INTRODUCTION

Diabetes mellitus is the most common metabolic disease in the world affecting an estimated 537 million adults worldwide between the age of 20 to 79 in the year 2021 (1,2). Overall, the number of adults globally living with diabetes has more than tripled over the past 20 years, predicted to increase to 784 million by 2045 (2). With millions of new cases being identified each year and costing billions of dollars in treatment; it has been rightly called the “modern pandemic” by certain authors (3).

Traditionally, T2DM was regarded as a disease which is associated with high incomes and affluence. However, over the last several decades there have been profound changes in the food habits and lifestyle of many low- and middle-income countries, leading to a substantial increase in the prevalence of diabetes across populations throughout the world.

The prevalence of diabetes has been increasing worldwide, with developing nations such as India accounting for a sizable portion of the global burden. With a prevalence as high as 10% in our country, India has earned the ominous title as the “Diabetic capital” of the world. According to a recent study (4), India was home to an estimated 77 million diabetic patients in 2019, a number that is expected to increase to more than 150 million by 2045. A recent survey by the ICMR (5) found that out of the 10000 individuals that were part of the study, nearly 9% were diabetics. However, even more concerning was the fact that an additional 24% of the people were found to be pre-diabetic. This is one of the more alarming aspects of diabetes mellitus - the disease remains undiagnosed in the majority of individuals for several years. As a result, patients often present for the first time with complications. The survey also pointed out that almost half of diabetics were unaware of their raised glycemic status.

This increasing burden of diabetes in developing countries such as India is fueled primarily by an increasing prevalence of overweight/obesity, sedentary lifestyles and unhealthy dietary habits.

The role of ethnicity and genetic factors is also extremely significant (6); studies have found a strong evidence of increased risk of insulin resistance and a stronger predisposition to the development of diabetes in the Indian population (6).

Even though a number of diseases and conditions have been identified such as chronic pancreatitis, drugs, certain viral infections etc., which can potentially lead to the development diabetes, the vast majority of cases can be attributed to obesity and the development of insulin resistance.

Irrespective of etiology, T2DM is characterized by a chronic state of hyperglycemia which leads to changes in capillary flow, vascular permeability, basement membrane thickening and other blood vessel changes. The risk of developing these vascular complications is directly correlated with both – the degree of hyperglycemia as well as the duration of disease (7) and is a significant cause for increased premature morbidity and mortality among diabetics, leading to reduced life expectancy and increased economic burden on the Indian health care system.

Although virtually all organs of the body are affected, Diabetic retinopathy (DR) is considered to be the most common micro vascular complication of diabetes (7). In the United States, T2DM is the most common cause of blindness, with roughly 10,000 new patients of DR going blind each year (8). The prevalence of DR in the US is estimated to reach 16 million by 2050, leading to life-threatening complications in around 3.4 million (9). The situation is no better in India. A cross-sectional population prevalence study from south India estimated that the prevalence of DR is 18.0% in urban areas and 10.3% among rural diabetics (10).The pathogenesis of diabetic retinopathy is complex and involve degenerative changes of capillary walls, leading to a reduction in capillary perfusion and hypoxia. The cellular hypoxia stimulates the release of cytokines and cell signaling molecules such as Vascular Endothelial Growth Factor, which lead to the development of new, immature blood vessels (11). These changes of Neo-vascularization

are characteristic of proliferative diabetic retinopathy and can also be found in other areas including nail fold capillaries (7).

As pointed out earlier, many patients remain unaware of their glycemic status, and often do not seek medical attention until they develop a diabetic complication. Therefore, it is imperative for medical practitioners to detect the presence of vascular complications in the patients at the point of first contact itself. Since many of the diabetics belong to the lower rungs of the socio-economic ladder, it might not be financially feasible for them to undergo expensive investigations such as a urine micro albumin levels or consult an ophthalmologist for fundus examination. This necessitates the development of a simple, effective, non-invasive and relatively inexpensive procedure which can not only be carried out in the OPD itself, but is also able to delineate the patients who are at an increased risk of having other micro vascular complications such as retinopathy or nephropathy.

Nailfold capillaroscopy (NFC) is one such promising method. It is a noninvasive method for the observation of the capillary microvasculature in the nail folds. Traditionally, it has been commonly employed by dermatologists for observing capillaries in connective tissue diseases such as systemic sclerosis, Raynaud's disease etc. However, recent studies have found that NFC is a valuable modality for assessing the risk of micro vascular complications such as DR.

The morphological changes in NFC are similar to those found in DR and include increased tortuosity, Neo-angiogenesis, development of avascular areas, ectatic capillaries and micro-hemorrhages among others (7). Some studies have even found a correlation between the severity of morphological changes in NFC and the severity of DR (7).

However, there is a paucity of literature supporting a relationship between the development of NFC changes and other diabetic complications or glycemic control, from India.

