PREVALENCE OF MICROVASCULAR COMPLICATIONS AMONG INDOOR DIABETIC PATIENTS

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By

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In the Name of Allah,
The Most Gracious,
The Most Merciful.

DEDICATED TO:

My Father, my Best Teacher

&

My Mother, my best well-wisher

CERTIFICATE

This thesis, submitted by **Dr. Shafiqur-Rahman** is accepted in its present form by the Department of Biological Sciences, Quaid-i-Azam University, Islamabad as satisfying the thesis requirements for the degree of MASTER OF PHILOSOPHY IN ENDOCRINOLOGY.

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ABBREVIATIONS

ACR	Albumin / Creatinine Ratio
ADA	American Diabetes Association
BMI	Body mass index
BP	Blood pressure
DCCT	Diabetes control and Complication Trial
DERI	Diabetes Epidemiology Research International
df	Degrees of freedom
DM	Diabetes mellitus
ESRD	End-stage renal disease
GFR	Glomerular filtration rate
HLA	Human leukocyte antigen
Hr.	Hour
HTN	Hypertension
IDDM	Insulin dependant diabetes mellitus
IFG	Impaired fasting glucose
IGT	Impaired glucose tolerance
kg	Kilogram
M:F	Male to female ratio
m^2	Square meter
mg	Milligram
mg/dl	Milligram per deciliter
mmol/l	Millimole per liter
NIDDM	Non-insulin dependent diabetes mellitus
N	Number
OD	Optical density
PMRC	Pakistan Medical Research Council
SD	Standard deviation
TNF α	Tumor necrosis factor α

UAE	Urinary albumin excretion
UKPDS	United Kingdom Prospective Diabetic Study
VA	Veteran Affairs
vs.	Versus
WESDR	Wisconsin Epidemiological Study of Diabetic Retinopathy
wl	Wavelength
yr.	Year
μg	Microgram
μmol/L	Micromole per litre

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FIGURES AND PLATES

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ABSTRACT

Uncontrolled diabetes mellitus with a longer duration leads to the development and progression of microvascular complications such as retinopathy, nephropathy and neuropathy. The present study was undertaken to estimate the number and proportion of diabetic patients having these complications and to understand the role of risk factors including age, BMI and diabetes duration responsible for the development and progression of these microvascular complications. Two hundred diabetic patients with different clinical problems, admitted to Endocrinology unit of Hayatabad Medical Complex, Peshawar were included in the study.

Age, sex, type of diabetes, past medical history (including heart attacks, strokes, smoking, foot ulcers and gangrenes), family history, duration since diagnosis of diabetes, treatment modality, and cause of admission of each patient were recorded. Weight and height were measured to determine the body mass index. Each patient was examined for the measurement of blood pressure and presence or absence of arterial pulses in the feet. Casual plasma glucose on the day of admission and subsequent days was also measured. Ophthalmic examination to detect retinopathy was performed with the help of direct ophthalmoscope. Neuropathy was diagnosed by the assessment of ankle reflexes, vibration, pain and touch sensations in the feet using appropriate instruments (tendon hammer, tuning fork, small soft brush and common-pin respectively). Nephropathy was diagnosed by the estimation of total proteins in a 24 hourly-collected urinary specimen taking 200mg/24 hours as the cut-off point. Renal function tests constituting measurements of serum urea and creatinine were also performed. Renal ultrasonography was performed, where appropriate.

The results showed considerably high prevalence of retinopathy (51%), nephropathy (56.5%) and neuropathy (68.5%) occurring singly or in combination of any two or all the three complications. Of 200 patients, only 35 patients were free from any complication. Most of these uncomplicated patients had a shorter diabetes

duration (3.3±2.1 vs 9.6±5.3 years), lesser body mass index (24.3±3.3 vs 26.9±3.9 kg/m^2) and younger age (37.3±7.4 vs 50.6±12.8 years). Each complication showed positive correlation with increasing age, diabetes duration and body mass index. The differences between various groups of age, duration and body mass index were statistically significant. Mean values for age, BMI and diabetes duration were found higher in patients with retinopathy, nephropathy or neuropathy than those not having them. Hypertension was found in 107 (53.5%) cases, and its relation with the occurrence of retinopathy, nephropathy and neuropathy was evident. Mean casual plasma glucose level on the day of admission was 331±111. Family histories demonstrated that 46% patients were positive for diabetes.

The results necessitate that the diabetic patients should have awareness of the problems associated with diabetes. Moreover, there is a critical need for conducting programs for the primary and secondary prevention of not only the related complications but also the disease itself.

INTRODUCTION

DIABETES MELLITUS

Literally, Diabetes means. "to pass through" and Mellitus means, "honey flavored". Physiologically, it represents a group of metabolic disturbances characterized by chronic hyperglycemia and glycosuria, with or without the typical symptoms of thirst, polyuria, weight loss, fatigue or blurred vision (Arthur, 2001). During the last decade, research has led to the recognition that diabetes mellitus (DM) is a syndrome and comprises a heterogeneous collection of disorders. Thus in addition to carbohydrate metabolism, often there are also disturbances of fat and protein metabolism. The basic defect of relatively or absolutely deficient insulin secretion from β-cells of pancreas leads to these disturbances. Decreased peripheral glucose utilization and increased hepatic glucose production cause hyperglycemia while an imbalance between the breakdown and synthesis of both triglycerides and protein leads to increased fatty acids, ketones and negative nitrogen balance. It has also been recognized that although different types of diabetes have different etiologies, their pathologic effects are similar (Arthur, 2001).

CLASSIFICATION

There are two main types of diabetes mellitus (DM).

I. Type 1 or insulin dependant DM (IDDM) that usually occurs in the younger age group (<30 yr) and is characterized by absolute insulin deficiency.

II. Type 2 or Non-insulin dependant DM (NIDDM) occurring in older age group (>30 yr), is characterized by relative insulin deficiency due to defects in insulin secretion and/or its action (Gerich, 1998).

Overall ratio of type 1 and type 2 diabetes remains 1:9 (ADA, 1998). In the new classification of diabetes by American Diabetic Association, two more categories are described (ADA, 1997).

III. Specific type diabetes, which includes the following groups:
   i. Genetic defects of β-cell function (maturity onset diabetes of the young *i.e.* MODY 1,2,3,4)
   ii. Endocrinopathies (e.g. Cushing's syndrome, hyperthyroidism and acromegaly)
   iii. Drugs (e.g. nicotinic acid, beta-blockers and diuretics) and
   iv. Genetic syndromes (e.g. Down's syndrome, Turner syndrome and Pradder-Willi syndrome)

IV. Gestational diabetes, which is recognized for the first time during pregnancy.

ETIOLOGY

Whereas the exact cause of diabetes mellitus is not known, different etiological factors and associations have been described. For type 1 DM these include genetic factors (e.g. HLA class 2 antigens with DR3 and DR4 on chromosome 6), viruses (e.g. coxsackie B, cytomegalo, Epstein Barr viruses, mumps and rubella etc.) and autoimmunity which is supported by the presence of islet cell antibodies, insulin antibodies and glutamic acid decarboxylase (GAD) antibodies. Etiological factors that may lead to type 2 DM are; genetic abnormalities causing β-cell defects and insulin resistance, obesity and environmental factors e.g. physical inactivity, diet, stress and some drugs such as diuretics and steroids (Bingley and Gale, 1997).

Diabetes has grown epidemically to become a major health problem and an estimated population of 135 million diabetic patients worldwide (4% prevalence in 1995) is expected to rise to 300 million by the year 2025 (King *et al*, 1998). The rate of increase is expected to be 170% in developing countries as compared to 42% in developed countries (Venkat *et al*, 2000). Prevalence rates vary widely between countries and people from 0% in Papua New Guinea to more than 50% in Pima Indians (Knowler *et al*, 1993; Fujimoto, 1996). A survey conducted in the Kingdom of Tonga of Pacific Islands demonstrated a prevalence of (15%) of diabetes in an age and sex-standardized populations with higher rates for women than in men. More than 50 % people were previously undiagnosed (Colagiuri *et al*, 2002).

It has been estimated that on the average, over 10% people in Pakistan in the >25 yr age group have diabetes while the incidence of type 1 diabetes continues to be low (Shera *et al*, 1995; 1999). In a small scale study conducted in four villages near Peshawar, it was shown that nearly 3% of our local population in the age group 40-60 years suffer from diabetes mellitus with a subtle difference between males and females (Tasleem *et al*, 1995). Whereas, a comparatively large scale study conducted in Sukkur on more than four thousand people revealed a prevalence of approximately 31% diabetic cases (Badaruddin *et al*, 2002).

CLINICAL PRESENTATION

The diabetic patient may present in one of the following forms (Kumar and Clark, 1994).

Acute presentation: In young people with type 1 DM, the onset is usually acute and patient has the classic triad of urinary frequency (polyuria) due to osmotic diuresis, increased thirst (polydipsia) due to loss of fluid and electrolytes and weight loss due to accelerated fat and muscle breakdown. The patient may also present in the state of acute complications i.e. ketoacidosis. This is a medical emergency and carries very high mortality if not treated promptly by administration of adequate insulin and rehydration.

Subacute presentation: The clinical onset may be insidious particularly in older patients with type 2 DM. The classic triad of polyuria, polydipsia and weight loss may be present but usually the patients present with non-specific symptoms of bodyaches, headaches, vertigo and easy fatigability. A condition in which type 2 diabetes (or impaired glucose tolerance) is associated with obesity, hypertension and dyslipidemia is called metabolic syndrome or syndrome X. This condition is associated with a high mortality rate due to cardiovascular abnormalities (Marques *et al*, 2002).

Presentation with complications: Complication may be the presenting feature such as skin, chest or urinary tract infection, blurred vision, tingling and numbness in the feet, impotence or ischemic heart disease.

Asymptomatic presentation: Glycosuria or raised blood glucose may be detected on routine examination for some other illness without any signs and symptoms related to diabetes.

DIAGNOSTIC CRITERIA

The final confirmatory diagnosis of diabetes rests on biochemical determination of plasma glucose levels. The American Diabetic Association (1997) has defined diabetes as the fasting plasma glucose of at least 126mg/dl (7.0 mmol/l) while the World Health Organization (1999) has defined it, in addition to this, as a 2 hr post-glucose load glycemia of >200mg/dl (11.1mmol/l). Two transitional states have also been defined; impaired fasting glucose (IFG), a fasting plasma glucose levels between 110 and 126 mg/dl and impaired glucose tolerance (IGT), a post-challenge plasma glucose levels between 140 and 200 mg/dl.

MICROVASCULAR COMPLCATIONS OF DIABETES

An important characteristic of the chronic hyperglycemia in diabetes is the deposition of harmful substances in vascular endothelium causing endothelial dysfunction (King *et al*, 1996). Consequent development of end-organ damage especially in the eyes, kidneys and peripheral nerves occurs that is called micro-vascular complications or microangopathies (Oslen *et al*, 2000). Diabetes is also a risk factor in the development of atherosclerosis causing end organ damage in the heart (myocardial infarction), brain (stroke) and peripheral vessels (gangrene). These are called macrovascular complications or macroangiopathies (Stratton *et al*, 2000). These vascular complications are responsible for increased morbidity and mortality among diabetic population leading to much increased health care costs (ADA, 1998).

Many earlier and recent studies have concluded that poor glycemic control and duration of diabetes are the main determinants of microangiopathies in diabetes. Johnsson (1960) compared the occurrence of nephropathy in fifty six patients on strict diabetes control (achieving aglycosuria) with 104 patients on free diet (with ignored glycosuria) and obtained striking results as after >15 yrs diabetes duration, in "strict

control group" only 9% had nephropathy as compared to 61% in the " free diet group". It was later shown that good glucose control minimizes complications but it was only possible at the cost of dangerous hypoglycemia (Laurrence, 1963). Knowles (1964) reviewed retrospectively 85 studies on the degree of glucose control and prevalence of diabetic complications. He concluded that in 50 studies, results indicated that poor glucose control was positively correlated with vascular disease. Recently, Ramachandra *et al* (1999) in a study of 3010 type 2 DM patients noted a prevalence of 23.7% retinopathy, 19.7% nephropathy, 27.5% peripheral neuropathy and 38% hypertension. Duration of diabetes, poor glycemic control and hypertension were shown significantly associated with these complications.

