Neutrophil to Lymphocyte Ratio as a Risk Marker in Madhumehajanit NetraRoga (Diabetic Retinopathy)

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Abstract:

Ayurveda is a holistic ancient discipline of health management that describes numerous theories about ailments and how to control them. Present day lifestyle i.e. change in eating habits, progressive weight gain, stress, and a sedentary life style lead to increase in number of lifestyle disorders. One of such lifestyle related disorders is Diabetes Mellitus. Madhumeha is noted in Ayurveda as a form of Prameha that can be linked to diabetes in modern medical research. Ayurvedic scriptures contain information regarding the effects of Madhumeha, and some manifestations have been linked to modern nomenclature. If Diabetes Mellitus, Madhumeha not controlled later on leads to Neuropathy, Nephropathy, Cardiomyopathy and Retinopathy. Diabetic Retinopathy (DR) is considered as a most threatening complication of Diabetes Mellitus. In this regard, Ayurveda developed the idea of Madhumehajanit Netra Roga, which has been linked to diabetic retinopathy. The current study attempts to explore the similarities and differences between the various components of Madhumehajanit Netra Roga, i.e., diabetic retinopathy and its stages, based on Dosha Dushya in Madhumeha pathogenesis. The "Neutrophil Lymphocyte Ratio" has been described as a diagnostic sign in diabetic retinopathy by modern science. The purpose of this pilot study was to assess the Neutrophil to Lymphocyte Ratio as a Risk Marker in Madhumehajanit netraroga (Diabetic Retinopathy) in Madhumeha (Diabetes mellitus) patients.

Keywords:

Ayurveda, Cataract, Diabetes Mellitus, Diabetic Retinopathy, Neutrophil Lymphocyte Ratio, Netra Roga, Optic Neuropathy, ,etc.

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Introduction:

The neutrophil-lymphocyte ratio (NLR) is an inflammatory biomarker that can be used to detect systemic inflammation. It is calculated by dividing the absolute number of neutrophils by the absolute number of lymphocytes.

It is a simple measure that does not increase the cost of standard blood count laboratory checks in hospitals. The NLR has been used to predict the prognosis of several disorders, including cancer, community pneumonia, and sepsis. [1]

Diabetic retinopathy, optic neuropathy, cataract, and dry eye are among ocular consequences of diabetes mellitus (DM). Many researches have been published in order to detect the damage caused by diabetes in the optic nerve and visual pathway.

A systematic study and meta-analysis predicted that there were 132.12 million diabetic retinopathies worldwide in 2020, with that number expected to rise to 160.5 million by 2045. Diabetes retinopathy is most common in Africa (35.90%). [2]

Diabetic retinopathy (DR) was previously classified as a diabetic microvascular condition. Vascular modifications have been detected, including changes in retinal artery diameter, architectural indices, and blood flow. Elevated serum levels of certain blood markers and cytokines have been identified as early indicators of DR. DR was later identified as a neurovascular dysfunction that is not evident with an ophthalmoscope. [3] In addition to the caused microvascular damage, hyperglycemia and its associated metabolic disturbances have a number of negative consequences on the retinal neurovascular structure, including the optic nerve, glia, and immune cells. [4] It was discovered that in diabetic patients, neuronal dysfunction across the retina precedes clinical vasculopathy. This could open up new avenues for disaster recovery management. [5]

Pre-clinical diabetic retinopathy (P-DR) retinal neurodegenerative alterations have been documented. Apoptosis of retinal neuronal cells, along with activation of glial cells, resulted in a dramatic decrease in the thickness of the retinal nerve fiber layer, particularly at the edge of the optic disc, parapapillary. Diabetic papilopathy, optic disc neovascularization, and optic nerve atrophy are clinical manifestations of optic nerve changes in DR. [6]

Diabetic Nephropathy (DN) is a microvascular consequence of diabetes that is the major cause of end-stage renal disease (ESRD) in India. Diabetic nephropathy typically begins with microalbuminuria and progresses to macroalbuminuria. Microalbuminuria is thought to be an early indicator of DN. DN generally progresses irreversibly from overt proteinuria to renal failure. As a result, detecting microalbuminuria as early as feasible in the course of the disease becomes critical. [7]

