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ABOUT COVER

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ORIGINAL ARTICLE

Observational Study

MiRNA-200a and miRNA-200b expression, and vitamin-D level: Prognostic significance in obese non-diabetic and obese type 2 diabetes mellitus individuals

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Abstract

BACKGROUND

Obesity and type 2 diabetes mellitus (T2DM) are frequent co-occurring disorders that affect regular metabolic functions. Obesity has also been linked to an increased risk of developing diabetes. Obesity and diabetes are on the rise, increasing healthcare costs and raising mortality rates. Research has revealed that the expression profile of microRNAs (miRNAs) changes as diabetes progresses. Furthermore, vitamin D may have an anti-obesity effect and inverse association with body weight and body mass index (BMI). Low vitamin D levels do not solely cause obesity, which could be a factor in the etiology of T2DM.

AIM

To evaluate miRNA-200a and miRNA-200b expression, and vitamin-D levels in obese and obese T2DM individuals.

METHODS

This study included 210 participants, of which, 82 were obese (BMI > 30 kg/m^2) without T2DM, 28 were obese with T2DM, and 100 were healthy controls. BMI was evaluated and both fasting and postprandial blood glucose were used to confirm T2DM. Exosomal miRNA-200a and miRNA-200b expression were analyzed using real-time PCR using Taqman probes, and vitamin-D levels were

evaluated using an electrochemiluminescence-based immunoassay technique. All data analyses were performed using SPSS 20.0 and GraphPad Prism 5 software.

RESULTS

Overall, a 2.20- and 4.40-fold increase in miRNA-200a and miRNA-200b expression was observed among participants compared to healthy controls. MiRNA-200a and miRNA-200b expression among obese participants increased 2.40-fold and 3.93-fold, respectively, while in obese T2DM participants these values were 2.67-fold, and 5.78-fold, respectively, and these differences were found to be statistically significant ($P = 0.02$) ($P < 0.0001$). Obese participants showed a vitamin D level of 34.27 ng/mL, while in obese-T2DM participants vitamin D level was 22.21 ng/mL ($P < 0.0001$). Vitamin D was negatively correlated with miRNA-200a ($r = -0.22$, $P = 0.01$) and miRNA-200b ($r = -0.19$, $P = 0.04$). MiRNA-200a sensitivity was 75%, and specificity was 57%, with a cutoff value of 2.07-fold. MiRNA-200b sensitivity was 75%, and specificity was 71% with a cutoff value of 4.12-fold, suggesting that miRNA-200a and miRNA-200b with an increased expression of 2.07- and 4.12-fold could be predictive indicators for the risk of diabetes in obese participants.

CONCLUSION

MiRNA-200a and miRNA-200b were higher in diabetic obese participants *vs* non-diabetic obese participants, and insufficient vitamin D levels in obese T2DM participants may be involved in poor clinical outcome.

Key Words: Obesity; Type 2 diabetes mellitus; MiRNA200a; MiRNA200b; Vitamin-D; Prognosis

Core Tip: Obesity and being overweight are widespread health issues in modern societies, and their growing prevalence poses a serious public health challenge due to both the financial burden and health risks involved. In humans, the expression of miRNAs in adipose tissue has been demonstrated to be associated with various metabolic factors, including body mass index, adipogenesis, blood sugar levels, and leptin concentrations. The microRNA (miRNA)-200 family is reported to be more prevalent in human β-cells than in α-cells. The expression of miRNA-200a and miRNA-200b is regulated by thioredoxin-interacting protein, a proapoptotic regulator. It has been suggested that a deficiency in vitamin D is closely associated with obesity and plays a role in the development of insulin resistance and type 2 diabetes mellitus (T2DM). Studies have indicated that vitamin D influences the expression of miRNAs both in adipocytes in laboratory settings and in adipose tissue in living organisms. Therefore, the current work examines miRNA-200a and miRNA-200b expression, and vitamin-D level in obese non-diabetic and obese T2DM individuals.

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INTRODUCTION

Obesity and overweight are two widespread health-related problems in modern societies, and the spread of obesity is also associated with a real public health problem due to its costs and health effects. Indeed, excess body weight increases the likelihood of various metabolic diseases such as heart disease, type 2 diabetes mellitus (T2DM), dyslipidemia, osteoarthritis, and other related diseases[1]. Many microRNAs (miRNAs) are differentially regulated in the white adipose tissue in obese individuals *vs* non-obese individuals[2]. Fatty tissue from the visceral area is more critical for metabolism than subcutaneous tissue[3]. It has been suggested that several miRNAs are expressed differently in subcutaneous and visceral white adipose tissue[4]. In humans, a correlation between the expression of miRNAs in adipose tissue and different metabolic parameters [body mass index (BMI), adipogenesis, glycemia, leptinemia] has been demonstrated[5]. Obesity is associated with inflammation in adipose tissue. The production of inflammatory cytokines can interfere with insulin signaling and contribute to T2DM and other obesity-related diseases[6]. Endocrine activity in adipose tissue is crucial to maintaining an average weight and regulating energy homeostasis. Thus, it is not unexpected that miRNAs may be a new layer of regulation for the different functions of adipose tissue in obesity[7]. A growing body of evidence indicates that a specific set of miRNAs has an altered expression profile during the progression of DM[8]. It has been suggested that the miR-200 family is more abundant in human β-cells compared to α-cells[9]. The expression of miR-200a and miR-200b is controlled by the proapoptotic regulator Txnip (thioredoxin-interacting protein)[10]. The overexpression of miR-200b has been specifically shown to induce apoptosis in INS-1 cells (rat insulinoma cell line) by reducing Zeb1 (zinc finger E-box binding homeobox 1)[10]. Recent work in a mouse model has also suggested the involvement of a miR-

200–Zeb1 axis in regulating epithelial-to-mesenchymal transition and differentiation[11]. Additionally, in mice, the miR-200 family induces β-cell apoptosis by modulating JazF1 (juxtaposed with another zinc finger protein 1)[12]. Vitamin D modulates glycemic homeostasis and is involved in the modulation of glucose-mediated synthesis/secretion of insulin by β-cells, increasing both hepatic and peripheral glucose uptake by direct and indirect mechanisms, and blunting inflammation[13]. It has been shown that vitamin D deficiency is strongly linked to obesity, and it is involved in the development of insulin resistance and T2DM, However, data from intervention studies with vitamin D supplementation to recover insulin resistance and glucose tolerance are still controversial[14]. It has been demonstrated that vitamin D modulates the expression of miRNAs in adipocytes *in vitro* and in adipose tissue *in vivo*[15]. However, very little is known about the role of vitamin D as a mediator of miRNAs and *vice versa*. The functional implications of miRNA-200a and miRNA-200b in human islets are still unclear. It has been suggested that miRNAs are key regulatory factors in controlling gene expression and altering disease conditions. From this perspective, it is essential to understand the contribution of the epigenetic alteration of miRNA-200a and miRNA-200b expression and its relationship with vitamin D in obese and obese T2DM individuals.