Apart from the degree and duration of diabetes, other major risk factors for the development and progression of chronic diabetic complications are: obesity (Hendrick *et al*, 2002), hypertension (UKPDS group, 1998), smoking (Weil *et al*, 2001), inadequate screening of complications (Akbar *et al*, 2001), failure of life-style modification (Bray *et al*, 2002) and advancing age (Belmin and Valensi, 1996). Strong evidence exists that good diabetes management *i.e.* keeping blood glucose as close to normal as possible, results in significant benefits regarding the development and progression of microvascular complications as is evident from the Diabetes Control and Complication Trial for type 1 diabetes mellitus (DCCT research group, 1993) and United Kingdom Prospective Diabetic Study for type 2 diabetes mellitus (UKPDS group, 1998). United Kingdom Prospective Diabetic Study has been one of the major landmark studies including more than 5000 newly diagnosed type 2 DM patients. Basically designed to study the effect of intensive glucose control on the risk of diabetic complications, the final results showed that 50% patients had already had some complications at the time of diagnosis (retinopathy 55%, clinical proteinuria 4%, absent ankle jerks 13%, hypertension 39% and cardiovascular disease 8%). It also showed that after 10 years, the study population had a two-fold greater mortality than general population and one-third of the patients had a complication requiring medical attention including heart attack, stroke, retinopathy, renal failure and amputation (UKPDS group, 1991). In another landmark study, Diabetes Control and Complication

Trial (DCCT), consisting of 1441 type 1 diabetic patients, it was clearly shown that intensive blood glucose control caused remarkable reduction in the risk of complications i.e. 76% for retinopathy, 34% for microalbuminuria and 60% for neuropathy (DCCT Research group, 1993). Results similar to those of DCCT were reported in Stockholm Diabetes Intervention Study (Richard *et al*, 1993).

A variety of mechanisms contributing to the pathogenesis of one or more of the vascular complications have been suggested (Skyler, 2000). Chronic hyperglycemia leads to non-enzymatic glycation of amino acids essentially in all tissues. Cross linking of glycated proteins leads to the formation of advanced glycation end products (AGE's) which are responsible for profound alteration in protein structure and function. Accumulation of sorbitol and fructose occuring through the conversion of excessive glucose by aldose reductase inhibitors may cause changes in the nerve functions. The glucose induced activation of protein kinase C may lead to endothelial proliferation, increased cellular permeability to certain electrolytes and expression of certain growth factors such as vascular endothelium growth factor (VEGF) in the retina and transformation growth factor β (TGF β) in the renal glomeruli. Enzymatic glycation of basement membrane stimulated by hyperglycemia leads to capillary basement membrane thickening. Alteration in the platelet and endothelial function may lead to increased platelet aggregation, decreased fibrinolytic activity and thus hypercoagulability state. Hemodynamic alterations in the microcirculation especially renal glomeruli are important in initiating diabetic nephropathy (Skyler, 2000).

DIABETIC RETINOPATHY

Lundback (1954) was the first to describe microaneurysms (localized vessel dilatations) and venous beading on fundoscopic examination and termed them as the microvascular complications related to diabetes. Diabetic retinopathy is a leading cause of visual disability and is considered to be the most frequent cause of blindness among adults aged 20-74 years (ADA, 2002). In type 2 DM, 25% of the patients usually have retinopathy at the time of first diagnosis and most develop it over

subsequent decades (Shera, 1999). Diabetic retinopathy is caused by the occlusion of or damage to the small blood vessels of the retina, the inner most layer of the eyeball leading to progressive loss of vision. The basic pathology is increased thickness of basement membrane and increased permeability of the retinal capillaries to various blood constituents such as lipids and proteins etc. Aneurysmal dilatation may occur in some vessels while others become occluded (Rober *et al*, 1999).

Significant differences of prevalence of retinopathy among different races and ethnic groups have been reported. For example in a study conducted in the United States, diabetic Mexican-Americans had two times higher frequency of retinopathy than diabetic non-Hispanic Whites (Harris *et al*, 1998) after adjusting for all other influencing factors such as glycernic control, diabetes duration, age and hypertension. Data from population-based studies such as WISCONSIN Epidemiological Study of Diabetic Retinopathy (WESDR) provided valuable information on the prevalence of diabetic retinopathy. WESDR (study II) showed that in the younger onset group (<30 year), some degree of retinopathy was seen in 13% of the patients with <5 year diabetes duration and in 90 % of the patients with 10-15 year diabetes duration (Klein *et al*, 1984). Study III of the same series demonstrated that for the patients with onset of diabetes at ≥30 year age and <5 year diabetes duration, 40% of insulin taking and 24% of non-insulin taking patients had retinopathy. These rates increased to 84% and 53% respectively over 15-20 year diabetes duration (Klein *et al*, 1984). Similarly, Sheila *et al* (2001) conducted a population-based study on 4774 residents of Hispanic communities to determine the prevalence of retinopathy in diabetic population. Among 1044 diabetic patients, 2% being patients of type 1 DM, they showed a prevalence rate of 48% retinopathy out of which, 9.3% were newly diagnosed during the study.

After 20 years, all type 1 diabetic patients develop retinopathy while more than 80% of type 2 diabetic patients develop retinopathy, of which 20% show advanced changes (Kumar and Clark, 1994). A study on DCCT patients confirmed that retinopathy develops in about 10% of patients with type 1 diabetes under good metabolic control, whereas more than 40% of patients with type 1 diabetes remain

free of retinopathy despite poor metabolic control. It was suggested that such paradoxical situation might arise due to previous glycemic exposure and BMI (Zhang *et al*, 2001).

Retinopathy progresses through the stages of background, pre-proliferative and proliferative changes. Characteristically, no visual symptoms occur until the macula (area in the center of retina) is affected (diabetic maculopathy) or the newly grown fragile blood vessels in the proliferative stage rupture to cause hemorrhage leading to a considerable loss of vision. Different stages of diabetic retinopathy along with fundoscopy findings and visual symptoms at each stage are given in Table 1 (Kohner *et al*, 1996).

DIABETIC NEPHROPATHY

In an earlier study, 50 diabetic patients were followed for 25 years or more had been described. One-third of them died and in the remaining patients, "ominous signs" of hypertension, azotemia and proteinuria were reported indicating renal involvement in a significant number (Reuting, 1950). Diabetic nephropathy is a gradually progressing renal disease characterized by proteinuria, hypertension and subsequent loss of glomerular function (Group *et al*, 2001). It eventually leads to end-stage renal disease (ESRD), which implies that the kidney has lost the function to filter impurities from the blood to be excreted in urine. ESRD is a devastating medical condition requiring renal replacement therapy in the form of dialysis or kidney transplant for survival. 30-40% patients suffering from type 1 DM and 20-30% patients suffering from type 2 DM normally develop diabetic nephropathy. The risk of deaths in patients with diabetic nephropathy is nine times higher than those without this complication. Five-year survival of diabetic ESRD patients on dialysis has been shown to be poorer (17%) as compared to ESRD patients on dialysis due to glomerulonephritis (40-50%), (Intekhab, 1997). A prospective observational study in 150 diabetic patients with ESRD who began hemodialysis, revealed that better glycemic control was associated with longer survival while poor glycemic control increased the risk of death (Morioka *et al*, 2001). Similarly in another prospective observational study, it has been suggested that aggressive antihypertensive treatment

TABLE 1: DIFFERENT STAGES OF DIABETIC RETINOPATHY ALONG WITH FUNDOSCOPY FINDINGS AND VISUAL SYMPTOMS AT EACH STAGE

STAGE OF RETINOPATHY	RETINAL FINDINGS ON FUNDOSCOPY	VISUAL SYMPTOMS
Back ground	Microaneurysms[1], Hemorrhages, Hard exudates[2] } on a smaller area	No symptoms
Pre-proliferative	Background changes (on a larger area) Dilatation of the blood vessels Venous beading / loops Tortousity of the vessels Soft exudates (cotton-wool spots)[3]	No symptoms
Maculopathy	Background /preproliferative changes encroaching upon the macula Macular edema[4] Macular ischemia[5]	Central vision loss
Proliferative	New growth of blood vessels i. on the disc or ii. elsewhere	No symptoms
Advanced Diabetic Eye Disease	Vitreous hemorrhage Retinal detachment Thrombotic glaucoma	Considerable vision loss[6]

1. A small localized capillary dilatation.
2. Yellowish-white areas of exuded lipoprotein material.
3. Greyish-white areas representing microinfarcts of nerve fibre layer.
4. Detected only on slit-lamp examination.
5. Macula appears very pale.
6. Usually sudden in onset.

can induce long-lasting remission along with improved cardiovascular risk profile in a sizable fraction of type 1 diabetic patients with nephrotic range albuminuria (Hovind *et al*, 2001).

Diabetes causes glomerulosclerosis which leads to progressive renal glomerular damage through a number of stages (Table 2), (Mogensen, 1996). The earliest finding is the functional abnormality along with renal hypertrophy, hyperperfusion and hyperfiltration. The structural abnormality then ensues with glomerular thickening and consequent protein leak. Initially this manifests as microalbuminuria (urinary albumin excretion of 30-300mg/24hr) leading to persistent proteinuria after some years. At later stages, the glomerulus is replaced by hyaline material and the patient progresses to ESRD. A patient may remain at any stage of nephropathy for a variable time period (Kumar and Clark, 1994).

Clinical nephropathy means urinary albumin excretion of more than 300 mg/24hrs, usually manifests 15 years after diagnosis and affects 30-40 % patients diagnosed under the age of 30 years. It is the leading cause of premature death in young diabetic patients while the proportion of affected older individuals is smaller (Kumar and Clark, 1994). Diabetic nephropathy is responsible for a large proportion (25-35%) of all the patients treated for ESRD. Thus it is one of the causes of increased mortality and morbidity with increased health care costs (DERI group, 1995).

DIABETIC NEUROPATHY

It is possibly the commonest microvascular complication of diabetes of all the long-term complications of diabetes, as none affects so many organs or symptoms of human body as the diabetic neuropathy. Aaron and Dereck (2000) focusing on prevalence of diabetic neuropathy recently mentioned that depending upon the use of definition of neuropathy, it has resulted in a wide range of prevalence estimates ranging from almost 0% to 93%. Clinically diabetic neuropathy can be divided into three main types; polyneuropathy, autonomic neuropathy and mononeuropathy (Macleod, 1997).

TABLE 2: STAGES OF PROGRESSIVE DIABETIC NEPHROPATHY

STAGE	FINDINGS
Stage I **(Initial stage)**	Hyperperfusion (increased blood flow to the kidneys) Increased glomerular filtration rate (GFR) Renal Hypertrophy
Stage II **(Microalbumenuria or incipient nephropathy)**	Urinary albumin excretion (UAE), 20- 200 µg/min or 30-300 mg /24hrs
Stage III **(Macroalbumenuria or Clinical albumenuria or Overt nephropathy)**	UAE >200µg /min or >300 mg /24hrs Urea & Creatinine in the blood begin to rise GFR begins to decline
Stage IV **(Advanced clinical nephropathy)**	Proteinuria may reach >3 gram /24 hrs to cause nephrotic syndrome GFR decreases <75 ml /min Blood pressure rises Urea & creatinine levels rise further
Stage V **(End stage renal disease)**	GFR <10 ml /min Serum creatinine >5mg /dl Signs and symptoms of renal failure and uremia (edema, anemia, neuropathies and heart failure)

a. Polyneuropathy affecting the sensory neurons in the lower limbs is the most frequent presentation. The clinical features of peripheral sensory neuropathy are shown in Table 3 (Goran S, 2001). Frequently the symptoms produced by diabetic neuropathy are so distressing that it has considerable adverse effect on quality of life (Benbow *et al*, 1998). Its major clinical significance lies in its ability to cause loss of sensation and subsequent foot ulceration. When combined with peripheral vascular disease and superimposed infection, it may produce the syndrome of "diabetic foot" which may become an indication for amputation leading to serious disabilities and social, economic and rehabilitative implications (Onwuanyi and Lofor, 1999; Kasthuri *et al*, 2000; Andrew, 2001). It is believed that of all the component causes that, when combined, result in ulceration, sensory neuropathy is the commonest (Reiber *et al*, 1999).

b. Autonomic neuropathy may affect both sympathetic and parasympathetic supply of autonomic nervous system. The main clinical presentations include dizziness due to postural hypotension, sexual dysfunction, nausea and vomiting due to gastric dysfunction, urinary problems due to bladder dysfunction and sweating disturbances due to sweat glands dysfunction (Macleod, 1997).

c. In mononeuropathy mode, it mainly involves individual larger nerves e.g. cranial nerves (such as occulomotor or abducent nerve palsies), median nerve in the hands producing carpal tunnel syndrome and lateral cutaneous nerve of the thigh etc. (Macleod, 1997).

The mechanism underlying the abnormalities in diabetic neuropathy is not exactly known. However, available data and clues indicate that both metabolic and vascular changes in the nerve are possible factors. These include increased protein glycation, accumulation of polyols, altered lipid metabolism, decreased myo-inositol content, abnormal Schwan cell function, microangiopathy and lack of neurotrophic factors (Cameron and Coter, 1997). More commonly suggested pathogenic mechanism is hypoxia caused by the occlusion of smaller blood vessels supplying the

TABLE 3: SYMPTOMS AND SIGNS OF PERIPHERAL SENSORY NEUROPATHY[1]

SYMPTOMS	SIGNS
1. Pain (burning, aching, shooting, stabbing)	1. Absent ankle reflexes
2. Paresthesias (pins, needles, tingling)	2. Absent /altered vibration sense
3. Allodynia or hyperasthesia (increased skin sensitivity)	3. Absent /altered pressure sensation
4. Sensory loss or insensitivity	4. Absent /altered pin-prick sensation
5. Swelling of foot and ankle - charcot joints[2] - pseudo-fractures	5. Absent /altered touch perception
6. Planter ulcers	6. Foot deformities - pes cavus - callus formation - claw toes

1. The most frequently and commonly affected site is the lower limb.
2. Loss of sensation in the ankle joint leads to fractures/dislocations after unperceived trauma.

nerve fibers (vasa nervosa) which results in the loss of myelinated and non-myelinated axons with widespread segmental demyelination (Macleod, 1997).