The objective of this study is to evaluate whether evaluation of Nail Fold Capillary changes can be used as a marker of other micro vascular complications such as diabetic retinopathy. Being a non-invasive and relatively cheap investigation that can easily be performed in the OPD, it would facilitate early identification of patients who are at an increased risk of having underlying complications. An early detection and timely intervention might lead to a reduction in the end stage complications such as blindness and end stage renal failure. Thus, the findings of this study may aid in the development of screening and preventive strategies for diabetic patients, as well as the implementation of targeted interventions in those at an increased risk of having diabetic complications.

ROL

According to the American Diabetic Association (ADA) (12), diabetes mellitus may be diagnosed based on plasma glucose criteria, either the fasting plasma glucose (FPG) value or the 2-h plasma glucose (2-h PG) value during a 75-g oral glucose tolerance test (OGTT), or A1C criteria.

Criteria for the diagnosis of diabetes:

• FPG ≥126 mg/dL: Fasting is defined as no caloric intake for at least 8 h. OR

• 2-h PG ≥200 mg/dL during OGTT. The test should be performed as described by WHO, using a glucose load containing the equivalent to 75 g anhydrous glucose dissolved in water. OR

• A1C ≥6.5% : The test should be performed in a laboratory using a method that is NGSP certified and standardized to the DCCT assay OR

• In a patient with classic symptoms of hyperglycemia or hyperglycemic crisis, a random plasma glucose ≥200 mg/

Although the pathogenesis of diabetes is complex, a number of risk factors have been identified. Risk factors for type 1 diabetes include family history, ethnicity (with whites at higher risk than other racial or ethnic groups), and certain viral infections during childhood.

On the other hand, type 2 diabetes has a more diverse set of risk factors; some are modifiable while others are non-modifiable. Non-modifiable risk factors include age, family history and a history of gestational diabetes to name a few (13). Modifiable risk factors are mostly linked to lifestyle. Obesity is one of the most important modifiable risk factors for development of diabetes; others include physical inactivity, smoking and alcohol use (13). Additionally, the distribution of the body fat is also extremely relevant with central or abdominal obesity being linked to a strong risk for development of diabetes.

Broadly, Diabetes can be classified into the following general categories:

• Type 1 diabetes: It occurs due to autoimmune β-cell destruction.

• Type 2 diabetes: It occurs due to a progressive loss of adequate β-cell insulin secretion and is frequently associated with insulin resistance.

• Specific types of diabetes: It can be causes due to various causes such as monogenic diabetes syndromes, diseases of the exocrine pancreas (such as cystic fibrosis and pancreatitis), and drug- or chemical-induced diabetes (such as with glucocorticoid use) etc.

• Gestational diabetes mellitus: Diabetes diagnosed in the second or third trimester of pregnancy in individuals who were not clearly overt diabetes prior to gestation.

However, Type 2 diabetes accounts for the vast majority of the cases. Irrespective of type or etiology, the chronic state of hyperglycemia commonly produces systemic complications leading to multi-organ damage. The complications can broadly be divided into micro vascular and macro vascular complications.

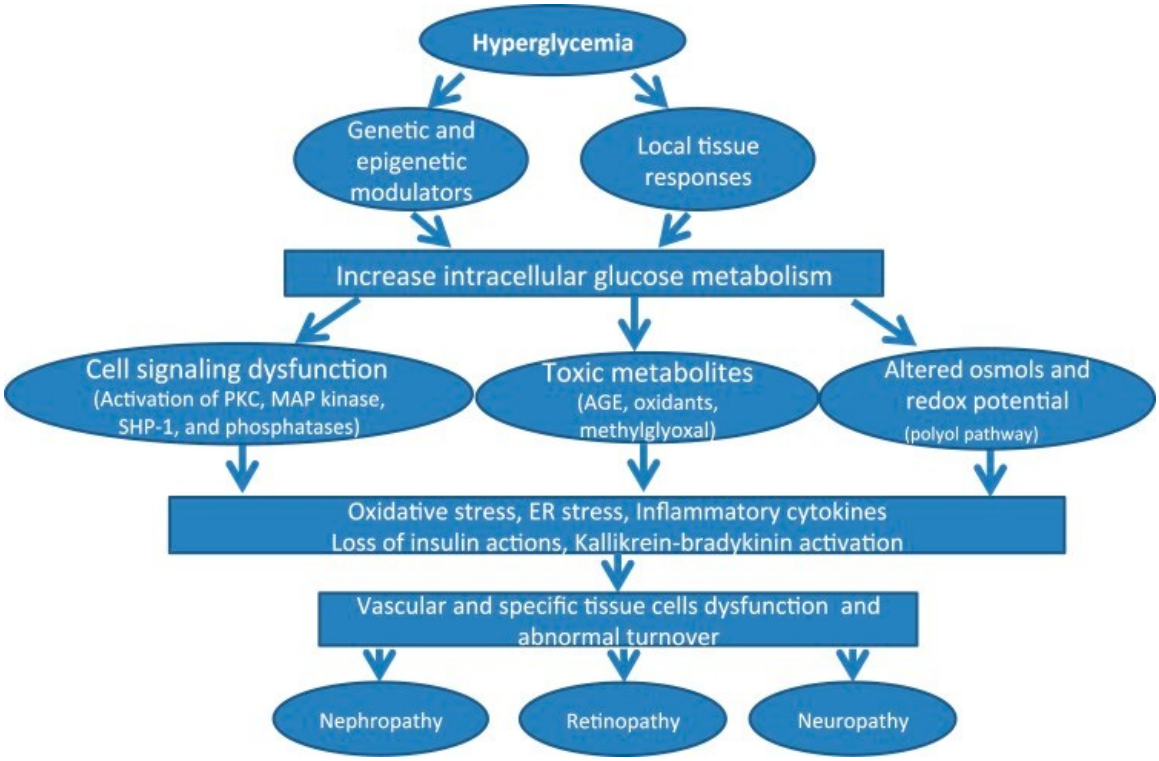
TABLE 419-1 DIABETES-RELATED COMPLICATIONS