MATERIALS AND METHODS

Subject selection, sample collection, and exosome isolation from serum

The current observational study included a total of 210 participants, of which 82 were obese (BMI > 30 kg/m^2) without T2DM, 28 were obese with T2DM, and 100 were healthy controls. Fasting and postprandial blood samples were collected in fluoride vials from all 100 participants to screen for T2DM with obesity. 1 mL of blood was collected in EDTA vials to measure hemoglobin (Hb)A1c and 1 mL of the blood sample was used to measure fasting blood glucose (glucose level 126 mg/dL) and postprandial blood glucose (two-hour blood glucose 200 mg/dL). Blood samples collected in plain vials were centrifuged at 1500 rpm to separate the serum, which was collected and stored at -70°C. Before exosome isolation, the samples were thawed and centrifuged at 3000 rpm to for 10–20 min to pellet and remove cells, debris, and platelets. 1.5 mL of serum was added into 600 µL of precipitation buffer A (miRCURY, Qiagen) and mixed for 30 s. The mixture was incubated for 60 min at 2–8°C and centrifuged at 1500 rpm for 25 min at 20°C. After centrifugation, the pellets were saved and resuspended in 200 µL of resuspension buffer and used for total exosomal RNA extraction. This research study was ethically approved by the Research Ethics Committee, Ministry of Health, (Reg. No: 607-43-1478), Saudi Arabia.

Exosomal RNA extraction, polyadenylation, and cDNA synthesis

Total RNA was extracted from exosomes suspended in resuspension buffer using Trizol and stored at -70°C in RNase-free Eppendorf tubes. Their A260/280 ratio determined the quality and purity of the RNA. According to the manufacturer's protocol, 100 ng of total RNA was used for polyadenylation and cDNA synthesis using the advanced microRNA cDNA Synthesis Kit (TaqMan, Thermo Scientific, United States). The reverse transcriptase enzyme and other essential reagents were subsequently added for cDNA synthesis to transcribe poly (A)-tailed miRNAs into cDNA using the universal RT primer supplied with the manufacturer's kit.

Quantitative real-time-PCR for miRNA-200a and miRNA-200b expression

The miRNA-200a and 200b expression level was calculated using quantitative real-time PCR (qPCR). To calculate the expression, qPCR was performed with TaqMan master mix (4444556), TaqMan probes for miRNA-200a (478752_mir), and TaqMan probes for miRNA-200b (478753_mir) for quantification, and U6 (001973) as an internal control.

Vitamin D level

Serum from all individuals was frozen and then thawed for the electrochemiluminescence-based immunoassay technique to evaluate serum vitamin D levels. Serum 25 (OH) D levels ≤ 20 ng/mL were used to designate vitamin D deficiency, whereas levels < 30 ng/mL were considered insufficient, and levels 30 ng/mL or more were considered sufficient.

Statistical analysis

All data analyses were performed using SPSS 20.0 and GraphPad Prism 5 software. The $2^{(-\text{delta delta ct})}$ method was applied to compute the fold change in expression of miRNA-200a and miRNA-200b in obese non-diabetic and obese T2DM individuals. Cases compared to controls and based on the expression level < 1-fold or > 1-fold were considered low and high, respectively. The χ^2 and Fisher's exact tests were used to compare the groups and vitamin D levels were compared among the obese and obese with T2DM participants. A receiver operating characteristic (ROC) curve was plotted to determine the prognostic importance of miRNA-200a and miRNA-200b among the participants, and $P < 0.05$ was considered statistically significant.

RESULTS

Demographics of the study participants

The study included 100 healthy controls, 82 obese, and 28 obese T2DM individuals. Among the healthy controls, 65% were male, and 35% were female; in obese participants, 70.7% were male, 29.3% were female; in obese T2DM participants,