Reviewing the literature, it becomes evident that from being a rarity a century ago, diabetes has grown to become a major cause of mortality in the developed countries and accelerated growth of diabetes constitutes a true epidemic in developing countries (Venkat, 2000). Nevertheless there is a wide range of variations in the prevalence rates of this disease among different countries and populations (Fujimoto, 1996). Clearly, as the disease grows, the prevalence and frequency of its complications also increase further adding to the rates of mortality and morbidity. Given these facts, intense efforts are going on to manipulate and explore different aspects of diabetes and its complications in relation to prevalence, etiology, pathogenesis, risk factors, prevention, treatment and socioeconomic implications on the communities.

The present study was carried out in the Department of Endocrinology of the Postgraduate Medical Institute, Hayatabad Medical Complex, Peshawar, which is a tertiary health care facility of provincial health, services. This is the only unit of its kind in the whole province of N.W.F.P. where a large percentage of diabetic patients with different clinical problems report and get admitted after clinical examination. Most of these patients have long lasting diabetes. Since consolidated data regarding underlying problems in diabetic patients of this area is not available, an attempt was made to assess microvascular complications among these patients with the following main objectives.

1. To find out the prevalence and age related distribution of retinopathy, nephropathy and neuropathy among the diabetic patients.
2. To investigate the correlation of diabetes duration and BMI with the development of retinopathy, nephropathy and neuropathy.
3. To search a possible relationship between hypertension and above mentioned complications.

MATERIALS AND METHODS

The present study was carried out in Endocrinology department of Hayatabad Medical Complex where diabetic and other endocrine patients having a variety of problems are routinely admitted through an out patient clinic and casualty department from various areas of N.W.F.P. The proportion of diabetic patients among these patients is very high. The duration of the study was six months starting from May 2002 to the end of October 2002.

PATIENTS

A total of 200 already diagnosed diabetic patients out of more than 250 consecutive admissions were prospectively selected during the study period. Most of the patients were admitted for metabolic control of their diabetic disease but other causes of admission included foot wounds, skin infections, body aches, chest infections and pre-operative cases. The patients with age ranging between 14 to 75 years included both type 1 and type 2 diabetics. No restriction of the duration was imposed and patients with newly diagnosed diabetes and those with >30 years duration were also included in the study. Age and duration was noted as that reported by the patient. Family history of diabetes was obtained by asking the patient whether any of his/her siblings, parents, cousins, uncles or aunts has or had diabetes.

Patients having age <10 years and >75 years, serious disabling illness, unconsciousness, psychiatric disorder, language/communication problem and repeat admission were excluded from the study. Similarly, patients having bilateral lens opacities (cataracts) were also excluded from ophthalmoscopic examination for retinopathy. Each patient was interviewed and examined after allowing him or her to settle down in the ward bed for a sufficient period of time.

PATIENTS' HISTORY

After noting the anthropometric history (*i.e.* name, age, sex, height and weight), address and type of diabetes, each patient was questioned about the duration

and treatment of diabetes, current symptomatology and previously associated medical conditions like hypertension, coronary artery disease, cerebrovascular accidents, foot ulcers and gangrenes. History of smoking (both current and previous) and the cause of admission were also recorded.

PHYSICAL EXAMINATION

The following examination was performed after taking the patient into confidence.

BODY MASS INDEX (BMI)

BMI is considered as the most linearly related parameter for reflection of the visceral body weight and obesity. Height and weight were measured with a height-weight machine and BMI was calculated as weight in kilograms divided by the square of height in meters (Lonneki *et al*, 2001). Normal BMI ranges from 20 to 25 kg/m^2. Grouping of diabetic patients was carried out on the basis of BMI values obtained.

BLOOD PRESSURE

Blood pressure was measured with the help of a mercury sphygmomanometer. Although the standard definition of hypertension is a blood pressure of more than 140/90 mmHg, evidence from clinical trails in diabetic patients suggests clinically significant benefits in outcomes with reduction in blood pressure below 140 or even further below 130 mmHg systolic and 80 mmHg diastolic (Carlos *et al*, 2002). In the present study, therefore, hypertension was defined as patients already taking antihypertensives or having systolic BP of >130 or diastolic BP of >80 mm Hg at the time of admission to the hospital.

PERIPHERAL PULSES

Poor blood circulation in the peripheral arteries is an indication of peripheral vascular disease. Pulsations of dorsalis pedis and posterior tibial arteries in the feet were felt and absence of any one of them was considered abnormal.

FUNDOSCOPIC EXAMINATION

Fundus is the area of retina under vision during direct ophthalmoscopic examination of the eye. Presence of retinopathy was assessed through dilated pupils with the help of a "Keeler" ophthalmoscope. Retinopathy was diagnosed if at least one microaneurysm, hemorrhage or exudate in at least one eye was observed. The condition was classified as absent, non-proliferative diabetic retinopathy or proliferative diabetic retinopathy (Aiello *et al*, 1998). If needed, the patient was sent to ophthalmologist for confirmation of retinopathy. If the patient had bilateral lens opacities, this examination was ignored and the patient was labeled as having bilateral cataracts.

NEUROLOGICAL EXAMINATION

Diabetic neuropathy may involve any part of the nervous system but peripheral sensory neurons in the feet and legs are the first and more severely affected. If a patient has both symptoms and signs of peripheral neuropathy, there is no doubt about the diagnosis of clinical peripheral neuropathy. The most frequent form of peripheral neuropathy is, however, asymptomatic type *i.e.* no symptoms but with positive signs of peripheral neuropathy (Table 3). On the other hand, diabetic patients with neurological symptoms do not always show signs of peripheral neuropathy. Assessment of sensory polyneuropathy was performed as below according to the method mentioned by Goran S (2001).

1. Ankle jerk

The presence or absence of ankle reflexes on both sides was noted using a tendon hammer. Reinforcement method was used to elicit the response when required.

2. Vibration sense

Vibration sense on lateral and medial malleoli of the ankle joint, first metatarsal head and big toe was assessed on both sides using a 128Hz tuning fork. Patient was asked whether he/she could perceive the buzzing sensation produced by

vibrating tuning fork on all the above sites. Failure to perceive on one or both sides was considered abnormal.

3. Pain

This was checked by pinprick. A clean common paper-pin was used to ask the patient if it was painful or not.

4. Touch

A piece of cotton or a small soft brush was used to ask the patient if he/she appreciated it or not.

5. Altered Sensation

Any reduced or abnormal sensation in the above examination was considered as altered sensation.

Evidence of neuropathy was obtained on the basis of at least one objective sign including absence of both ankle reflexes, absence of vibration sense, touch or pain on any side, with or without symptoms (Emmanuel *et al*, 2001).

NEPHROPATHIC EXAMINATION

Biochemical analysis of total proteins in 24hr urinary collection was used for nephropathic assessment while renal functions were assessed by estimation of urea and creatinine in the serum.

LABORATORY TESTS

The following tests were performed in the Pathology Department of this hospital.

1. Plasma Glucose: Plasma glucose at the time of admission as well as casual plasma glucose at subsequent days were used for overall assessment of glycemic status, but glucose levels taken at the time of admission were used as reference value.

Target levels of plasma glucose in diabetic patients are (Shera, 1999);

Ideal:	70-105 mg/dl (Fasting)	and	80-140 mg/dl (2hr post meal)
Acceptable:	80-120 mg/dl (Fasting)	and	80-160 mg/dl (2hr post meal)

2. 24 Hour Urine Protein: This was used for the assessment of nephropathy. Although albumin/creatinine ratio (ACR) and microalbuminuria are considered as the standard markers for incipient nephropathy, the conventional parameter of 24hr proteinuria was brought to application presently because of the general non-availability of the above tests in the local laboratories. The normal level of 24hr urine protein, according to Mogyorosi and Fuad (2000), is <150 mg but due to the effect of a variety of physiological and pathological factors such as posture, exercise, fever, infections etc, the values may be variable. A value of 200 mg/24hr proteinuria was considered as the cut-off value for detection of nephropathy, which is equivalent to ACR of 0.2 mg protein/mg of creatinine (Fairley, 2000).

3. Serum Urea and

4. Serum Creatinine: These parameters were used for assessment of renal function. Normal ranges of urea and creatinine were taken as 10-50 mg/dl and 0.5-1.1 mg/dl respectively according to the laboratory kits used.

SAMPLING

Blood

i. A separate laboratory request form for each patient was prepared mentioning the date, patient's name, age, sex, bed number, ward along with the request for plasma glucose, urea and creatinine.

ii. 3 ml of blood was taken from each patient in a 5ml disposable syringe using ante-cubical vein, on first day of admission.

iii. The syringe was labeled with appropriate particulars of the patient along with name of tests.

iv. After collection of samples from all newly admitted patients for each particular day, they were immediately sent to the laboratory on the same day.

Urine

i. A separate laboratory request form for each patient was prepared mentioning the date, patient's name, age, sex, bed number, and ward name along with request for 24-hour urinary protein.

ii. 24-hour urinary collection was obtained from each patient in an appropriately labeled clean plastic jar after giving him/her the following instructions.

a. To empty bladder by passing and discarding urine at 8.00 A.M.

b. To start collecting urine in the jar each time he/she passed urine thereafter, for the whole day and night.

c. The last installment to be added to the jar at 8.00 A.M. next day.

iii. After closing each jar tightly with its lid, the collected samples were sent to the laboratory on the same day.

PROCESSING

1. The blood from syringes was transferred to appropriately labeled centrifuge test tubes.
2. The tubes were centrifuged for 5 minutes in a centrifuge machine to separate the serum.
3. Each serum sample was then preserved and put in a tube-rack for further use.
4. Volume of 24-hour samples of urine was determined by pouring it into a graduated container and a 5 ml sample was preserved in a separate test tube for further use.

ANALYSIS

Plasma Glucose

Method: A laboratory reagent kit of "Elitech Diagnostics" was used for estimation of glucose levels in the plasma. Enzymatic colorimetric method was used according to manufacturer's instructions. Using the "Photometer 4010" the following steps were carried out.

	BLANK	STANDARD	SAMPLE
Working reagent	1mL	1mL	1 mL
Distilled water	10 µL	--	--
Standard	--	10µL	--
Sample	--	--	10µL

1. Using appropriate justers, solutions of blank, standard and samples were prepared in separate test tubes according to above specifications.
2. All the solutions were incubated at 37° C for 10 minutes.
3. The wavelength was set at 546 nm.
4. After making zero adjustment with reagent blank, optical density of standard solution was determined.
5. Similarly the optical density of each test solution was found one by one.
6. Finally concentration of glucose in each sample was determined by the following equation. OD of Sample / OD of standard x n
   where n = standard concentration of glucose = 100mg/dl

24 Hour Urine protein

Method: The method used was the Pyrogallol-Red Method. The laboratory reagent kit used for this purpose was "Ultra-Sensitive Proteins Fluitest, USP" from "Biocon Diagnostik, Germany". Using the " Photometer 4010" the following steps were carried out.

	BLANK	STANDARD	SAMPLE
Reagent	3 ml	3ml	3 ml
Standard	---	50 µL	---
Sample	---	---	50 µL

1. Using appropriate justers, solutions of blank, standard and samples were prepared in separate test tubes according to above specifications.
2. All the solutions were incubated at 37° C for 10 minutes.

3. The wavelength was set at 604 nm.
4. After making zero adjustment with reagent blank, optical density of standard solution was determined.
5. Similarly the optical density of each test solution was found one by one.
6. Concentration of protein in each sample was determined by the following equation. OD of Sample / OD of standard x n
   where n = standard concentration of protein = 100mg/dl
7. Finally, concentration of protein in 24 hr urine was determined by working out the equation. Protein conc./ dl ÷ 100 x total volume of urine in 24 hours.

Serum Urea

Method: Serum urea was estimated by urease-glutamate dehydrogenase (GLDH) method using reagent kit of "Human Diagnostics, Germany". Using the " Photometer 4010" the following steps were carried out.

	BLANK	STANDARD	SAMPLE
Working reragent	1 mL	1 mL	1 mL
Distilled water	10 µL	--	--
Standard	--	10 µL	--
Sample	--	--	10 µL

1. Using appropriate justers, solutions of blank, standard and samples were prepared in separate test tubes according to above specifications.
2. All the solutions were incubated at 37° C for 5 minutes.
3. The wavelength was set at 340 nm (K 20).
4. After making zero adjustment with reagent blank, optical density of standard solution was determined.
5. Similarly the optical density of each test solution was found one by one.
6. Finally, concentration of urea in each sample was determined by the following equation. OD of Sample / OD of standard x n
   where n = standard concentration of urea = 50mg / dl

Serum Creatinine

Serum creatinine estimation was carried out by alkaline picrate method using reagent kit of "RANDOX Laboratories Ltd". Using the " Photometer 4010" the following steps were carried out.

