Th Edition. 2017.
6. Triolo TM, Armstrong TK, McFann K, Yu L, Rewers MJ, Klingensmith GJ, et al. Additional autoimmune disease found in 33% of patients at type 1 diabetes onset. Diabetes Care. 2011;34(5):1211-3.
7. Stitt AW, Anderson HR, Gardiner TA, Archer DB. Diabetic retinopathy: quantitative variation in capillary basement membrane thickening in arterial or venous environments. The British journal of ophthalmology. 1994;78(2):133-7.
8. Cogan DG, Kuwabara T. The mural cell in perspective. Archives of ophthalmology (Chicago, Ill : 1960). 1967;78(2):133-9.
9. Bolton SJ, Anthony DC, Perry VH. Loss of the tight junction proteins occludin and zonula occludens-1 from cerebral vascular endothelium during neutrophil-induced blood-brain barrier breakdown in vivo. Neuroscience. 1998;86(4):1245-57.
10. Patel V, Rassam S, Newsom R, Wiek J, Kohner E. Retinal blood flow in diabetic retinopathy. BMJ (Clinical research ed). 1992;305(6855):678-83.
11. Comer GM, Ciulla TA. Pharmacotherapy for diabetic retinopathy. Current opinion in ophthalmology. 2004;15(6):508-18.
12. Joussen AM, Poulaki V, Qin W, Kirchhof B, Mitsiades N, Wiegand SJ, et al. Retinal vascular endothelial growth factor induces intercellular adhesion molecule-1 and endothelial nitric oxide synthase expression and initiates early diabetic retinal leukocyte adhesion in vivo. The American journal of pathology. 2002;160(2):501-9.
13. Kizub IV, Klymenko KI, Soloviev AI. Protein kinase C in enhanced vascular tone in diabetes mellitus. International journal of cardiology. 2014;174(2):230-42.
14. Nerlich AG, Sauer U, Kolm-Litty V, Wagner E, Koch M, Schleicher ED. Expression of glutamine:fructose-6-phosphate amidotransferase in human tissues: evidence for high variability and distinct regulation in diabetes. Diabetes. 1998;47(2):170-8.
15. Stitt AW, Hughes SJ, Canning P, Lynch O, Cox O, Frizzell N, et al. Substrates modified by advanced glycation end-products cause dysfunction and death in retinal pericytes by reducing survival signals mediated by platelet-derived growth factor. Diabetologia. 2004;47(10):1735-46.

16. Du XL, Edelstein D, Dimmeler S, Ju Q, Sui C, Brownlee M. Hyperglycemia inhibits endothelial nitric oxide synthase activity by posttranslational modification at the Akt site. The Journal of clinical investigation. 2001;108(9):1341-8.
17. Ishii H, Jirousek MR, Koya D, Takagi C, Xia P, Clermont A, et al. Amelioration of vascular dysfunctions in diabetic rats by an oral PKC beta inhibitor. Science (New York, NY). 1996;272(5262):728-31.
18. Obrosova IG, Julius UA. Role for poly(ADP-ribose) polymerase activation in diabetic nephropathy, neuropathy and retinopathy. Current vascular pharmacology. 2005;3(3):267-83.
19. Peti-Peterdi J, Kang JJ, Toma I. Activation of the renal renin-angiotensin system in diabetes--new concepts. Nephrology, dialysis, transplantation : official publication of the European Dialysis and Transplant Association - European Renal Association. 2008;23(10):3047-9.
20. Kolb H, Mandrup-Poulsen T. The global diabetes epidemic as a consequence of lifestyle-induced low-grade inflammation. Diabetologia. 2010;53(1):10-20.
21. Medzhitov R. Origin and physiological roles of inflammation. Nature. 2008;454(7203):428-35.
22. Bates DO, Curry FE. Vascular endothelial growth factor increases hydraulic conductivity of isolated perfused microvessels. The American journal of physiology. 1996;271(6 Pt 2):H2520-8.
23. Brownlee M. Biochemistry and molecular cell biology of diabetic complications. Nature. 2001;414(6865):813-20.
24. Nishikawa T, Edelstein D, Du XL, Yamagishi S, Matsumura T, Kaneda Y, et al. Normalizing mitochondrial superoxide production blocks three pathways of hyperglycaemic damage. Nature. 2000;404(6779):787-90.
25. Eizirik DL, Colli ML, Ortis F. The role of inflammation in insulitis and beta-cell loss in type 1 diabetes. Nature reviews Endocrinology. 2009;5(4):219-26.
26. Schram MT, Chaturvedi N, Schalkwijk C, Giorgino F, Ebeling P, Fuller JH, et al. Vascular risk factors and markers of endothelial function as determinants of inflammatory markers in type 1 diabetes: the EURODIAB Prospective Complications Study. Diabetes Care. 2003;26(7):2165-73.
27. Roy MS, Gunkel RD, Podgor MJ. Color vision defects in early diabetic retinopathy. Archives of ophthalmology (Chicago, Ill : 1960). 1986;104(2):225-8.
28. Tyrberg M, Lindblad U, Melander A, Lovestam-Adrian M, Ponjavic V, Andreasson S. Electrophysiological studies in newly onset type 2 diabetes without visible vascular retinopathy. Documenta ophthalmologica Advances in ophthalmology. 2011;123(3):193-8.
29. Fagrell B, Jorneskog G, Intaglietta M. Disturbed microvascular reactivity and shunting - a major cause for diabetic complications. Vascular medicine (London, England). 1999;4(3):125-7.
30. Sangiorgi S, Manelli A, Reguzzoni M, Ronga M, Protasoni M, Dell'Orbo C. The cutaneous microvascular architecture of human diabetic toe studied by corrosion casting and scanning electron microscopy analysis. Anatomical record (Hoboken, NJ : 2007). 2010;293(10):1639-45.