The American Diabetes Association (ADA) requires regular screening for diabetic nephropathy. Inflammatory mechanisms are important in the pathophysiology of type 2 diabetes complications. They also significantly anticipate the development of the disease and may serve

as an early signal. Subclinical inflammation worsens metabolic abnormalities and contributes to the development of vascular problems. [8]

There is additional evidence that chronic inflammation may have a role in the development and progression of micro and macro angiopathy in diabetes patients. Microalbuminuria detection is the current gold standard marker for early detection of DN, but it is not a sufficiently reliable predictor of DN due to some of its own limitations. Many diabetic nephropathy biomarkers (classified as glomerular indicators, tubular markers, oxidative stress markers, inflammation, and miscellaneous markers) have been found, but they still lack clinical value in detecting the condition. As a result, it is probable that renal impairment began long before the identification of microalbuminuria, and appropriate therapies may be delayed in such patients. [9]

As a result, there is an urgent need for credible early predictors of diabetic nephropathy. TNF- and IL-1 biomarkers, which are implicated in the initiation and progression of DN, have been found to play a predictive role in the early detection of DN. However, these are not frequently utilized in India because they are highly expensive, difficult to obtain, and unapproachable in the basic care context. As a result, there is a need for a low-cost, easily accessible research that can serve as a marker for the inflammatory process. [10]

The Neutrophil - Lymphocyte ratio (NLR), which can be computed using readily available blood counts, is one such investigation that has been assessed by this study. NLR could be regarded as a sign of persistent inflammation. It is made up of two markers: neutrophils, which are the active nonspecific mediators that initiate the first line of defense, and lymphocytes, which are the regulatory or protective components of inflammation. In Egyptian patients, NLR has been shown to be a simple and cost-effective study. As a result, the predictive ability of NLR in the diagnosis of diabetic nephropathy among Indian patients was investigated in this study. [11]

The Institutional Ethics Committee approved the project. The sample size was determined as 127 (considering a prevalence of diabetic nephropathy of 27% from the previous study, an error of 5%, and an error of 20%). Over a three-month period, the study was conducted as a single-center, cross-sectional observational study. Patients were regarded as having diabetes mellitus if they had a HbA1c level of 6.5% or more at any point in time and were eligible to participate in the trial. The study excluded patients with type 1 diabetes, gestational diabetes, secondary diabetes, or other forms of monogenic diabetes.

Also excluded were patients with known coronary artery disease, heart failure, acute or chronic infections, acute cerebrovascular illnesses, acute and chronic poisonings, malignancies, haematological diseases, and chronic inflammatory diseases. Pregnant women, patients with other diseases that affect urinary protein excretion, such as nephritic / nephrotic syndrome, urolithiasis, chronic kidney disease caused by causes other than diabetes, urinary tract infections, diseases affecting renal blood flow, and patients on medications known to alter white blood cell counts and urinary protein excretion, were also excluded from the study. Blood

samples were taken for complete blood counts with differential counts, blood urea, serum creatinine, and HbA1c calculation after a detailed history and clinical examination. The TPUC GEN 3, Cobas C, Integra kit was used to calculate urine PCR (Protein Creatinine Ratio) in a spot urine sample. The NLR was computed based on the blood counts. The estimated Glomerular Filtration Rate (EGFR) was computed using the MDRD equation: GFR (in mL/min/1.73 m2) = 175 (Serum creatinine)-1.154 (Age)-0.203 (0.742 if female). [12]