- Microvascular
  - Eye disease
    - Retinopathy (nonproliferative/proliferative)
    - Macular edema
  - Neuropathy
    - Sensory and motor (mono- and polyneuropathy)
    - Autonomic
  - Nephropathy (albuminuria and declining renal function)
- Macrovascular
  - Coronary heart disease
  - Peripheral arterial disease
  - Cerebrovascular disease
- Other
  - Gastrointestinal (gastroparesis, diarrhea)
  - Genitourinary (uropathy/sexual dysfunction)
  - Dermatologic
  - Infectious
  - Cataracts
  - Glaucoma
  - Cheiroarthropathy[a]
  - Periodontal disease
  - Hearing loss

Pathogenesis of Vascular Complications

Vascular complications are the most important cause of morbidity and mortality in diabetic patients. These result from a complex interaction between various systemic metabolic abnormalities, such as sustained hyperglycemia, dyslipidemia as well as locally produced cell molecules which are produced in response to these toxic metabolites (11). Although hyperglycemia is the most important risk factor for the development of these complications, it is not the only relevant cause.

Figure showing the pathogenesis of micro vascular complications in T2DM



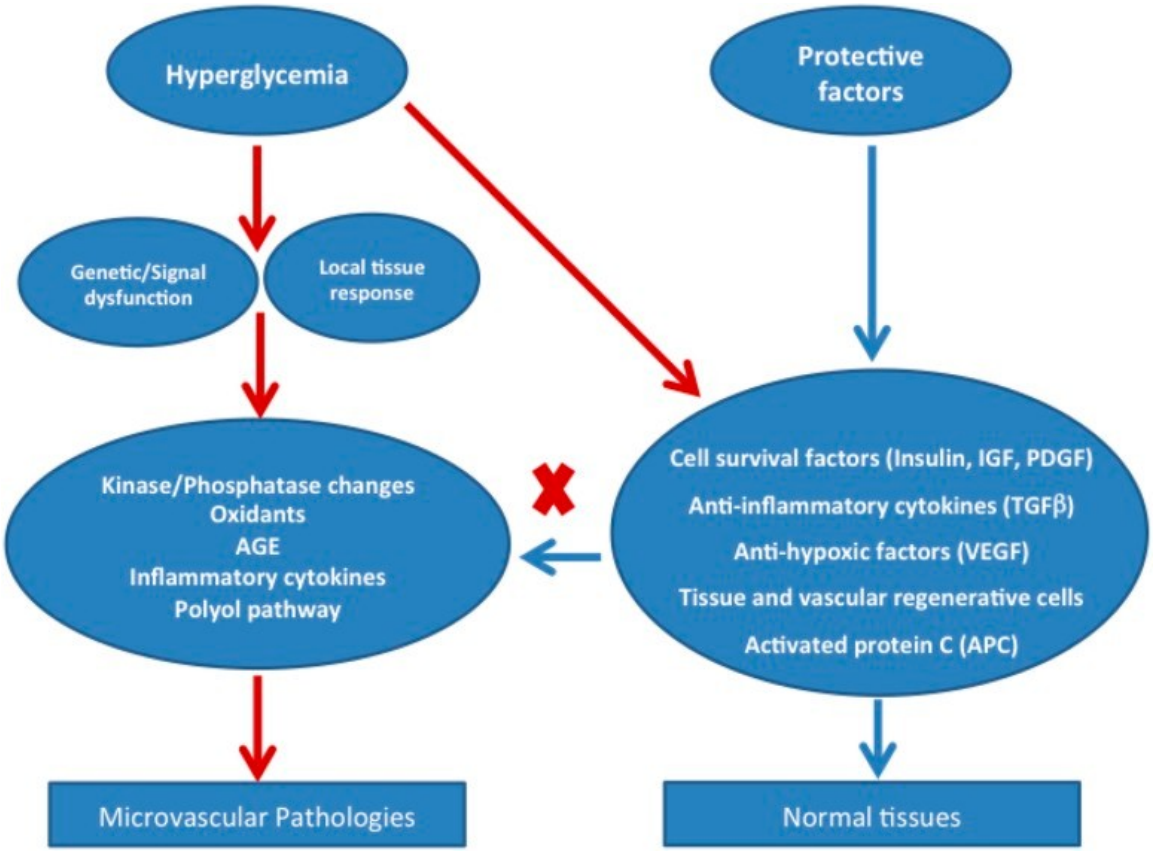
Some of the cellular mechanisms which have been proposed as mediators in the development of vascular anomalies include (11):

