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Clinical Value of Serum BMP-4, BMP-2, GDF-15, MMP-9, GP39 Levels in Pregnant Women with Obesity and the Related Comorbidities Diabetes Mellitus and Gestational Hypertension

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ABSTRACT

Aims We evaluated the clinical value of selected serum biomarkers BMP-4, BMP-2, GDF-15, MMP-9, and GP39 in pregnant women with obesity and the comorbidities diabetes mellitus (DM) and gestational hypertension (GHT).

Methods This observational study had groups of controls, including healthy pregnant women; women with only obesity, including pregnant women with BMI ≥ 30 kg/m^2; women with gestational DM (GDM) with obesity, including pregnant women with GDM and obesity; women with pregestational DM (PGDM) with obesity, including pregnant women with PGDM and obesity; and women with GHT with obesity, including pregnant women with GHT and obesity. We measured serum levels of selected biomarkers by ELISA.

Results Obesity increased serum levels of all the biomarkers; GDM developed in obese women caused a more pronounced increase in the serum levels of BMP-4 and BMP-2, and GHT developed in obese women caused a more pronounced increase in the serum levels of GDF-15. In the women with GDM-, PGDM-, and GHT-complicated obesity, serum levels of MMP-9 and GP39 did not change meaningfully.

Conclusions Obesity and its comorbidities DM and GHT lead to meaningful changes in the studied serum biomarkers. Since obesity has a causal effect on developing numerous conditions, reliable clinical biomarkers are needed to improve the early prediction and diagnosis of high-risk conditions during pregnancy.

Introduction

Maternal obesity is a growing public health concern, and it is widely understood that diet and metabolism play a critical role in the health and well-being of both the mother and the fetus [1]. Obese women have a higher risk of antepartum problems than normal-weight women, including spontaneous miscarriage, congenital abnormalities, proteinuria, nonalcoholic fatty liver disease, gestational diabetes mellitus (GDM) [2], pregestational diabetes mellitus (PGDM) [3], and gestational hypertension (GHT) [4]. Obese pregnant women are more likely to have a cesarean section, have a failed labor trial, develop endometritis, have a wound rupture, or have children with autism spectrum disorders, developmental delays, or attention deficit hyperactivity disorder [5]. They are more likely to experience excessive gestational weight gain, which raises the risk of having metabolic syndrome later in life [6].

Obesity is a multifactorial condition, in essence, defined clinically by a body mass index (BMI) of greater than 30 kg/m^2 because of an excess of white adipose tissue (WAT) [7]. WAT is essential for homeostasis, but excessive amounts can predispose people to severe insulin resistance and diabetes [8]. Obesity is a condition that occurs when long-term caloric intake exceeds energy expenditure and is one of the primary comorbidities of type 2 diabetes mellitus (DM) [9].

During pregnancy, obesity and its comorbidities, DM and GHT, are antenatal disorders that have increased in incidence worldwide as a remarkably significant health burden [10]. So intertwined are these seemingly independent conditions that one needs to consider the challenging impact of obesity with increasing gestational age. To better manage these conditions in pregnancy, clinicians and researchers need to broaden their understanding of the pathophysiology of obesity and its combined effects with DM and GHT.

For women with high-risk pregnancies, a better understanding of the pathophysiology is necessary to guide the development of more efficacious and cost-effective diagnostic modalities for their management during pregnancy. Obesity is not only a disease but also a metabolic risk factor associated with other complicated pregnancies, such as DM and GHT. Having antenatal obesity and excessive weight gain during pregnancy is related to the development of GHT [11, 12]. The current epidemic of obesity during pregnancy and its related antenatal complications like DM and GHT are increasingly at the point of attention to understand the differential physiology of fat tissue types producing bioactive peptides and proteins to maintain their development and to crosstalk with other pathways with benefits as well as harmful effects [13, 14]. In addition to the secretion of hundreds of bioactive molecules from adipose tissue as an active endocrine organ, due to the mobilization of other protective mechanisms that try to reduce the harms caused by obesity, a different environment is formed in the human body than usual [14, 15].

Adipose derivatives have a strict relationship with insulin resistance and DM type 2 development, an increase in adipose tissue, inflammation, and other pathogenic conditions [16]. For this reason, the observation of changes in the factors that change in adipose derivatives in obese subjects is very important to better understand the basic pathophysiology of health problems encountered antenatally. In accordance with the growth of pregnancy, the increasing pregnancy-related bioactive molecules make these conditions even more complex. A pathological increase in fat mass and/or an unhealthy distribution of body fat may cause cellular dysfunctions throughout the body, resulting in an unhealthy metabolic profile as well as underlying states of insulin resistance and chronic low-grade inflammation caused by adipose tissue dysfunction, contributing to an excess workload for organs and systems and eventually leading to organ damage and dysfunction [8]. In pregnant women, it seems to be necessary to investigate the status of serum BMP-4, BMP-2, GDF-15, MMP-9, and GP39 for the development of new diagnostic test panels and treatment options.