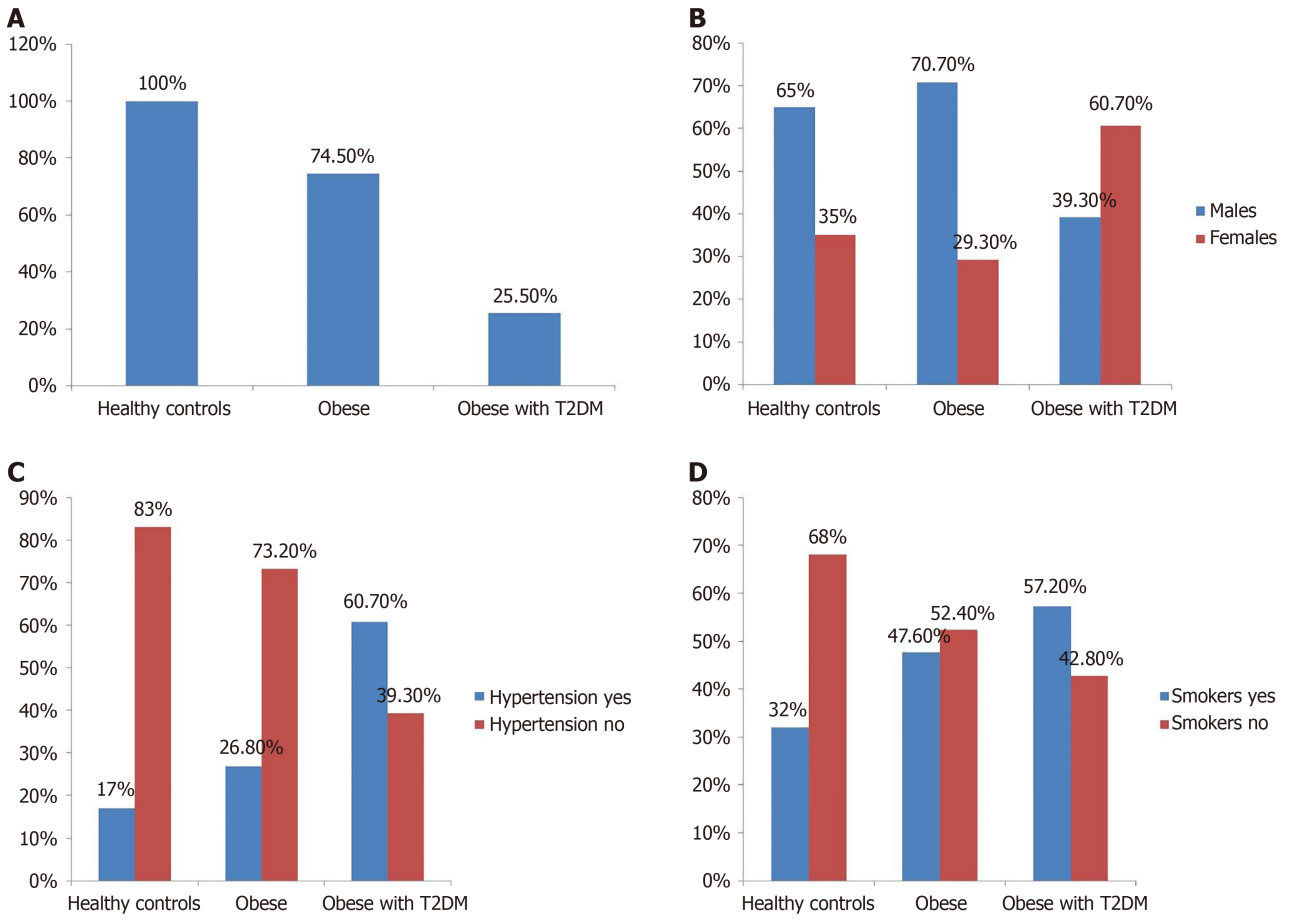

Figure 1 Demographic parameters of the participants. A: Healthy controls, obese and obese with type 2 diabetes mellitus participants; B: Male and female participants in the healthy controls, obese and obese with type 2 diabetes mellitus; C: Hypertensive and non-hypertensive participants in the healthy controls, obese and obese with type 2 diabetes mellitus; D: Smoking and nonsmoking participants in the healthy controls, obese and obese with type 2 diabetes mellitus. T2DM: Type 2 diabetes mellitus.

39.3% were male, and 60.7% were female. Among the healthy controls, 17% were hypertensive, and 83% were non-hypertensive; among the obese participants, 26.8% were hypertensive, 73.2% were non-hypertensive, while in obese T2DM participants, 60.7% were hypertensive, and 39.3% were non-hypertensive. Among the healthy controls, 32% were smokers, and 68% were non-smokers; among the obese participants, 47.6% were smokers, and 52.4% were non-smokers, while in obese T2DM participants, 57.2% were smokers, and 42.8% were non-smokers (Figure 1).

Comparison of biochemical parameters between healthy control, obese, and obese with diabetes participants

Biochemical parameters were compared between the healthy control, obese, and obese with T2DM individuals (Table 1). We compared glucose and lipid parameters, and observed significant differences in fasting blood glucose (P< 0.0001), postprandial glucose (P< 0.0001), and HbA1c (P< 0.0001) among the healthy controls, obese and obese T2DM participants. Lipid parameters such as high-density lipoprotein (HDL) (P< 0.0001), low-density lipoprotein (LDL) (P< 0.0001), triglycerides (TG) (P< 0.0001), cholesterol (P< 0.0001), and very low-density lipoprotein (VLDL) (P< 0.0001) showed significant differences between the three groups.

Comparison of miRNA-200a and miRNA-200b expression in obese and obese T2DM participants

The miRNA-200a and miRNA-200b expression was compared in obese and non-obese T2DM participants (Figure 2). Overall, a 2.20-fold increase in miRNA-200a and a 4.40-fold increase in miRNA-200b were observed in the obese and obese T2DM groups compared to healthy controls. It was observed that miRNA-200a was lower in obese participants (2.40 + 1.04) and higher in obese T2DM participants (2.67 + 1.10) (P = 0.02). A comparison of miRNA-200b was also conducted between obese and non-obese T2DM participants; obese participants had a 3.93-fold increase in miRNA-200b expression while obese T2DM participants had a 5.78-fold increase in miRNA-200b expression (P < 0.0001).

MiRNA-200a and miRNA-200b expression, hypertension, and smoking

We compared miRNA-200a expression with regard to hypertension and smoking status (Figure 3). It was observed that miRNA-200a expression was high in hypertensive participants (2.66 + 1.41) compared to non-hypertensive participants (1.92 + 1.26) and the difference was statistically significant (P = 0.008). An important difference was observed in miRNA-200a with regard to smoking status among the participants. We also compared miRNA-200b expression in terms of

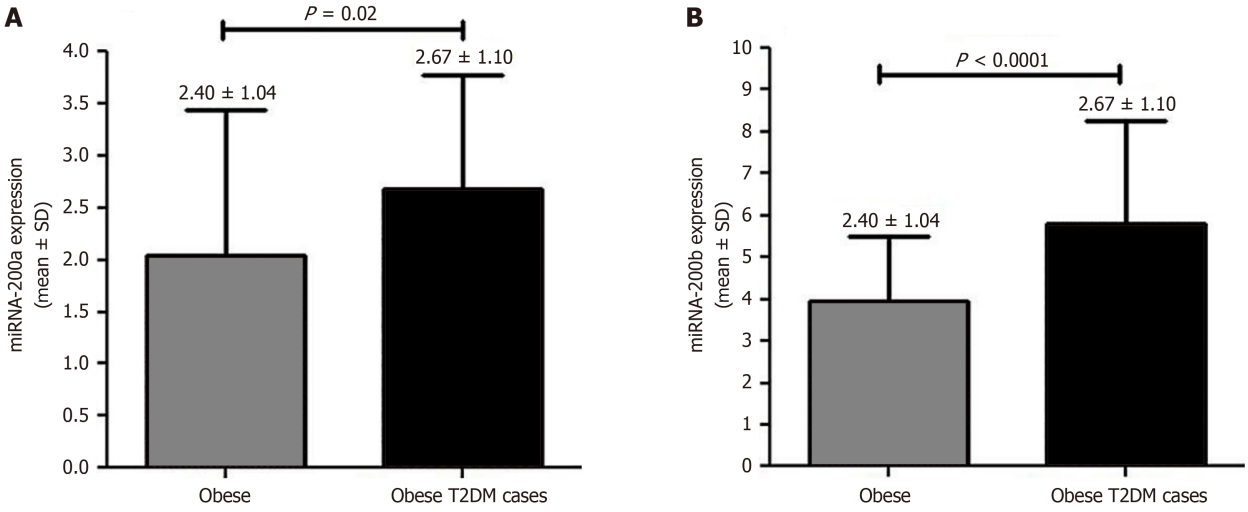

Figure 2 MiRNA-200a and miRNA-200b expression. A: Obese; B: Obese with type 2 diabetes mellitus. T2DM: Type 2 diabetes mellitus.