- Non-enzymatic glycation and the formation of advanced glycation end products (AGEs)
- Enhanced reactive oxygen stress
- Activation of the polyol pathway, Diacylglycerol (DAG)–Protein kinase C pathway
- Involvement of the Src homology-2 domain-containing phosphatase-1 (SHP-1)
- Imbalances in the Renin-angiotensin system (RAS) and kallikrein-bradykinin (BK) systems.

However, the deleterious effects of imbalances in the above pathways and molecules is negated to a certain extent by a few factors that have been found to have a protective role (11):

- Cellular survival factors such as Platelet derived Growth Factor (PDGF), Insulin and Insulin Derived Growth Factor (IDGF)
- Anti hypoxic molecules such as Vascular Endothelial Growth Factor (VEGF)
- Anti inflammatory molecules such as Transforming Growth Factor Beta (TGF Beta) etc.
- Protective factors such as Activated Protein C

Figure Showing the complex interplay between protective and damaging cellular pathways



Nail Fold Capillaroscopy

Nail Fold capillaroscopy (NFC) is a non-invasive diagnostic test which can be used to study micro vascular abnormalities in the nail fold. It consists of powerful magnifying glass, which can be used to visualize the amplified images of skin microcirculation. Till date, a variety of instruments have been used to visualize the nail fold capillaries – ranging from the simple, ordinary hand-held dermatoscope, ophthalmoscope, all the way to light microscope (stereomicroscope) and dedicated video capillaroscopes which have the capability of recording the video of the entire procedure as well(14). Even though experienced practitioners can identify early changes in capillaries using a simple dermatoscope, Video Capillaroscopes are increasingly being utilized for the procedure. Not only do they allow the digital recording and reproduction of pictures taken, they can also be used to identify dynamic parameters such as blood flow patterns (15). Til now, it was mainly being used in rheumatological disorders such as Systemic Sclerosis (SSc) and Raynauds disease. The importance of NFC in SSc is evidenced by the fact that certain NFC patterns have even been included in the latest classification criteria for SSc (16). However, its utility in non rheumatological indications such as diabetes is relatively less well studied.

NFC can be used to evaluate the measurements of individual capillaries such as length, shape, and diameter as well as any abnormal patterns such as avascular areas and Neo vascularization etc (15).

Presence of capillary ectasia and giant capillaries represents the early stage of peripheral microangiopathy and are one of the initial response by the body to maintain perfusion in the face of reduced vascular supply (15). Endothelial cell dysfunction leads to an increased transition of endothelial cells into active myofibroblasts. Additionally, there is an overproduction of vasoconstrictors such as endothelins, and a simultaneous decrease of serum levels vasodilators (as nitric oxide) (15). The sustainedvascular dysfunction leads to the opening up of endothelial junctions leading to increased vascular permeability and progressive vascular leakage, producing

microhemorrhages. Furthermore, the development of hypoxia of downstream tissues is a potent activator for the release of angiogenetic factors such as VEGF. The consequent overproduction of VEGF leads to the formation of chaotic vessels, called neo-angiogenesis, and is seen in the NFC as meandering loop or bushy and branched capillaries organized in clusters. Finally, the chronic hypoxia leads to irreversible microangiopathy, which is characterized in NFC by the presence of avascular areas. Interestingly, tortuosity is not a pathognomic feature of microangiopathy and can even be found in some otherwise healthy individuals as capillaries have a tendency to became tortuous and dilated with age (17).

The advantages of NFC include the simplicity and ease in obtaining images in a reproducible fashion in an OPD setting. Certain drawbacks include the need for a specialized equipment and trained personnel to perform the procedure.

Since the role of NFC in non rheumatological conditions such as DM is still less clear, we hereby present a brief review of the previous literature regarding the same.

Maldonado et al (18)

Theirs was an observational, cross-sectional study which was conducted on 65 diabetic subjects. The study also included 50 age and sex matched controls who were non diabetic. The mean age of subjects in their study was 57 ± 8.94 years and females (75%) outnumbered males (25%). All subjects underwent a capillaroscopy examination and the following NFC changes were recorded - Capillary diameter (ectasia and giant capillaries), cross-linked, tortuous, arborified capillaries, avascular zones and haemorrhages. Overall, NFC changes were detected in 83% of the diabetics. The most common change was the presence of tortous capillaries, followed by cross-linked capillaries, avascular zone, ectasia and giant capillaries. As compared to the control group, the diabetic group had a higher prevalence of tortuous capillaries (63% of diabetics versus 20%

controls, p value 0.008), avascular areas (48% of diabetic versus 4% of controls, p value <0.001) and ramified capillaries (11% of diabetics versus 4% in controls, p value 0.02). Both the groups were comparable with respect to the rest of the NFC changes. They also observed that patients with capillaroscopic changes had a longer duration of disease as compared to the diabetics who had no changes on NFC (12.8 years versus 8.5 years).