Bone morphogenetic proteins (BMPs) are a subset of the transforming growth factor (TGF) superfamily of signaling molecules that play a wide range of biological functions. BMP-4 is a key regulator of white/beige adipogenic differentiation in vivo, with implications for thermogenesis, energy homeostasis, and obesity development. Human adipose tissue expresses and releases BMP-4 in significant amounts, and serum levels rise with obesity [17]. BMP-4 is one of the most studied BMPs in adipose tissue, with major roles in white and brown adipogenesis, respectively, but BMP-2 has also been shown to affect adipogenesis [18]. Growth differentiation factor 15 (GDF-15) is a member of the TGF-β superfamily. Functionally, its roles in appetite regulation, metabolism, cell and tissue survival, and immune tolerance have been described [19]. GDF-15 levels are impacted by a plethora of metabolic factors: exercise, tissue injury, pregnancy, hypoxia, and drugs. GDF-15 is emerging as a premier biomarker to determine prognosis in several health disorders, including type 1 DM [20]. Matrix metalloproteinase-9 (MMP-9) increases in obese subjects [21]. The knowledge about the physiological function and the mechanisms by which human cartilage glycoprotein 39 (GP39) mediates its effects is still scarce. It has been demonstrated that patients with type 1 diabetes as well as patients with type 2 diabetes have elevated plasma GP39 levels [22, 23]. The upstream and downstream signaling pathways of these biomolecules are thus far less well clarified in pregnant women with obesity. Despite several important advances in deciphering the signaling modalities of these biomolecules, much remains to be fully understood, in particular the different effects observed in obesity-related disorders. To what extent this is due to differences in the signaling pathways and expression of these biomolecules is still not clear.

Nevertheless, despite the high prevalence of obesity and its comorbidities, such as diabetes and GHT, in pregnant women, we still know little about their exact pathogenesis. Furthermore, there is also a growing scientific interest in the study of novel biomarkers that can predict and diagnose these conditions. Studying new possible biomarkers may not only provide insight into the intricate pathophysiology of obesity and its comorbidities in pregnant women, but also implicates the measurement of several target proteins that may provide useful information about the severity of these conditions, their possible complications, and prognoses. In our study, we therefore endeavored to assess the clinical value of serum levels of selected biomarkers such as BMP-4, BMP-2, GDF-15, MMP-9, and GP39 in pregnant women with obesity and related comorbidities such as GDM, PGDM, and GHT.

Material and Methods

Study groups

Following the approval of the local ethics committee, this hospital-based observational case-control study was conducted in the Gynecology and Obstetrics Services at Haseki Training and Research Hospital affiliated with the University of Health Sciences in the Sultangazi district of İstanbul, over one year, following the appropriate clinical ethical guidelines and the valid Declaration of Helsinki. Informed written consent was obtained from each pregnant woman once the purpose and nature of all the procedures used were thoroughly explained. Care was taken to ensure that cases were included in research groups one by one. There was no feature of the study that could jeopardize the participant's pregnancy care.

The study groups were the following: controls, including pregnant women who had no obesity (BMI 18.5–24.9 kg/m2) and no GDM, no PGDM, or no GHT (n = 35); a group of only obesity, including pregnant women with BMI ≥ 30 kg/m2 (n = 34); a group of GDM with obesity, including pregnant women who had GDM and obesity (n = 34); a group of PGDM with obesity, including pregnant women who had PGDM and obesity (n = 22); and a group of GHT with obesity, including pregnant women who had GHT and obesity (n = 33). The inclusion criteria were as follows: being between the ages of 18–42, gestational age of 24–41 weeks, and absence of labor. Multiple pregnancies, congenital fetal infections, fetal congenital malformations, pregnancy without a one-year interval, those with severe systemic disease and chronic drug usage, and those with infection and inflammation results are all excluded.

The diagnosis of GDM was made when at least one of the three criteria of 75-g, 2-hour oral glucose tolerance test (OGTT) thresholds were met or exceeded: fasting 92 mg/dL, 1-hour 180 mg/dL, or 2 hours 153 mg/dL at 24–28 weeks of gestation [24]. The term "PGDM" refers primarily to type 1 or type 2 DM diagnosed prior to pregnancy [25, 26]. In this study, all the PGDM cases had DM type 2. GHT is defined as hypertension that develops spontaneously after 20 weeks of pregnancy, without proteinuria or biochemical or hematological abnormalities [27].