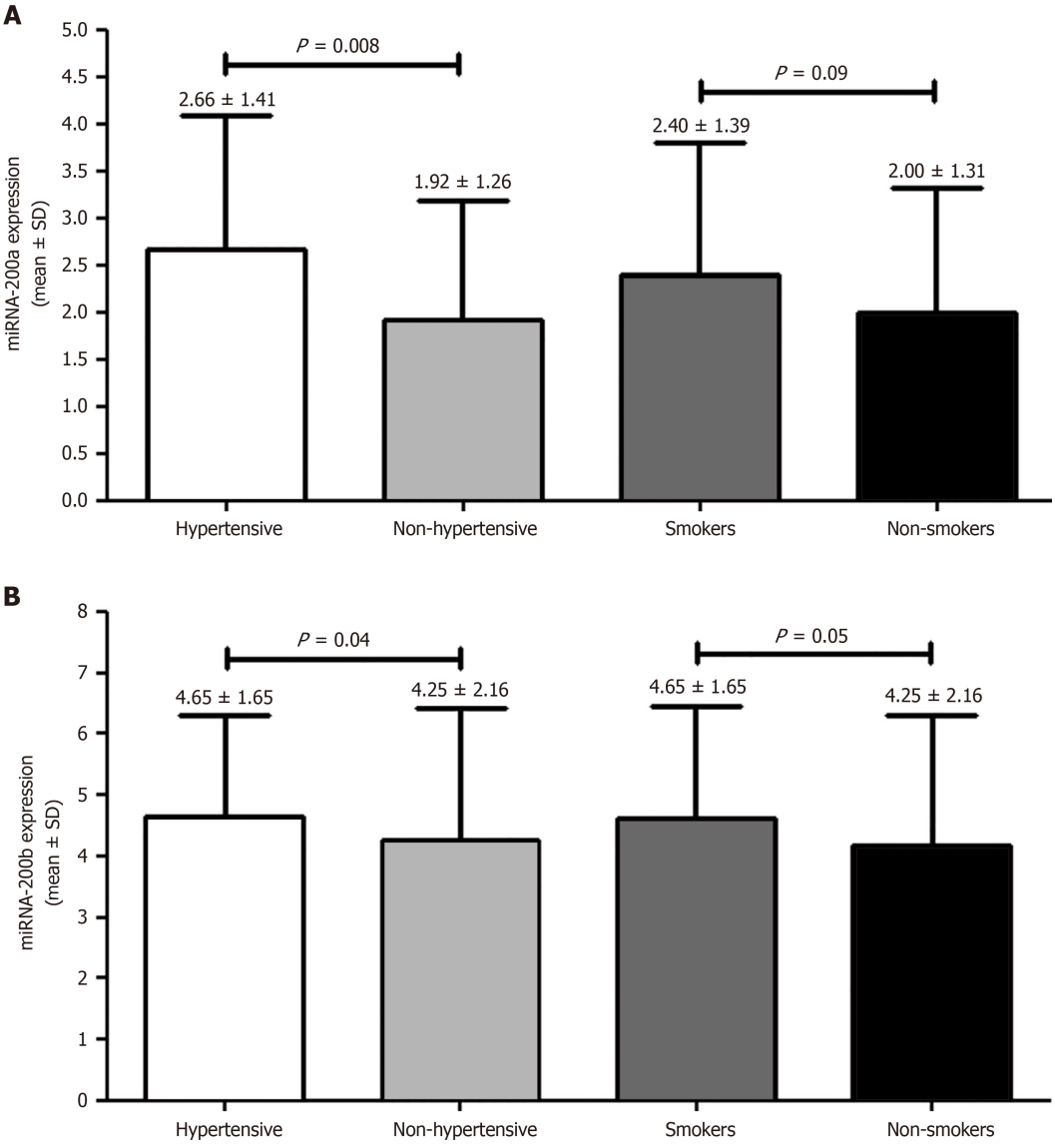

Figure 3 Comparison of miRNA-200a and miRNA-200b expression with regard to hypertension and smoking in obese and obese with type 2 diabetes mellitus participants. A: MiRNA-200a; B: MiRNA-200b.

Table 1 Comparison of biochemical parameters in healthy controls, obese, and obese with type 2 diabetes mellitus participants, mean ± SD

Biochemical parameters	Healthy controls	Obese	Obese type 2 diabetes	*P* value
Fasting blood glucose (mg/dL)	82.91 ± 8.34	87.48 ± 5.50	185.4 ± 39.50	< 0.0001
Postprandial blood glucose (mg/dL)	128.3 ± 8.44	135.0 ± 5.54	239.7 ± 43.21	< 0.0001
HbA1c (%)	5.18 ± 4.47	5.65 ± 0.54	8.28 ± 0.55	< 0.0001
HDL (mg/dL)	56.14 ± 5.90	45.91 ± 13.49	43.32 ± 11.66	< 0.0001
LDL (mg/dL)	98.35 ± 10.89	203.6 ± 18.65	202.3 ± 13.71	< 0.0001
TG (mg/dL)	124.7 ± 15.27	282.3 ± 76.87	424.7 ± 76.32	< 0.0001
Cholesterol (mg/dL)	161.9 ± 23.80	237.7 ± 34.73	279.7 ± 27.58	< 0.0001
VLDL (mg/dL)	25.27 ± 2.40	34.89 ± 5.68	35.50 ± 4.79	< 0.0001

HDL: High-density lipoprotein; LDL: Low-density lipoprotein; TG: Triglycerides; VLDL: Very low-density lipoprotein.

