

# Is There An Association Between Kisspeptin Levels And Gestational Diabetes Mellitus?

Emine ARSLAN<sup>1</sup>, Umit GORKEM<sup>1</sup>, Cihan TOGRUL<sup>1</sup>

Corum, Turkey

## ABSTRACT

**OBJECTIVE:** To find out the relationship of maternal serum levels of kisspeptin with gestational diabetes mellitus status.

**STUDY DESIGN:** A total of 158 pregnant women between 24 and 28 weeks of gestation were divided into two groups according to gestational diabetes mellitus status: (i) Pregnant women with gestational diabetes mellitus (gestational diabetes mellitus group, n=76) and (ii) Healthy pregnant women (control group, n=82). Maternal serum concentrations of kisspeptin, insulin, and homeostasis model assessment-insulin resistance were assessed.

**RESULTS:** In both groups, there were no difference in terms of age and gestational age ( $p=0.058$  and  $p=0.820$ , respectively). The median of body mass indices of both groups at 24 to 28 weeks of gestation were statistically similar ( $p=0.062$ ). The serum concentrations of kisspeptin did not demonstrate significant differences between the gestational diabetes mellitus and control groups ( $p=0.28$ ). There was a significant difference in terms of serum level of insulin and homeostasis model assessment-insulin resistance between the gestational diabetes mellitus and control groups ( $p<0.001$ ).

**CONCLUSION:** No differences were found in serum kisspeptin levels between pregnant women with GDM and healthy pregnant women. Further prospective studies will be essential to elucidate the contribution of kisspeptin to gestational diabetes mellitus.

**Keywords:** Gestational diabetes, Kisspeptin, Metabolism

*Gynecol Obstet Reprod Med* 2020;26(3):179-183

## Introduction

Kisspeptin, also called metastin, was first discovered from a metastasis suppressor gene, KISS1 in 1996 (1). Since the time it was first discovered, it has been understood that kisspeptin was involved in many issues including central control of the gonadotropic axis, placentation, pregnancy, energy homeostasis and cardiovascular function (2,3). Therefore, many studies from different fields like cancer biology, repro-

ductive neuroendocrinology, reproductive biology and metabolism have started to focus on kisspeptin.

Kisspeptin is also highly expressed in the placenta (1,4,5). During a normal pregnancy, measurement of its levels revealed 1000 to 7000 fold increase (6). In vitro, kisspeptin inhibits the migration and invasion of trophoblast cells. Therefore, it was concluded that kisspeptin plays an important role in restricting trophoblast invasion, regulating implantation and subsequent placental development (3,7,8). There was a correlation between kisspeptin levels and perinatal outcomes. The decreased kisspeptin levels at first trimester of pregnancy resulted in the delivery of a small for gestational age fetus. In addition, pregnancies with decreased kisspeptin levels at second trimester were complicated with fetal growth restriction, and preeclampsia (9,10).

Gestational diabetes mellitus (GDM) is defined as a carbohydrate intolerance which occurs during pregnancy (11). Up to 6-7% of pregnancies are thought to be complicated by diabetes mellitus and approximately 86% of these cases are gestational diabetes mellitus (12). In a large-sized study from Turkey, the authors stated that the national prevalence of GDM was found to be 16.2% without a significant difference between urban and rural regions (13). In another cross-

<sup>1</sup> Hitit University Faculty of Medicine, Department of Obstetrics and Gynecology, Corum, Turkey

Address of Correspondence: Emine Arslan  
Hitit University Faculty of Medicine,  
Department of Obstetrics and Gynecology,  
19040 Corum, Turkey  
ekaranfildr@gmail.com

Submitted for Publication: 31.03.2019

Accepted for Publication: 27.05.2019

ORCID IDs of the authors: EA:0000-0003-2259-8376  
UG:0000-0002-0848-9731, CT: 0000-0003-3814-3632

Quick Response Code:	Access this article online
	Website: <a href="http://www.gorm.com.tr">www.gorm.com.tr</a>
	e-mail: <a href="mailto:info@gorm.com.tr">info@gorm.com.tr</a>
	DOI:10.201613/GORM.2018.946

**How to cite this article:** Arslan E, Gorkem U, Togrul C. Is There An Association Between Kisspeptin Levels And Gestational Diabetes Mellitus? *Gynecol Obstet Reprod Med*. 2020;26(3):179-183

tional study from Turkey, 4684 (6.07%) of 77227 patients who were screened for GDM were diagnosed as gestational diabetes mellitus (14). In that study, the patients were evaluated by either two step or one step glucose tolerance tests. Fasting plasma glucose (FPG) testing was evaluated in 144 113 women and 21% of these were between 92-126 mg/dL, in GDM diagnosis range according to International Association of Diabetes and Pregnancy Study Group recommendations. In this study FPG dependent GDM prevalence was almost four times higher than two-step glucose screening test.