	BLANK	STANDARD	SAMPLE
Working reagent	1mL	1mL	1 mL
Distilled water	100 µL	--	--
Standard	--	100µL	--
Sample	--	--	100µL

1. Using appropriate justers, solutions of blank, standard and samples were prepared in separate test tubes according to above specifications.
2. All the solutions were incubated at 37° C for 10 minutes.
3. The wavelength was set at 546 nm.
4. After making zero adjustment with reagent blank, optical density of standard solution was found out.
5. Similarly the optical density of each test solution was found one by one.
6. Finally, concentration of creatinine in each sample was determined by the following equation. OD of Sample / OD of standard x n
   where n = standard concentration = 1mg / dl

RENAL ULTRASONOGRAPHY

Renal ultrasound of a group of patients having nephropathic range proteinuria was performed in the Radiology Department for detection of renal size and morphology. The kidney may increase in size during initial stages of nephropathy while in the final stage it may become smaller in size.

DATA ANALYSIS

All relevant data were entered into a pre-designed proforma. After scrutinizing more than 250 consecutive cases, 200 cases were selected for study who

met the inclusion criteria. A database was created in the DOS based statistical program "EPI info version 6.4" and data analysis was carried out by application of relevant tests. Means, standard deviations and standard errors for various characteristics were calculated.

$$\text{Mean } (\bar{x}) = \Sigma x / n$$

where Σ = sum, x = element value, n = total number of elements.

$$\text{Standard deviation (SD)} = \sqrt{\Sigma (x - \bar{x})^2 / n}$$

where Σ = sum, $x - \bar{x}$ = deviation score (element value minus mean), n = total number of elements.

$$\text{Standard error (SE)} = SD / \sqrt{n}$$

P value determination was carried out either by finding 't' statistics using student's 't' test for the difference between two variables or by using chi-square test to evaluate differences between different groups of diabetes duration, age, BMI and hypertensive / non-hypertensive cases . The 't' value expresses the number of standard errors by which the sample mean lies above or below the hypothesized mean.

$$t = \bar{x}_1 - \bar{x}\,2 \div \sqrt{(SD_1)^2 / n_1 + (SD_2)^2 / n_2}$$

where $\bar{x}_1$= mean of one sample, $\bar{x}_2$= mean of second sample, SD_1= standard deviation of one sample, SD_2= standard deviation of second sample, n_1= number of elements in one sample, n_2= number of elements in the second sample. Degrees of freedom (DF) are caculated by the formula

$$DF = n_1 + n_2 - 2$$

The 't' value is then compared with critical value of 't' corresponding for the degrees of freedom and level of probability in the prescribed 't' tables.

Chi-square, basically a test of proportions, shows whether the proportions of observations falling in different categories differ significantly from the proportions that would be expected by chance.

$$\text{Chi- square } (\chi^2) = (O - E)^2 / E$$

where O = observed frequency and E = expected frequency.

It is then compared with critical value corresponding for the required degrees of freedom and level of probability in the prescribed Chi–square tables. In Yate's correction, (O – E) values are reduced by 0.5 before calculating χ^2.

Most of the above statistics were reproduced through computer in statistical program " EPI info" by appropriate commands. Differences were considered significant at $P < 0.1$. Results were expressed as the mean ± SD/SE for quantitative variables and as percent proportions for categorical findings.

RESULTS

PATIENTS' CHARACTERISTICS

The total number of patients studied was 200. They were already diagnosed diabetic patients with male to female ratio of 1:2 (34.5% and 65.5% respectively). At presentation, the mean age of the study group was 48.3±13.3 years with ages between 14 and 75 years. Mean height and weight were 1.57±0.09 meters and 65.2±13.4 kg respectively with heights ranging from 1.40 to 1.81 meters and weights ranging from 27 to 100 kg. 32 out of 200 patients presented as type 1 DM and 168 were identified as type 2 DM. BMI ranged from 12.3 to 31.6 kg/m^2 for type 1 DM (21.3± 5.6) and from 15.1 to 42.9 kg/m^2 for type 2 DM (27.3±4.7). Duration of diabetic diagnosis ranged from newly diagnosed to more than thirty years with a mean duration of 8.5± 5.6 years. 46% patients had a positive family history of diabetes. 8.5% patients were found as being smokers. Clinically, on measurement of blood pressures (BP), it was found that mean systolic BP ranged from 70 to 210 mm Hg (131 ± 25 mm Hg) while diastolic BP ranged from 30 to 110 mm Hg (79 ± 13 mm Hg). A total of 53.5% patients were recorded as hypertensives of which 31.5% had an already diagnosed hypertension while in the remaining 22% patients, high blood pressure was detected for the first time. Blood glucose measured at the time of admission ranged from 34 to 583 mg/dl with a mean value of 331±111. The above general and clinical characteristics are represented in Table 4.

PREVALENCE OF MICROVASCULAR COMPLICATIONS

Determination of the prevalence of micro-vascular complications *i.e.* retinopathy, nephropathy and neuropathy was one of the main objectives of the present study. Retinopathy was found in 51% cases with a male to female ratio of ~1:2. Nephropathy was found to have a prevalence rate of 56.5%, male to female ratio being ~1:2. Maximum prevalence was noted in case of neuropathy, which was 68.5% with a male to female ratio of 1:2. Fifteen patients who had bilateral lens opacities (cataracts) required surgery and retinopathy was therefore not possible to be assessed among

them. By excluding these 15 patients from total number of patients for the purpose of retinopathy, the prevalence of retinopathy was 102 out of 185 patients *i.e.*55.2 % (Table 5). Since the microangiopathies mostly have a common underlying pathological mechanism, they frequently occur simultaneously, hence their presence in different possible combinations was also assessed. All the three complications occurred in 30.5% patients. The percentage of patients affected with only one complication (either retinopathy or nephropathy or neuropathy) was 19.5%. Neuropathy and nephropathy were detected in 9% and 8% cases respectively and retinopathy alone was detected in only 2.5% cases. The proportion of patients having two complications was 32.5%. The association between nephropathy and neuropathy and also between retinopathy and neuropathy occurred in 14.5% cases. However combination of retinopathy and nephropathy was unexpectedly less frequent and was only 3.5%. The patients without any complication represented 17.5% of the study population (Table 6).

DURATION OF DIABETES AND MICROVASCULAR COMPLICATIONS

To assess a possible relationship between duration of diabetes and the prevalence of retinopathy, nephropathy and neuropathy, the patients were categorized into five groups; **1**= ≤5.0 years, **2**= 5.1-10 years, **3**= 10.1-15 years, **4**= 15.1-20 years, **5**= >20 years (Table 7). Distribution of complications was studied in all five groups. The number of patients was maximum in group 1 *i.e.* 66 while the number of patients in group 5 was minimal. The positive relationship of retinopathy, nephropathy and neuropathy was significantly associated with increasing duration of diabetes ($p < 0.001$, < 0.01 and < 0.001 respectively). As expected, least proportions of complications (16.66% for retinopathy, 37.88% for nephropathy and 36.36% for neuropathy) were observed in the ≤5 year group and highest proportions (88.89% for retinopathy, 77.78% for nephropathy and 83.33% for neuropathy) were observed in the 15.1-20 year group (Table 7). For >20 year duration group, the resultant proportions were almost 100% but it cannot be considered conclusive since the number of patients in that group was very small *i.e.* only 3. Comparing mean values of diabetes duration between patients with and without retinopathy, nephropathy or

neuropathy, the values were higher in every case for the former group (10.9 vs 5.1 year, 10.0 vs 6.7 years and 10.4 vs 4.5 years respectively) the results being highly significant, $p = 0.001$ (Table 11). Similarly by comparing the mean values in patients having any one or more complications (9.6±5.3 years) with those having no complication (3.3±2.1 years), there was a highly significant statistical difference, $p = 0.001$ (Table 12). Male to female ratio was approximately 1:2 for each duration group. Figure 1 shows relationship between diabetes duration and the three complications.

BODY MASS INDEX AND MICROVASCULAR COMPLICATIONS

To determine relationship between body mass index (BMI) and the development of retinopathy, nephropathy and neuropathy, the patients were divided into six groups taking BMI (in kg/m^2) into consideration. **1**= <20, **2**= 20-24.9, **3**= 25-29.9, **4**= 30-34.9, **5**= 35-39.9, **6**= ≥40 (Table 8). Distribution of complications was assessed in all six groups. Maximum number of patients *i.e.* 36% (n=72) fell in group 3 indicating "mild obesity". Only three patients fell in group 6 indicating "severe or morbid obesity". It can be observed from Table 8 that as BMI increases, the proportion of patients having retinopathy, nephropathy and neuropathy also increase until a paradoxical decline (33.33%) in highest BMI group. The results were significant for retinopathy and neuropathy *($p < 0.01$)* but non-significant for nephropathy *($p = 0.664$)*. The mean values of BMI in patients with either retinopathy or neuropathy were higher as compared to those patients who were without that complication (26.5 vs 26.0 and 27.1 vs 26.9 respectively) although the difference did not reach statistical significance, $p = 0.1$ (Table 11). Mean BMI values in patients with and without nephropathy were almost equal (26.4 vs 26.5). Similarly when mean BMI values were compared in patients having any one or more complications (26.9±3.9 kg/m^2) with those having no complication (24.3±3.3 kg/m^2), a highly significant statistical difference was observed with $p = 0.001$ (Table 12). Male to female ratio in different groups for any complication remained approximately 1:2. Figure 2 shows a relationship pattern between body mass index and retinopathy, nephropathy and neuropathy.

AGE AND MICROVASCULAR COMPLICATIONS

To determine age-wise distribution of retinopathy, nephropathy and neuropathy in the presently studied patients, the patients were grouped on the basis of age presented at the time of admission; 1= 10-19 years, 2= 20-29 years, 3= 30-39 years, 4= 40-49 years, 5= 50-59 years, 6= 60-69 years, 7= >70 years (Table 9). Highest number of patients, 33.5% fell in group 5 while group 1 contained least number of patients (3.5%). Distribution of complications was studied in all seven groups. As shown in Table 9, it was observed that as age increased, the proportion of patients having complications also increased even upto >80% in 60-70 years age group and above. The results were statistically significant ($p < 0.01$) for retinopathy and nephropathy and highly significant ($p < 0.001$) for neuropathy. The mean age in patients with either retinopathy or nephropathy or neuropathy was higher than those without these complications (51.3 vs 42.8, 51.1 vs 44.6 and 51.8 vs 40.6 years respectively), the difference being highly significant with $p < 0.001$ (Table 11). Similarly comparing mean age in patients having any one or more complications (50.6 ± 12.8 yrs) with those having no complication (37.3 ± 7.4 yrs), the result showed a statistically highly significant difference, $p < 0.001$ (Table 12). Overall ratio of male and female distribution again remained 1:2. The relationship between age and the three complications is shown in Figure 3.

HYPERTENSION AND MICROVASCULAR COMPLICATIONS:

Hypertension was observed in 53.5% cases out of which 22% cases were newly diagnosed at the time of admission. Hypertensive and non-hypertensive patients were compared for retinopathy, nephropathy and neuropathy. The results showed that prevalence of retinopathy was 57.94% in hypertensive patients and 43.01% in non-hypertensive patients. Nephropathy prevailed in 58.88% cases among hypertensive patients and in 53.76% cases among normotensive patients. Neuropathy occurred in 76.64% cases in patients having hypertension and in 59.14% in those having normal blood pressure (Table 10). The results were statistically significant in case of retinopathy ($p < 0.01$) and neuropathy ($p < 0.05$) and non-significant in case of nephropathy ($p = 0.558$). The proportion of hypertensive cases among patients having

either retinopathy or nephropathy or neuropathy was greater than among those who did not have that complication (60.78 vs 35.71%, 55.75 vs 50.57%, and 59.85 vs 39.68% respectively) (Table 11). Similarly there were 56.35% cases of hypertension in patients having any one or more complications as compared to 40% cases in those who had no complication. Statistical analysis revealed highly significant difference ($p < 0.001$) (Table 12). Figure 4 represents relationship between hypertension and the three complications.

RENAL FUNCTION TESTS

Renal functions of the patients were assessed by serum creatinine and urea. Serum urea ranged from 14 to 80 mg/dl with a mean value of 29.9 ± 12.6 and serum creatinine ranged from 0.4 to 1.1 mg/dl with a mean value of 0.73 ± 0.19 in patients without nephropathy. In contrast, mean serum urea level was 38.7 ± 25.4 (range=12-180 mg/dl) and mean serum creatinine level was 0.94 ± 0.53 (range= 0.4-3.9 mg/dl) in patients with nephropathy ($p < 0.01$ *and* < 0.05 respectively). Mean proteinuria level in 24hr was 102 ± 48 (range=21-200mg) in patients without nephropathy and 945 ± 1109 (range= 207-6200mg) in patients with nephropathy, $p < 0.001$ (Table 13).