Table 2 Correlation analysis of miRNA-200a and miRNA200b with biochemical parameters

Biochemical parameters	miRNA-200a		miRNA-200b	
	r	*P* value	*r*	*P* value
Fasting blood glucose	0.16	0.09	0.26	0.005
Postprandial blood glucose	0.25	0.007	0.28	0.003
HbA1c	0.21	0.02	0.23	0.01
HDL	0.01	0.88	-0.27	0.004
LDL	0.14	0.13	0.03	0.73
TG	0.11	0.24	0.31	0.001
Cholesterol	0.24	0.009	0.34	< 0.0001
VLDL	0.08	0.40	0.26	0.006

HDL: High-density lipoprotein; LDL: Low-density lipoprotein; TG: Triglycerides; VLDL: Very low-density lipoprotein.

hypertension and smoking status (Figure 3B). Higher miRNA-200b expression was observed in hypertensive participants (4.65 + 1.65) compared to non-hypertensive participants (4.25 + 2.16), and the difference was statistically significant (P = 0.04). A substantial difference in miRNA-200b was observed with regard to smoking status.

Correlation of biochemical parameters with miRNA200a and miRNA200b

A correlation analysis was performed to determine the association between miRNA-200a and miRNA-200b with glucose and lipid parameters (Table 2). MiRNA-200a showed a positive correlation with postprandial blood glucose (r = 0.25, P = 0.007), HbA1c (r = 0.21, P = 0.02), and cholesterol (r = 0.24, P = 0.009). MiRNA-200b also showed a positive correlation with fasting blood glucose, postprandial blood glucose, HbA1c (r = 0.23, P = 0.01), TG (r = 0.31, P = 0.001), cholesterol (r = 0.34, P < 0.0001), VLDL (r = 0.26, P = 0.006), and a negative correlation with HDL (r = -0.27, P = 0.004).

Comparison of vitamin D levels among obese and obese T2DM participants

The level of vitamin D was analyzed in the obese and obese T2DM groups (Figure 4). The level of vitamin D in obese participants was 34.27 ng/mL, while in obese T2DM participants, the vitamin D level was 22.21 ng/mL, and the difference was statistically significant (P < 0.0001).

Association of vitamin D with miRNA-200a and miRNA-200b expression

A correlation analysis was also performed to determine the association between vitamin D with miRNA-200a and miRNA-200b (Figure 5). Vitamin D was found to be negatively correlated with miRNA-200a (r = -0.22, P = 0.01) and miRNA-200b (r = -0.19, P = 0.04). It is suggested that the decrease in vitamin D levels was due to the increase in miRNA-200a and miRNA-200b in the participants.

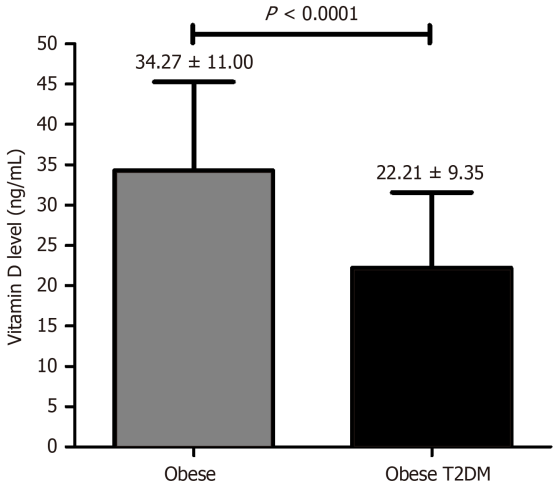

Figure 4 Level of vitamin D in obese and obese with type 2 diabetes mellitus participants. T2DM: Type 2 diabetes mellitus.

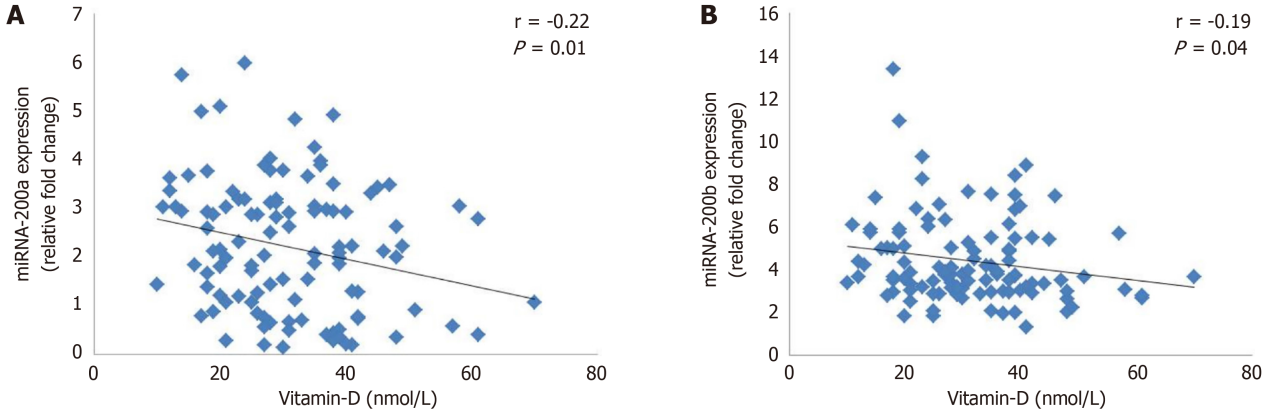

Figure 5 Correlation of vitamin D with miRNA-200a and miRNA-200b in obese and obese with type 2 diabetes mellitus participants. A: MiRNA-200a; B: MiRNA-200b.

Prognostic significance of miRNA-200a and miRNA-200b expression

To understand the prognostic importance of miRNA-200a and miRNA-200b, a ROC was plotted between obese and obese T2DM participants (Figure 6, Table 3). The area under the curve (AUC) for miRNA-200a was 0.64, sensitivity was 75%, and specificity was 57%, with a cutoff value of 2.07-fold. For miRNA-200b, the AUC was 0.75, sensitivity was 75%, and specificity was 71%, with a cutoff value of 4.12-fold. This suggests that miRNA-200a and miRNA-200b with increased expression of 2.07- and 4.12-fold could be predictive indicators for the risk of diabetes in obese individuals, respectively.

DISCUSSION

Obesity or excessive weight gain is recognized as the most critical and significant risk factor for the development and progression of T2DM across all age groups. Obesity and diabetes are chronic conditions that are rising worldwide, necessitating innovative strategies to manage and prevent diabetes in obese individuals[16]. The complex interplay and shared pathophysiological mechanisms between obesity and T2DM contribute to the increased prevalence and incidence of insulin resistance, dyslipidemia, non-alcoholic fatty liver disease, and a range of metabolic abnormalities in obese individuals and increases in BMI and abdominal fat distribution directly elevate the risk of T2DM, as changes in adipose tissue biology connect obesity to insulin resistance and beta cell dysfunction[17]. The present study investigated the role of miRNA-200a and miRNA-200b in obese participants with and without T2DM, Higher miRNA-200a and miRNA-200b expression were observed and significant differences in miRNA-200a and miRNA-200b were found between obese participants without T2DM and with T2DM. Higher miRNA-200a expression was observed among those with hypertension and smokers while miRNA-200b was significantly higher in hypertensive participants.