Gestational diabetes mellitus is associated with variable severity of maternal and perinatal complications such as fetal macrosomia, shoulder dystocia, birth injuries, neonatal hypoglycemia, respiratory distress syndrome, perinatal death, childhood obesity, preeclampsia, and cesarean delivery (15). According to that, it is critical to identify gestational diabetes risk factors, predisposing factors, appropriate preventive measures, and treatment strategies.

Even though numerous functions of kisspeptin in the reproductive pathways have been emphasized in the previous studies (2,16,17), some other studies have recently started to focus on the peripheral functions of kisspeptin. In the existing literature, there is a body of evidence showing that kisspeptin has important roles in regulating glucose homeostasis, insulin secretion, food intake and body composition. It is also known that inadequate kisspeptin signaling results in decreased locomotor activity and increased adiposity (18-21).

Since GDM is one of the most important metabolic problems seen in pregnancy and a close association between glucose metabolism and the kisspeptin has been detected, we aimed to evaluate the relationship between serum kisspeptin levels and GDM status in pregnant women.

## Material and Method

### *Study population*

We carried out this cross-sectional study between March 31, 2015 and September 30, 2015 at the Hitit University Hospital. The study was approved by the ethics committee of Ankara Numune Hospital which was in accordance with the Declaration of Helsinki, 2013 Brazil version (20796219-724.131). The written informed consents for the study were gathered from all participants just prior to the study.

In the study, all pregnant women between 18 to 35 years of age had singleton pregnancies. Advanced maternal age (>35 years of age), personal history of diabetes mellitus and/or GDM in previous pregnancies, history of chronic disease affecting carbohydrate metabolism, history of congenital malformation, family history of diabetes mellitus, and smoking were accepted as the exclusion criteria for the present study. Patients between 24 and 28 weeks of gestation who admitted to our obstetrics outpatient clinic were screened for GDM according to the recommendations of the American College of Obstetricians and Gynecologists (11).

All pregnant women in the low-risk group were evaluated with a 50-g glucose challenge test (GCT). Women with serum glucose  $\geq 140$  mg/dL at 1 h after GCT were subjected to a 100-g oral glucose tolerance test (OGTT). Serum glucose concentrations were measured at 0, 1, 2, and 3 h after glucose ingestion. The diagnosis of GDM was based on the criteria of Carpenter and Coustan (21), in which, after a 100-g oral glucose load, 2 or more of the following plasma values must be obtained: fasting  $\geq 95$  mg/dL, 1h  $\geq 180$  mg/dL, 2h  $\geq 155$  mg/dL, and 3h  $\geq 140$  mg/dL (22). The estimation of pregnancy duration was based on routine ultrasonographic examination performed in the first trimester. BMI was calculated using pregnancy weight and height, which were recorded at the time of blood sampling.

A total of 158 pregnant women who met the inclusion criteria were included. The study population was divided into two groups according to GDM status: (i) Pregnant women with GDM (GDM group, n=76) and (ii) Healthy pregnant women (control group, n=82). The demographic and biochemical characteristics of the study population, including age, BMI, and gestational age, were recorded in the second trimester.

### *Sample Collection and Assays*

The blood samples of the participants were collected after overnight fasting and between 08 AM and 10 AM for biochemical measurements. To measure kisspeptin levels, the sera were frozen at 20 °C within 2 h, for a maximum of 7 days and then analyzed. The kisspeptin 54 levels were determined with enzyme-linked immunosorbent assay (ELISA) method using Human KISS-54 kits (Biotek Synergy HT, Mybiosource, San Diego, CA). Maternal serum concentrations of kisspeptin, insulin and homeostasis model assessment-insulin resistance (HOMA-IR) were assessed. Serum glucose levels were determined daily using the glucose hexokinase method (Siemens Healthcare Diagnostic Limited; Camberley, UK). Serum insulin concentrations were measured by chemiluminescence assay (Advia Centaur, Siemens Medical Solutions Diagnostics; Tarrytown, USA). HOMA-IR was calculated using the following formula: Plasma glucose (mg/dL)  $\times$  fasting plasma insulin (IUmg/L) in the fasting state divided by 405. All data were compared and correlation analyses were performed.