Recorded characteristics of the study population included maternal age, gravidity, parity, ethnicity (native, Arabic), smoking status (yes or no), requiring assisted reproductive technology (ART) for conception (yes or no), pre-pregnancy body mass index (BMI), gestational weight gain, mode of delivery (vaginal or cesarean), gestational age at delivery (week), the ratio of fetal gender (female or male), newborn birthweight, Apgar scores at 1 and 5 min, cord blood pH, and the ratio of neonatal intensive care unit admission (NICU) (yes or no).

Blood collection and analysis

For each study participant, fasting blood samples were collected during their first perinatal examination in the second or third trimester of pregnancy. Maternal hematological and biochemical tests were performed. Serum human BMP-4, BMP-2, GDF-15, MMP-9, and GP39 were determined using commercial enzyme-linked immunosorbent assay (ELISA) kits (Elabscience, Houston, Texas, USA) according to the manufacturer's protocol. Serum samples were not diluted with those kits, and the standards were serially diluted from starting concentrations of 2000 to 31.25 pg/mL for BMP-4, from 4000 to 62.5 pg/mL for BMP-2, from 15000 to 23.44 pg/mL for GDF-15, from 10 to 0.16 ng/mL for MMP-9, and from 4000 to 62.5 pg/mL for GP39, in the sample diluent supplied with the kit. The intra- and inter-assay coefficient of variation for the assays is < 10 %.

Statistical analysis

Analysis of data was conducted with the IBM SPSS v23 (IBM Corp., Armonk, New York, USA). Graphical presentations were prepared using the GraphPad Prism v9 (Graphpad, San Diego, California, USA). Descriptive analyses of numerical data as mean, standard deviation, median, interquartile range, and percentage were presented. After the normality test was done with the Kolmogorov–Smirnov test, the comparison of parametric data with ANOVA followed by post hoc Tukey test and the comparison of non-parametric data with Kruskal–Wallis with post hoc Mann – Whitney U test was performed. If the p-value was below 0.05 after the analysis of the data, it was considered that there was a significant difference as a result of the comparison.

Results

The maternal and perinatal clinical characteristics of the controls and groups of only obesity, GDM with obesity, PGDM with obesity, and GHT with obesity are presented in ▶ **Table 1**. The median age of the group of PGDM with obesity was significantly higher than those of the other groups ($p < 0.05$). The groups of only obesity and GDM with obesity had significantly higher median ages compared to the controls ($p < 0.05$). The ratio of being Arabic was significantly higher in the controls than in those of the other groups ($p < 0.05$). The median value of pre-pregnancy BMI was significantly lower in the controls than in those of the other groups ($p < 0.05$). We found a significantly higher median value of gestational weight gain in the group of only obese people than in those of the other groups ($p < 0.05$). The controls and groups of GDM with obesity and GHT with obesity had a significantly higher median value of gestational weight gain compared to the PGDM with obesity ($p < 0.05$). The ratio of vaginal deliveries was higher in the controls than in those of the other groups ($p < 0.05$). The median values of birthweight in the groups of only obesity, GDM with obesity, and PGDM with obesity were significantly higher compared to the controls and the group of GHT with obesity ($p < 0.05$). There were no significant differences between the study groups regarding gravidity, parity, smoking status, having natural pregnancies, gestational age at delivery, the ratio of female newborns, Apgar scores at 1 and 5 min, cord blood pH, or the ratio of neonatal intensive care unit NICU admission ($p > 0.05$).

▶ **Table 2** shows the maternal hematological and biochemical findings of the controls and groups of only obesity, GDM with obesity, PGDM with obesity, and GHT with obesity. The median values of fasting plasma glucose and HbA1c were significantly higher in the groups of GDM with obesity and PGDM with obesity than in those of the controls and groups of only obesity and GHT with obesity ($p < 0.05$). There were no significant differences between the study groups regarding the mean values of hematocrit, creatinine, high-density lipoprotein cholesterol, and low-density lipoprotein

▶ **Table 1** Maternal and perinatal clinical characteristics of the study population.