Rajaei et al (19)

This cross-sectional study was conducted with the aim to determine the NFC changes in diabetic subjects. The study was conducted on 235 patients of type 1 and 2 diabetes from a single center in Iran. All patients underwent a nailfold capillaroscopy examinations using video capillaroscopy. In their study, the mean age of subjects was 59.91 ± 12.39 years and females outnumbered males (77.9% versus 22.1%). Normal and early scleroderma patterns were observed in 83.0% and 40 cases 17.0%, respectively. The most common NFC change observed was tortuosity (100%) followed by neoangiogenesis (72%). Based on the NFC changes, subjects were classified into two classes – normal pattern or scleroderma pattern. They found that except for neoangiogenesis, all other NFC changes were significantly more common in patients with scleroderma pattern than patients in the normal group (P value < 0.05).

Bakirci et al (20)

This cross-sectional study was conducted in patients of T2DM to compare the NFC changes in the subjects who were diagnosed with DR versus those who did not have DR. After ophthalmological evaluation, 44 subjects were found to have DR and 20 subjects did not have DR. NFC was performed to evaluate the diameters of apical, arterial, and venous loop of capillariesand the microvascular changes of capillaries. The subjects with DR were significantly older as compared to those without DR (61.4 years versus 56.6 years). Both the genders were

equally represented (Males 47.7% in DR and 55% in non DR group). As compared to the subjects without DR, the subjects with DR had been suffering from diabetes for a significantly longer duration (14.6 years versus 8 years, p value <0.001). Surprisingly, the subjects without DR had a higher prevalence of Diabetic nephropathy, as compared to those with DR (80% versus 31.8%, p value <0.001). Both the groups were comparable with respect to prevalence of hypertension (p value 0.53), dyslipidemia (p value 0.22) or diabetic neuropathy (p value 1.0). Both the groups were also comparable with respect to HbA1c (p value 0.5) and BMI (p value 0.72) as well. The most common NFC finding was Cross-linked capillaries (97.7% in DR and 100% in non DR) followed by increased tortuosity (79.5% in DR and 75% in non DR) and ectatic (54.5% in DR and 40% in non DR) or giant capillaries (40.9% in DR and 25% in non DR). Surprisingly, none of the NFC findings were more common in the DR group as compared to the non DR.

Kuryliszyn-Moskal et al (21)

Theirs was a cross-sectional observational study to evaluate nailfold videocapillaroscopic changes in diabetics. They included 106 patients of Type 1 diabetes and 40 healthy controls. Additionally, they also evaluated various biomarkers such as VEGF, sTM and ET-1. The mean age of cases and controls was 37.6 ± 12.83 years and 33.25 ± 10.06 years. Females outnumbered males in both the groups (66% in cases and 74% in controls). Both the groups were matched with respect to age (p value >0.05), gender (p value >0.05), and BMI (p value >0.05). In their study, none of the controls had any NFC abnormality. All the videocapillaroscopic changes were scored on a scale from 0 to 3.Morphological changes were observed by NVC in 86 out of 106 (81%) cases. Normal capillaroscopic patterns (score 0) were observed in 19%, minor changes (score 1) – in 27%, moderate or severe abnormalities in 19% and 35% of participants with diabetes, respectively. Severe capillaroscopic changes (score 3) were seen in 32 out of 54 (59%) people with microangiopathy, but in only seven out of 52 (13%) individuals without microangiopathy. Higher serum concentration of VEGF (p<0.001), ET-1 (p<0.001) and sTM (p<0.05) were

demonstrated in people with diabetes complicated with microangiopathy compared to healthy controls. Moreover, comparison between people with and without microangiopathic complications showed a significantly higher capillaroscopic score and sTM serum concentration in the group with retinopathy ($p<0.001$) nephropathy ($p<0.001$) and neuropathy ($p<0.01$).

Mohanty et al (22)

This was a hospital based cross-sectional study which included 250 patients of diabetes mellitus. All subjects were evaluated for diabetic retinopathy and nailfold capillaroscopy. Based on the presence or absence of DR, subjects were divided into 2 groups. Both the groups included 125 patients each. Majority of the subjects (52.8%) were middle aged to old, between the ages of 40-60 years. Males outnumbered females in a ratio of 1:2 (64.8% males versus 35.2% females). 80% The percentage of subjects with poorly controlled sugars was 20% in those without DR, 75.8 % in the subjects with NPDR and 97.6 % of the patients with PDR (P -value < 0.001). The mean duration of illness was significantly (p value <0.001) higher in those with PDR (12.8 ± 4.8 years), as compared to the subjects with NPDR (10.4 ± 3.4 years), which in turn was higher than the subjects without DR (5.5 ± 3.2 years) in the subjects without DR. Reduced capillary density was the most common NFC finding and was present in 55.6% of all subjects. This was followed increased tortuosity (52.8%), microhaemorrhages (28.4%), avascular areas (28.4%) and neoangiogenesis (6%). Except for increased tortuosity (p value 0.28), all other NFC changes including reduced nailfold capillary density (p value 0.006), neoangiogenesis (p value <0.001), microhaemorrhages (p value <0.001), abnormal forms (p value <0.001) as well as avascular areas (p value <0.001) were more common in PDR and NPDR group, as compared to those without DR.