### *Statistical analysis*

Data were analyzed using SPSS (Statistical Package for the Social Sciences) software version 21 (IBM Corp. Armonk, NY, USA). Continuous variables were first evaluated for normality of statistical distribution by the Shapiro-Wilk test. As the continuous variables were not normally distributed, a non-parametric method (Mann-Whitney U test) was used to perform the statistical analysis. Spearman correlation tests were used to determine the correlations of continuous variables. p values <0.05 were considered to be significant in all analyses.

## Results

The maternal demographic and biochemical characteristics of the study, participants are summarized in table I. No difference was confirmed in maternal age and gestational duration between the GDM and the control groups ( $p=0.058$  and  $p=0.820$ , respectively). The median values of body mass indices (BMI) of both groups were statistically similar ( $p=0.062$ ). The significant increase in serum insulin and HOMA concentrations were confirmed in GDM group ( $p<0.001$  and  $p<0.001$ , respectively). There was no statistically significant difference of serum kisspeptin level between healthy pregnant women and pregnant women with GDM ( $p=0.280$ ). We demonstrated that kisspeptin levels were higher in GDM group, but the difference did not reach statistical significance ( $p>0.05$ ).

We evaluated the correlation of kisspeptin with other study parameters including maternal age, BMI, gestational duration, insulin, HOMA-IR in both groups, as presented in table II. We did not find any correlation between kisspeptin levels and maternal age, body mass index, gestational age, insulin level, and HOMA-IR value in GDM and control groups ( $p>0.05$ , for all).

## Discussion

This study mainly focused on the association of serum kisspeptin level with GDM status. Kisspeptin levels were found to be higher in GDM group, but the difference was not statistically significant. Kisspeptin has several functions in

metabolic pathways as participation in islet hormone crosstalk (18,23-26), glucose-stimulated insulin secretion, and insulin sensitivity (27,28). Studies investigating the effect of kisspeptin on metabolism have shown that impaired glucose tolerance, higher body weight, leptin levels, and adiposity were found in kisspeptin deficient rats (29,30). Kisspeptin has also been shown to be responsible for the formation of an obese phenotype independent of gonadal estrogen deficiency (19).

There are numerous studies demonstrating the relationship between kisspeptin and diabetes mellitus (DM) in the known literature. Increased circulating plasma kisspeptin concentrations and increased liver kisspeptin expression is detected in high-fat diet fed obese and glucose-intolerant mice, as well as from a mouse model of Type 2 DM (T2DM) (18). Furthermore, the levels of kisspeptin in liver biopsies were higher than those without T2DM. However, in patients with T2DM, the levels of kisspeptin may vary, regardless of the condition of treatment for diabetes mellitus. At this point, it is not obvious whether there is a subgroup of T2DM with elevated kisspeptin expression and it is unclear how this subgroup can be characterized.

In a normal human pregnancy, the time when the insulin resistance develops and reaches its peak is considered to be between the 34-36th weeks of the gestation (31). This increase in insulin resistance is thought to be an adaptive mechanism of organism to provide a slight excess of energy substrates for the developing fetus. There are two theoretical mechanisms of kisspeptin on insulin-glucose metabolism in pregnancy

**Table I:** Comparison of clinical and biochemical parameters in control and gestational diabetes groups

	Control Group (n=82, 51.9%)	GDM Group (n=76, 48.1%)	<i>p</i>
Maternal age (years)	27.9±5.2	30.5±5.8	0.058
BMI (kg/m <sup>2</sup> )	28.6±4.3	30.6±3.7	0.062
Gestational duration (weeks)	25.0±5.0	23.0±7.9	0.820
Insulin (mLU/mL)	10.5±4.9	16.2±6.5	<0.001*
HOMA-IR	1.8±0.9	4.1±1.9	<0.001*
Kisspeptin (ng/mL)	161.3±78.2	187.6±132.3	0.280

Values are shown as mean ± standard deviation. GDM; gestational diabetes, BMI; Body mass index, HOMA-IR; Homeostasis model assessment of insulin resistance. \**p*-values indicate statistically significant ( $p<0.05$ ).

**Table II.** Correlation of kisspeptin with other study parameters in control and gestational diabetes groups

	Control Group (n=82, 51.9%)	GDM Group (n=76, 48.1%)	<i>p</i>
Maternal age (years)	-0.277	0.004	0.526
BMI (kg/m <sup>2</sup> )	-0.172	-0.118	0.740
Gestational duration (weeks)	-0.003	-0.086	0.625
Insulin (mLU/mL)	-0.222	-0.254	0.518
HOMA-IR	-0.216	-0.127	0.840

Values are shown as Pearson's correlation coefficient (*r*). GDM; gestational diabetes, BMI; Body mass index, HOMA-IR; Homeostasis model assessment of insulin resistance.