Diabetic retinopathy is the most frequent type of diabetic eye disease and can result in visual loss. Diabetic retinopathy is a type of diabetic retinopathy in which fluctuations in blood glucose levels cause changes in retinal blood vessels. These vessels will become damaged and will leak fluid into the back of the eye. This can result in haemorrhages, exudates, and even retinal edoema. Diabetic retinopathy causes platelet stickiness, capillary pericyte loss, retinal ischemia, haemorrhage, and retinal edoema. Medical research has described various criteria that can aid in the diagnosis of diabetes complications such as retinopathy. The "Neutrophil Lymphocyte Ratio" is a helpful inflammatory marker that predicts negative outcomes in diabetes and associated comorbidities, such as diabetic retinopathy. The neutrophil lymphocyte ratio (NLR), an effective and persistent inflammatory marker, can be used to predict the occurrence of microvascular diabetes problems. [13] In diabetic patients with microvascular problems (retinopathy, nephropathy, and peripheral neuropathy), Neutrophil Lymphocyte Ratio levels will be much greater. Several epidemiological studies have previously found a link between persistent low-grade inflammation and diabetes mellitus. It is made up of two markers: neutrophils, which are the active nonspecific mediators that initiate the first line of defense, and lymphocytes, which are the regulatory or protective components of inflammation. [14]

Objectives:

Neutrophil to Lymphocyte Ratio as a Risk Marker in Madhumehajanit NetraRoga (Diabetic Retinopathy) in Madhumeha (Diabetes mellitus) patients.

To analyze Ayurveda's preventive and therapeutic measures for diagnosis Diabetic Retinopathy.

Review of Literature:

According to the International Diabetes Federation, 463 million persons worldwide were diagnosed with diabetes mellitus (DM) in 2019, with the disease burden expected to reach 700 million by 2045. India currently ranks second after China in terms of diabetes prevalence, with an estimated 77 million diabetics. Diabetes is the leading cause of chronic kidney disease (CKD) in adults in India, and with diabetes prevalence expected to rise in the coming years, the magnitude of both CKD and End Stage Renal Disease (ESRD) is also likely to rise, contributing to morbidity and mortality increases.2 In the Global Burden of Disease study, CKD was ranked globally 16th and 8 th in India among the causes of deaths. [15]

Prameha/Madhumehajanya (Diabetic retinopathy) and its stages. To establish a probable etiopathogenesis of the disease from Ayurveda perspective, all the important literature of both modern medicine and Ayurveda along with online sources were searched and analyzed. All the three dosha along with Raktadosha and Saptadhatu with four internal Dristipatals of eye are affected in Madhumehajanya timir in different stages of the disease. Avarana and Dhatu kshaya play important role in development of diabetic retinopathy due to prolonged and uncontrolled hyperglycemia. [16] Agnimandya related Ama formation has a role in pathology of diabetic retinopathy which is quite similar to oxidative theory of diabetic retinopathy explained in modern pathology. Urdhwaga raktapitta, Ojas kshaya, Raktavritta vata, and Pranavritta vyana are other causes in development of diabetic retinopathy. [17]

Samprapti Madhumehajanit Netra Roga:

Netra is Tejo mahabhoota pradhana i.e. pitta dominant in origin. According to Acharya Charaka and Vagbhata, eye has a fear from Kapha dosha.[18,19]

Any organ with Pitta origin, if get Avrudha by Kapha dosha will definitely lead to Srotoavarodha. We can see the involvement of all type of Sroto dusti i.e, Atipravruthi, Sanga, Siragranthi, Vimarga gamana in modern pathogenesis of NPDR and PDR as: Any occlusion in theretinal vessels considered as Sanga. Development of micro-aneurysms can be correlated to Siragranthi, Retinal hemorrhage and Neovascularisation to Vimarga Gamana and Maculopathy and Retinal oedema to Atipravruthi.

Thickening of capillary:

Kapha dosha plays a significant role in Madhumeha. The Panchbotika composition of Kapha consists of Prithvi and Jal component.[20] With the increase in Kapha i.e. Prithvi and Jal component there is thickening of capillary basement membrane.

Capillary endothelial cell damage and loss of capillary pericytes: Diabetic Retinopathy is a microangiopathy which affects the retinal precapillary arterioles, capillaries and venules. Precapillary arterioles, capillaries and venules are the types of vessels. Vessels are made up of tissue, elastin fibres and smooth muscle cells.[21] Kapha dosha has a property of Sandhibandhan that is why all these factors are intact with each other.[22] The vitiation of Kapha dosha hampers the normal functioning of Kapha dosha which in turn causes Sandhibandhan vikriti.[23] It means there will be damage to endothelial cells of retinal capillaries and loss of capillary pericytes due to vitiated Kapha dosha.