Hosking et al (23)

Theirs was a small pilot study to study the changes in NFC in Type 1 diabetics. Being a pilot study, they included only 26 subjects. The mean age of participants was 14.3 ± 2.3 years and both the genders were equally represented (50% males). The mean duration of type 1 diabetes of was 7.9 ± 3.4 years and the mean HbA1c was 8.1 ± 1.1%. 3 participants had microalbuminuria and one had early signs of retinopathy. Participants with microvascular complications had more avascular areas on nailfold capillaroscopy ($p = 0.03$). Recent HbA1c was positively associated with the number of nailfold microhaemorrhages ($p = 0.03$).

Lisco et al (24)

This was a cross-sectional case-control study conducted on Type 1 and type 2 diabetics. Their main aim was to study NFC changes in diabetics.using a video capillaroscope. The study included 63 subjects of T2DM, 22 subjects of T1DM and 31 healthy controls. The mean age was 63.0 ± 10.7 years in T2DM group, 44 ± 15.9 years in T1DM group, and 44.4 ± 15.7 years in controls. The T2DM group included 65.7% males, the T1DM group included 50% males and the control group included 33.3% males. Good glycemic control (HbA1c < 8%), was observed in 69.1% of diabetics. A higher mean HbA1c level was observed in T1DM than in T2DM patients (8.4 ± 1.51% vs. 7.2 ± 1.25%, $p < 0.01$). Nailfold alterations were found to be more prevalent in diabetics, including tortuosity (84% vs. 50%, $p < 0.01$), avascular zones (60% vs. 38.6%, $p < 0.01$), ectasiae (63.8% vs. 31.6%, $p < 0.01$) and capillary with bizarre shape (65.1% vs. 34.2%, $p < 0.01$). At least two of these patterns were found with a higher prevalence in diabetics as compared to controls ($p < 0.01$). A univariate logistic regression analysis found diabetic status to be a significant predictor of tortuosity (OR: 4.7; $p < 0.01$), bizarre shape (OR: 3.3, $p < 0.01$), avascular areas (OR: 3.1, IC: 1.4–7, $p < 0.01$), and ectasia (OR:2.9, $p < 0.01$).

Ahmad et al (7)

This cross-sectional observational case-control study was conducted to evaluate the NFC changes in patients of diabetes mellitus. Their study included 262 cases and 150 controls. The subjects underwent ophthalmological testing and were divided into two groups based on the presence of DR. NFC changes such as tortuosity, increased capillary density, neoangiogenesis, microhaemorrhages, avascular areas, crossing and meandering capillaries and receding capillaries were recorded. The mean age of subjects was 50 ± 13 years in controls, 56 ± 13 years in DR+ group and 53 ± 12 years in the DR- group. Males were the predominant gender in all 3 groups – 57.3% in controls, 65.25% in DR+ group and 60.41% in DR- group. All the three groups were comparable with respect to age (p value 0.283) and gender (p value 0.418). The duration of disease was significantly higher in the DR+ve group as compared to the DR–ve group (6.9 ± 4.4 years versus 5 ± 3.8 years, p value <0.001). The HbA1c value was also significantly higher for the DR+ve group (9.4 ± 2.2 versus 7.3 ± 1.7, p value <0.001). Tortuosity, increased capillary density and neoangiogenesis were among the most common finding in both groups. As compared to the healthy controls, all NFC parameters were significantly (p value <0.001) more common in the diabetics. Patients with diabetic retinopathy had significant nailfold capillaroscopic features as compared to patients without DR (P value < 0.001). Neoangiogenesis, receding capillaries and avascular area were significantly higher in proliferative DR as against nonproliferative DR (P < 0.001). A positive association was found between the duration of DM and HbA1c values and NFC features.

Uyar et al (25)

This was an observational case-control study was conducted with the objective of evaluating nailfold capillaries in type 2 diabetes mellitus patients. 216 patients with type 2 diabetes mellitus and 101 healthy controls were included. Retinopathy was detected in 43.05% of diabetic patients (n = 93). The mean age was comparable in all three groups (60.89 ± 8.281 years in the DR+