The first possible mechanism is that: Increased kisspeptin serves to suppress increased insulin secretion to provide a necessary level of redundancy in blood levels of energy substrates by a KISS1R dependent mechanism. The other thought is that kisspeptin serves to increase insulin secretion to compensate for increased insulin resistance in late pregnancy through a receptor-independent mechanism (6,31).

As mentioned above, high levels of kisspeptin in maternal circulation during normal pregnancy may have a physiological role to protect the mother and fetus from increasing glucose levels and the development of GDM. Cetkovic et al. revealed decreased levels of kisspeptin in GDM in their study which is, to the best of our knowledge, the first study that investigated the relationship between kisspeptin and GDM (32).

When the literature about the kisspeptin is investigated, there are conflicting results about the effect of kisspeptin on insulin secretion. This conflict can be due to the unreliability of the commercially available methods. The reason for non-reliable methods might be the variations in the assay methods, their ranges of detection, and uncertainty about which forms of kisspeptin (i.e., KP10, KP15, KP54) is detected.

The main limitation of the current study is the lack of postpartum follow-up. Also, we did not investigate the complications associated with GDM and newborn follow up is not included in our study. However, in the known literature, there is limited number of studies involving kisspeptin levels and GDM status. Especially, a small number of human studies were published. This issue can be considered a strong side of the study.

In summary, we have demonstrated no association of serum kisspeptin concentration with GDM status in pregnant women. According to the literature, kisspeptin is associated with glucose metabolism. We suggest that long term observation of kisspeptin during pre-pregnancy, pregnancy and postpartum periods would increase our understanding of the pathogenesis of GDM. So, further large-sized prospective studies will be essential to elucidate the contribution of kisspeptin to GDM.

*Conflict of Interest: The authors declare that they have no competing interests.*

*Financial Support: None*

*Acknowledgement: This study was presented as oral presentation at Perinatal Medicine 2017 Congress 28-29 April 2017, Izmir, Turkey*

*Authors' contributions: EA: Hypothesis development, planning methodology, data collection, data analysis and interpretation, article writing, UG: Data collection, planning methodology, critical review, CT: Data collection.*

## References

1. Lee JH, Miele ME, Hicks DJ, Phillips KK, Trent JM, Weissman BE, et al. KiSS-1, a novel human malignant melanoma metastasis suppressor gene. *J Natl Cancer Inst.* 1996;88(23):1731-7. doi: 10.1093/jnci/88.23.1731
2. Seminara SB, Messenger S, Chatzidaki EE, Thresher RR, Acierno JS Jr, Shagoury JK, et al. The GPR54 gene as a regulator of puberty. *N Engl J Med.* 2003;349(17):1614-27. doi: 10.1056 / NEJMoa035322.
3. Bilban M, Ghaffari-Tabrizi N, Hintermann E, Bauer S, Molzer S, Zoratti C, et al. isspeptin-10, a KiSS-1/ metastatin-derived decapeptide, is a physiological invasion inhibitor of primary human trophoblasts. *J Cell Sci.* 2004;117(Pt 8):1319-28. doi: 10.1242/jcs.00971
4. Kotani M, Detheux M, Vandenbogaerde A, Communi D, Vanderwinden JM, Le Poul E, et al. The metastasis suppressor gene KiSS-1 encodes kisspeptins, the natural ligands of the orphan G protein-coupled receptor GPR54. *J Biol Chem.* 2001;276(37):34631-6. doi: 10.1074/jbc.M104847200.
5. Ohtaki T, Shintani Y, Honda S, Matsumoto H, Hori A, Kanehashi K, et al. Metastasis suppressor gene KiSS-1 encodes peptide ligand of a G-protein coupled receptor. *Nature.* 2001;411(6837):613-7. doi: 10.1038/35079135.
6. Horikoshi Y, Matsumoto H, Takatsu Y, Ohtaki T, Kitada C, Usuki S, et al. Dramatic elevation of plasma metastatin concentrations in human pregnancy: metastatin as a novel placenta-derived hormone in humans. *J Clin Endocrinol Metab.* 2003;88(2):914-9. doi: 10.1210/jc.2002-021235
7. Janneau JL, Maldonado-Estrada J, Tachdjian G, Miran I, Motte N, Saulnier P, et al. Transcriptional expression of genes involved in cell invasion and migration by normal and tumoral trophoblast cells. *J Clin Endocrinol Metab.* 2002;87(11):5336-9. doi: 10.1210/jc.2002-021093.
8. Hiden U, Bilban M, Knofler M, Desoye G. Kisspeptins and the placenta: regulation of trophoblast invasion. *Rev Endocr Metab Disord.* 2007;8(1):31-9. doi: 10.1007/s11154-007-9030-8.
9. Smets E, Deurloo KL, Go AT, van Vugt JM, Blankenstein MA, Oudejans CB. Decreased plasma levels of metastatin in early pregnancy are associated with small for gestational age neonates. *Prenat Diagn.* 2008;28(4):299-303. doi: 10.1002/pd.1969.
10. Armstrong RA, Reynolds RM, Leask R, Shearing CH, Calder AA, Riley SC. Decreased serum levels of kisspeptin in early pregnancy are associated with intrauterine growth restriction and pre-eclampsia. *Prenat Diagn.* 2009;29(10):982-5. doi: 10.1002/pd.2328
11. Committee on Practice Bulletins-Obstetrics. ACOG Practice Bulletin. No. 190: gestational diabetes mellitus. *Obstet Gynecol.* 2018;131(2):e49-e64.
12. Correa A, Bardenheier B, Elixhauser A, Geiss LS, Gregg E. Trends in prevalence of diabetes among delivery hospi-

