Research Article

Elevated Maternal Serum Folate Concentrations are Associated with Risk of Developing Gestational Diabetes Mellitus

Rehnuma Tasnim[1*], Md. Nurullah[2], A. K. M Shahidur Rahman[3], Moushumi Akhter[4], Laila Israt Jahan[5], Tania Patwary[6], Mohammad Monzurul Alam Bhuiyan[7], Sheuly Ferdoushi[8], Md. Saiful Islam[9], Md Quddusur Rahman[9]

Abstract

Background: Gestational diabetes mellitus (GDM) is a metabolic disorder and one of the common complications in all pregnancies. Folate serves in synthesis of methionine from homocysteine. High folate level inhibits DNA synthesis and elevates homocysteine. An elevated serum homocysteine level linked with insulin resistance which is associated with GDM.

Objective: This study was aimed to examine the association of serum folate concentrations with its risk of developing GDM.

Methods: This study was conducted at the Department of Laboratory Medicine, Bangabandhu Sheikh Mujib Medical University (BSMMU), Dhaka, Bangladesh from March 2021 to February 2022. A total of 43 pregnant women with GDM were selected as cases and 43 healthy pregnant women were taken as control group. The demographic profile and a full clinical history of the study population were recorded accordingly. Maternal serum folate concentrations were estimated following standard procedure. Data were analyzed and compared by statistical tests.

Results: The mean(±SD) age of the GDM women was 31.35±3.02 years and that was 25.19±2.65 years in control group. It was found that mean(±SD) serum folate concentrations was significantly high in GDM women (23.36±10.20 nmol/L versus 10.86±8.34 nmol/L, p= 0.001). In multivariate logistic regression analysis the odd ratio of serum folate with GDM was 4.977 (95% CI; p= 0.031). There was a significant positive correlation observed between serum folate concentrations of GDM women and their plasma glucose levels [fasting plasma glucose (r=+0.597, p<0.001); plasma glucose 1 hour after 75 gm glucose (r=+0.492, p= 0.001) and plasma glucose 2 hours after 75 gm glucose (r=+0.449, p=0.003)].

Conclusion: Maternal serum folate concentration is significantly higher in patients with GDM women than healthy pregnant women. High maternal serum folate concentrations are associated with risk of developing GDM. Serum folate may be used as a predictive tool to identify risk of developing GDM.

Keywords: Gestational Diabetes Mellitus (GDM), Pregnant Women, Risk, Serum Folate Concentrations

Introduction

Gestational diabetes mellitus (GDM) is a common condition in pregnancy which is defined as any degree of glucose intolerance with onset or first recognition during pregnancy [1]. Worldwide approximately 17% pregnancies

Affiliation:

[1]Junior Consultant of Laboratory Medicine, Labaid Limited (Diagnostics), Dhaka, Bangladesh

[2]Assistant Surgeon, Officer on Special Duty (OSD), Directorate General of Health Services (DGHS), Dhaka, Bangladesh

[3]Medical Officer, Department of Nephrology, Bangabandhu Sheikh Mujib Medical University (BSMMU), Dhaka, Bangladesh

[4]Consultant of Laboratory Medicine, Obstetrical and Gynaecological Society of Bangladesh (OGSB) and Institute Rotary Cancer Hospital (IRCH), Dhaka, Bangladesh

[5]Consultant of Pathology, ENT and Head Neck Cancer Hospital and Institute, Dhaka, Bangladesh

[6]Consultant, COVID- 19 Unit, Department of Microbiology, Mugda Medical College and Hospital, Dhaka, Bangladesh

[7]Assistant Professor, Department of Laboratory Medicine, Bangabandhu Sheikh Mujib Medical University (BSMMU), Dhaka, Bangladesh

[8]Associatet Professor, Department of Laboratory Medicine, Bangabandhu Sheikh Mujib Medical University (BSMMU), Dhaka, Bangladesh

[9]Professor, Department of Laboratory Medicine, Bangabandhu Sheikh Mujib Medical University (BSMMU), Dhaka, Bangladesh

*Corresponding author:
Dr. Rehnuma Tasnim, Junior Consultant of Laboratory Medicine, Labaid Limited (Diagnostics), Dhaka, Bangladesh,

