

Original Research

Study to find out the role of neutrophil lymphocyte ratio and platelet lymphocyte ratio as diagnostic biomarkers for gestational diabetes mellitus – A cross-sectional study

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Abstract

Introduction: Gestational diabetes mellitus (GDM) is a condition leading to the development of glucose intolerance for the first time in pregnancy. Early detection of this condition can prevent the morbidity and mortality of women with GDM. Aim: To find out the role of NLR ratio and PLR ratio as a diagnostic marker in the development of GDM. **Materials and methods:** It was a cross-sectional study done among patients attending Chettinad Hospital and Research Institute among 156 pregnant women. They were divided into two groups based on the OGTT 2 hour values into post glucose values into those with values >140 mg/dl (60) and those with values <140 mg/dl (96). HbA1c, post glucose values total blood count, platelet count, RBC count, PCV, neutrophils, lymphocytes, eosinophils, basophils, lymphocytes, and monocytes were measured among all the patients. **Results:** Mean and SD were calculated for all the variables. An independent sample t-test was done between the groups. Post glucose, TLC (total leucocyte count) MCV (mean corpuscular volume), MCHC (mean corpuscular hemoglobin concentration), NLR ratio and PLR ratio showed a significant differences between the two groups. **Conclusion:** There was a significant difference in the levels of NLR ratio and PLR ratio between GDM women and normal pregnant women. These parameters can be used as a diagnostic tool for GDM.

Key words: gestational diabetes mellitus, neutrophil lymphocyte ratio, platelet lymphocyte ratio.

Introduction

Gestational diabetes mellitus (GDM) is defined as carbohydrate intolerance resulting in hyperglycemia with the first onset during pregnancy [1]. GDM affects approximately 10% of pregnant women with the percentages varying based on the country and the diagnostic criteria [2].

GDM increases the chances of developing perinatal complications such as shoulder dystocia, perinatal asphyxia, macrosomia, neonatal complications such as hypoglycemia, hyperbilirubinemia, and hypocalcemia, and maternal problems like pre-eclampsia and urinary tract

infections. So the early detection and management of GDM are very necessary to decrease the morbidity among the mother and fetus.

Cost-effective and feasible tests which can be done easily will help in improving the detection rate of GDM among pregnant women. GDM is usually diagnosed by the OGTT (oral glucose tolerance test) using the one-step or two-step procedure.

Previous studies have shown the role of inflammation in the development of GDM. The role of inflammatory markers like IL6, TNF α , resistin, visfatin have been studied, but all these tests are very costly and not feasible to



people belonging to low socioeconomic status especially in a country like India.

NLR (neutrophil-lymphocyte ratio) ratio and PLR (platelet lymphocyte ratio) ratio are very inexpensive and can be done very easily in all the clinical laboratories in India. These ratios have been studied as markers for inflammation over the past few years. They are proven to have a role in the detection of metastasis of cancers, inflammatory bowel disease, in the detection of heart diseases and also to assess the risks associated with diabetes mellitus [3–5].

NLR ratio alone has been studied as an independent marker for all the complications of pregnancy like pre-eclampsia, hyperemesis gravidarum, ectopic pregnancy and pre-term delivery and also in GDM but still, its role is inconclusive in the diagnosis of many of these diseases [6–10].

This study was aimed at finding out the role of NLR ratio and PLR ratio in the diagnosis of GDM patients and to find out the changes in the erythropoietic parameters among pregnant women and their relationship to the development of GDM.

Material and methods

Ethical committee approval

This study was conducted after obtaining consent from Institute Ethical Committee. It was a cross-sectional study done among 156 pregnant women of the age group of 20–40 years attending the Obstetrics and Gynaecology OPD of Chettinad Hospital and Research Institute, Chengalpattu from July 2019 to December 2019 for a period of 6 months. **Inclusion criteria:** All the patients who came to perform OGTT between the gestational period of 24–28 weeks were included in the study. **Exclusion criteria:** Those patients on steroid, anemic patients, patients having associated renal disease or hemoglobin variants and those with any other inflammatory diseases, those who were not willing for the study were excluded from the study.