RENAL ULTRASONOGRAPHY

Renal ultrasonography for assessment of renal morphology was performed in 43 patients having proteinuria in nephropathic range. The result showed abnormal increase in the size of kidney in only two patients. In the remaining forty one patients, ultrasound examination showed normal results.

CONCURRENCE OF MACROANGIOPATHIES

Coronary artery disease was found in 9% patients, while history of cerebro-vascular disease was positive in only 2.5% patients. Peripheral vascular disease as defined by absence of pulsations in dorsalis pedis and posterior tibial arteries of the foot, was recorded in 4% patients while lower limb amputations (including loss of leg, foot or a part of foot) were found in 7% cases (Table 14).

SEX DISTRIBUTION

The ratio of male to female in the subjects studied was nearly 1:2 (absolute values, M:F= 69:131) in the study subjects. The same ratio was reflected proportionately in case of every type of complication whether alone or in combination. This pattern was also observed for different groups formed on the bases of age, duration of diabetes and body mass index and also when these were compared among themselves in relation with retinopathy, nephropathy or neuropathy (Tables 7,8 & 9). On the other hand, in case of macroangiopathies, female patients showed slight preponderance over male patients as compared to microangiopathies (Table 14).

TABLE 4: GENERAL AND CLINICAL CHARACTERISTICS OF THE STUDY POPULATION

CHARACTER	NUMBER (N)	PERCENT -AGE(%)	RANGE	MEAN±SD
Number	200	--	--	--
Male	69	34.5	--	--
Female	131	65.5	--	--
Age (years)	--	--	14-75	48.3±13.3
Height (meters)	--	--	1.40-1.81	1.57±0.09
Weight (kgs)	--	--	27-100	65.2±13.4
BMI (kg/m^2)				
•Overall	--	--	12.3-42.9	26.4±5.4
• For type 1	--	--	12.3-31.6	21.3±5.6
• For type 2	--	--	15.1-42.9	27.3±4.7
Patients with Type 1 DM	32	16	--	--
Patients with Type 2 DM	168	84	--	--
Duration of Diabetes (years)	--	--	0.1-32.0	8.5±5.6
Positive family history	92	46	--	--
Smokers	17	8.5	--	--
Systolic BP (mm Hg)	--	--	70-210	131±25
Diastolic BP (mm Hg)	--	--	30-110	79±13
Hypertension				
• before admission	63	31.5	--	--
• on admission	44	22	--	--
•Total	107	53.5	--	--
Plasma Glucose (mg/dl)	--	--	34-583	331±111

TABLE 5: PREVALENCE OF MICROVASCULAR COMPLICATIONS

COMPLICATION	N	M:F	%age
Retinopathy	102	38:64	51%
Nephropathy	113	39:74	56.5%
Neuropathy	137	45:92	68.5%
Bilateral cataracts	15	4:11	7.5%

TABLE 6: DIFFERENT COMBINATIONS OF RETINOPATHY, NEPHROPATHYAND NEUROPATHY

PATIENT GROUP	COMPLICATIONS IN COMBINATION	N	% age
1. With All three complications	Retinopathy+ Nephropathy+ Neuropathy	61	30.5%
2. With Two complications	Retinopathy+Nephropathy Retinopathy+Neuropathy Nephropathy+Neuropathy Total	7 29 29 =65	32.5%
3. WithOnly one complication	Only Retinopathy Only Nephropathy Only Neuropathy Total	5 16 18 =39	19.5%
4. Without any complication	No retinopathy No nephropathy No neuropathy	35	17.5%
TOTAL		200	100%

TABLE 7: MICROVASCULAR COMPLICATIONS IN DIFFERENT DIABETES DURATION GROUPS

DURATION (yrs)	NO. OF PTS.	RETINOPATHY[1]			NEPHROPATHY[2]			NEUROATHY[3]		
		N	M:F	%	N	M:F	%	N	M:F	%
1. ≤5.0	66	11	4:7	**16.66**	25	8:17	**37.88**	24	6:18	**36.36**
2. 5.1-10	64	42	14:28	**65.62**	38	12:26	**59.38**	50	18:32	**78.13**
3. 10.1-15	49	31	12:19	**63.26**	33	13:20	**67.35**	45	13:32	**91.84**
4. 15.1-20	18	16	7:9	**88.89**	14	4:10	**77.78**	15	6:9	**83.33**
5. >20.0	3	2	1:1	**66.67**	3	2:1	**100**	3	2:1	**100**
TOTAL	**200**	**102**	**38:64**	--	**113**	**39:74**	--	**137**	**45:92**	--

1. $p = \sim 0$ (χ^2=72.46)
2. $p = 0.0036$ (χ^2=17.50)
3. $p = \sim 0$ (χ^2=49.92)

TABLE 8: MICROVASCULAR COMPLICATIONS IN DIFFERENT BMI GROUPS

BMI (Kg/m^2)	NO. OF PTS.	RETINOPATHY[1]			NEPHROPATHY[2]			NEUROPATHY[3]		
		N	M:F	%	N	M:F	%	N	M:F	%
1. *<20*	22	8	4:4	**36.36**	12	5:7	**54.54**	9	3:6	**40.90**
2. *20-24.9*	61	29	10:19	**47.54**	32	11/21	**52.45**	37	12:25	**60.66**
3. *25-29.9*	72	45	20:25	**62.50**	46	19:27	**63.89**	56	23:33	**77.78**
4. *30-34.9*	30	15	3:12	**50.00**	14	3:11	**46.67**	25	5:18	**83.33**
5. *35-39.9*	12	5	1:4	**41.67**	7	1:6	**58.33**	9	2:7	**75.00**
6. *≥40*	3	0	--	--	2	0:2	**66.67**	1	0:1	**33.33**
TOTAL	**200**	**102**	**38:64**		**113**	**39:74**		**137**	**45:92**	

1. $p = 0.0065$ $(\chi^2 = 27.50)$
2. $p = 0.644$ $(\chi^2 = 4.24)$
3. $p = 0.0057$ $(\chi^2 = 18.20)$

TABLE 9: MICROVASCULAR COMPLICATIONS IN DIFFERENT AGE GROUPS

AGE (years)	NO. OF PTS.	RETINOPATHY[1]			NEPHROPATHY[2]			NEUROPATHY[3]		
		N	M:F	%	N	M:F	%	N	M:F	%
1. *10-19*	7	1	1:0	**14.28**	3	2:1	**42.86**	1	1:0	**14.28**
2. *20-29*	16	4	2:2	**25.00**	7	3:4	**43.75**	4	1:3	**25.00**
3. *30-39*	17	7	2:5	**41.18**	4	2:2	**23.53**	12	4:8	**70.58**
4. *40-49*	50	24	9:17	**48.00**	25	8:17	**50.00**	32	10:22	**64.00**
5. *50-59*	67	41	14:25	**61.20**	46	13:33	**68.66**	51	16:35	**76.12**
6. *60-69*	32	19	6:13	**59.37**	19	7:12	**59.37**	28	9:19	**87.50**
7. *≥70*	11	6	4:2	**54.55**	9	4:5	**81.82**	9	4:5	**81.82**
TOTAL	**200**	**102**	**38:64**	--	**113**	**39:74**	--	**137**	**45:92**	--

1. $p = 0.0013$ $(\chi^2 = 31.97)$
2. $p = 0.0093$ $(\chi^2 = 16.97)$
3. $p = 0.00015$ $(\chi^2 = 32.13)$

TABLE 10: MICROVASCULAR COMPLICATIONS IN HYPERTENSIVE AND NON-HYPERTENSIVE PATIENTS

CONDITION	NO. OF PTS.	RETINOPATHY[1]			NEPHROPATHY[2]			NEUROPATHY[3]		
		N	M:F	%	N	M:F	%	N	M:F	%
With Hyper tension	107	62	25:37	57.94	63	24:39	58.88	82	27:55	76.64
Without Hyper-tension	93	40	13:27	43.01	50	15:35	53.76	55	18:37	59.14
TOTAL	200	102	38:64	--	113	39:74	--	137	45:92	--

1. $p = 0.023$ $(\chi^2 = 7.50)$
2. $p = 0.558$ $(\chi^2 = 0.34)$
3. $p = 0.012$ $(\chi^2 = 6.27)$

TABLE 11: COMPARISON OF MEANS OF AGE, BMI, DIABETES DURATION AND HYPERTENSIVE CASES BETWEEN PATIENTS WITH AND WITHOUT RETINOPATHY, NEPHROPATHY OR NEUROPATHY

	RETINOPATHY*		NEPHROPATHY*		NEUROPATHY*	
	ABSENT	PRESENT	ABSENT	PRESENT	ABSENT	PRESENT
AGE mean±SD (yrs.)	42.8±14.1	51.3±11.4	44.6±13.7	51.1±12.4	40.6±14.4	51.8±11.2
BMI mean±SD (Kg/m^2)	26.0±6.3	26.5±4.7	26.4±5.5	26.5±5.3	26.9±5.9	27.1±4.9
DIABETES DURATION mean±SD (Yrs)	5.1±4.8	10.9±4.9	6.7±5.0	10.0±5.7	4.5±4.5	10.4±5.1
HTN (No./out of)	35/98 (35.71%)	62/102 (60.78%)	44/87 (50.57%)	63/113 (55.75%)	25/63 (39.68%)	82/137 (59.85%)

* $p < 0.001$ (highly significant) for age, diabetes duration and hypertension when compared for present and absent state of each complication but not for BMI results having yielded non- significant difference ($p > 0.1$) for all the three complications.

TABLE 12: COMPARISON OF MEANS OF AGE, BMI, DIABETES DURATION AND HYPERTENSIVE CASES BETWEEN PATIENTS HAVING SOME COMPLICATION AND THOSE HAVING NO COMPLICATION

GROUP	N	AGE[1] (mean±SD) yrs	BMI[2] (mean±SD) kg/m^2	DIABETES DURATION[3] (mean±SD) yrs	HYPER TENSION[4] N (%)
WITH SOME COMPLICATION	165	50.6±12.8	26.9±3.9	9.6±5.3	93 (56.35%)
WITHOUT ANY COMPLICATION	35	37.3±7.4	24.3±3.3	3.3±2.1	14 (40%)

1. $p < 0.001$ 2. $p < 0.05$ 3. $p < 0.001$ 4. $p < 0.001$

TABLE 13: RENAL FUNCTION TESTS AND PROTEINURIA IN NEPHROPATHIC AND NON-NEPHROPATHIC SUBJECTS

TEST	WITH NEPHROPATHY			WITHOUT NEPHROPATHY			*p*-value
	Range	Mean±SD	SE	Range	Mean±SD	SE	
Blood Urea (mg/dl)	12-180	38.7±25.4	2.39	14-80	29.9±12.6	1.34	*<0.01*
Serum Creatinine (mg/dl)	0.4-3.9	0.94±0.53	0.05	0.4-1.1	0.73±0.19	0.02	*<0.05*
24Hr Proteinuria (mg)	207-6200	945±1109	104	21-200	102±48	5	*<0.001*

TABLE 14: CONCURRENCE OF MACROANGIOPATHIES

CONDITION	N	M:F	%
Coronary artery disease	18	4:14	9
Cerebro-vascular disease	5	1:4	2.5
Peripheral vascular disease	8	3:5	4
Amputations	14	4:10	7