Classification of Diabetic Retinopathy:

Diabetic Retinopathy has been variously classified. Presently followed classification is as follows[24]:

I. Non Proliferative Diabetic Retinopathy (NPDR)

- Mild NPDR
- Moderate NPDR
- Severe NPDR
- Very severe NPDR

II. Proliferative Diabetic Retinopathy (PDR)

III. III. Diabetic Maculopathy

IV. IV. Advanced Diabetic eye diseases

Diabetic retinopathy is micro-angiopathy affecting retinal pre capillary, arterioles, capillaries and venules.

Research Methodology:

This study's overall design was exploratory. The purpose of this pilot study was to assess the role of "Neutrophil Lymphocyte Ratio" in the diagnosis of Madhumehajanit Netra Roga (Diabetic Retinopathy) in Madhumeha (Diabetes mellitus) patients. The patients in the control group had normal NLR ratios and no indications of diabetic retinopathy (visual problems). However, patients in the test (case) group had a greater NLR ratio and some indications of diabetic retinopathy. Investigations indicated mild to severe diabetic retinopathy in the test group with a greater NLR ratio. Diabetic retinopathy abnormalities in test group participants can be related to vascular consequences of persistent diabetes mellitus. The varying NLR ratio ranges in the control and test groups revealed that this ratio may increase in diabetic retinopathy. As a result, the NLR ratio can be utilised as a diagnostic sign to validate the presence of diabetic retinopathy. This study concluded that the NLR ratio has diagnostic use in diabetic retinopathy; nevertheless, a large-scale population study is needed to investigate the role of other parameters. [25]

Result and Discussion:

Neutrophil–Lymphocyte Ratio as a Reliable Marker to Predict Pre-Clinical Retinopathy Among Type 2 Diabetic Patients:

The neutrophil-lymphocyte ratio, band neutrophils, and total leukocytes were all measured to indicate sepsis. The ROC curve values for NLR were 0.62 (95% CI 0.55 - 0.69), 0.98 (95% CI 0.97 - 1.0), and 0.51 (95% CI 0.44 - 0.59) for total leukocytes. Table 3 shows the performance of the NLR, total leucocytes, and band neutrophils in predicting sepsis in ICUs. The best performance was recorded for the band neutrophil count, followed by the NLR (Table 1). [26]

Table1: Sensitivity, specificity and positive and negative predictive values for the prediction of sepsis on admission to the intensive care unit.

Parameter	Sensitivity %(95%CI)	Specificity %(95%CI)	PPV %(95%CI)	NPV %(95%CI)
NLR > 5.0	81.0 (75.3 –	35.5(29.3 – 42.0)	55.6(52.8 –	65.1(57.6 –

	85.8)		58.4)	71.9)
Band neutrophils > 10%	98.8 (95.9 – 99.9)	63.7(52.6 – 74.1)	85.4(81.4 – 88.6)	96.4(86.9 – 99.1)
Total leukocytes> 12,000	36.7 (31.6 – 42.1)	72.13(66.6 – 77.1)	59.5(53.9 – 64.9)	50.6(47.9 – 53.2)
Total leukocytes> 4,000	6.9 (4 - 11)	95.2(91.6 – 97.6)	59.3(40.8 – 75.4)	40.6(49.4 – 51.7)
95%CI– 95% confidence interval; PPV– positive predictive value; NPV- negative predictive value; NLR- neutrophil-lymphocyte ratio.				