31. Gabbay IE, Gabbay M, Gabbay U. Diabetic foot cellular hypoxia may be due to capillary shunting--a novel hypothesis. Medical hypotheses. 2014;82(1):57-9.
32. Bek T, Stefansson E, Hardarson SH. Retinal oxygen saturation is an independent risk factor for the severity of diabetic retinopathy. The British journal of ophthalmology. 2018.
33. Duke-Elder S. System of Ophtalmology. Volume X. London: Henry Kimpton; 1967. p. 417.
34. Antonetti DA, Barber AJ, Bronson SK, Freeman WM, Gardner TW, Jefferson LS, et al. Diabetic retinopathy: seeing beyond glucose-induced microvascular disease. Diabetes. 2006;55(9):2401-11.
35. Ballatyne AJ LA, editor The pathology of diabetic retinopathy1943 1943.
36. Bearse MA, Jr., Han Y, Schneck ME, Barez S, Jacobsen C, Adams AJ. Local multifocal oscillatory potential abnormalities in diabetes and early diabetic retinopathy. Investigative ophthalmology & visual science. 2004;45(9):3259-65.
37. Bengtsson B, Heijl A, Agardh E. Visual fields correlate better than visual acuity to severity of diabetic retinopathy. Diabetologia. 2005;48(12):2494-500.
38. Hellgren KJ, Agardh E, Bengtsson B. Progression of early retinal dysfunction in diabetes over time: results of a long-term prospective clinical study. Diabetes. 2014;63(9):3104-11.
39. Holm K, Larsson J, Lovestam-Adrian M. In diabetic retinopathy, foveal thickness of 300 µm seems to correlate with functionally significant loss of vision. Documenta ophthalmologica Advances in ophthalmology. 2007;114(3):117-24.
40. Holm K, Ponjavic V, Lovestam-Adrian M. Using multifocal electroretinography hard exudates affect macular function in eyes with diabetic retinopathy. Graefe's archive for clinical and experimental ophthalmology = Albrecht von Graefes Archiv fur klinische und experimentelle Ophthalmologie. 2010;248(9):1241-7.
41. Milkiewicz M, Ispanovic E, Doyle JL, Haas TL. Regulators of angiogenesis and strategies for their therapeutic manipulation. The international journal of biochemistry & cell biology. 2006;38(3):333-57.
42. Early Treatment of Diabetic Retinopathy Study Group Photocoagulation for diabetic macular edema. early treatment diabetic retinopathy study report number 1. Arch Ophthtalmol. 1985;103(12):1796-806.
43. Gustavsson C, Agardh CD, Agardh E. Profile of intraocular tumour necrosis factor-alpha and interleukin-6 in diabetic subjects with different degrees of diabetic retinopathy. Acta ophthalmologica. 2013;91(5):445-52.
44. Patel JI, Tombran-Tink J, Hykin PG, Gregor ZJ, Cree IA. Vitreous and aqueous concentrations of proangiogenic, antiangiogenic factors and other cytokines in diabetic retinopathy patients with macular edema: Implications for structural differences in macular profiles. Experimental eye research. 2006;82(5):798-806.
45. Stefansson E. Ocular hypotony: what is the mechanism of effusion and oedema? Acta Ophthalmol Scand. 2007;85(6):584-5.
46. Bresnick GH, Palta M. Predicting progression to severe proliferative diabetic retinopathy. Archives of ophthalmology (Chicago, Ill : 1960). 1987;105(6):810-4.