Laboratory, anthropometric and clinical data collection

Obstetric history and family history were taken from all patients. About 5 ml of blood was collected from all the patients in violet-colored vacutainer tubes for assessing the levels of HbA1c levels and complete blood count. DIPSI (Diabetes in a pregnancy study group of India) guidelines are followed for diagnosing GDM in our hospital. All the pregnant women were given 75 g of anhydrous glucose mixed in 300 ml of water to drink over 5–10 minutes irrespective of their previous meal status. All women were advised to come after 2 hours and give their blood sample for taking post-prandial blood sugar levels. If the PPBS values were ≥ 140 mg/dl, they were investigated for gestational diabetes mellitus. HbA1c was measured using a chromatographic method using D-10 machine in Biochemistry laboratory. Complete blood count was measured in the pathology laboratory in Beckman Coulter LH 780 machine. Total blood count, platelet count, RBC count, and PCV (packed cell volume) were measured using the impedance method. Neutrophils, lymphocytes, eosinophils, basophils, lymphocytes, and monocytes were measured using the colorimetric method. MCV, MCHC, MCH and RDW were calculated using formulas. MCV values were calculated using the formula $PCV/RBC \times 10$, MCH using $Hb/RBC \times 10$, MCHC using $Hb/PCV \times 100$. Hemoglobin was measured using the cyanmethemoglobin method.

Statistical analysis

The mean \pm SD of all the variables was taken. The sample was divided based on the post glucose values into two groups as those with values above and less than 140 mg/dl. Ninety-six patients belonged to the group with post glucose values < 140 mg/dl and 60 patients were present with post glucose values > 140 mg/dl. An independent sample t-test was done between the two groups. NLR ratio was calculated by taking the ratio of neutrophil count to the lymphocyte count. Platelet lymphocyte count (PLR) was

Table 1: Show the mean±SD of all the variables in the study.

Variables	Mean	SD
Hemoglobin (g/dl)	11.156	1.3596
Post-glucose	120.017	23.0700
HbA1c	5.075	0.4115
TLC (total leucocyte count)	9190.517	2451.0860
RBC	4.0293	0.41599
PCV (packed cell volume)	34.224	4.2001
MCV (mean corpuscular volume)	84.067	10.4339
MCH (mean corpuscular hemoglobin)	28.297	6.4484
MCHC (mean corpuscular hemoglobin concentration)	32.653	0.8847
RDW (red cell distribution width)	15.525	2.7980
Platelet count	2.7553	0.63759
Neutrophils	67.372	9.4181
Eosinophils	2.122	1.8150
Basophils	.364	0.2148
Lymphocytes	22.401	5.1809
Monocytes	6.699	2.0696
NLR ratio	3.3	1.3
PLR ratio		

NLR – neutrophil lymphocyte ratio, PLR – platelet lymphocyte ratio.

Table 2: Pearson’s correlation analysis between NLR ratio and post-glucose values.

		NLR ratio	PLR ratio
Post-glucose	R value	-0.14	0.009
	P value	0.08	0.91

calculated by taking the ratio of platelet to lymphocytes. Correlation analysis was done between NLR ratio, PLR ratio, and post glucose values.

Result

Discussion

GDM is a clinical condition characterized by the development of hyperglycemia during pregnancy. The exact cause for this condition is not known still. Theories suggested the relationship of inflammation with the development of

GDM. The main characteristic feature of GDM is insulin resistance. The secretion of hormones like human chorionic gonadotrophin (HCG), cortisol, etc., plays an important role in the development of insulin resistance [11, 12]. Additionally, the excess deposition of adipose tissue also contributes to the above condition by the excess secretion of cytokines like resistin, leptin TNF alpha [13].

NLR ratio is routinely done the test to assess the presence of systemic inflammation and to assess neoplastic diseases [14, 16, 17]. It’s very cheap and can be easily calculated. Studies done by Park JJ, Jang HJ, Oh IY, et al. showed a

Table 3: Independent sample t-test comparing the variables in the two groups.