	Controls (n = 35)	Only Obesity (n = 34)	GDM with Obesity (n = 34)	PGDM with Obesity (n = 22)	GHT with Obesity (n = 33)	P value
Maternal age (years)	26 (18–42)[a]	29 (19–42)[b]	31 (20–41)[b]	34 (25–42)[c]	31 (22–41)[b]	0.001
Gravidity	2 (1–7)	3 (2–8)	3 (1–7)	3 (1–8)	3 (1–7)	0.504
Parity	1 (0–6)	2 (1–4)	2 (0–5)	2 (0–7)	1 (0–5)	0.375
Ethnicity, n (%)						0.001
Native	20 (57.1%)[a]	25 (73.5%)[b]	31(91.2%)[b]	21 (95.5%)[b]	32 (97%)[b]	
Arabic	15 (42.9%)	9 (26.5%)	3 (8.8%)	1 (4.5%)	1 (3%)	
Smoking, n (%)						0.512
Yes	2 (5.7%)	4 (11.8%)	2 (5.9%)	1 (4.5%)	5 (15.2%)	
No	33 (94.3%)	30 (88.2%)	32 (94.1%)	21 (95.5%)	28 (84.8%)	
ART pregnancy, n (%)						0.117
Yes	0 (0%)	2 (5.9%)	0 (0%)	0 (0%)	0 (0%)	
No	35 (100%)	32 (94.1%)	34 (100%)	22 (100%)	33 (100%)	
Pre-pregnancy BMI (Kg/m2)	23.5 (19–24.8)[a]	32.8 (30–45.5)[b]	33.4 (30–40.7)[b]	34.9 (30.9–43)[b]	34.5 (30–47.7)[b]	0.001
Gestational weight gain (kg)	9 (0–20)[b]	13.5 (2–25)[c]	11.5 (3–18)[b]	7 (5–20)[a]	9 (0–35)[b]	0.001
Mode of delivery, n (%)						0.001
Vaginal	19 (54.3%)	7 (20.6%)	8 (23.5%)	8 (36.4%)	11 (33.3%)	
Cesarean	16 (45.7%)[b]	27 (79.4%)[a]	26 (76.5%)[a]	14 (63.6%)[a]	22 (66.7%)[a]	
Gestational age at delivery (week)	38 (28–42)	39 (36–40)	39 (34–40)	38 (35–38)	37 (33–39)	0.223
Fetal gender, n (%)						0.366
Female	18 (51.4%)	22 (64.7%)	14 (41.1%)	11 (50%)	20 (60.6%)	
Male	17 (48.6%)	12 (35.3%)	20 (58.9%)	11 (50%)	13 (39.4%)	
Birth weight (g)	2970 (770–3825)[a]	3300 (2355–4410)[b]	3565 (2335–4685)[b]	3362 (3081–4895)[b]	2915 (1990–4060)[a]	0.001
Apgar score						
At 1 min	9 (1–9)	9 (6–9)	9 (2–9)	9 (7–9)	9 (3–9)	0.588
At 5 min	10 (6–10)	10 (8–10)	10 (6–10)	10 (8–10)	10 (6–10)	0.617
Cord blood pH	7.34 (6.96–7.49)	7.34 (7.25–7.42)	7.37 (7.19–7.45)	7.34 (7.2–7.4)	7.35 (7.21–7.49)	0.914
NICU admission, n (%)						0.258
Yes	3 (8.6%)	11 (32.4%)	10 (29.4%)	6 (27.3%)	16 (48.5%)	
No	32 (91.4%)	23 (67.6%)	24 (70.6%)	16 (72.7%)	17 (51.5%)	

Data were presented as median with minimum and maximum values or counts with percentages and analyzed with the Kruskal–Wallis test followed by the Mann–Whitney test for pairwise comparisons or chi-square test as appropriate. Results of the pairwise comparisons were denoted with a letter, and there was no significant difference between/among the study groups marked with the same letter. There was a significant difference between/among the study groups marked with different letters ($p < 0.05$). GDM, gestational diabetes; PGDM, pregestational diabetes; GHT, gestational hypertension; ART, assisted reproductive technology; BMI, body mass index; NICU, neonatal intensive care unit.

cholesterol ($p > 0.05$). The median values of white blood cell count, hemoglobin, platelets, aspartate aminotransferase, alanine aminotransferase, and triglyceride were found as similar in the study groups ($p > 0.05$).