47. Klein R, Klein BE, Moss SE, Davis MD, DeMets DL. The Wisconsin Epidemiologic Study of Diabetic Retinopathy. IX. Four-year incidence and progression of diabetic retinopathy when age at diagnosis is less than 30 years. Archives of ophthalmology (Chicago, Ill : 1960). 1989;107(2):237-43.
48. Aldington SJ, Kohner E, Methodology for retinal photography and assessment. Diabetologia. 1995;38:437-44.
49. Klein R, Klein BE, Magli YL. An alternative method of grading. Ophtalmology. 1986;93:1183-7.
50. Klein R, Klein BE, Moss SE, Davis MD, DeMets DL. The Wisconsin Epidemiologic Study of Diabetes Retinopathy IX. Four -year incidence. Archives of ophthalmology (Chicago, Ill : 1960). 1989(107):237-43.
51. Fong DS, Aiello L, Gardner TW, King GL, Blankenship G, Cavallerano JD, et al. Retinopathy in diabetes. Diabetes Care. 2004;27 Suppl 1:S84-7.
52. Klein R, Klein BE, Moss SE, Davis MD, DeMets DL. The Wisconsin epidemiologic study of diabetic retinopathy. II. Prevalence and risk of diabetic retinopathy when age at diagnosis is less than 30 years. Archives of ophthalmology (Chicago, Ill : 1960). 1984;102(4):520-6.
53. Klein R, Klein BE, Moss SE, Davis MD, DeMets DL. The Wisconsin epidemiologic study of diabetic retinopathy. III. Prevalence and risk of diabetic retinopathy when age at diagnosis is 30 or more years. Archives of ophthalmology (Chicago, Ill : 1960). 1984;102(4):527-32.
54. Klein R, Klein BE, Moss SE, Linton KL. The Beaver Dam Eye Study. Retinopathy in adults with newly discovered and previously diagnosed diabetes mellitus. Ophthalmology. 1992;99(1):58-62.
55. Keenan TD, Johnston RL, Donachie PH, Sparrow JM, Stratton IM, Scanlon P. United Kingdom National Ophthalmology Database Study: Diabetic Retinopathy; Report 1: prevalence of centre-involving diabetic macular oedema and other grades of maculopathy and retinopathy in hospital eye services. Eye (London, England). 2013;27(12):1397-404.
56. McLeod BK, Thompson JR, Rosenthal AR. The prevalence of retinopathy in the insulin-requiring diabetic patients of an English country town. Eye (London, England). 1988;2 (Pt 4):424-30.
57. Morgan CL, Currie CJ, Stott NC, Smithers M, Butler CC, Peters JR. The prevalence of multiple diabetes-related complications. Diabet Med. 2000;17(2):146-51.
58. Sparrow JM, McLeod BK, Smith TD, Birch MK, Rosenthal AR. The prevalence of diabetic retinopathy and maculopathy and their risk factors in the non-insulin-treated diabetic patients of an English town. Eye (London, England). 1993;7 (Pt 1):158-63.
59. Henricsson M, Nilsson A, Groop L, Heijl A, Janzon L. Prevalence of diabetic retinopathy in relation to age at onset of the diabetes, treatment, duration and glycemic control. Acta Ophthalmol Scand. 1996;74(6):523-7.
60. Kohner EM, Aldington SJ, Stratton IM, Manley SE, Holman RR, Matthews DR, et al. United Kingdom Prospective Diabetes Study, 30: diabetic retinopathy at diagnosis of non-insulin-dependent diabetes mellitus and associated risk factors. Archives of ophthalmology (Chicago, Ill : 1960). 1998;116(3):297-303.

61. Field RA MJSW, Schepens CL symposium on the treatment of diabetic retinopathy. Goldberg MF FS, editor. Washington: Government Printing Office; 1969.
62. Meyer-Schwickerath G. History and Development of Photocoagulation. Am J Ophtamol. 1967; Jun 63 (6):1812-14.
63. Meyer-Schwickerath G. The treatment of Ophtalmic Vascular Disease by Argon Laser Photocoagulation. Trans Am Acad Ophthalmol Otolaryngol. 1969(158):605.
64. Diabetic Retinopathy Study Group. Photocoagulation treatment. Trans Am Acad Ophthalmol Otolaryngol. 1978;85:82.
65. Photocoagulation for diabetic macular edema. Early Treatment Diabetic Retinopathy Study report number 1. Early Treatment Diabetic Retinopathy Study research group. Archives of ophthalmology (Chicago, Ill : 1960). 1985;103(12):1796-806.
66. Zein WM, Noureddin BN, Jurdi FA, Schakal A, Bashshur ZF. Panretinal photocoagulation and intravitreal triamcinolone acetonide for the management of proliferative diabetic retinopathy with macular edema. Retina (Philadelphia, Pa). 2006;26(2):137-42.
67. Virgili G, Parravano M, Evans JR, Gordon I, Lucenteforte E. Anti-vascular endothelial growth factor for diabetic macular oedema: a network meta-analysis. The Cochrane database of systematic reviews. 2018;10:Cd007419.
68. Blumenkranz MS. Optimal current and future treatments for diabetic macular oedema. Eye (London, England). 2010;24(3):428-34.
69. Lewis H, Abrams GW, Blumenkranz MS, Campo RV. Vitrectomy for diabetic macular traction and edema associated with posterior hyaloidal traction. Ophthalmology. 1992;99(5):753-9.
70. Whitehead M, Wickremasininghe S OA. Diabetic retinopthy: a complex Expert Opin Biol Ther. 2018;18(12):1257-70.
71. Yanko L, Goldbourt U, Michaelson IC, Shapiro A, Yaari S. Prevalence and 15-year incidence of retinopathy and associated characteristics in middle-aged and elderly diabetic men. The British journal of ophthalmology. 1983;67(11):759-65.
72. Klein R KB, Moss SE, Cruickshanks KJ. The Wisconsin Epidemiologic´Study of Diabetic Retinopathy. Arch Ophthlmol. 1994;112(Sep):1217.
73. Pryce TD. A case of perforating ulcers of booth feet associated with diabetes and ataxic symptoms. The Lancet. 1887(July 2):11-2.
74. Walsh CH, Soler NG, Fitzgerald MG, Malins JM. Association of foot lesions with retinopathy in patients with newly diagnosed diabetes. Lancet (London, England). 1975;1(7912):878-80.
75. Steel JM, Shenfield GM, Duncan LJ. Rapid onset of proliferative retinopathy in young insulin-independent diabetics. British medical journal. 1976;2(6040):852.
76. Glynn JR, Carr EK, Jeffcoate WJ. Foot ulcers in previously undiagnosed diabetes mellitus. BMJ (Clinical research ed). 1990;300(6731):1046-7.
77. Singh N, Armstrong DG, Lipsky BA. Preventing foot ulcers in patients with diabetes. Jama. 2005;293(2):217-28.