Variables	PG<140		PG>140		p-Value
	Mean	SD	Mean	SD	
Hemoglobin (g/dl)	11.12	1.37	11.335	1.32	0.34
Post-glucose (mg/dl)	112.3	16.2	156.9	13.12	0.0001
HbA1c	5.06	0.39	5.15	0.5	0.09
TLC (total leucocyte count)	9008	2500	10065	1999.7	0.006
RBC	4.2335	0.36	4.007	0.42	0.05
PCV (packed cell volume)	34.107	4.3683	34.956	2.9403	0.15
MCV (mean corpuscular volume)	84.643	7.8461	80.469	20.3057	0.005
MCH (mean corpuscular hemoglobin)	27.743	3.0149	31.763	15.6187	0.001
MCHC (mean corpuscular hemoglobin concentration)	32.68	0.81	32.5	1.16	0.37
RDW (red cell distribution width)	15.52	2.89	15.54	2.27	0.75
Platelet count (PC)	2.71	0.64	2.97	0.6	0.97
Neutrophils (%)	67.82	8.78	65.21	11.84	0.17
Eosinophils (%)	2.16	1.91	1.92	1.16	0.39
Basophils (%)	0.370	0.23	0.33	0.09	0.89
Lymphocytes (%)	22.14	5.4	23.63	3.9	0.06
Monocytes (%)	6.69	1.86	6.813	2.9	0.66
NLR ratio	2.9	0.8	3.4	1.4	0.001
PLR ratio	0.1	0.1	0.3	0.1	0.001

PG – post glucose, NLR – neutrophil lymphocyte ratio, PLR – platelet lymphocyte ratio.

relationship between patients presenting with myocardial infarction and NLR ratio [15]. Buyuk-kaya et al. showed a relationship between metabolic syndrome and NLR ratio. Hence this simple ratio is being investigated in the diagnosis of a wide range of diseases.

In our study, we found out a significant difference in the values of NLR/PLR ratio between GDM women and normal pregnant women. A study done by Fahima SM, et al. showed that there was no significant difference in the levels of NLR ratio between women with and without GDM, a finding similar to our study [18]. Sargin MA, et al. did a study among 762 pregnant women that showed that there was no statistically significant difference in the levels of NLR ratio between the GDM subjects and those without GDM [19]. But a study done by Yilmaz, et al. showed a statistically significant difference in the levels of NLR ratio between the two groups [20].

Pearson's correlation analysis showed a negative correlation between NLR ratio and post-glucose values and a positive correlation between PLR ratio and post-glucose values but was not statistically significant.

In this study, the levels of total leucocyte counts were higher among GDM patients compared to normal pregnant women and the difference was statistically significant. A study done by Wolf, et al. [21] showed a significant difference in the levels of leucocyte counts in the first trimester between GDM patients and non-GDM patients and they did a linear correlation between the NLR ratio and abnormal GCT's and OGTT in the second trimester. Through their findings, they ascertained the role of inflammation in the development of GDM.

The findings in our study point towards the role of inflammation in the development of GDM, some of the previous studies have

supported the findings in our study and some are not on par with our findings.

Conclusion

This study has shown that there is a significant difference in the values of NLR ratio and PLR ratio between GDM and normal pregnant women but further studies are required to prove its role as a diagnostic marker for gestational diabetes mellitus.

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Conflict of Interest

No conflict of interest.