The study by Crépin *et al* in 2014 revealed that the overexpression of pre-miR-200a in a human neuroblastoma cell line disrupted both insulin and leptin signaling, this suggested a connection between altered leptin and insulin signaling and the up-regulation of hypothalamic miR-200a, which may serve as a potential target for obesity treatment[18]. Overexpression of the miRNA-200 family in mice triggers apoptosis of pancreatic β cells, leading to reduced insulin production

Table 3 Area under the curve for miRNA-200a, miRNA-200b, sensitivity, specificity and cutoff value

Variables	AUC	95%CI	Sensitivity	Specificity	Cutoff	*P* value
miRNA-200a	0.64	0.53-0.75	75%	57%	2.07 fold	0.02
miRNA-200b	0.75	0.64-0.75	75%	71%	4.12 fold	< 0.0001

AUC: Area under the curve.

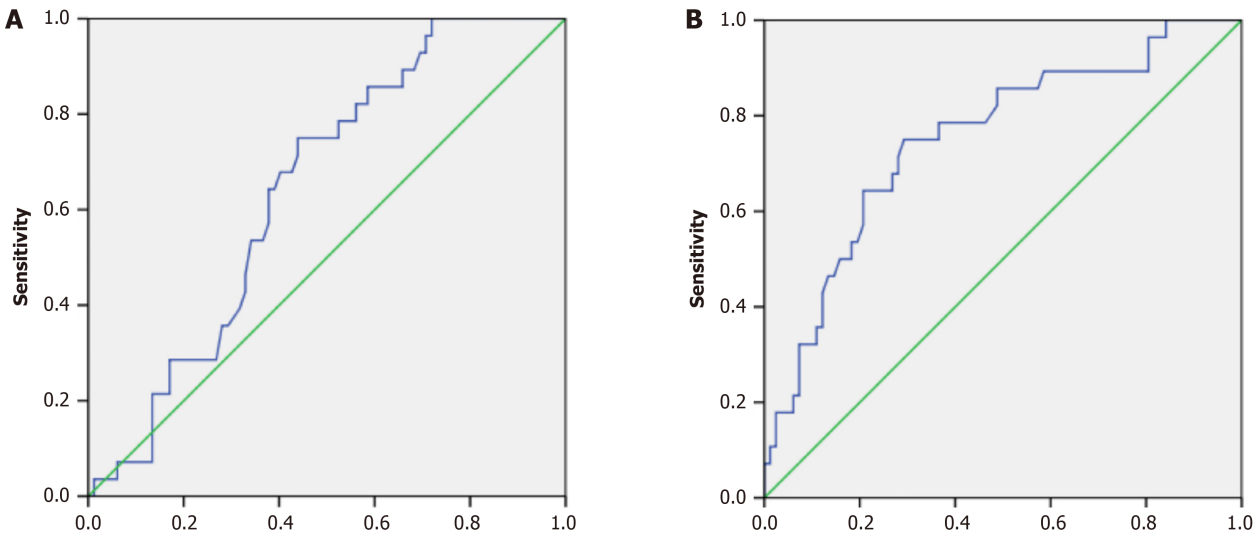

Figure 6 Prognostic importance of miRNA-200a and miRNA-200b in obese *vs* obese with type 2 diabetes mellitus participants. A: MiRNA-200a; B: MiRNA-200b.

and resulting in severe type 2 diabetes[19]. Similarly, the study by Martino *et al*[20] in 2015 suggested that expression of the miRNA-200 family may reduce insulin production by inducing apoptosis in pancreatic β cells[20]. Correlation analysis was carried out between biochemical parameters and both miRNA-200a and miRNA-200b, and a positive correlation was observed between miRNA-200a and postprandial glucose, HbA1c, and cholesterol. In addition, a positive correlation was observed between miRNA200b and fasting blood glucose, postprandial glucose, HbA1c, TG, cholesterol, and VLDL, and a negative correlation with HDL.

We compared the vitamin D level in obese *vs* obese with T2DM participants and observed that those with obesity and T2DM had lower vitamin D levels. Abed *et al*[21] in 2024, proposed a link between vitamin D and insulin resistance, particularly in individuals with T2DM. Obesity, combined with insulin resistance, may further increase the risk of other health-related complications[21]. Individuals with obesity often have low vitamin D levels due to factors such as a greater volume of distribution, vitamin D being tightly bound in fat tissues, reduced absorption, and diets deficient in vitamin D [22]. In a 2023 study, Vijay *et al*[23] observed vitamin D deficiency in patients with T2DM and suggested that consistent vitamin D supplementation could help prevent further complications in these patients[23]. Most individuals with T2DM in a study conducted at the King Faisal University Health Center in Saudi Arabia were found to have below-normal vitamin D levels[24]. Bhatt *et al*[25] in 2020, observed significant reductions in fasting blood glucose, 2-hour blood glucose (post-OGTT), HbA1c, and truncal subcutaneous fat after 78 weeks of vitamin D supplementation in overweight/obese, prediabetic, and vitamin D-deficient Asian Indian women[25]. In patients with T2DM, who are overweight or obese, serum 25 (OH) D levels are inversely correlated with TG. Therefore, providing vitamin D supplementation is crucial, especially for obese individuals with T2DM[26]. Those with the lowest vitamin D levels also had the greatest BMI. Furthermore, the degree of vitamin D insufficiency was found to be negatively linked with being overweight/obese. Patients with vitamin D insufficiency were more insulin resistant and had higher oxidative stress (particularly those with severe deficiency)[27]. MiRNAs control dynamic posttranscriptional regulatory networks. Not only do miRNAs alter vitamin D signaling, but vitamin D regulates miRNA networks throughout homeostasis and illness across species[28].