E-mail: rehnumatasnim63@gmail.com

are affected by GDM [2, 3]. GDM is diagnosed when fasting plasma glucose is 5.1 mmol/L, or 1 hour plasma glucose is 10.0 mmol/L and 2 hours plasma glucose is 8.5 mmol/L following a 75g oral glucose load [4]. Hormones secreted during pregnancy (like estrogen, progesterone, cortisol and placental lactogen etc) thought to prevents the body from using insulin effectively [5]. It was reported that, GDM and impaired glucose tolerance (IGT) during pregnancy are associated with future metabolic dysfunction and diabetes [6]. There are several risk factors responsible for developing GDM which include- obesity, increased maternal age, previous history of GDM, history of diabetes in 1st degree relatives and certain Asian and African ethnic women etc [7]. GDM has many adverse consequences on the maternal and fetal health like- abortion, preeclampsia, polyhydramnios and increased risk of perinatal morbidity [8]. Folic acid is a water soluble member of vitamin B family used to prevent neural tube defects in fetus [9]. It aids in the one-carbon unit transfer for DNA methylation and synthesis of purine and thymidine neucleotides which is essential for cell division and fetal growth during pregnancy [10]. High serum folate levels lead to elevate body homocysteine level [11]. Elevated homocysteine concentrations impair endothelial function in skeletal muscles, adipose tissue and liver, thus reducing insulin delivery to these insulin-sensitive tissues [12-15]. Therefore, homocysteine at elevated concentrations has been linked with insulin resistance [12-13]. Elevated homocysteine concentrations also play a harmful role in pancreatic β-cell metabolism and insulin secretion [13-16]. GDM is a common complication in pregnancy and is associated with increase maternal and neonatal morbidity [17]. During recent years it has been suggested that high folate concentrations associated with higher risk of GDM [16-19]. Folic acid consumption in early pregnancy may increase the risk of GDM [20]. Identifying and treating women with increased risk of GDM is important to improve the pregnancy outcomes. It may be helpful to prevent maternal and fetal complications due to GDM by monitoring serum folate level. In this background, current study aimed to evaluate the association of serum folate with GDM.

Methods

Study design

This cross-sectional study was conducted at the Department of Laboratory Medicine, Bangabandhu Sheikh Mujib Medical University (BSMMU), Dhaka, Bangladesh from March 2021 to February 2022. This study was approved by the Institutional Review Board (IRB) of BSSMU, Dhaka, Bangladesh.

Study population

A total of 43 pregnant women with gestational diabetes mellitus (GDM) were selected as case (group I) according to

the selection criteria. At the same time 43 normal pregnant women were taken as control group (group II). Diagnosed cases of GDM women from 28th weeks onward of gestation and apparently normal pregnant women from 28th weeks onward of gestation were included. Patient with history of preeclampsia, diagnosed cases of chronic renal disease/liver disease/cardiovascular disease/diabetes mellitus, patient with history of previous GDM and patient with malabsorption syndrome were excluded from the study.

Study procedure

Informed written consent was taken from each study subject prior to enrollment. A detail case history, clinical examination and relevant investigations of all study population were done accordingly. Their body mass index (BMI) was calculated along with last menstrual period, expected date of delivery, gestational age by ultrasonogram and antenatal check up were noted. In both groups gestational age was estimated from last menstrual date and confirmed by early ultrasonogram. A separate data collection sheet was used for each subject to maintain confidentiality. The highest levels of confidentiality and ethical standards were maintained during the analysis of the data.

Diagnosis of GDM

Among the study population GDM was leveled by following criteria [4]:

- Fasting plasma glucose: 5.1 mmol/L

- 1 hour plasma glucose: 10.0 mmol/L following a 75gm oral glucose load

- 2 hours plasma glucose: 8.5 mmol/L following a 75gm oral glucose load

Blood sample collection and analysis

With all aseptic precaution an overnight fasting (8-12 hours) venous blood (5ml) from ante-cubital vein was collected from each study subject. Each subject was then allowed to drink glucose water (75 gm glucose in 300 ml of water). They were requested not to take any food or beverage and be rested for two hours. Then after 1 hour and 2 hours of glucose intake the 2nd and 3rd blood samples (3 ml venous blood each time) were taken. The blood samples were transferred into clean tubes. Then the test tubes were taken to the Laboratory medicine of BSMMU within half an hour of collection of blood. All collected blood samples were kept in upright position for 30 minutes, thereafter each blood sample was centrifuged at 3000 rpm for 5 minutes (in room temperature: 22˚C - 24˚C). Then separated serum/plasma was collected in a sample cup or eppendorf and marked accordingly. All separated serum/plasma was stored in -20˚C temperature until analysis was done. Blood glucose was assayed by glucose-oxidase method in automated biochemistry analyzer SIEMENS Dimension EXL with LM

on the principle of photometric technique. Serum folate level was measured by immunochemistry auto-analyzer (Cobas e411) using the Electro-chemiluminescence Immunoassay (ECLIA) with commercially available cartridge.