In ▶ **Fig. 1**, the median serum BMP-4 and BMP-2 levels of controls and groups of only obesity, GDM with obesity, PGDM with obesity, and GHT with obesity are presented. Regarding BMP-4 analysis, the median serum BMP-4 levels in the groups of only obesity, GDM with obesity, PGDM with obesity, and GHT with obesity were significantly higher than those of the controls ($p < 0.05$); the median serum BMP-4 levels in the group of GDM with obesity was significantly higher than those of the groups of only obesity and GHT with obesity ($p < 0.05$); and other pairwise comparisons provided no meaningful difference ($p > 0.05$). Regarding BMP-2 analysis, the groups of only obesity, GDM with obesity, PGDM with obesity, and GHT with obesity had significantly higher values of median serum BMP-2 levels compared to the controls ($p < 0.05$); the group of GDM with obesity had significantly higher value of medi-

▶ **Table 2** Maternal hematological and biochemical findings of the study population.

	Controls	Obese	GDM with Obesity	PGDM with Obesity	GHT with Obesity	P value
WBC (10^3 uL)	11.5 (6.1–17)	12.5 (7–18.2)	11.7 (5.7–21.2)	10.2 (7.9–16.9)	10.1 (6.4–24.9)	0.062
Hb (g/dL)	11.1 (8.4–13.9)	10.8 (7.5–13.7)	11.7 (8.3–13.9)	11.9 (9.2–13.4)	11.1 (9–14.2)	0.107
Hct (%)	33.9 ± 3.3	32.8 ± 3.8	33.6 ± 4.8	35.1 ± 3.1	33.4 ± 3.7	0.324
PLT (10^3 uL)	217 (125–3429	200 (8100–325)	207 (119–348)	247 (155–355)	239 (146–417)	0.078
Fasting plasma glucose (mg/dL)	82 (65–96)[a]	84 (66–126)[a]	95 (78–225)[b]	118 (82–200)[b]	83 (70–100)[a]	0.001
HbA1c (%)	5.2 (4.5–6.1)[a]	5.3 (4.3–6)[a]	5.7 (4.8–6.4)[b]	6 (4.3–8.2)[b]	5.5 (4.7–5.7)[a]	0.001
Creatinine (mg/dL)	0.49 ± 0.1	0.48 ± 0.09	0.4 ± 0.09	0.48 ± 0.07	0.52 ± 0.12	0.170
AST (IU/L)	17 (12–38)	17 (9–50)	15 (6–37)	15 (5–25)	16 (7–47)	0.115
ALT (IU/L)	11 (5–83)	10 (5–34)	11 (7–30)	9 (5–20)	10 (5–44)	0.625
Triglyceride (mg/dL)	194 (106–412)	206 (112–597)	227 (79–435)	225 (143–378)	206 (86–522)	0.278
HDL-C (mg/dL)	62 ± 12.7	55 ± 12.1	54 ± 11.1	58 ± 10.3	58 ± 12.2	0.089
LDL-C (mg/dL)	118 ± 29	104 ± 33	107 ± 36	101 ± 40	123 ± 54	0.223

Parametric data were presented as mean with standard deviation and analyzed with the ANOVA test followed by the Tukey test for pairwise comparisons as appropriate. Non-parametric data were presented as median with minimum and maximum values and analyzed with the Kruskal–Wallis test followed by the Mann–Whitney test for pairwise comparisons as appropriate. Results of the pairwise comparisons were denoted with a letter, and there was no significant difference between/among the study groups marked with the same letter. There was a significant difference between/among the study groups marked with different letters ($p < 0.05$). GDM, gestational diabetes; PGDM, pregestational diabetes; GHT, gestational hypertension; WBC, white blood cell count; Hb, hemoglobin; Hct, hematocrit; PLT, platelet; HbA1c, hemoglobin A1C; AST, aspartate aminotransferase; ALT, alanine aminotransferase; IU, international unit; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol.

an serum BMP-2 levels compared to the group of only obesity ($p < 0.05$); and other pairwise comparisons provided no meaningful difference ($p > 0.05$).