Neutrophil-lymphocyte ratio and band neutrophils had a weak (r = 0.2), positive and statistically significant correlation with length of hospital stay ($p < 0.05$); however, with time of hospitalization in the unit, a weak, positive and statistically significant correlation was found only with NLR ($r = 0.3$ and $p < 0.05$). [27]

The diabetic patients were then split into two groups: those with P-DR (28 patients, 46.66%) and those without (32 patients, 53.33%) (Table 2). There was no statistically significant difference between the two groups in terms of gender ($\chi 2 = 0.424$ at $p = 0.515$) or BMI ($t = 1.577$ at $p = 0.120$). [28] Patients with P-DR, on the other hand, were older ($t = 2.67$, $p = 0.010$) and had a longer diabetes duration ($U = 283.0$, $p = 0.014$) than those without P-DR. Smoking was found to be substantially linked with P-DR ($\chi 2 = 6.687$ at $p = 0.010$). The use of oral hypoglycemic medications was found to have a significant connection with P-DR, although insulin users did not ($p = 0.042$; $\chi 2 = 4.115$). [29]

Table 2 Comparison between diabetics with and without P-DR according to PRVEPs P100 wave latency and IOLD:

PRVEP P100 (Latency)	Diabetics with P-DR (n-28)	Diabetics without P-DR (n=32)	Test of sig	*p*
Binocular field				
Mean ± SD	113 ± 7.70	99.63 ± 3.20	*t= 8.565**	<0.001*
Right monocular				
Mean ± SD	114 ± 8.83	101 ± 2.92	t =7.254*	<0.001*
Left monocular				
Mean ± SD	116 ± 6.91	101 ± 3.14	t =10.967*	<0.001*
IOLD				
Mean ± SD	4.0(0-16)	1.4(0-3)	u =213.5*	<0.001*
SD Standard deviating, *t* Student *t*-test, *U* Mann-Whitney test, IOLD Interocular latency difference, *pp*-value for comparing diabetics with and without P-DR *Statistically significant at p≤0.05				

Furthermore, patients with P-DR scored higher on the Toronto clinical neuropathy score scale (U = 220.0, p = 0.001). The Toronto severity grade was higher in the P-DR group (2 = 11.292, p < 0.001). [30]

Table 3 compares the diabetic group with P-DR to the diabetic group without P-DR in terms of laboratory results. Patients with P-DR had significantly higher HbA1C and NLR levels.

Table 3 compares diabetics with and without P-DR based on test results. [31]

	P-DR (*n*=28)	without P-DR (*n*=32)	Test of Sig	*p*
HbA1C (%)				
Mean ± SD	8.46 ± 1.66	7.27 ± 1.28	*t= 3.152**	0.003*
NLR				
Mean ± SD	2.33(1.48-6.79)	1.48(1.08-5.30)	*U* = 112.5*	<0.001*
SD Standard deviating, *t* Student *t*-test, *U* Mann-Whitney test, *pp*-value for comparing between diabetics with and without P-DR *Statistically significant at p ≤ 0.05				

Females comprised 47% of the study population (Figure 1). The majority of the patients were between the ages of 51 and 60. (See Figure 1) [32]

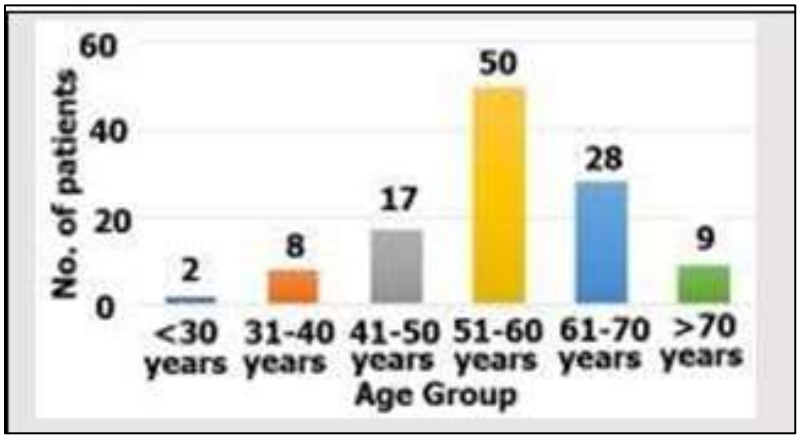

Figure 1: Most of the patients were in the age group of 51- 60 years.