Normal range of serum folate concentrations during pregnancy [18]

The normal reference range of serum folate concentrations during pregnancy was: 4.1-22.3 nmol/L

Statistical analysis

All collected data were cross-checked and verified to reduce inconsistency. Then data were edited, coded and entered into computer. Data was analyzed by windows based software program Statistical Package for Social Sciences (SPSS) version- 26. All qualitative data were expressed as mean with standard deviation (±SD) and quantitative data were expressed as frequency with percentage. Unpaired t-test, Chi-squared Test (χ^2), Pearson's correlation coefficient test and multivariate logistic regression analysis were performed for the statistical analysis of data. A p value <0.05 was considered as statistically significant.

Results and Observations

This study was intended to assess the association of maternal serum folate level with GDM. A total of 86 pregnant women were enrolled in this study. Among the 86 study subjects 43 pregnant women who had GDM from 28 weeks onward was categorized as group I and another 43 pregnant women who had normal pregnancy from 28 weeks onward was categorized as group II. The mean age was 31.35±3.02 years in group I and that was 25.19±2.65 years in group II, the age difference was statistically significant (p<0.001). It was observed that no patient 0(0.0%) belonged to age 20-25 years in group I and 22(51.2%) patients were belonged to that age in group II. Age belonged to 26-30 years was found 19(44.2%) pregnant women in group I and 21(48.8%) in group II. Age belonged to 31-35 years was found 19(44.2%) and 0(0.0%) in group I and group II respectively. In group I; 5(11.6%) pregnant women were belonged to 36-40 years but no pregnant women in group II belonged to this age limit (Table- 1).

Table- 2 shows the distribution of the study patients according to gestational age confirmed by ultrasonogram (USG). It was observed that majority of the study subjects [30(69.8%)] had 28-36 weeks of gestation in group I and 36(83.7%) study subjects had that age of gestation in group II. But 13 (30.2%) subjects had ≥37 weeks of gestation in group I and 7(16.3%) subjects had ≥37 weeks of gestation in group II. The mean gestational age was found 33.7±3.19 weeks in group I and that was 32.9±3.01 weeks in group II. The difference was not significant between two groups (p= 0.228) (Table- 2).

Table 1: Age distribution of the study subjects (N= 86)

Age (years)	Group I (n=43)		Group II (n=43)		p value
	n	%	n	%	
20-25	0	0	22	51.2	
26-30	19	44.2	21	48.8	
31-35	19	42.2	0	0	
36-40	5	11.6	0	0	
Mean±SD (years)	31.35±3.02		25.19±2.65		
Range (minimum – maximum)	(27-38) years		(20-30) years		<0.001s

Data were expressed as frequency with percentage and mean±SD, Unpaired Student t-test was performed to compare between two groups, s= significant

Table 2: Distribution of the study subjects according to the gestational age by ultrasonogram (N= 86)

Variable	Group I (n=43)		Group II (n=43)		p value
	n	%	n	%	
28-36 weeks of gestation	30	69.8	36	83.7	
≥ 37 weeks of gestation	13	30.2	7	16.3	
Mean gestational age (weeks)	33.7±3.19		32.9±3.01		
Range (minimum – maximum)	(28-39) weeks		(28-38) weeks		0.228ns

Data were expressed as frequency with percentage and mean±SD, Unpaired Student t-test was performed to compare between two groups, ns= not significant

It was observed that; 22(51.2%) study subjects had normal (4.1-22.3 nmol/L) serum folate concentration in group I and 36(83.7%) study subjects had normal (4.1-22.3 nmol/L) serum folate concentration in group II. On the other hand, 21(48.8%) study subjects had increased (>22.3 nmol/L) serum folate concentration in group I and only 7(16.3%) study subjects had increased (>22.3 nmol/L) serum folate concentration in group II, which was statistically significant (p= 0.001). The mean serum folate concentration was significantly higher in group I than group II (23.36±10.20 nmol/L versus 10.86±8.34 nmol/L, p=0.001) (Table- 3).