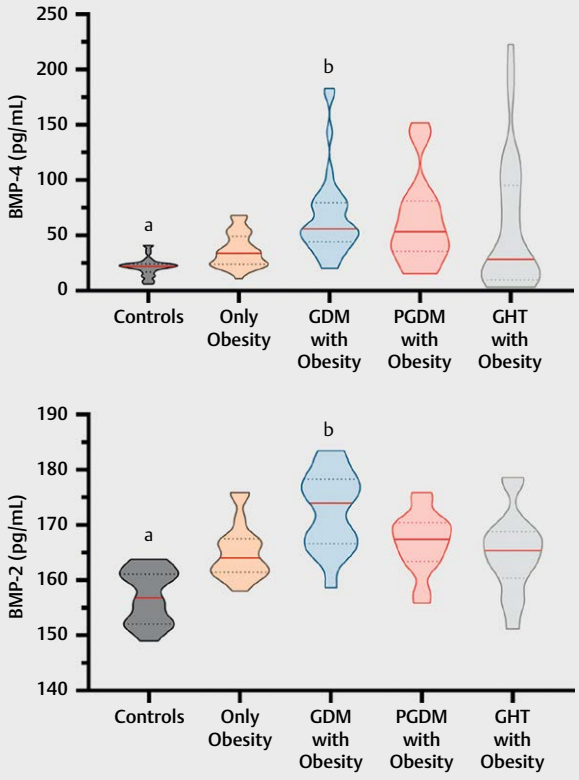
In ▶ **Fig. 2**, the median serum GDF-15, MMP-9, and GP39 levels of controls and groups of only obesity, GDM with obesity, PGDM with obesity, and GHT with obesity are shown. Regarding GDF-15 analysis, the median serum GDF-15 levels in the groups of only obesity, GDM with obesity, PGDM with obesity, and GHT with obesity were significantly higher than those of the controls ($p < 0.05$); the median serum GDF-15 levels in the group of GHT with obesity was significantly higher than that of the group of only obesity ($p < 0.05$); and other pairwise comparisons provided no meaningful difference ($p > 0.05$). Regarding MMP-9 analysis, the median serum MMP-9 levels in the groups of only obesity, GDM with obesity, PGDM with obesity, and GHT with obesity were significantly higher than those of the controls ($p < 0.05$); and other pairwise comparisons provided no meaningful difference ($p > 0.05$). Regarding GP39 analysis, the median serum GP39 levels in the groups of only obesity, GDM with obesity, PGDM with obesity, and GHT with obesity were significantly higher than those of the controls ($p < 0.05$); and other pairwise comparisons provided no meaningful difference ($p > 0.05$).

Discussion

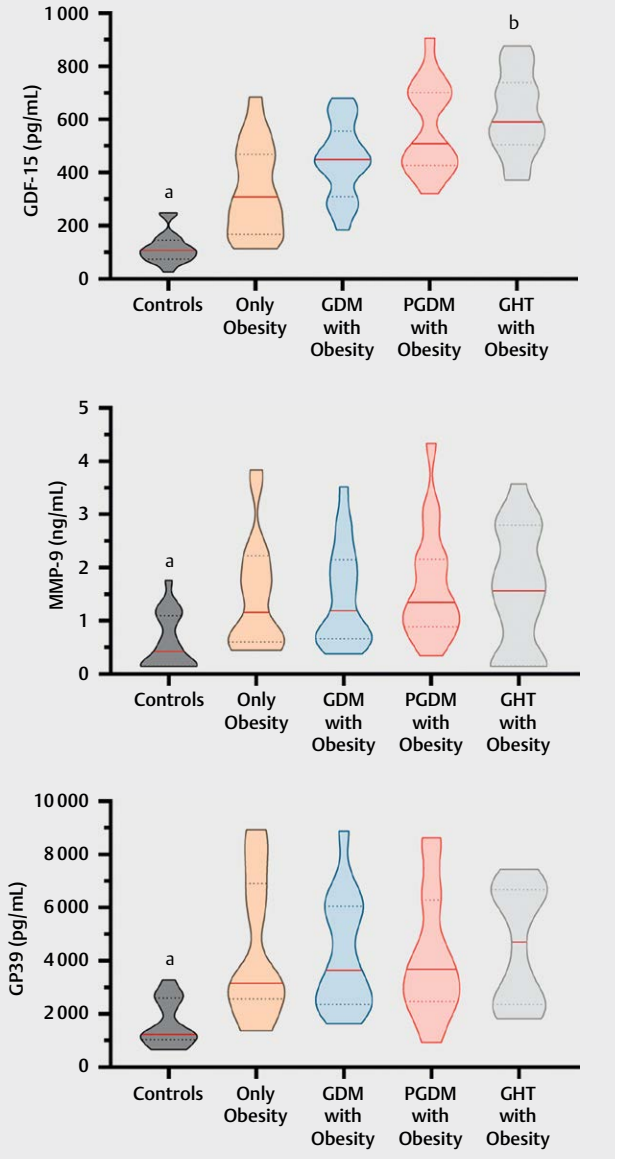
In this study investigating the clinical value of serum BMP-4, BMP-2, GDF-15, MMP-9, and GP39 levels in pregnant women with obesity and related comorbidities such as GDM, PGDM, and GHT, analyses of maternal and perinatal clinical and laboratory variables revealed that there were significant differences among the study groups regarding some of the variables. Overall, these differences were related to the nature of this study, which included pregnant women of a wide range of reproductive age (18–42 years), clinical background, and the type of disease-related care that they received. In addition, these results supported the clinical burden of obesity and obesity-related morbidities, which focused in this study on the clinical care of the mothers and their babies. During antenatal care, medical comorbidities including obesity, DM, and hypertension are increasingly prevalent [28], and as supported by the findings of this study, such as increased birthweight and cesarean rate, and NICU admission despite an effort to reduce the effects of obesity, the results of the current study highlight the importance of improved follow-up of these pregnant women as a requirement to reduce maternal and fetal/neonatal morbidities.