This study observed insufficient levels of vitamin D among obese T2DM participants; however, the level of vitamin D was sufficient among obese participants. A study carried out in a population in Lagos, Nigeria, demonstrated that vitamin D deficiency was 63.2% in T2DM patients, which is in a similar range[29]. The prevalence of vitamin D deficiency and insufficiency was 38.4% and 21.9%, respectively, in another study conducted in a referral hospital in Kenya with only T2DM patients as the study group[30]. Most individuals in a study conducted at the King Faisal University Health Center in Saudi Arabia who had T2DM had lower vitamin D levels[24].

We observed a negative correlation between miRNA-200a and miRNA-200b with vitamin D level, which suggested that increasing miRNA-20a and miRNA-20b would decrease vitamin-D level, influencing disease severity. Prognostic efficacy was evaluated in obese T2DM *vs* obese participants, and it was observed that miRNA-200a and miRNA-200b can

be used as predictive markers using cutoff values of 2.07-fold and 412-fold, respectively. Vitamin D insufficiency has been associated with pregnancy issues such as gestational diabetes, hypertension, and bacterial vaginosis[31]. Although many of these responses may involve transcriptional control, it appears that differences in vitamin D-mediated miRNA expression may also affect pregnancy health outcomes at the posttranscriptional level[32]. Our study concluded that miRNA-200a and miRNA-200b were higher among diabetic obese participants *vs* non-diabetic obese participants, and higher miRNA-200a and miRNA-200b were observed among hypertensive individuals. Overexpression of miRNA-200a and miRNA-200b was linked to altered glucose and lipid parameters, which could lead to severe disease and other health-related complications. Insufficient vitamin D among obese T2DM individuals may be involved in poor clinical outcome, disease progression, and worsening of disease. The ROC was evaluated and it was found that higher miRNA-200a and miRNA-200b expression could be used as prognostic indicators for disease worsening and for the prediction of obesity with diabetes.

CONCLUSION

Monitoring dysregulation of miRNAs is very important in controlling disease outcomes and the evaluation of vitamin D levels could be used to prevent disease progression and further health-related complications.

FOOTNOTES

Author contributions: Alshahrani AF, Ashfaq F contributed to the concept, methodology, experimentation and data collection, corrections after revision, and reading before submission; Alsayegh AA, Bajahzer M contributed to data entry, produced the first draft, and reading before submission; Khan MI, Beg MMA contributed to data analysis, writing and corrections after revision, and reading before submission.

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REFERENCES

1 **Haslam DW**, James WP. Obesity. *Lancet* 2005; **366**: 1197-1209 [PMID: 16198769 DOI: 10.1016/S0140-6736(05)67483-1]
2 **Arner P**, Kulyté A. MicroRNA regulatory networks in human adipose tissue and obesity. *Nat Rev Endocrinol* 2015; **11**: 276-288 [PMID: 25732520 DOI: 10.1038/nrendo.2015.25]
3 **Maurizi G**, Babini L, Della Guardia L. Potential role of microRNAs in the regulation of adipocytes liposecretion and adipose tissue physiology. *J Cell Physiol* 2018; **233**: 9077-9086 [PMID: 29932216 DOI: 10.1002/jcp.26523]
4 **Vohl MC**, Sladek R, Robitaille J, Gurd S, Marceau P, Richard D, Hudson TJ, Tchernof A. A survey of genes differentially expressed in subcutaneous and visceral adipose tissue in men. *Obes Res* 2004; **12**: 1217-1222 [PMID: 15340102 DOI: 10.1038/oby.2004.153]
5 **Ortega FJ**, Moreno-Navarrete JM, Pardo G, Sabater M, Hummel M, Ferrer A, Rodriguez-Hermosa JI, Ruiz B, Ricart W, Peral B, Fernández-

Real JM. MiRNA expression profile of human subcutaneous adipose and during adipocyte differentiation. *PLoS One* 2010; **5**: e9022 [PMID: 20126310 DOI: 10.1371/journal.pone.0009022]

6 **Landrier JF**, Karkeni E, Marcotorchino J, Bonnet L, Tourniaire F. Vitamin D modulates adipose tissue biology: possible consequences for obesity? *Proc Nutr Soc* 2016; **75**: 38-46 [PMID: 26564334 DOI: 10.1017/S0029665115004164]

7 **Vienberg S**, Geiger J, Madsen S, Dalgaard LT. MicroRNAs in metabolism. *Acta Physiol (Oxf)* 2017; **219**: 346-361 [PMID: 27009502 DOI: 10.1111/apha.12681]

8 **Vaishya S**, Sarwade RD, Seshadri V. MicroRNA, Proteins, and Metabolites as Novel Biomarkers for Prediabetes, Diabetes, and Related Complications. *Front Endocrinol (Lausanne)* 2018; **9**: 180 [PMID: 29740397 DOI: 10.3389/fendo.2018.00180]

9 **Klein D**, Misawa R, Bravo-Egana V, Vargas N, Rosero S, Piroso J, Ichii H, Umland O, Zhijie J, Tsinoremas N, Ricordi C, Inverardi L, Domí nguez-Bendala J, Pastori RL. MicroRNA expression in alpha and beta cells of human pancreatic islets. *PLoS One* 2013; **8**: e55064 [PMID: 23383059 DOI: 10.1371/journal.pone.0055064]

10 **Filios SR**, Xu G, Chen J, Hong K, Jing G, Shalev A. MicroRNA-200 is induced by thioredoxin-interacting protein and regulates Zeb1 protein signaling and beta cell apoptosis. *J Biol Chem* 2014; **289**: 36275-36283 [PMID: 25391656 DOI: 10.1074/jbc.M114.592360]

11 **Title AC**, Silva PN, Godbersen S, Hasenöhrl L, Stoffel M. The miR-200-Zeb1 axis regulates key aspects of β-cell function and survival in vivo. *Mol Metab* 2021; **53**: 101267 [PMID: 34116231 DOI: 10.1016/j.molmet.2021.101267]

12 **Belgardt BF**, Ahmed K, Spranger M, Latreille M, Denzler R, Kondratiuk N, von Meyenn F, Villena FN, Herrmanns K, Bosco D, Kerr-Conte J, Pattou F, Rülicke T, Stoffel M. The microRNA-200 family regulates pancreatic beta cell survival in type 2 diabetes. *Nat Med* 2015; **21**: 619-627 [PMID: 25985365 DOI: 10.1038/nm.3862]

13 **Park S**, Kim DS, Kang S. Vitamin D deficiency impairs glucose-stimulated insulin secretion and increases insulin resistance by reducing PPAR-γ expression in nonobese Type 2 diabetic rats. *J Nutr Biochem* 2016; **27**: 257-265 [PMID: 26522682 DOI: 10.1016/j.jnutbio.2015.09.013]