The study population's average age was 57.39 10.6 years. The majority of the patients had diabetes for 6 to 15 years (Figure 2). Diabetes mellitus had been present in 83% of the study population for less than ten years. Diabetes lasted an average of 7.9 5.1 years. The patients' mean BMI was 25.8 4.7 kg/m2. The patients' mean blood creatinine level was 1.1 1.0 mg/dL. [33]

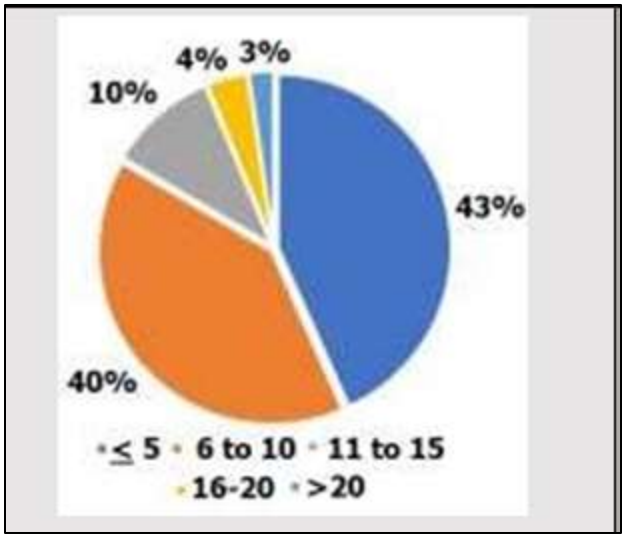

Figure 2: Age of The Study Population

The study population's mean eGFR was 91.6 48.7 mL/min. The mean protein creatinine ratio (PCR) was 0.8 0.4, with 66% of patients taking oral hypoglycemic medications and 18% taking both insulin and oral medications. Figure 3 depicts a scatter pattern for the Urine PCR and NLR. [34]

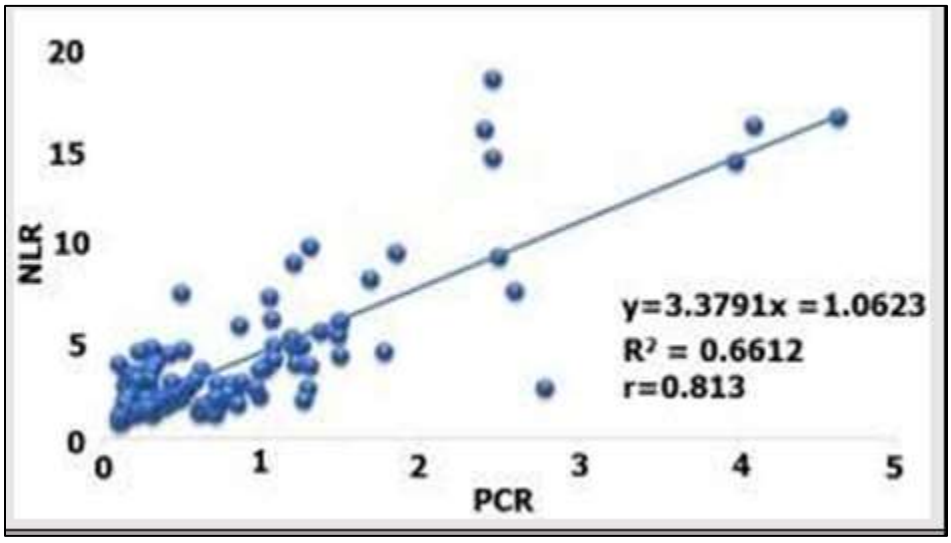

Figure 3: Scatter Plot for the Urine PCR and NLR

Association study demonstrated a statistically significant positive association between urine PCR and NLR (r=0.813, p0.0001). The relationship between eGFR and NLR was statistically significant (r = -0.214, p = 0.006). Using the ROC curve, an NLR with a cut off of 7 exhibited a sensitivity and specificity of 88.89% and 94.9% in predicting diabetic nephropathy (AUC = 93.7%) with a positive likelihood ratio of 17.481 and a negative likelihood ratio of 0.117. [35]

Diabetic Retinopathy:

Diabetic retinopathy is a condition that may occur in people who have diabetes. It causes progressive damage to the retina, the light-sensitive lining at the back of the eye.