FIGURE 1: RELATIONSHIP BETWEEN DIABETES DURATION AND MICROVASCULAR COMPLICATIONS

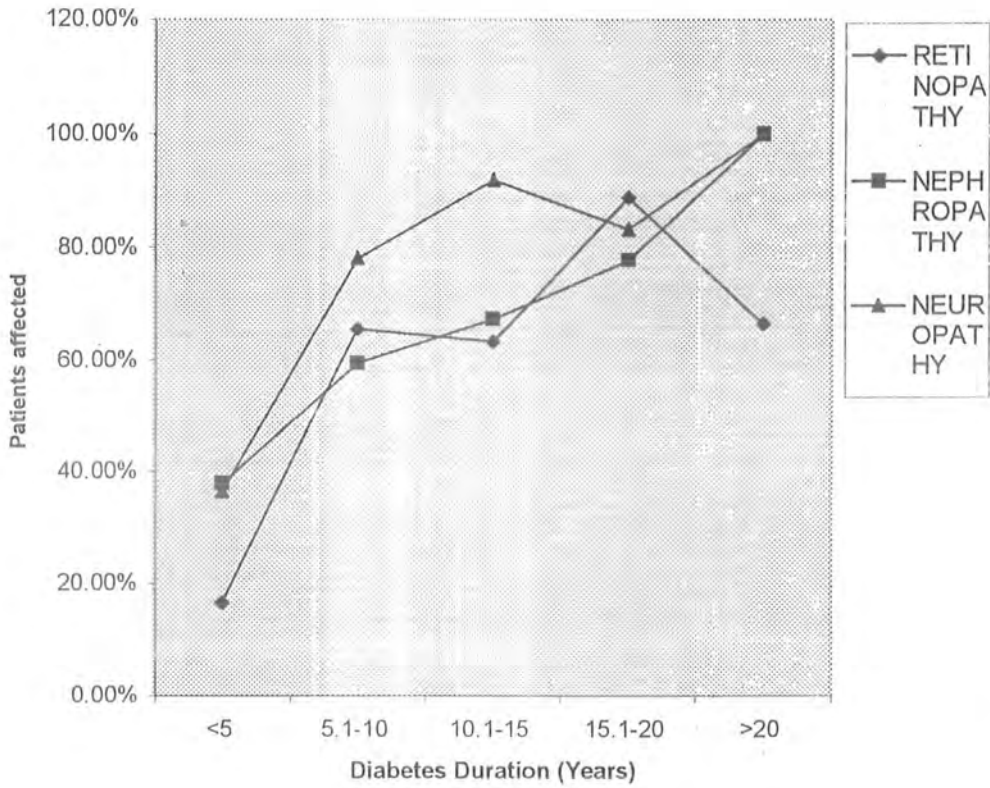


FIGURE 2: RELATIONSHIP BETWEEN BMI AND MICROVASCULAR COMPLICATIONS

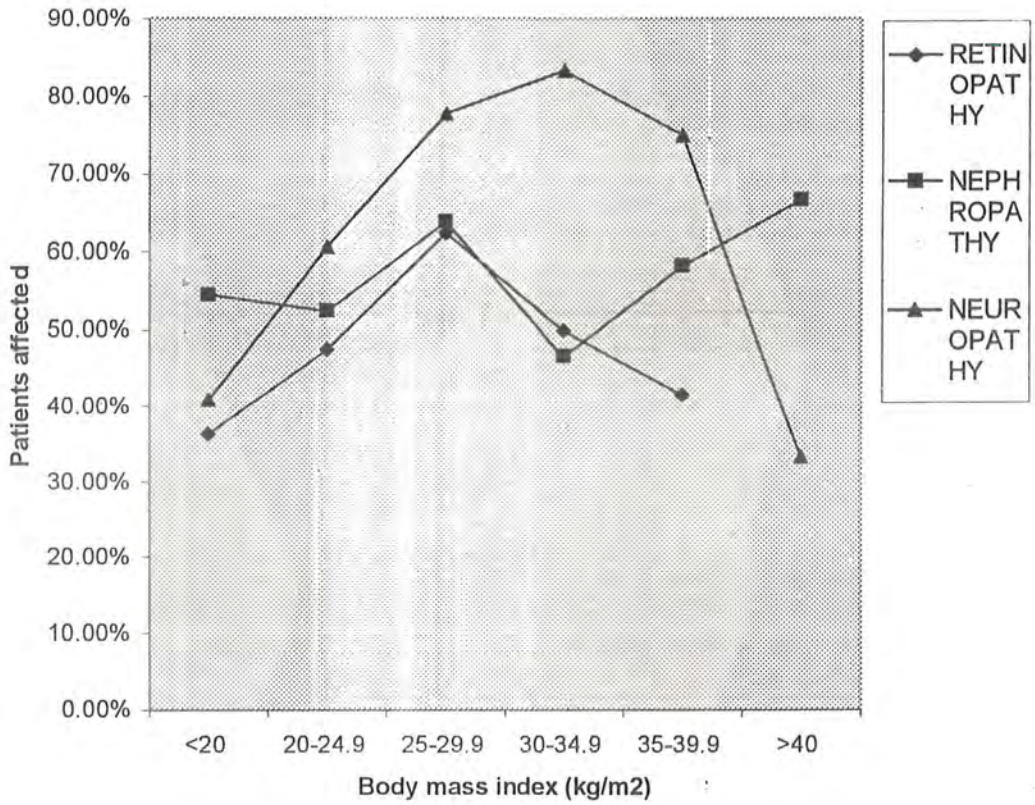


FIGURE 3: RELATIONSHIP BETWEEN AGE AND MICROVASCULAR COMPLICATIONS

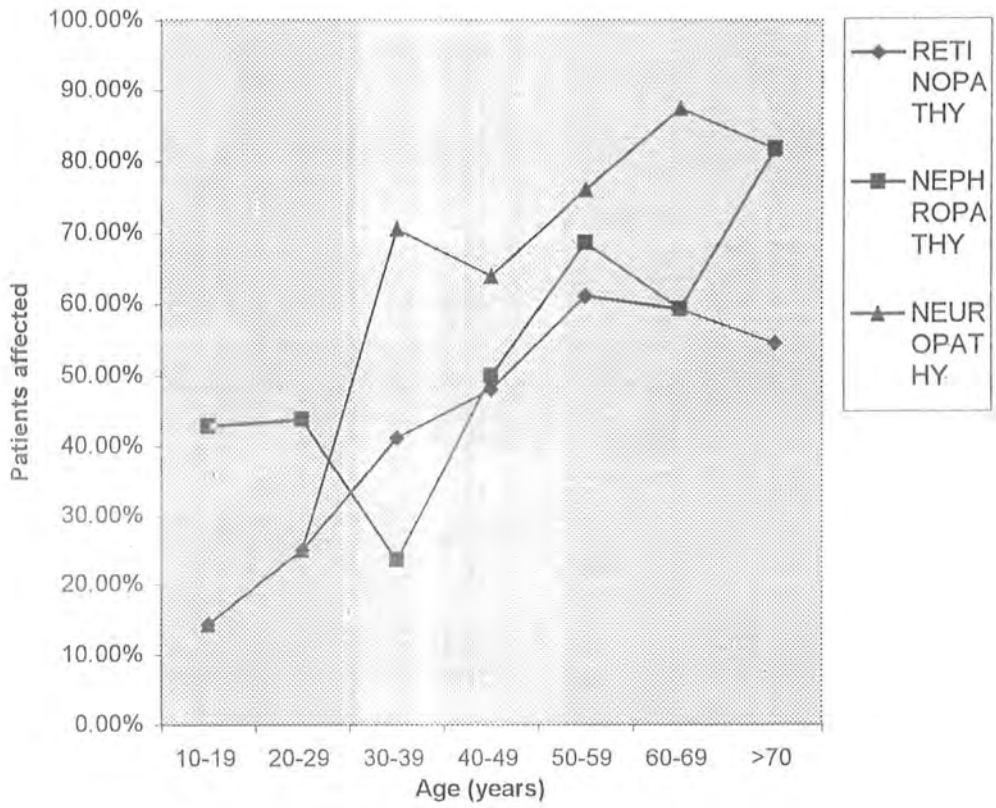
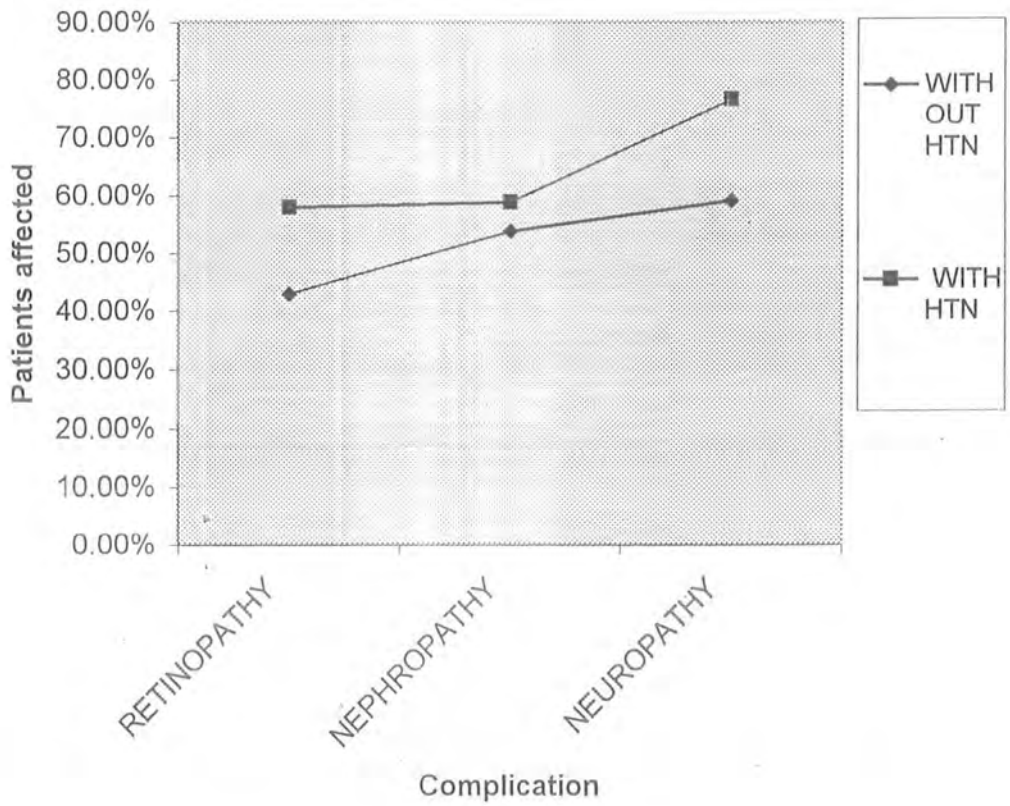


FIGURE 4: RELATIONSHIP BETWEEN HYPERTENSION AND MICROVASCULAR COMPLICATIONS



DISCUSSION

Diabetes mellitus is usually an irreversible disease and although patients can reasonably have a normal life-style, complications that develop later result in reduced life expectancy and considerable uptake of health resources. Its microvascular complications lead to retinopathy, nephropathy and neuropathy while macrovascular complications lead to coronary artery disease, cerebro-vascular disease, peripheral vascular disease and amputations. It has been argued with certainty that all diabetic patients are at risk for developing complications like retinopathy and others. Although one cannot precisely predict progress of complications to an advanced stage, certain factors such as degree of hyperglycemia, disease duration, advancing age, hypertension, pre-existing counterpart, and chronically poor glycemic control increase the likelihood that complications will arise (Gamel, 1998). The present work carried out in the Department of Endocrinology, Postgraduate Medical Institute, Hayatabad Medical Complex, encompasses some of the above mentioned factors *i.e.* diabetes duration, obesity, age and hypertension, studied in relation with the frequency of retinopathy, nephropathy and neuropathy. The patients studied were categorized into different groups each time on the basis of above parameters. To avoid bias that might be caused by the unequal number of patients in different groups, they were compared according to percent proportions in each group.

PREVALENCE OF MICROVASCULAR COMPLICATIONS

Diabetes mellitus is growing worldwide like an epidemic and our population is not an exception. Being inhabitants of a developing country, we are at higher risk of contracting this disease (Venkat, 2000). Increasing prevalence of diabetes, lack of proper education in the population about the nature and course of the disease and general apathy of the patients towards necessary metabolic control are the main factors paving way for an earlier onset of microvascular complications. Khan (1991) conducted a pilot study in Karachi on 3000 diabetic patients for screening of retinopathy. The study showed that 780 (26%) patients had retinopathy. The incidence was 0.7% in 21-30 year age group and 66.1% in >51 year age group for males and

1.1% and 54.2% respectively for females. In contrast the present study demonstrated 51% patients with retinopathy which is almost double the rate of aforementioned study.

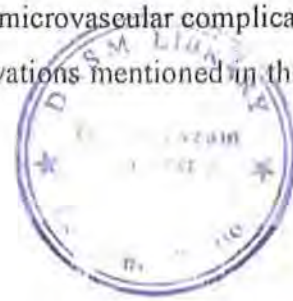
Both in type 1 and type 2 DM, it has become increasingly clear that multiple risk factors may be as important as hyperglycemia. Karamanos *et al* (2000), using the cross sectional data of the EURODIAB IDDM project to identify the factors associated with early onset and late protection from microangipathy in type 1 diabetes, studied 300 cases and compared the extreme groups i.e. those with microangiopathy within 5 year duration with those without microangipathy even after 14 year duration. He concluded that the former group had higher prevalence of hypertension, hyprelipidemia, poor glycemic control, obesity, smoking and cardiovascular disease indicating their positive relation with the development of microvascular complications. Thus the more these factors are present concurrently in a diabetic patient, greater is the possibility of developing complications. The present study included more patients with older age (>50 / 60 years), longer diabetes duration, poor glycemic control and hypertension. Obviously, these patients are more prone to develop microangiopathies. Moreover, this older age group has relatively lower level of education and awareness and present themselves to the health care system at a time when the disease has already progressed to advanced stage and the complications have shown their presence. This is another reason that the study has shown a relatively higher frequency for each complication as compared to studies reported before (Cathelineau *et al*, 1997). Yet, these results are approximately in accordance to those reported for retinopathy by Klein *et al* (1996), who monitored a group of 996 IDDM patients and another group of 1780 NIDDM patients for almost 10 years. Among these patients including both younger and older individuals, they observed that some degree of retinopathy occurred in 82 –100% of the patients depending upon the levels of HbA1c. In another study, Akbar *et al* (2001) screened 572 admitted diabetic patients in a leading hospital of Saudi Kingdom and showed that 268 (41%) were hypertensive, 99 (38%) had retinopathy, 174 (30%) had proteinuria with 81(47%) among them in renal failure and 54% had neuropathy. They attributed the frequency of complications to inadequate screening. Results of the present study are not different and have shown

even higher frequencies than above. It is reasonable to believe that in our diabetic patients too, inadequate screening is another factor leading to unacceptably high prevalence of above complications.