group; 58.92 ± 8.506 years in the DR- group and 59.41 ± 11.867 years in the control group, p value 0.336). Males were relatively more prevalent in the control group as compared to the diabetic group (Males 41.6% in diabetics versus 53.5% in controls, p value 0.033). Median disease duration of DR+ group was significantly higher than DR- group [Median 14 years versus 4 years; $p < 0.001$]. HbA1c levels of patients with DR was significantly higher than patients without DR group [Median 8.7% versus 7.2% ; $p < 0.001$]. Capillaroscopic findings including tortuosity ($p < 0.001$), bushy capillary ($p < 0.001$), neoangiogenesis ($p < 0.001$), bizarre capillary ($p < 0.001$), microhemorrhage ($p = 0.001$), capillary ectasia ($p = 0.002$), and aneurysm ($p = 0.004$) were significantly higher in diabetic group as compared to the control group. As compared to the subjects who did not have any tortuosity on NFC, the subjects with tortuosity had significantly longer duration of disease (Median 10 years versus 3 years, p value <0.001). The same was also true for bushy capillary (Median 12.50 years versus 5 years; $p < 0.001$), aneurysm (Median 14 years versus 6 years; $p = 0.001$), neoangiogenesis (12 years versus 5.50 years ; $p = 0.002$), and bizarre capillary (Median 10 years versus 5.50 years; $p = 0.049$). In logistic regression analysis, only tortuosity was shown to be a significant predictor of DR (OR, 2.16; $p = 0.036$).

Okabe et al (26)

This observational, cross-sectional case-control study was conducted with the aim to determine NFC changes are associated with the presence and severity of DR. The study included a total of 83 diabetics and 63 age and sex matched healthy controls. Based on the presence and severity of DR, they classified the cases into three groups - non-DR (DR-), non-proliferative DR (NPDR), and proliferative DR (PDR). Using an automated capillary microscope with Capillary Analysis System program, which enabled automatic quantification of NC. The number of capillaries, along with the length, width and Turbidity were recorded. The mean age of subjects in the case and control groups was comparable (57.9 ± 8.6 years in cases and 57.8 ± 7.5 years in controls, p value

0.65). Males outnumbered females in both the groups (Males 60.2% versus 69.8% in controls, p value 0.23). As compared to controls, diabetics had significantly higher values of Total cholesterol (195.9 ± 45.9 mg/dl versus 219.8 ± 35.5 mg/dl, P < 0.001), HDL (53.1 ± 13.9 mg/dl versus 59.8 ± 16.3 mg/dl, P = 0.03), LDL (111.9 ± 37.4 mg/dl versus 134.2 ± 28.8 mg/dl, P < 0.001) and eGFR (65.5 ± 24.8 ml/min/1.73 m 2 versus 73.2 ± 12.2 ml/min/1.73 m 2, P = 0.02). As compared to the controls, the diabetic patients had significantly lower number of capillaries (5.52 ± 1.28 versus 4.24 ± 1.57, p value <0.001), length of capillaries (1445 ± 760 versus 774 ± 569 um, p value <0.001), width (28.4 ± 7.9 um versus 23.6 ± 8.4 um, p value <0.001) as well as turbidity (0.471 ± 0.164 versus 0.354 ± 0.162, p value <0.001). As compared to those with NPDR, subjects with PDR had a statistically significant decrease in length of capillaries (546 ± 479 versus 538 ± 384 um, p value <0.001), width (21.3 ± 7.6 versus 20.3 ± 6.4 um, p value <0.001) as well as turbidity (0.291 ± 0.157 versus 0.308 ± 0.141 ± 0.162, p value <0.001). Logistic regression analysis revealed that combining the systemic characteristics of age, sex, systolic blood pressure, estimated glomerular filtration rate, hemoglobin A1c level, and history of hypertension and dyslipidemia could indicate the presence of DR (AUC = 0.81, P = 0.006). Furthermore, the discriminative power of DR was significantly improved (P = 0.03) by adding NC length to the systemic findings (AUC = 0.89, P < 0.001).

STATISTICAL ANALYSIS

The data was entered in Microsoft Excel spreadsheet and analysis was done using Epi-Info, JASP and Statistical Package for Social Sciences (SPSS) version 23.0.

Continuous variables are represented as mean ± SD or medians with Inter-quartile range. Categorical variables are represented as number and percentage (%). The variables were tested for normality with the Shapiro Wilk test for normality, Q-Q plots, visual inspection of the histograms and the z-scores for the degree of skewness and kurtosis. Spearman Rank correlation test was used to assess correlation between continuous quantitative variables. Scatter diagrams were used to describe the relationship between two quantitative variables. Not all variables met the assumptions required for parametric; therefore, non-parametric tests (i.e., Mann-Whitney test, Spearman correlation) were used for all analyses for consistency. Appropriate graphs such as pie charts, bar diagrams and histograms have been constructed. All tests of significance were two-tailed and statistical significance was defined as $P < 0.05$.

AIM

To study Nailfold Capillary (NFC) changes in patients of Type 2 Diabetes Mellitus.