Table- 4 delineates the independent predictors for gestational diabetes mellitus (GDM). A multivariate logistic regression analysis was applied to assess the risk factors for developing GDM. Serum folate levels with age and body mass index (BMI) were found to be independent predictors for GDM. The result showed that serum folate was 4 times more risk of developing GDM, OR of 4.977 (95% CI .992-24.975; p= 0.031), older age was 2 times more risk for developing GDM, OR of 2.786 (95% CI 1.188-5.546; p=0.021) and body mass index (BMI) was 2 times more risk for developing GDM, OR of 2.567 (95% CI 1.182-5.573; p=0.017) (Table- 4).

Correlation between plasma glucose levels and serum folate concentrations were analyzed with Pearson's Correlation co-efficient test. It was observed that, there was a significant positive correlation between fasting plasma glucose level with serum folate concentrations in GDM women (Pearson's Correlation co-efficient, r=+0.597, p<0.001). There was a significant positive correlation between plasma glucose level 1 hour after 75 gm glucose and serum folate concentrations in GDM women (Pearson's Correlation co-efficient, r=+0.492, p=0.001). There was also a significant positive correlation between plasma glucose level 2 hours after 75 gm glucose and serum folate concentrations in GDM women (Pearson's Correlation co-efficient, r=+0.449, p=0.003) (Table- 5).

Table 3: Comparison of serum folate concentrations between two groups of study subjects (N= 86)

Serum folate concentrations (nmol/L)	Group I (n=43)		Group II (n=43)		p value
	N	%	n	%	
Normal (4.1-22.3 nmol/L)	22	51.2	36	83.7	*0.001s
Raised (>22.3 nmol/L)	21	48.8	7	16.3	
Mean±SD	23.36±10.20		10.86±8.34		**0.001s

Data were expressed as frequency with percentage and mean±SD, *Chi-square test and **unpaired Student t-test were performed to compare between two groups, s= significant

Table 4: Independent predictors for assessing risk factor of developing GDM by multivariate logistic regression analysis (N= 86)

Variables	Odds ratio	95% CI	p-value
Age (>30 years)	2.786	1.188-5.546	0.021s
Body mass index BMI (>25 kg/m²)	2.567	1.182-5.573	0.017s
Gestational age (≥28 weeks)	0.805	0.165-3.93	0.789ns
Serum folate (>22.3 nmol/L)	4.977	0.992-24.975	0.031s

s=significant, ns= not significant

Table 5: Correlation of serum folate concentration with fasting plasma glucose, plasma glucose 1 hour after 75 gm glucose and 2 hours after 75 gm glucose levels in pregnant women with GDM (n= 43)

Variables	r value	p value
Fasting plasma glucose	0.597	<0.001
Plasma glucose 1 hour after 75 gm glucose	0.492	0.001
Plasma glucose 2 hours after 75 gm glucose	0.449	0.003

Pearson's correlation test was performed

Discussion

Gestational diabetes mellitus (GDM) refers to any degree of glucose intolerance that is first recognized during pregnancy [1]. GDM is one of the common complications of pregnancy that has harmful effect on maternal and fetal health [21-22]. Folic acid which is a member of water soluble vitamin B family and is widely used in early pregnancies to prevent neural tube defects of fetus [23]. It has been reported that maternal serum folate level linked to GDM [18-20, 24]. However, most of the previous studies yielded with inconsistent results. In this background, current study aimed to assess the association of serum folate concentrations with its risk of developing GDM. A total of 86 pregnant women with 28th weeks onward gestational ages were enrolled; of them 43 had GDM and rest 43 were normal pregnant women. It was observed that the mean age was significantly high among GDM women (31.35±3.02 years versus 25.19±2.65 years, p<0.001). This result was consistent with similar previous studies [18-20, 25]. In this current study the mean gestational age was 33.7±3.19 weeks in GDM women and that was 32.9±3.01 weeks in normal pregnant women. The mean gestational age was not significantly different between the groups (p= 0.228). In this context, Mustary F *et al.* found that mean gestational age of their GDM cases was 36.58 ± 2.34 weeks and 35.9±2.01 weeks in normal pregnant women which was not significant (p= 0.59), that was consistent with this present study [26].