An interesting and fortunate finding in the present study is that in spite of very high prevalence rates, most of the patients with retinopathy and nephropathy did not reach the end-stage disease *i.e.* advanced diabetic eye disease and ESRD which are the principal causes of blindness and death in such type of patients. Diabetic retinopathy is a condition which ranges from minor incipient changes needing only observation to severe proliferative changes needing laser treatment (Plate 1) (Kohner *et al*, 1996). Five patients in this study suffered from proliferative stage. Similar situation occurred with nephropathy, which also progresses from the initially benign condition of microalbuminuria to the fatal ESRD and renal failure needing renal replacement or dialysis for survival. Only one patient was found to have ESRD who was started on dialysis but was unable to survive (Plate 2). Various other population-based studies have shown such type of findings for retinopathy. However, the reason for a large variation in the prevalence of total and proliferative retinopathy could possibly be due to the fact that the studies were not performed under the same conditions (Khan, 1991). Especially in this study, it can also be explained by the relatively smaller value for the mean duration of diabetes, which has not been able to prevent the initiation of complications but is still not enough to cause progression. In contrast, diabetic foot problem, which is commonly due to combination of neuropathy and peripheral vascular disease (deficient blood supply), is very common presentation causing considerable morbidity and threat to amputations. Given the lack of education and inadequate self-care on part of the patients, it is reasonable to expect that a large percentage of diabetic patients may develop serious disabilities concerned with these complications. Concurrent presence of retinopathy, nephropathy and neuropathy in various combinations that was observed with a reasonably high prevalence rate in this study is another matter of concern. This situation sets up a vicious cycle with one complication predisposing to the other. This may leave the patient in a highly morbid state.

DURATION OF DIABETES

The prevalence and severity of retinopathy, nephropathy and neuropathy have a positive relationship with duration of diabetes. For example, the younger the age at onset of diabetes and the longer the duration of diabetes, the greater is the risk of developing neuropathy during the following years in type 1 DM. On the other hand, type 2 DM begins insidiously and may be preceded by a long period of undiagnosed glucose intolerance. Neuropathy may be a presenting feature or appearance of another complication may be the incidental finding during routine examination for some other illness. This was also confirmed by the study of Kasthuri *et al* (2000), in which it was shown that both prevalence and severity of neuropathy increased with duration of diabetes: prevalence from 7.50% at the time of diagnosis to 66.35% after 25 years and severity from 17.39% to 48.28% as the duration increased. In the present study, the interplay between the duration of diabetes comprising different groups and the frequency of complications followed the above mentioned principle. This observation is also in agreement with the study of Khan (1991) who observed a remarkable correlation between diabetes duration and retinopathy i.e. 182 (23.3%) had diabetes for 1-5 years and 598 (76.6%) had diabetes for 6-16 years. Results of the present study show that after >20 years diabetes duration, the occurrence of microvascular complications is nearly 100 percent. Although these figures cannot be regarded as truly conclusive due to small number of patients representing this duration group, they are in agreement with the study of Intekhab (1997). He described that the peak incidence of diabetic nephropathy occurs after 16 years in type 1DM and after 20 years in type 2 DM. Another study suggested that diabetes duration and degree of metabolic control are the most important predictors of occurrence and progression of developing retinopathy (Agardh *et al*, 1994). Similarly Zhang *et al* (2001), analyzed from their study in DCCT that only three variables were significantly related to the development of diabetic retinopathy after adjustment for metabolic control; *i.e.* duration of participation, HbA_{1c} and BMI at baseline. A considerably high prevalence of retinopathy and other microvascular complications in the present study is probably a reflection of the observations mentioned in the above



studies namely the effect of combination of poor glycemic control and relatively longer diabetes duration.

The higher mean values of diabetes duration in patients having one or other complication as compared to those without that complication is also an indication that there exists a positive relationship between the duration of diabetes and the microvascular complications. Likewise detection of higher mean values of diabetes duration for patients having one or more complication as compared to those having no complication also favours this relationship. A meager number of patients in the group of >20 yr duration, which is only three, may indicate that the prevalence of patients with a very long diabetes duration is low in the general diabetic pool. This in turn, may be due to the fact that diabetes was much less common 20 years ago than it is today or because of the possibility that mortality may be high in patients approaching this much duration.

BODY MASS INDEX

Obesity is highly associated with insulin resistance and is the biggest risk factor for type 2 DM. Although the molecular basis of this common syndrome is still poorly understood, a possible key role of tumor necrosis factor (TNF α) produced by adipose tissue has been suggested. Katsuki *et al* (1998) have concluded that serum TNF α levels are influenced by body fat accumulation and that they contribute to insulin resistance associated with obesity and type 2 DM. Obesity is defined and classified on the basis of body mass index (Baron, 1989): mild obesity with BMI between 25 and 30 kg/m^2, moderate obesity with BMI of 30-40 kg/m^2 and severe or morbid obesity with BMI of >40 kg/m^2. However in this study, as evaluation of possible relationship between BMI and microangiopathies was desired, patients were grouped according to uniformly increasing BMI from lower to higher values.

Body mass index (BMI) is not only a reliable index of body weight but diabetic microangoipathies are also positively related to it. Type 2 DM is generally more common in obese adults where insulin resistance is considered to be the chief mechanism responsible for the causation of diabetes (Masud *et al*, 1992). On

evaluation of relationship between different BMI groups and microvascular complications in diabetic patients, the present study has shown a rise in percent proportions of retinopathy, nephropathy and neuropathy with the increase in range of body mass index. This is generally in agreement with studies reported previously (Hendrick *et al*, 2002). The apparent decline of complications in the higher BMI group ($\geq$40 kg/m^2) could possibly be due to a smaller representative group. The finding in this study that the mean BMI value for patients having retinopathy or neuropathy was higher as compared to that for patients without them replicates the results of EURODIAB IDDM Complication Study, in which Karamanos *et al* (2000) found that those who had microangiopathy, in addition to other outcomes, also had higher BMI levels. Higher mean BMI value for patients having one or more complications as compared to those having no complication is yet another indication of positive relationship between BMI and the above complications. Another interesting observation of the present study was that patients with type 2 DM showed lesser tendency for obesity. The mean BMI values fell in the high normal to mild obesity range, which was 27.38 kg/m^2. Firstly, it may indicate that obesity genetics is not involved in most of these diabetic patients. Secondly, it may indicate the state of under-nutrition prevailing in our population due to low socio-economic conditions.

PATIENTS' AGE

In developing countries, majority of people with diabetes are in age range of 45-64 years. In the developed countries, majority of people with diabetes are aged >65 yrs. This pattern is expected to be accentuated through the year 2025 (King *et al*, 1998). Apart from being a risk factor for diabetes, advancing age causes increase in the frequency and severity of diabetic complications. As the age of diabetic patient increases, the duration of existent diabetes increases, capacity of resistance to infections and other stress conditions decreases, diet adjustments become more disturbed, physical activity declines and metabolic control disturbances become more evident. In addition, aging itself is detrimental to the vessel wall integrity. All these factors contribute to the acceleration of diabetic complications including microangiopathies (Najib, 1999). In one study, elderly subjects with impaired glucose

tolerance (IGT) were shown to have already increased levels of several endothelin-dependant haemostatic factors indicating endothelial damage in this group. One of these factors was found to be restricted to microvasculature only (Leurs *et al*, 2001). In the present study maximum number of patients were above the age of 50 years. Distribution of retinopathy, nephropathy and neuropathy among different age groups in this study has shown the above trend positively.

Diabetic nephropathy and neuropathy both occur more frequently in older patients with type 2 DM. In nephropathy, microalbuminuria is the first indication followed by clinical proteinuria. Diabetic neuropathy causes paresthesias, tingling, burning and diminished pain sensation in the feet. This predisposes the patient to injury, ulceration, infection, gangrene and amputation if significant peripheral vascular insufficiency is present. In such patients, diabetic proliferative retinopathy is a leading cause of blindness, which can be detected by regular ophthalmoscopical examinations and is treated with Laser photocoagulation (Rosenstock and Julio, 2001). In the present study, mean age values were higher in patients with the complications under discussion as compared to those not having them showing a positive inter-relation between increasing age and microvascular complications. These findings replicate the results of Rosenstock and Julio (2001).

It is an established fact that old age also predisposes to the development of lens opacities. Therefore deteriorating vision in aging patient associated with lens opacities on one hand and retinopathy on other, may be a factor to initiate a vicious cycle leading to poor metabolic control and predisposition to development of complications. This is due to the fact that good vision is essentially required for self-care such as self-administration of insulin injections or other medicines and foot care during walking.

HYPERTENSION

Hypertension is a co-morbid condition in diabetic patients. For example, hypertension affects 20-60% patients with diabetes depending upon age, obesity and

ethnicity. It substantially increases the risk of both macrovascular and microvascular complications (ADA, 2002).

In Pakistan, 25% people with diabetes have hypertension as compared to 10% in non-diabetic counterparts (Diabetic Association of Pakistan, 2000). In contrast, the figures of hypertension are very high (53.5%) in the present study. The main reason for this discrepancy could possibly be inadequate representation of the general diabetic pool because only hospitalized patients have been studied here who are usually more ill having greater chance of associated conditions. Lower levels of blood pressure taken for diagnosis of hypertension could be another reason. Instead of standard definition of more than 140/90 mm Hg, hypertension was defined as blood pressure levels of >130/80 mm Hg. The primary intention is to increase the level of awareness about the concept of blood pressure levels because it has been proved by several studies that bringing BP levels to below 130/80 mm Hg leads to a fall in the incidence of complications (Carlos *et al*, 2002). In the present study, the results of interaction between hypertension and microvascular complications have revealed that the presence of high blood pressure has significantly increased the likelihood of all the three complications. This indication follows the findings of United Kingdom Prospective Diabetic Study Group, who in one of their major studies published in 1998, has shown that hypertension is an independent risk factor for the development of microalbuminuria and retinopathy (UKPDS, 1998). This study also showed that long-term blood pressure control in hypertensive patients with diabetes results in significant reduction in all diabetes related end points including microvascular and cardiovascular complications.

Combination of high blood glucose levels and high blood pressure in type 2 DM has been referred to as "double jeopardy" and the latest strategy is to combine strict glucose and blood pressure control to optimize the patient outcomes (Mogensen CE, 1998). From the present study it appears that hypertension is a common co-morbidity in this population and needs to be diagnosed and managed promptly and adequately.

MACROANGIOPATHIES

Macroangiopathies in the form of coronary artery disease, cerebrovascular disease and peripheral vascular disease are other extremely co-morbid conditions in diabetic patients. Macroangiopathies have occurred in this study relatively infrequently. However, the figures regarding cerebrovascular and coronary artery diseases cannot be regarded as the actual representative of the above fact because patients with these conditions are not usually admitted to Endocrinology unit where this study was conducted. These patients are usually referred to medical or cardiology units for management of their primary events. On the other hand, foot wounds and ulcers were a common cause of presentation for hospital admission (Plate 3). Syndrome of "diabetic foot" is usually due to combination of several factors including neuropathy, sluggish circulation and infection. Improper footwear and unhygienic behavior towards feet (Plate 4) further add to the problem. This condition which at times poses an annoying health problem, is very difficult to treat and requires an organized therapeutic approach involving bacteriological and radiological investigations, surgical debridements, appropriate antibiotic cover and good nursing care in addition to optimum glycemic control. The slight preponderance of females over males in case of macroangiopathies is in agreement with other studies, in which relation and absolute excess of cardiovascular mortality was greater in women than in men particularly for type 1 DM suggesting that diabetes is a more important risk factor in women than men as far as macrovascular complications are concerned (Roper *et al*, 2002). In contrast, observation in case of microangiopathies showed that although the total number of female diabetic patients was greater than their male counterparts, there was a proportionate occurrence of complications among both sexes.

ROLE OF METABOLIC CONTROL

Chronic hyperglycemia, which is the hallmark of diabetes mellitus, is the main contributor to the development of microvascular complications. Diabetes is an incurable disease and the changes it produces in small blood vessels are not reversible. However, It is now an established fact that upto 60% of the complications can be prevented or delayed by keeping blood glucose levels close to normal (DCCT, 1993

and UKPDS, 1998). Ohkubo *et al* (1995), in Kumamoto Study, randomized 110 patients to intensive or conventional insulin treatment and monitored them for 6 years. The study proved that intensively treated group had less retinopathy (13% Vs 38%), nephropathy (10% Vs 30%) and neuropathy (12% Vs 65%), the results again being similar to DCCT. Similarly, in the feasibility trial of Veteran Americans cooperative study, Abraira *et al* (1995) after 27 months follow up of 153 men with type 2 diabetes, showed that in the standard therapy group, the urinary albumin excretion rose significantly (from 14 to 158 mg) while no significant difference occurred in the intensive group. However there was no significant difference for retinopathy.