Maternal obesity and DM are two prevalent metabolic disorders that are often detected at the same time during pregnancy and are both connected to poor pregnancy outcomes, with their contribution to the state of metabolic homeostasis [29]. Glucose homeostasis is typically disrupted during normal pregnancy by metabolic changes that cause a physiologic form of insulin resistance [30]. This raises insulin demand, which is frequently related to obesity and other risk factors [31], and this progressive change may lead to GDM.

It is well established that GDM increases the likelihood of unfavorable outcomes in mothers and newborns. Interestingly, the incidence of GDM has increased concurrently with the growth in obesity, as well as the trend toward longer maternal age, less physical



▶ **Fig. 1** Median serum BMP-4 and BMP-2 levels of controls and only obesity, GDM with obesity, PGDM with obesity, and GHT with obesity. Non-parametric data were presented as median with minimum and maximum values and analyzed with the Kruskal–Wallis test followed by the Mann – Whitney test for pairwise comparisons as appropriate. [a]For BMP-4 and BMP-2, controls were significantly different compared to only obesity, GDM with obesity, PGDM with obesity, and GHT with obesity ($p < 0.05$). [b]For BMP-4, GDM with obesity was significantly different compared to only obesity and GHT with obesity ($p < 0.05$). [b]For BMP-2, GDM with obesity was significantly different compared to only obesity ($p < 0.05$).



▶ **Fig. 2** Median serum GDF-15, MMP-9, and GP39 levels of controls and only obesity, GDM with obesity, PGDM with obesity, and GHT with obesity. Non-parametric data were presented as median with minimum and maximum values and analyzed with the Kruskal–Wallis test followed by the Mann – Whitney test for pairwise comparisons as appropriate. [a]For GDF-15, MMP-9, and GP39, controls were significantly different compared to only obesity, GDM with obesity, PGDM with obesity, and GHT with obesity ($p < 0.05$). [b]For GDF-15, GHT with obesity was significantly different compared to only obesity ($p < 0.05$).

activity, and a greater prevalence of DM type 2 worsening during pregnancy due to the added burden of insulin resistance.

The pathophysiology of hypertension in pregnancy is not completely clarified. Preeclampsia occurs in around 15 %–25 % of women with GHT. Although numerous physiologic and biochemical screening tests have been developed, none have been shown to be highly predictive and just a few are currently utilized in clinical practice [32].

Molecular elucidation of the underlying pathways in obesity and obesity-associated comorbidities is currently one of the most active study subjects. Within this perspective, serum BMP-4, BMP-2, GDF-15, MMP-9, and GP39 levels in pregnant women with obesity and related comorbidities of GDM, PGDM, and GHT were examined to assess their status under the influence of obesity, obesity with DM, and obesity with GHT. According to the findings of our study, obesity increased serum levels of all the studied biomarkers; GDM developed in obese women caused a more pronounced increase in the serum levels of BMP-4 and BMP-2; GHT developed in obese women caused a more pronounced increase in the serum levels of GDF-15. In the women with GDM-, PGDM-, and GHT-complicated obesity, serum levels of MMP-9 and GP39 did not change meaningfully.

Serum BMP-4 levels were measured in 104 non-diabetic participants in a recent study to determine whether they may be utilized as a biomarker for metabolic syndrome in the non-diabetic state [33]. In that study, the serum BMP-4 levels were considerably increased in obese or metabolic syndrome individuals. It was discovered that elevated serum BMP-4 levels were associated with obesity, insulin resistance, and the existence of metabolic syndrome, implying that BMP-4 may contribute to the development of obesity and metabolic syndrome. Cai et al. performed an animal study to investigate the role of BMP-4 in GDM-related vascular endothelial dysfunction, which leads to hypertension. In that study, a GDM rat model was used to determine the expression levels of BMP-4 pathway-related proteins and vascular cell adhesion molecule 1 proteins in the rat abdominal aorta endothelium. They noted that BMP-4 might contribute to vascular endothelial dysfunction in pregnant rats by impairing endothelium-dependent vasodilation, resulting in increased blood pressure [34]. Kim et al. focused on determining changes in serum BMP-4 and pro-inflammatory cytokines following Roux-en-Y gastric bypass (RYGB), and RYGB was performed on 57 patients with DM type 2 [35]. Before and 12 months after RYGB, they found that serum levels of BMP-4 were considerably lower one year after RYGB.