In this study, among the GDM women 48.8% had high serum folate concentrations and among the normal pregnant women only 16.3% had high serum folate concentrations. It was found that the mean serum folate concentration was significantly higher in GDM women than that of normal pregnant women (23.36±10.20 nmol/L versus 10.86±8.34 nmol/L, p= 0.001). This finding was supported by a couple of previous study [27-28]. A multivariate logistic regression analysis was done to investigate the association of maternal serum folate concentrations with risk of developing GDM after adjusting for the effect of age, gestational age and body mass index (BMI). Serum folate with age and BMI were found to be independent predictors for GDM. The result showed that serum folate was 4 times more risk of developing GDM, OR= 4.977 (95% CI .992-24.975; p= 0.031). Xie K *et al.* showed that women with high serum folate concentrations had high odd ratio and increased risk of GDM (OR = 2.76, 95% CI = 1.56–4.89, p<0.001) [25]. This present study showed older age was 2 times more risk for developing GDM [OR=2.786 (95% CI 1.188-5.546; p=0.021)]. Lai JS *et al.* found that older women had a significantly higher risk for GDM [OR=2.67, 95%CI 2.13–3.34; p<0.001)] [18]. In present study BMI was 2 times more risk for developing GDM [OR of 2.567 (95% CI 1.182-5.573; p=0.017)]. Similarly, Lai JS *et al.* found that increased BMI had 5 times more risk of developing GDM

Citation: Tasnim R, Nurullah M, Rahman AKMS, Akhter M, Jahan LI, Patwary T, Bhuiyan MMA, Ferdoushi S, Islam MS, Rahman MQ. Elevated Maternal Serum Folate Concentrations are Associated with Risk of Developing Gestational Diabetes Mellitus. Fortune Journal of Health Sciences. 6 (2023): 521-526.

[OR=5.36, (95% CI 4.53–6.36; p<0.001)] [18]. The results of this current study were comparable with these previous studies. In this current study, Pearson's correlation tests revealed that, there was a significant positive correlation between fasting plasma glucose, 1 hour after 75 gm glucose and 2 hours after 75 gm glucose levels with serum folate concentrations (Pearson's Correlation co-efficient r=+0.597, r=+0.492 and r=+0.449 respectively; p<0.001, p= 0.001 and p= 0.003 respectively).

Conclusion

This study concluded that GDM women had higher serum folate concentrations than normal pregnant women. Present study indicates that high maternal serum folate increase the risk of developing GDM. Serum folate concentrations may be used as a predictive tool to identify risk of developing GDM.

Limitations of the study

It was a single centre study with a small sample size.

Recommendation

A multicentre study with large population size is recommended for the better evaluation of serum folate concentrations in GDM patients.

Conflicts of interest

Authors declared no conflict of interest regarding this publication.

References

1. Care D. Standards of medical care in diabetes 2019. Diabetes Care 42 (2019):S124-38.

2. McIntyre HD, Catalano P, Zhang C, Desoye G, Mathiesen ER, Damm P. Gestational diabetes mellitus. Nature reviews Disease primers 11 (2019):47.

3. Jesmin S, Akter S, Akashi H, Al-Mamun A, Rahman MA, Islam MM, et al. Screening for gestational diabetes mellitus and its prevalence in Bangladesh. Diabetes research and clinical practice 103 (2014):57-62.

4. American Diabetes Association 14. Management of diabetes in pregnancy: standards of medical care in diabetes—2021. Diabetes care 44 (2021):S200-10.

5. Lipschitz DA, Cook JD, Finch CA. A clinical evaluation of serum ferritin as an index of iron stores. New England Journal of Medicine 290 (1974):1213-6.

6. Stuebe AM, Mantzoros C, Kleinman K, Gillman MW, Rifas-Shiman S, Seely EW, et al. Gestational glucose tolerance and maternal metabolic profile at 3 years postpartum. Obstetrics & Gynecology 18 (2011):1065-73.

7. Kiani F, Naz MS, Sayehmiri F, Sayehmiri K, Zali H. The risk factors of gestational diabetes mellitus: a systematic review and meta-analysis study. diabetes 10 (2017):17.

8. Ornoy A. Prenatal origin of obesity and their complications: Gestational diabetes, maternal overweight and the paradoxical effects of fetal growth restriction and macrosomia. Reproductive toxicology 32 (2011):205-12.

9. Atta CA, Fiest KM, Frolkis AD, Jette N, Pringsheim T, St Germaine-Smith C, et al. Global birth prevalence of spina bifida by folic acid fortification status: a systematic review and meta-analysis. American journal of public health 106 (2016):e24-34.

10. Ly A, Hoyt L, Crowell J, Kim YI. Folate and DNA methylation. Antioxidants & redox signaling 17 (2012):302-26.

11. Selhub J, Morris MS, Jacques PF. In vitamin B12 deficiency, higher serum folate is associated with increased total homocysteine and methylmalonic acid concentrations. Proceedings of the National Academy of Sciences 104 (2007):19995-20000.