References

- Metzger, B. E., Buchanan, T. A., Coustan, D. R., et al. (2007). Summary and recommendations of the Fifth International Workshop-Conference on Gestational Diabetes Mellitus. *Diabetes Care*. 30(SUPPL. 2).
- Ryan, E. A. (2011). Diagnosing gestational diabetes. *Diabetologia*. 54(3):480–486.
- Wang, D., Yang, J. X., Cao, D. Y., et al. (2013). Preoperative neutrophil-lymphocyte and platelet-lymphocyte ratios as independent predictors of cervical stromal involvement in surgically treated endometrioid adenocarcinoma. *Onco Targets Ther*. 6:211–216.
- Celikbilek, M., Dogan, S., Ozbakir, O., et al. (2013). Neutrophil-lymphocyte ratio as a predictor of disease severity in ulcerative colitis. *J Clin Lab Anal*. 27(1):72–76.
- Aygün, F., Efe, D. (2015). Association of neutrophil/lymphocyte ratio with obstructive coronary artery disease and coronary artery calcium score detected by multislice computed tomography in type 2 diabetes mellitus patients. *Patient Prefer Adherence*. 9:1023–1031.
- Yilmaz, H., Celik, H. T., Namuslu, M., et al. (2014). Benefits of the neutrophil-to-lymphocyte ratio for the prediction of gestational diabetes mellitus in pregnant women. *Exp Clin Endocrinol Diabetes*. 122(1):39–43.
- Sifnaios, E., Mastorakos, G., Psarra, K., et al. (2019). Gestational diabetes and T-cell(Th1/Th2/Th17/Treg) immune profile. *In Vivo (Brooklyn)*. 33(1):31–40.
- Hai, L., Hu, Z.-D. (2020). The clinical utility of neutrophil to lymphocyte ratio in pregnancy related complications: a mini-review. *J Lab Precis Med*. 5:1–1.
- Akin, S., Aydin, Z., Yilmaz, G., Aliustaoglu, M., Keskin, O. (2019). Evaluation of the relationship between glycaemic regulation parameters and neutrophil-to-lymphocyte ratio in type 2 diabetic patients. *Eur Med J*. 91–96.
- Kim, M. A., Lee, B. S., Park, Y. W., Seo, K. (2011). Serum markers for prediction of spontaneous preterm delivery in preterm labour. *Eur J Clin Invest*. 41(7):773–780.
- Di Cianni, G., Miccoli, R., Volpe, L., et al. (2003). Intermediate metabolism in normal pregnancy and in gestational diabetes. *Diabetes Metab Res Rev*. 19:259–270.
- Asemi, Z., Samimi, M., Tabassi, Z., et al. (2013). A randomized controlled clinical trial investigating the effect of DASH diet on insulin resistance, inflammation, and oxidative stress in gestational diabetes. *Nutrition* 29:619–624.
- Festa, A., D'Agostino, R. Jr., Howard, G., et al. (2000). Chronic subclinical inflammation as part of the insulin resistance syndrome: the Insulin Resistance Atherosclerosis Study (IRAS). *Circulation* 102:42–47.
- de Jager, C. P., van Wijk, P. T., Mathoera, R. B., et al. (2010). Lymphocytopenia and neutrophil-lymphocyte count ratio predict bacteremia better than conventional infection markers in an emergency care unit. *Crit Care*. 14:R192.
- Park, J. J., Jang, H. J., Oh, I. Y., et al. (2013). Prognostic value of neutrophil to lymphocyte ratio in patients presenting with ST-elevation myocardial infarction undergoing primary percutaneous coronary intervention. *Am J Cardiol*. 111:636–642.
- Dan, J., Zhang, Y., Peng, Z., et al. (2013). Postoperative neutrophil-to-lymphocyte ratio change predicts survival of patients with small hepatocellular carcinoma undergoing radiofrequency ablation. *PLoS One*. 8:e58184.
- Buyukkaya, E., Karakas, M. F., Karakas, E., et al. (2012). Correlation of neutrophil to lymphocyte ratio with the presence and severity of metabolic syndrome. *Clin Appl Thromb Hemost*. 18.
- Fahima, S. M., De Silvab, W. V., Abeykoona, W. (2021). Role of neutrophil to lymphocyte ratio at booking visit in prediction of gestational diabetes. *Sri Lanka J Obstetr Gynaecol*. 43:16–21.
- Yilmaz, H., Celik, H. T., Namuslu, M., et al. (2014). Benefits of the neutrophil-to-lymphocyte ratio for the prediction of gestational diabetes mellitus in pregnant women. *Exp Clin Endocrinol Diabetes*. 122(1):39–43.
- Sargin, M. A., Yassa, M., Taymur, B. D., Celik, A., Ergun, E., Tug, N. (2016). Neutrophil-to-lymphocyte and platelet-to-lymphocyte ratios: are they useful for predicting gestational diabetes mellitus during pregnancy? *Ther Clin Risk Manag*. 12:657.
- Wolf, M., Sauk, J., Shah, A., et al. (2004). Inflammation and glucose intolerance: a prospective study of gestational diabetes mellitus. *Diabetes Care*. 27(1):21–27.