OBJECTIVES

- To study the NFC changes in type 2 diabetes mellitus
- To study association of NFC changes with microvascular and macro-vascular complications of type 2 diabetes

OUTCOME VARIABLES

Primary Outcome Variables:

- Prevalence and type of NFC changes in type 2 diabetes mellitus

Secondary Outcome Variables:

- Association of NFC changes with microvascular and macro-vascular complications of type 2 diabetes
- Association of NFC changes with ASCVD* risk score[8]

*ASCVD - atherosclerotic cardiovascular disease, defined as a nonfatal myocardial infarction, coronary heart disease, or stroke

MATERIALS AND METHODS

This was a cross-sectional observational study was conducted in Department of General Medicine, Maulana Azad Medical College and associated Lok Nayak Hospital, New Delhi from 1st January 2021 to 31st May 2022. Patients presenting to the Medicine or Diabetic OPD were screened for participation in our study. Subjects were categorized as diabetic based on the ADA 2021 criteria. The subjects who met the ADA criteria for Type 2 DM (fasting plasma glucose of ≥126 mg/dl or an HbA1c of ≥6.5%, as measured on 2 separate occasions) and were willing to give informed consent for participation in the study, were included. Subjects with the following conditions were excluded :

- History of Raynaud phenomenon, connective tissue disease
- Current use of drugs such as glucocorticoids and oral contraceptives
- Occupations with a risk of micro-trauma (e.g., farmer and gardener)
- Cancer
- Active liver disease
- Current pregnancy
- Active infection
- Any skin disease precluding the use of nailfold capillaroscopy

After the application of appropriate inclusion and exclusion criteria, a total of 65 subjects were included in the study. Subjects were evaluated as per standardized protocol:

SOCIO-DEMOGRAPHIC ASSESSMENT

Subjects were interviewed by the examiner using a standardized proforma. They were asked details such as age, date of birth, gender, occupation, address and contact details.

CLINICAL ASSESSMENT

Subjects were evaluated by taking exhaustive history related to diabetes such as age of onset of diabetes, major events, level of glycemic control, drugs being used for treatment, past history of stroke, coronary artery disease, angina, coronary interventions, family history of premature coronary artery disease, smoking and alcohol use. A general physical examination including assessment of pulse, BP measurement, Height, Weight, measurement and calculation of Waist:Hip ratio was performed for each subject. This was followed by a comprehensive systemic examination.

ASSESSMENT FOR ASCVD RISK FACTORS AND DIABETIC COMPLICATIONS

The ASCVD Risk Score was calculated using the online ACC/AHA ASCVD risk calculator. We screened for various macrovascular and micro vascular complications on the basis of history, examination and laboratory investigations. Cerebrovascular accident was determined on the basis of history provided by the subjects. The presence of ischemic heart disease was determined on the basis of history of coronary event or intervention in the past or 2D echocardiography. Diabetic nephropathy was determined on the basis of albumin to creatinine ratio > 30 in spot urine samples and eGFR < 60 (CKD EPI 2021 equation). The presence of diabetic retinopathy was made on the basis of a fundus examination by a trained ophthalmologist at our affiliated hospital - Guru Nanak Eye Centre. The severity of diabetic retinopathy was recorded according to the Early Treatment of Diabetic Retinopathy Study (ETDRS) classification. The presence of diabetic neuropathy was determined on the basis of absence of vibration perception using a 128-Hz tuning fork, absence of

bilateral ankle reflexes and inability to perceive pressure with a 10-g monofilament on the dorsal aspect of the great toe.

NAILFOLD VIDEO CAPILLAROSCOPY

After 20 minutes resting at a room temperature of 20–24 degrees Celsius, immersion oil was applied on the nailfold of all participants for better visualization. A physician who was blinded to the patient's condition, performed the Capillaroscopy on 8 digits (excluding the thumbs) of all participants at 200x magnification using a capillaroscopy device. 4 images (1 × 1 mm in size) from the middle of the nailfold in each finger was recorded on the video capillaroscopy device for each patient. The nailfold capillaries were assessed for various characteristics such as Capillary distribution, Density and morphology according to the Maricq criteria modified by Bergman et al.

The abnormalities in capillaroscopic findings were defined as follows:

- Tortuosity: 2 or more cross capillaries, each over 1 mm in length
- Neoangiogenesis: Tortuous, bush-like capillaries with marked heterogeneity in size (a) as the presence of extremely tortuous, bushy, branching, ramified and coiled capillaries(b) ≥4 capillaries within a single dermal papilla, and (c) thin and branching interconnected capillaries originating from a single loop
- Microhemorrhages: ≥2 punctate bleeds around a single capillary in at least 2 fingers (separate or confluent microhemorrhage areas)
- Extravasation: Leakage of capillary content
- Avascular area: Loss of at least 2 consecutive capillaries or ≤6 capillaries over each 1 mm length

- Ectatic capillaries: Capillary wall diameter between 0.02 and 0.05 micrometers (regular or irregular)
- Megacapillary: Capillary wall diameter >0.05 micrometers.

LABORATORY EVALUATION

A 10 mL of venous blood sample was obtained using venipuncture in each subject and subjected to the following biochemical investigations

- Fasting blood sugar
- HbA1c
- Renal function tests (Blood urea and serum creatinine level)
- Lipid levels (Total cholesterol, LDL cholesterol, Triglycerides, HDL cholesterol)
- hsCRP
- Serum Uric acid
- Lipoprotein (a)