78. Muller IS, de Grauw WJ, van Gerwen WH, Bartelink ML, van Den Hoogen HJ, Rutten GE. Foot ulceration and lower limb amputation in type 2 diabetic patients in dutch primary health care. Diabetes Care. 2002;25(3):570-4.
79. Monteiro-Soares M, Boyko EJ, Ribeiro J, Ribeiro I, Dinis-Ribeiro M. Predictive factors for diabetic foot ulceration: a systematic review. Diabetes/metabolism research and reviews. 2012;28(7):574-600.
80. Moulik PK, Mtonga R, Gill GV. Amputation and mortality in new-onset diabetic foot ulcers stratified by etiology. Diabetes Care. 2003;26(2):491-4.
81. Kumar S, Ashe HA, Parnell LN, Fernando DJ, Tsigos C, Young RJ, et al. The prevalence of foot ulceration and its correlates in type 2 diabetic patients: a population-based study. Diabet Med. 1994;11(5):480-4.
82. Nyamu PN, Otieno CF, Amayo EO, McLigeyo SO. Risk factors and prevalence of diabetic foot ulcers at Kenyatta National Hospital, Nairobi. East African medical journal. 2003;80(1):36-43.
83. Walters DP, Gatling W, Mullee MA, Hill RD. The distribution and severity of diabetic foot disease: a community study with comparison to a non-diabetic group. Diabet Med. 1992;9(4):354-8.
84. Abbott CA, Vileikyte L, Williamson S, Carrington AL, Boulton AJ. Multicenter study of the incidence of and predictive risk factors for diabetic neuropathic foot ulceration. Diabetes Care. 1998;21(7):1071-5.
85. Abbott CA, Carrington AL, Ashe H, Bath S, Every LC, Griffiths J, et al. The North-West Diabetes Foot Care Study: incidence of, and risk factors for, new diabetic foot ulceration in a community-based patient cohort. Diabet Med. 2002;19(5):377-84.
86. Wagner FW, Jr. The dysvascular foot: a system for diagnosis and treatment. Foot & ankle. 1981;2(2):64-122.
87. Lavery LA, Armstrong DG, Harkless LB. Classification of diabetic foot wounds. Ostomy/wound management. 1997;43(2):44-8, 50, 2-3.
88. Ince P, Abbas ZG, Lutale JK, Basit A, Ali SM, Chohan F, et al. Use of the SINBAD classification system and score in comparing outcome of foot ulcer management on three continents. Diabetes Care. 2008;31(5):964-7.
89. Hyperbaric oxygen therapy as adjunctive treatment of diabetic foot ulcers. In: Boulton AJ, editor. The diabetic foot: elsevier; 2013. p. 957-80.
90. Jain K.K. (Ed), Textbook of hyperbaric medicine. Cambridge, MA, USA: Hogrefe; 2009.
91. Covin AP. Human factors in decompression sickness in compressed air workers in the United Kingdom 1986-2000. London: Health and security executive; 2003.
92. Moon RE, Camporesi EM, Kisslo JA. Patent foramen ovale and decompression sickness in divers. Lancet (London, England). 1989;1(8637):513-4.
93. Hagen PT, Scholz DG, Edwards WD. Incidence and size of patent foramen ovale during the first 10 decades of life: an autopsy study of 965 normal hearts. Mayo Clinic proceedings. 1984;59(1):17-20.
94. McCallum RI. Decompression sickness: a review. British journal of industrial medicine. 1968;25(1):4-21.