Diabetes is an under-diagnosed and under-recorded cause of death. In Pakistan only 36.3% of diabetics are aware of their condition (PMRC, 1994). Diabetes is an under-treated disease too since it was observed during the course of this study that a large number of patients had very poor metabolic control (mean blood glucose =331±111) often with inadequate treatment. No patient could be controlled with diet alone. Even most of the type 2 diabetic patients required insulin for the control of their badly managed blood glucose. Non-compliance to anti-diabetic treatment was usually a common finding. It was realized that poor socio-economic conditions did not allow many patients to fulfil the cost of their medicines. Another reason of poor metabolic control is believed to be the frequent changing of consulting doctors and even resorting to the use of traditional "hakeemi" medicines.

Endothelial dysfunction is one of the several abnormalities which are commonly associated and inter-linked in diabetic patients (Tooke, 1996). In one study, the endothelial dysfunction causing microvascular complications was found well correlated with metabolic control along with other factors such as hypertension, obesity, age, sex, positive family history, hyperlipidemia, treatment modalities and smoking (Ak *et al*, 2001). Similarly, retrospective studies have shown that patients with poor glycemic control tend to develop neuropathy at an earlier age and are more severely affected than those with better glycemic control (Rehman and Akhtar, 1999). It may be inferred from these observations that aside from control of blood pressure,

lipids and obesity, currently the only effective way to prevent and/or delay the development and progression of retinopathy, nephropathy and neuropathy remains to be the achievement of blood glucose levels as close to normal values as possible.

RENAL FUNCTION AND MORPHOLOGY

As the severity of nephropathy progresses with increasing duration of diabetes and manifests itself as clinical proteinuria (>500 mg/24 hr), urea and creatinine levels in the blood begin to rise until the renal functions become compromised (Table 2). Impaired renal function as determined by rising serum creatinine and urea levels in diabetic patients is a late feature, although the rate of progression varies widely between individuals (Kumar and Clark, 1994). Since the mean diabetes duration was not longer enough (8.5±5.6 years), renal function was not affected to a considerable extent in most of the patients examined during this study. Although mean serum creatinine and urea levels were higher in patients having nephropathy as compared to those in non-nephropathic patients (0.94 vs 0.73 mg/dl and 38.7 vs 29.9 mg/dl respectively), yet they remained within normal limits. End-stage renal failure occurred in only one patient.

Renal size as assessed by ultrasonic examination in a subgroup of 43 patients with nephropathy, was shown to have increased only in two patients. This finding is usually seen in initial stages of nephropathy. No patient showed a decrease in renal size, which is usually a feature of final stages. Thus the role of ultrasonography in assessment of nephropathy was not evident.

The primary aim of this study was to determine prevalence of chronic complications in diabetic patients and if there occurred a correlation between these complications and demographic aspects like age, sex, BMI, duration since diagnosis and hypertension. Such an information may prove a useful tool in application to the management and prevention programs of the aforementioned diabetic complications.

Conclusions and Recommendations

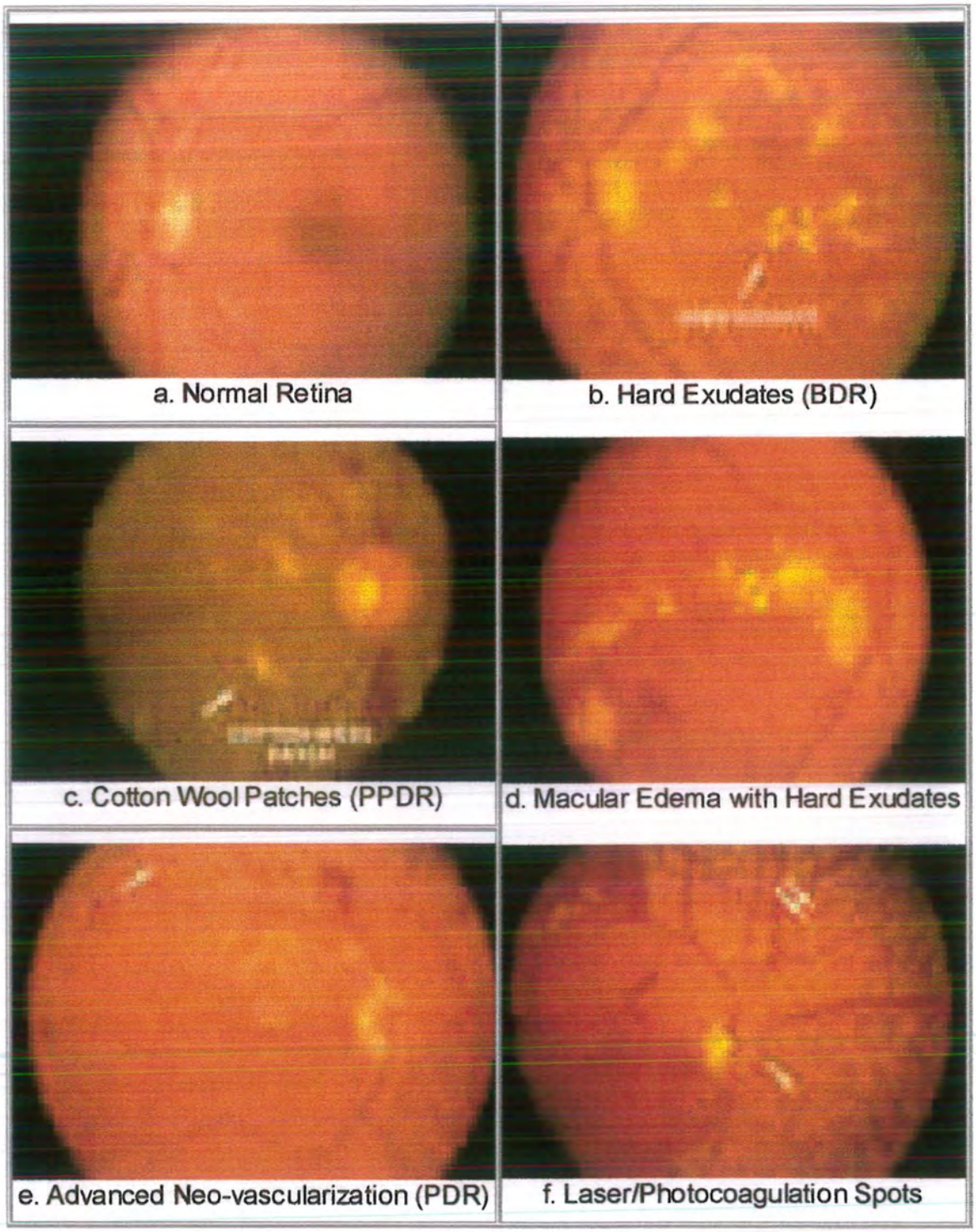
In conclusion, the study shows that considerably high proportion of diabetic patients develop complications in one form or other and in different combinations

before they consult the tertiary care hospital. Furthermore, Increasing age, BMI and diabetes duration were all found positively related to the development and progression of microvascular complications as was high blood pressure. These results may be well interpreted in perspective of the reports of King *et al* (1998) on prevalence, numerical estimates and projection of diabetes mellitus, which have supported the prediction of its epidemic nature during the first quarter of 21st century. Intense realization is therefore emerging about the fact that worldwide surveillance of diabetes and its complications is needed towards their prevention and control. Education, guidance and counseling to the patients may help reducing the occurrence of diabetic complications. Examples of at least three patients from the present study can be quoted in this regard. First, a doctor himself; second, a doctor's wife and third, my own mother; all of them had type 2 diabetes for more than 15 years and yet they had no evidence of any complication.

This study represents a relatively smaller group of diabetic patients the participants having been examined at tertiary care hospital, which is usually a less accessible facility for general population. As many diabetic patients consult general practitioners at primary care level, more studies are required to be launched at that level. This will not only ensure a better knowledge and understanding about the magnitude of problems sustained by diabetic patients but also pave way for prevention and management protocols for diabetes mellitus and ensuing complications. The most important aspect of these protocols involve education of doctors and para-medical staff at primary level who will in turn educate the patients on one hand and ensure early identification and referral of high risk patients on other.

There is a need to realize that a higher rate of microvascular complications prevails in our diabetic population. As it is almost impossible to revert these complications once they appear, the only treatment is to prevent them. Achievement of optimum glycemic control remains the only effective way to prevent or delay the development of these complications or slow their progression. Natural history of diabetes shows that it is much easier to control it if detected in early stages.

PLATE 1: RETINAL IMAGES AT DIFFERENT STAGES OF DIABETIC RETINOPATHY



BDR= Background Diabetic Retinopathy
PPDR= Pre-proliferative Diabetic Retinopathy
PDR= Proliferative Diabetic Retinopathy

PLATE 2: A PATIENT WITH DIABETIC NEPHROPATHY HAVING PUFFY FACE AND SWOLLEN RIGHT LEG. HIS LEFT LEG WAS AMPUTATED. LATER, THE PATIENT DIED OF END-STAGE RENAL DISEASE (ESRD).

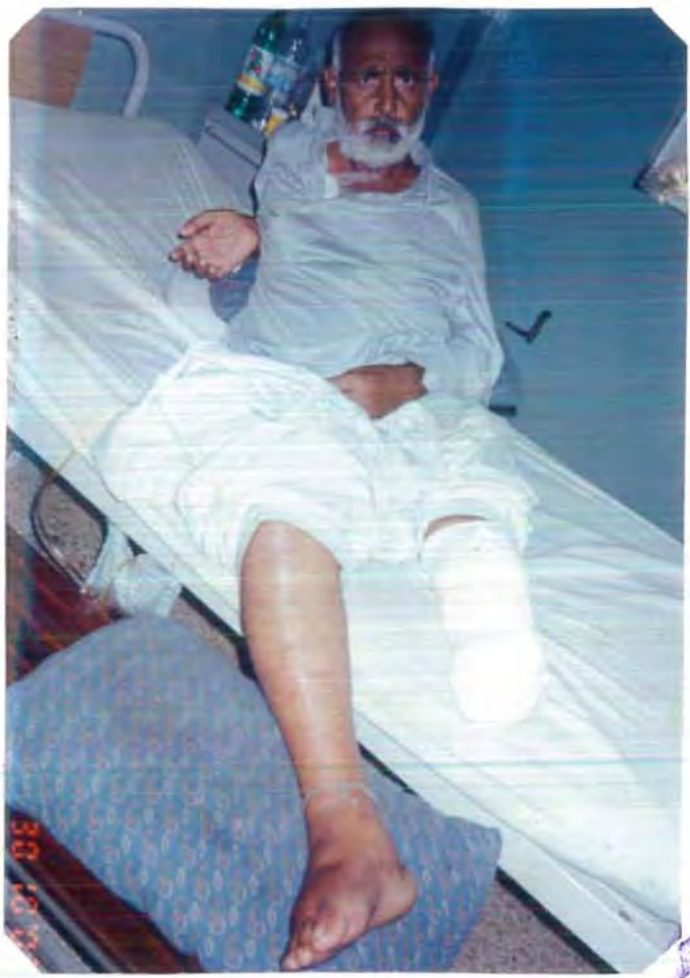
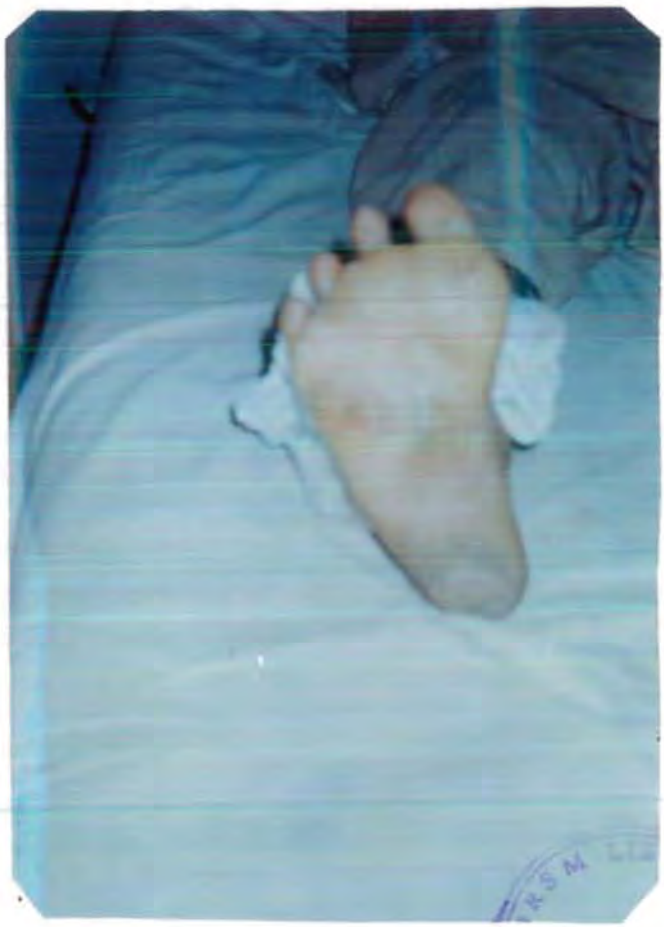


PLATE 3: DIABETIC NEUROPATHY WITH PLANTER FOOT ULCERS. A PREVIOUSLY AMPUTATED TOE IS ALSO NOTED.



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