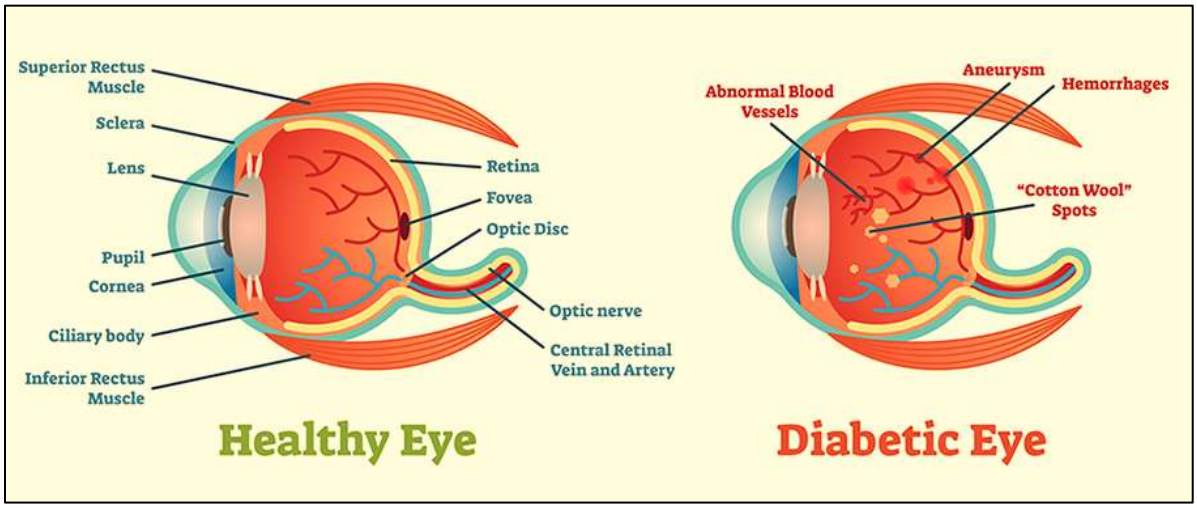

Figure 4: Diabetic Retinopathy

Diabetic retinopathy is a potentially blinding consequence of diabetes. Diabetes impairs the body's capacity to utilize and store sugar (glucose). Too much sugar in the blood causes the condition, which can cause damage throughout the body, including the eyes. Diabetes causes small blood vessels throughout the body, including the retina, to deteriorate over time. When these tiny blood vessels leak blood and other fluids, diabetic retinopathy develops. The retinal tissue swells as a result, resulting in foggy or blurred vision.

Diabetes retinopathy almost always affects both eyes. Diabetes retinopathy is more likely to develop as a person's diabetes progresses. Diabetic retinopathy can lead to blindness if left untreated. When persons with diabetes have high blood sugar levels for an extended length of time, fluid can build up in the lens inside the eye, which governs focusing. This alters the curvature of the lens, resulting in visual alterations. Once blood sugar levels are under control, the lens usually returns to its former shape and eyesight improves. Diabetic patients who can better control their blood sugar levels will have a slower onset and progression of diabetic retinopathy. [36]

Conclusion:

Our study provided evidence that the neutrophil-lymphocyte ratio and band neutrophils can be used in conjunction with other indicators to detect sepsis early. The criteria described in this study are low-cost and simple for health-care providers to adopt, which encourages their usage in low- and middle-income countries like Brazil. Sepsis is a complex physiological illness, and other laboratory criteria, such as blood culture and inflammatory indicators such as C-reactive protein or prolactin, should be assessed to ensure a valid diagnosis. Visual evoked potentials play an important role in evaluating the visual pathway in diabetics and diagnosing pre-clinical

diabetic retinopathy before structural damage occurs. The neutrophil-lymphocyte ratio has a high sensitivity (89.29%) and specificity (84.37%) for detecting pre-clinical diabetic retinopathy. Finding an accurate and widely available laboratory test to predict P-DR could help diabetic individuals avoid major ocular problems.

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