95. Swan JG, Wilbur JC, Moodie KL, Kane SA, Knaus DA, Phillips SD, et al. Microbubbles are detected prior to larger bubbles following decompression. Journal of applied physiology (Bethesda, Md : 1985). 2014;116(7):790-6.
96. Freyssinet JM. Cellular microparticles: what are they bad or good for? Journal of thrombosis and haemostasis : JTH. 2003;1(7):1655-62.
97. Madden LA, Chrismas BC, Mellor D, Vince RV, Midgley AW, McNaughton LR, et al. Endothelial function and stress response after simulated dives to 18 msw breathing air or oxygen. Aviation, space, and environmental medicine. 2010;81(1):41-5.
98. Singh CK, Chhabra G, Ndiaye MA, Garcia-Peterson LM, Mack NJ, Ahmad N. The Role of Sirtuins in Antioxidant and Redox Signaling. Antioxidants & redox signaling. 2018;28(8):643-61.
99. B C. Dykarkonstens utveckling i Sverige fram till 1850-talet (Development in Sweden of the art of diving until 1850) Sjöhistorisk Årsbok 1975-1976 (Year book of Maritime History 1975-1976). Stockholm: Föreningen Sveriges Sjöfartsmuseum i Stockholm (Society of the Swedish Maritime Museum in Stockholm); 1977. p. 9-24.
100. Behnke A S. US Navy Bulletins. 1937. p. 66-73.
101. Grover CA, Grover DH. Albert Behnke: nitrogen narcosis. The Journal of emergency medicine. 2014;46(2):225-7.
102. Lavoisier A-L, Memoires sur la respiration et la transpiration des animaux. Les maîtres de la pensee scientifique. Paris: Gauthier-Villars et Cie éditeurs; 1920.
103. Dejours P, Dejours S. The effects of barometric pressure according to Paul Bert: the question today. International journal of sports medicine. 1992;13 Suppl 1:S1-5.
104. Palmquist BM, Philipson B, Barr PO. Nuclear cataract and myopia during hyperbaric oxygen therapy. The British journal of ophthalmology. 1984;68(2):113-7.
105. Fledelius HC, Jansen E. Hypermetropic refractive change after hyperbaric oxygen therapy. Acta Ophthalmol Scand. 2004;82(3 Pt 1):313-4.
106. Lyne AJ. Ocular effects of hyperbaric oxygen. Transactions of the ophthalmological societies of the United Kingdom. 1978;98(1):66-8.
107. Evanger K, Haugen OH, Irgens A, Aanderud L, Thorsen E. Ocular refractive changes in patients receiving hyperbaric oxygen administered by oronasal mask or hood. Acta Ophthalmol Scand. 2004;82(4):449-53.
108. Evanger K, Vaagbo G, Thorsen E, Haugen OH. Phakic and pseudophakic eyes in patients during hyperbaric oxygen therapy. Optometry and vision science : official publication of the American Academy of Optometry. 2011;88(6):691-6.
109. Evanger K, Pierscionek BK, Vaagbo G, Thorsen E, Haugen OH. Myopic Shift during Hyperbaric Oxygenation Attributed to Lens Index Changes. Optometry and vision science : official publication of the American Academy of Optometry. 2015;92(11):1076-84.
110. Londahl M, Katzman P, Nilsson A, Hammarlund C, Sellman A, Wykman A, et al. A prospective study: hyperbaric oxygen therapy in diabetics with chronic foot ulcers. Journal of wound care. 2006;15(10):457-9.

111. Sellman A KP, Andréasson S, Löndah M. Long-term Effects of Hyperbaric OxygenTtherapy on Visual Acuity and Retinopathy. SUBMITTED. 2019.
112. Sellman A LM, Andréasson S, Katzman P. Transcutaneous oximetry but not arterial toe blood pressure or ankle-brachial index is related to macular thickness in patients withchronic diabetic foot ulcers. Journal of Experimental and Integrative Medicine. 2013;3 (2):81-5.
113. Sellman A, Katzman P, Andreasson S, Londahl M. Presence of chronic diabetic foot ulcers is associated with more frequent and more advanced retinopathy. Diabet Med. 2018;35(10):1364-70.
114. Kaiser PK. Prospective evaluation of visual acuity assessment: a comparison of Snellen versus ETDRS charts in clinical practice (An AOS Thesis). Transactions of the American Ophthalmological Society. 2009;107:311-24.
115. Grading diabetic retinopathy from stereoscopic color fundus photographs--an extension of the modified Airlie House classification. ETDRS report number 10. Early Treatment Diabetic Retinopathy Study Research Group. Ophthalmology. 1991;98(5 Suppl):786-806.
116. Diabetic retinopathy study. Report Number 6. Design, methods, and baseline results. Report Number 7. A modification of the Airlie House classification of diabetic retinopathy. Prepared by the Diabetic Retinopathy Study Group. Invest Ophthalmol Vis Sci. 1981;21(1 Pt 2):1-226.
117. Coscas G, Cunha-Vaz J, Soubrane G. Macular Edema: Definition and Basic Concepts. Developments in ophthalmology. 2017;58:1-10.
118. Levey AS, Coresh J, Greene T, Stevens LA, Zhang YL, Hendriksen S, et al. Using standardized serum creatinine values in the modification of diet in renal disease study equation for estimating glomerular filtration rate. Ann Intern Med. 2006;145(4):247-54.
119. C H. In: A S, editor. 2017.
120. McNeely MJ, Boyko EJ, Ahroni JH, Stensel VL, Reiber GE, Smith DG, et al. The independent contributions of diabetic neuropathy and vasculopathy in foot ulceration. How great are the risks? Diabetes Care. 1995;18(2):216-9.
121. Day MR, Harkless LB. Factors associated with pedal ulceration in patients with diabetes mellitus. Journal of the American Podiatric Medical Association. 1997;87(8):365-9.
122. Prompers L, Schaper N, Apelqvist J, Edmonds M, Jude E, Mauricio D, et al. Prediction of outcome in individuals with diabetic foot ulcers: focus on the differences between individuals with and without peripheral arterial disease. The EURODIALE Study. Diabetologia. 2008;51(5):747-55.
123. Yan BP, Zhang Y, Kong AP, Luk AO, Ozaki R, Yeung R, et al. Borderline ankle-brachial index is associated with increased prevalence of micro- and macrovascular complications in type 2 diabetes: A cross-sectional analysis of 12,772 patients from the Joint Asia Diabetes Evaluation Program. Diabetes & vascular disease research. 2015;12(5):334-41.