12. Weiss N, Heydrick SJ, Postea O, Keller C, Keaney Jr JF, Loscalzo J. Influence of hyperhomocysteinemia on the cellular redox state–impact on homocysteine-induced endothelial dysfunction.

13. Meigs JB, Jacques PF, Selhub J, Singer DE, Nathan DM, Rifai N, et al. Fasting plasma homocysteine levels in the insulin resistance syndrome: the Framingham offspring study. Diabetes care 24 (2001):1403-10.

14. Serné EH, Stehouwer CD, Ter Maaten JC, Ter Wee PM, Rauwerda JA, Donker AJ, et al. Microvascular function relates to insulin sensitivity and blood pressure in normal subjects. Circulation 99 (1999):896-902.

15. Pinkney JH, Stehouwer CD, Coppack SW, Yudkin JS. Endothelial dysfunction: cause of the insulin resistance syndrome. Diabetes 46 (1997):S9-13.

16. Engel SM, Joubert BR, Wu MC, Olshan AF, Håberg SE, Ueland PM, et al. Neonatal genome-wide methylation patterns in relation to birth weight in the Norwegian Mother and Child Cohort. American journal of epidemiology 179 (2014):834-42.

17. Metzger BE, Lowe LP, Dyer AR, Trimble ER, Sheridan B, Hod M, et al. Hyperglycemia and Adverse Pregnancy Outcome (HAPO) Study: associations with neonatal anthropometrics. Diabetes 58 (2009):453-9.

18. Lai JS, Pang WW, Cai S, Lee YS, Chan JK, Shek LP, et al. High folate and low vitamin B12 status during pregnancy is associated with gestational diabetes mellitus. Clinical nutrition 37 (2018):940-7.

19. Yang Y, Cai Z, Zhang J. Association between maternal

folate status and gestational diabetes mellitus. Food science & nutrition 9 (2021):2042-52.

20. Zhu B, Ge X, Huang K, Mao L, Yan S, Xu Y, et al. Folic acid supplement intake in early pregnancy increases risk of gestational diabetes mellitus: evidence from a prospective cohort study. Diabetes Care 39 (2016):e36-7.

21. Catalano PM, McIntyre HD, Cruickshank JK, McCance DR, Dyer AR, Metzger BE, et al. The hyperglycemia and adverse pregnancy outcome study: associations of GDM and obesity with pregnancy outcomes. Diabetes care 35 (2012):780-6.

22. Damm P, Houshmand-Oeregaard A, Kelstrup L, Lauenborg J, Mathiesen ER, Clausen TD. Gestational diabetes mellitus and long-term consequences for mother and offspring: a view from Denmark. Diabetologia 59 (2016):1396-9.

23. Bibbins-Domingo K, Grossman DC, Curry SJ, Davidson KW, Epling JW, García FA, et al. Folic acid supplementation for the prevention of neural tube defects: US preventive services task force recommendation statement. Jama 317 (2017):183-9.

24. Li S, Hou Y, Yan X, Wang Y, Shi C, Wu X, et al. Joint effects of folate and vitamin B12 imbalance with maternal characteristics on gestational diabetes mellitus. Journal of diabetes 11 (2019):744-51.

25. Xie K, Xu P, Fu Z, Gu X, Li H, Cui X, et al. Association of maternal folate status in the second trimester of pregnancy with the risk of gestational diabetes mellitus. Food Science & Nutrition 7 (2019):3759-65.

26. Mustary F, Chowdhury TA, Begum F, Mahjabeen N. Maternal and perinatal outcome in gestational diabetes mellitus compared to pregestational diabetes mellitus. BIRDEM Medical Journal 9 (2019):127-32.

27. Berglund SK, García-Valdés L, Torres-Espinola FJ, Segura MT, Martínez-Zaldívar C, Aguilar MJ, et al. Maternal, fetal and perinatal alterations associated with obesity, overweight and gestational diabetes: an observational cohort study (PREOBE). BMC public health 16 (2016):1-2.

28. Saravanan P, Sukumar N, Adaikalakoteswari A, Goljan I, Venkataraman H, Gopinath A, et al. Association of maternal vitamin B 12 and folate levels in early pregnancy with gestational diabetes: a prospective UK cohort study (PRiDE study). Diabetologia64 (2021):2170-82.