Luna-Luna et al. conducted a study in young adults with obesity and overweight to measure adipokines, chemokines, and BMP-2 and their associations with clinical, biochemical, and anthropometric parameters. They noticed that overweight and obese participants had greater plasma levels of chemokines and adipokines, but there was no difference between the groups with regard to the plasma levels of BMP-2 [36]. Another study examined whether BMP-2 is linked to the distribution of adipose tissue (AT) in obesity. Serum BMP-2 levels were evaluated in paired visceral and subcutaneous AT samples. BMP-2 protein in both depots was significantly higher in obese persons when compared to healthy and lean individuals. Serum BMP-2 levels were significantly higher in DM type 2 individuals who were moderately obese but not morbidly obese. Their findings indicated that as the demand for energy storage grows, AT BMP-2 levels increase, possibly contributing to the partitioning of energy storage into visceral and subcutaneous AT depots [37].

Serum GDF-15 levels have been found to be elevated in patients with insulin resistance or DM type 2 in previous cross-sectional investigations [38, 39], and a study of GDF-15 in Chinese pregnant women related elevated GDF-15 to GDM [40]. In a recent study that was conducted to investigate how GDF-15 levels changed during and after pregnancy in women of normal weight and women who were obese, they also looked at how changes in beta cell activity were related to variations in GDF-15 levels [41]. In that study, GDF-15, insulin, and fasting glucose were all measured. GDF-15 levels increased significantly throughout pregnancy and were significantly greater than in the postpartum condition. They found that during pregnancy, GDF-15 levels were higher in normal-weight women than in obese women but were reversed postpartum. They revealed that increasing serum GDF-15 levels throughout pregnancy, and to a larger extent in normal-weight women than in obese women, was associated with decreased glucose and higher insulin secretory function in normoglycemic obese pregnancies. Contrary to their finding, we could not find a more pronounced increase in serum GDF-15 levels in healthy pregnant women compared to those with obesity and obesity-complicated diseases.

In several studies, MMP-9 levels in obesity and GHT are inconclusive, and in some studies, there were increased MMP-9 levels in obese or hypertensive pregnant women [42, 43], whereas others found no difference [44], or lower MMP-9 levels [45, 46]. Our findings support that serum MMP-9 level had a relationship with obesity, GDM, PGDM, and GHT.

Hampen et al. investigated the levels of GP39 in serum samples from morbidly obese patients and healthy controls before and after bariatric surgery. They demonstrated for the first time that increased levels of GP39 dropped following substantial weight loss via bariatric surgery [47]. In a systemic review that explored the relationship between GP39 and DM, it was suggested that the serum level of GP39 is increased in the DM population [48].

This study had some limitations related to its design including the measurement of studied serum biomarkers only in the first perinatology examination at the second or third trimester of pregnancy but not during each trimester of pregnancy. Since the study included disease states of GDM, PGDM, and GHT in obese women, there may be the impact of other clinical variables as a confounding factor that may mediate the serum levels of studied biomarkers. However, this study examined considerably new serum biomarkers as a panel to determine their significance for obesity and its important comorbidities including GDM, PGDM, and GHT in a broad perspective. Overall, our findings support the hypothesis that obesity was the main effector in increasing the serum levels of these biomarkers. BMP-4 and BMP-2 provided additional increases in GDM and GDF-15 had an additional increase in GHT. Their pattern in obese pregnant women needs to be examined in further studies measuring them serially during pregnancy.

In conclusion, in obese women with or without GDM, PGDM, and GHT, BMP-4, BMP-2, GDF-15, MMP-9, and GP39 have a potential to be used as a predictive and diagnostic test panel after further studies are performed with more homogeneously enrolled participants. Because obesity plays a central role in the development of numerous complications and comorbidities, robust and reliable clinical biomarkers would be beneficial in improving disease diagnosis and classification, early diagnosis or prediction of the risk of secondary events, and more personalized and precise medical treatment. Future studies should look into the benefits and drawbacks of various screening approaches and diagnostic thresholds on maternal and newborn outcomes as well as their impact on maternal well-being and health expenses.

Conflict of Interest

The authors declare that they have no conflict of interest.

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