124. Boyko EJ, Ahroni JH, Cohen V, Nelson KM, Heagerty PJ. Prediction of diabetic foot ulcer occurrence using commonly available clinical information: the Seattle Diabetic Foot Study. Diabetes Care. 2006;29(6):1202-7.
125. Boyko EJ, Ahroni JH, Stensel V, Forsberg RC, Davignon DR, Smith DG. A prospective study of risk factors for diabetic foot ulcer. The Seattle Diabetic Foot Study. Diabetes Care. 1999;22(7):1036-42.
126. Monteiro-Soares M, Dinis-Ribeiro M. External validation and optimisation of a model for predicting foot ulcers in patients with diabetes. Diabetologia. 2010;53(7):1525-33.
127. Sriussadaporn S, Mekanandha P, Vannasaeng S, Nitiyanant W, Komoltri C, Ploybutr S, et al. Factors associated with diabetic foot ulceration in Thailand: a case-control study. Diabet Med. 1997;14(1):50-6.
128. Monami M, Vivarelli M, Desideri CM, Colombi C, Marchionni N, Mannucci E. Pulse pressure and prediction of incident foot ulcers in type 2 diabetes. Diabetes Care. 2009;32(5):897-9.
129. Gonzalez JS, Vileikyte L, Ulbrecht JS, Rubin RR, Garrow AP, Delgado C, et al. Depression predicts first but not recurrent diabetic foot ulcers. Diabetologia. 2010;53(10):2241-8.
130. Lavery LA, Armstrong DG, Vela SA, Quebedeaux TL, Fleischli JG. Practical criteria for screening patients at high risk for diabetic foot ulceration. Archives of internal medicine. 1998;158(2):157-62.
131. Ndip A, Lavery LA, Lafontaine J, Rutter MK, Vardhan A, Vileikyte L, et al. High levels of foot ulceration and amputation risk in a multiracial cohort of diabetic patients on dialysis therapy. Diabetes Care. 2010;33(4):878-80.
132. Karam T, Kamath YS, Rao LG, Rao KA, Shenoy SB, Bhandary SV. Diabetic retinopathy in patients with diabetic foot syndrome in South India. Indian journal of ophthalmology. 2018;66(4):547-50.
133. Hwang DJ, Lee KM, Park MS, Choi SH, Park JI, Cho JH, et al. Association between diabetic foot ulcer and diabetic retinopathy. PloS one. 2017;12(4):e0175270.
134. Pearce I, Simo R, Lovestam-Adrian M, Wong DT, Evans M. Association between diabetic eye disease and other complications of diabetes: Implications for care. A systematic review. Diabetes, obesity & metabolism. 2019;21(3):467-78.
135. Chen SC, Hsiao PJ, Huang JC, Lin KD, Hsu WH, Lee YL, et al. Abnormally Low or High Ankle-Brachial Index Is Associated with Proliferative Diabetic Retinopathy in Type 2 Diabetic Mellitus Patients. PloS one. 2015;10(7):e0134718.
136. Chen YW, Wang YY, Zhao D, Yu CG, Xin Z, Cao X, et al. High prevalence of lower extremity peripheral artery disease in type 2 diabetes patients with proliferative diabetic retinopathy. PloS one. 2015;10(3):e0122022.
137. Apelqvist J, Agardh CD. The association between clinical risk factors and outcome of diabetic foot ulcers. Diabetes research and clinical practice. 1992;18(1):43-53.
138. Tomita M, Kabeya Y, Okisugi M, Katsuki T, Oikawa Y, Atsumi Y, et al. Diabetic Microangiopathy Is an Independent Predictor of Incident Diabetic Foot Ulcer. Journal of diabetes research. 2016;2016:5938540.