14 **Greco EA**, Lenzi A, Migliaccio S. Role of Hypovitaminosis D in the Pathogenesis of Obesity-Induced Insulin Resistance. *Nutrients* 2019; **11** [PMID: 31266190 DOI: 10.3390/nu11071506]

15 **Karkeni E**, Bonnet L, Marcotorchino J, Tourniaire F, Astier J, Ye J, Landrier JF. Vitamin D limits inflammation-linked microRNA expression in adipocytes in vitro and in vivo: A new mechanism for the regulation of inflammation by vitamin D. *Epigenetics* 2018; **13**: 156-162 [PMID: 28055298 DOI: 10.1080/15592294.2016.1276681]

16 **Chandrasekaran P**, Weiskirchen R. The Role of Obesity in Type 2 Diabetes Mellitus-An Overview. *Int J Mol Sci* 2024; **25** [PMID: 38339160 DOI: 10.3390/ijms25031882]

17 **Klein S**, Gastaldelli A, Yki-Järvinen H, Scherer PE. Why does obesity cause diabetes? *Cell Metab* 2022; **34**: 11-20 [PMID: 34986330 DOI: 10.1016/j.cmet.2021.12.012]

18 **Crépin D**, Benomar Y, Riffault L, Amine H, Gertler A, Taouis M. The over-expression of miR-200a in the hypothalamus of ob/ob mice is linked to leptin and insulin signaling impairment. *Mol Cell Endocrinol* 2014; **384**: 1-11 [PMID: 24394757 DOI: 10.1016/j.mce.2013.12.016]

19 **Magenta A**, Ciarapica R, Capogrossi MC. The Emerging Role of miR-200 Family in Cardiovascular Diseases. *Circ Res* 2017; **120**: 1399-1402 [PMID: 28450364 DOI: 10.1161/CIRCRESAHA.116.310274]

20 **Martino F**, Carlomosti F, Avitabile D, Persico L, Picozza M, Barillà F, Arca M, Montali A, Martino E, Zanoni C, Parrotto S, Magenta A. Circulating miR-33a and miR-33b are up-regulated in familial hypercholesterolaemia in paediatric age. *Clin Sci (Lond)* 2015; **129**: 963-972 [PMID: 26229086 DOI: 10.1042/CS20150235]

21 **Abed MN**, Alassaf FA, Qazzaz ME. Exploring the Interplay between Vitamin D, Insulin Resistance, Obesity and Skeletal Health. *J Bone Metab* 2024; **31**: 75-89 [PMID: 38886966 DOI: 10.11005/jbm.2024.31.2.75]

22 **Alzohily B**, AlMenhali A, Gariballa S, Munawar N, Yasin J, Shah I. Unraveling the complex interplay between obesity and vitamin D metabolism. *Sci Rep* 2024; **14**: 7583 [PMID: 38555277 DOI: 10.1038/s41598-024-58154-z]

23 **Vijay GS**, Ghonge S, Vajjala SM, Palal D. Prevalence of Vitamin D Deficiency in Type 2 Diabetes Mellitus Patients: A Cross-Sectional Study. *Cureus* 2023; **15**: e38952 [PMID: 37313077 DOI: 10.7759/cureus.38952]

24 **Al Ghadeer HA**, AlRamadan MS, Al Amer MM, Alshawaf MJ, Alali FJ, Bubshait AA, Alramadhan MA, Almurayhil Z, Aldandan NS, AlKhamis MA, AlHaddad HA, AlOmair A. Vitamin D Serum Levels in Type 2 Diabetic Patients: A Cross-Sectional Study. *Cureus* 2022; **14**: e22558 [PMID: 35345711 DOI: 10.7759/cureus.22558]

25 **Bhatt SP**, Misra A, Pandey RM, Upadhyay AD, Gulati S, Singh N. Vitamin D Supplementation in Overweight/obese Asian Indian Women with Prediabetes Reduces Glycemic Measures and Truncal Subcutaneous Fat: A 78 Weeks Randomized Placebo-Controlled Trial (PREVENT-WIN Trial). *Sci Rep* 2020; **10**: 220 [PMID: 31937856 DOI: 10.1038/s41598-019-56904-y]

26 **Gong T**, Di H, Han X, Hu X, Liu C, Chen G. Vitamin D is negatively associated with triglyceride in overweight/obese patients with type 2 diabetes. *Endocrine* 2022; **76**: 304-311 [PMID: 35247144 DOI: 10.1007/s12020-022-03009-8]

27 **Cătoi AF**, Iancu M, Pârvu AE, Cecan AD, Bidian C, Chera EI, Pop ID, Macri AM. Relationship between 25 Hydroxyvitamin D, Overweight/Obesity Status, Pro-Inflammatory and Oxidative Stress Markers in Patients with Type 2 Diabetes: A Simplified Empirical Path Model. *Nutrients* 2021; **13** [PMID: 34445049 DOI: 10.3390/nu13082889]

28 **Zhao H**, Forcellati M, Buschittari D, Heckel JE, Machado CJ, Ramulu N. Pullagura S, Lisse TS. Vitamin D and microRNAs. In: Feldman and Pike' s Vitamin D. Academic Press, 2024 [DOI: 10.1016/b978-0-323-91386-7.00011-8]

29 **Anyanwu AC**, Olopade OB, Onung SI, et al. : Serum vitamin D levels in persons with type 2 diabetes mellitus in Lagos, Nigeria. *Int J Diabetes Clin Res* 2020; **7**: 133-10 [DOI: 10.23937/2377-3634/1410133]

30 **Karau PB**, Kirna B, Amayo E, Joshi M, Ngare S, Muriira G. The prevalence of vitamin D deficiency among patients with type 2 diabetes seen at a referral hospital in Kenya. *Pan Afr Med J* 2019; **34**: 38

31 **Mulligan ML**, Felton SK, Riek AE, Bernal-Mizrachi C. Implications of vitamin D deficiency in pregnancy and lactation. *Am J Obstet Gynecol* 2010; **202**: 429.e1-429.e9 [PMID: 19846050 DOI: 10.1016/j.ajog.2009.09.002]

32 **Liu NQ**, Hewison M. Vitamin D, the placenta and pregnancy. *Arch Biochem Biophys* 2012; **523**: 37-47 [PMID: 22155151 DOI: 10.1016/j.abb.2011.11.018]

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