139. Baba M, Davis WA, Davis TM. A longitudinal study of foot ulceration and its risk factors in community-based patients with type 2 diabetes: the Fremantle Diabetes Study. Diabetes research and clinical practice. 2014;106(1):42-9.
140. Leymarie F, Richard JL, Malgrange D. Factors associated with diabetic patients at high risk for foot ulceration. Diabetes & metabolism. 2005;31(6):603-5.
141. Bruun C, Siersma V, Guassora AD, Holstein P, de Fine Olivarius N. Amputations and foot ulcers in patients newly diagnosed with type 2 diabetes mellitus and observed for 19 years. The role of age, gender and co-morbidity. Diabet Med. 2013;30(8):964-72.
142. Parisi MC, Moura Neto A, Menezes FH, Gomes MB, Teixeira RM, de Oliveira JE, et al. Baseline characteristics and risk factors for ulcer, amputation and severe neuropathy in diabetic foot at risk: the BRAZUPA study. Diabetology & metabolic syndrome. 2016;8:25.
143. Harris Nwanyanwu K, Talwar N, Gardner TW, Wrobel JS, Herman WH, Stein JD. Predicting development of proliferative diabetic retinopathy. Diabetes Care. 2013;36(6):1562-8.
144. Hamalainen H, Ronnemaa T, Halonen JP, Toikka T. Factors predicting lower extremity amputations in patients with type 1 or type 2 diabetes mellitus: a population-based 7-year follow-up study. Journal of internal medicine. 1999;246(1):97-103.
145. Krogh A, The number and distribution of capillaries in muscles with calculations of the oxygen pressure head necessary for supplying the tissue. Journal od Physiology. 1919(52):409-15.
146. Londahl M. The diabetic foot. Boulton AJ, editor 2013.
147. Margolis DJ, Gupta J, Hoffstad O, Papdopoulos M, Glick HA, Thom SR, et al. Lack of effectiveness of hyperbaric oxygen therapy for the treatment of diabetic foot ulcer and the prevention of amputation: a cohort study. Diabetes Care. 2013;36(7):1961-6.
148. Fedorko L, Bowen JM, Jones W, Oreopoulos G, Goeree R, Hopkins RB, et al. Hyperbaric Oxygen Therapy Does Not Reduce Indications for Amputation in Patients With Diabetes With Nonhealing Ulcers of the Lower Limb: A Prospective, Double-Blind, Randomized Controlled Clinical Trial. Diabetes Care. 2016;39(3):392-9.
149. Tesfaye S, Harris N, Jakubowski JJ, Mody C, Wilson RM, Rennie IG, et al. Impaired blood flow and arterio-venous shunting in human diabetic neuropathy: a novel technique of nerve photography and fluorescein angiography. Diabetologia. 1993;36(12):1266-74.
150. Hunt TK, Hopf HW. Wound healing and wound infection. What surgeons and anesthesiologists can do. The Surgical clinics of North America. 1997;77(3):587-606.
151. Hammarlund C, Sundberg T. Hyperbaric oxygen reduced size of chronic leg ulcers: a randomized double-blind study. Plastic and reconstructive surgery. 1994;93(4):829-33; discussion 34.
152. Jain KK. Textbook of Hyperbaric Medicine. Cambridge, MA, USA: Hogrefe 2009.

153. Winstanley J. Treatment of neo-vascularization with oxygen at high pressure. The British journal of ophthalmology. 1963;47:542-6.
154. Haddad HM, Leopold IH. Effect of hyperbaric oxygenation on microcirculation: use in therapy of retinal vascular disorders. Investigative ophthalmology. 1965;4(6):1141-51.
155. Sindlerova E. [Diabetic retinopathy and its therapy using hyperbaric oxygenation]. Ceskoslovenska oftalmologie. 1978;34(4):289-95.
156. Ogura Y, Kiryu J, Takahashi K, Honda Y. [Visual improvement in diabetic macular edema by hyperbaric oxygen treatment]. Nippon Ganka Gakkai zasshi. 1988;92(9):1456-60.
157. Dumitru R. [The hyperbaric method in the treatment of diabetic retinopathy, an alternative to laser therapy?]. Oftalmologia (Bucharest, Romania : 1990). 1993;37(1):12-6.
158. Caamaño JV, de Lara A, editor HBO treatment of 525 cases of diabetic retinopathy; HBO applications in other types of ocular pathology. Proceedings of the IX congress of European Undersea Biomedical Society; 1983: 169-178. Barcelona.
159. Tran V, Smart D. Proliferative retinopathy during hyperbaric oxygen treatment. Diving and hyperbaric medicine. 2017;47(3):203.
160. Yonekawa Y, Hypes SM, Abbey AM, Williams GA, Wolfe JD. Exacerbation of macular oedema associated with hyperbaric oxygen therapy. Clinical & experimental ophthalmology. 2016;44(7):625-6.
161. Löndahl M. Hyperbaric oxygen therapy as adjunctive treatment of diabetic foot ulcers. In The diabetic foot. Ed Boulton AJM; 2013 p 969-971.
162. Londahl M, Katzman P, Nilsson A, Hammarlund C. Hyperbaric oxygen therapy facilitates healing of chronic foot ulcers in patients with diabetes. Diabetes Care. 2010;33(5):998-1003.
163. Railton R, Newman P, Hislop J, Harrower AD. Reduced transcutaneous oxygen tension and impaired vascular response in Type 1 (insulin-dependent) diabetes. Diabetologia. 1983;25(4):340-2.
164. Uccioli L, Monticone G, Russo F, Mormile F, Durola L, Mennuni G, et al. Autonomic neuropathy and transcutaneous oxymetry in diabetic lower extremities. Diabetologia. 1994;37(10):1051-5.
165. Huang K, Ma Y, Wang J, Shi S, Fu L, Liu J, et al. The correlation between transcutaneous oxygen tension and microvascular complications in type 2 diabetic patients. Journal of diabetes and its complications. 2017;31(5):886-90.
166. Fagher K, Katzman P, Londahl M. Transcutaneous oxygen pressure as a predictor for short-term survival in patients with type 2 diabetes and foot ulcers: a comparison with ankle-brachial index and toe blood pressure. Acta diabetologica. 2018;55(8):781-8.
167. Pirart J. [Diabetes mellitus and its degenerative complications: a prospective study of 4,400 patients observed between 1947 and 1973 (3rd and last part) (author's transl)]. Diabete & metabolisme. 1977;3(4):245-56.

168. Klein R, Klein BE, Moss SE, Davis MD, DeMets DL. The Wisconsin Epidemiologic Study of Diabetic Retinopathy. X. Four-year incidence and progression of diabetic retinopathy when age at diagnosis is 30 years or more. Archives of ophthalmology (Chicago, Ill : 1960). 1989;107(2):244-9.
169. Klein R, Klein BE, Moss SE, Cruickshanks KJ. The Wisconsin Epidemiologic Study of diabetic retinopathy. XIV. Ten-year incidence and progression of diabetic retinopathy. Archives of ophthalmology (Chicago, Ill : 1960). 1994;112(9):1217-28.
170. Cusick M, Meleth AD, Agron E, Fisher MR, Reed GF, Knatterud GL, et al. Associations of mortality and diabetes complications in patients with type 1 and type 2 diabetes: early treatment diabetic retinopathy study report no. 27. Diabetes Care. 2005;28(3):617-25.
171. Henricsson M, Nilsson A, Heijl A, Janzon L, Groop L. Mortality in diabetic patients participating in an ophthalmological control and screening programme. Diabet Med. 1997;14(7):576-83.
172. Jones CD, Greenwood RH, Misra A, Bachmann MO. Incidence and progression of diabetic retinopathy during 17 years of a population-based screening program in England. Diabetes Care. 2012;35(3):592-6.
173. Gu K, Cowie CC, Harris MI. Mortality in adults with and without diabetes in a national cohort of the U.S. population, 1971-1993. Diabetes Care. 1998;21(7):1138-45.
174. Gregg EW, Gu Q, Cheng YJ, Narayan KM, Cowie CC. Mortality trends in men and women with diabetes, 1971 to 2000. Annals of internal medicine. 2007;147(3):149-55.
175. Klein R, Moss SE, Klein BE, DeMets DL. Relation of ocular and systemic factors to survival in diabetes. Archives of internal medicine. 1989;149(2):266-72.
176. Rubio J A JS, Álvarez J. Caracteristicas clinicas y mortalidad de los pacientes atendidos en una unidad Multidiciplinar de Pie Diabético. Endocrinología, Diabetes y Nutrición. 2017;64(5):241-9.
177. Mundet X, Pou A, Piquer N, Sanmartin MI, Tarruella M, Gimbert R, et al. Prevalence and incidence of chronic complications and mortality in a cohort of type 2 diabetic patients in Spain. Primary care diabetes. 2008;2(3):135-40.
178. Freeman EE, Egleston BL, West SK, Bandeen-Roche K, Rubin G. Visual acuity change and mortality in older adults. Investigative ophthalmology & visual science. 2005;46(11):4040-5.
179. Felson DT, Anderson JJ, Hannan MT, Milton RC, Wilson PW, Kiel DP. Impaired vision and hip fracture. The Framingham Study. Journal of the American Geriatrics Society. 1989;37(6):495-500.
180. Ivers RQ, Mitchell P, Cumming RG. Sensory impairment and driving: the Blue Mountains Eye Study. American journal of public health. 1999;89(1):85-7.
181. Fong CS, Mitchell P, Rochtchina E, de Loryn T, Tan AG, Wang JJ. Visual impairment corrected via cataract surgery and 5-year survival in a prospective cohort. American journal of ophthalmology. 2014;157(1):163-70.e1.

182. Kulmala J, Era P, Tormakangas T, Parssinen O, Rantanen T, Heikkinen E. Visual acuity and mortality in older people and factors on the pathway. Ophthalmic epidemiology. 2008;15(2):128-34.
183. Lovestam-Adrian M, Hansson-Lundblad C, Torffvit O. Sight-threatening retinopathy is associated with lower mortality in type 2 diabetic subjects: a 10-year observation study. Diabetes research and clinical practice. 2007;77(1):141-7.
184. Klein BE, Klein R, Knudtson MD, Lee KE. Frailty, morbidity and survival. Archives of gerontology and geriatrics. 2005;41(2):141-9.
185. Kulmala J, Nykanen I, Hartikainen S. Frailty as a predictor of all-cause mortality in older men and women. Geriatrics & gerontology international. 2014;14(4):899-905.
186. Siantar RG, Cheng CY, Gemmy Cheung CM, Lamoureux EL, Ong PG, Chow KY, et al. Impact of Visual Impairment and Eye diseases on Mortality: the Singapore Malay Eye Study (SiMES). Scientific reports. 2015;5:16304.

Printed by Media-Tryck, Lund 2019 NORDIC SWAN ECOLABEL 3041 0903

FACULTY OF MEDICINE

Lund University, Faculty of Medicine
Doctoral Dissertation Series 2019:31
ISBN 978-91-7619-760-8
ISSN 1652-8220