- talizations, United States, 1993-2009. *Matern Child Health J.* 2015;19(3):635-42. doi: 10.1007/s10995-014-1553-5.
13. Aydın H, Celik O, Yazıcı D, Altunok C, Tarcın O, Deyneli O, et al. Prevalence and predictors of gestational diabetes mellitus: a nationwide multicentre prospective study. *Diabet Med.* 2019;36(2):221-7. doi: 10.1111/dme.13857.
  14. Ozgu-Erdinc AS, Sert UY, Buyuk GN, Engin-Ustun Y. Prevalence of gestational diabetes mellitus and results of the screening tests at a tertiary referral center: A cross-sectional study. *Diabetes Metab Syndr.* 2019;13(1):74-7. DOI: 10.1016/j.dsx.2018.08.019
  15. Ayhan S, Altinkaya SO, Gungor T, Ozcan U. Prognosis of pregnancies with different degrees of glucose intolerance. *Gynecol Obstet Reprod Med.* 2016;19(2):76-81.
  16. Smith JT, Dungan HM, Stoll EA, Gottsch ML, Braun RE, Eacker SM, et al. Differential regulation of KiSS-1 mRNA expression by sex steroids in the brain of the male mouse. *Endocrinology.* 2005;146(7):2976-84. doi: 10.1210/en.2005-0323. Epub 2005 Apr 14.
  17. Smith JT, Cunningham MJ, Rissman EF, Clifton DK, Steiner RA. Regulation of kiss1 gene expression in the brain of the female mouse. *Endocrinology.* 2005;146(9):3686-92. doi: 10.1210/en.2005-0488.
  18. Song WJ, Mondal P, Wolfe A, Alonso LC, Stamateris R, Ong BW, et al. Glucagon regulates hepatic kisspeptin to impair insulin secretion. *Cell Metab.* 2014;19(4):667-81. doi: 10.1016/j.cmet.2014.03.005.
  19. Tolson KP, Garcia C, Yen S, Simonds S, Stefanidis A, Lawrence A, et al. Impaired kisspeptin signaling decreases metabolism and promotes glucose intolerance and obesity. *J Clin Invest.* 2014;124(7):3075-9. doi: 10.1172/JCI171075.
  20. Katugampola H, King PJ, Chatterjee S, Meso M, Duncan AJ, Achermann JC, et al. Kisspeptin is a novel regulator of human fetal adrenocortical development and function: a finding with important implications for the human fetoplacental unit. *J Clin Endocrinol Metab.* 2017;102(9):3349-59. doi: 10.1210/jc.2017-00763.
  21. Taylor J, Pampillo M, Bhattacharya M, Babwah AV. Kisspeptin/KISS1R signaling potentiates extravillous trophoblast adhesion to type-I collagen in a PKC- and ERK1/2-dependent manner. *Mol Reprod Dev.* 2014; 81(1):42-54. doi: 10.1002/mrd.22279.
  22. Carpenter MW, Coustan DR. Criteria for screening tests for gestational diabetes. *Am J Obstet Gynecol.* 1982; 144(7):768-73. doi: 10.1016/0002-9378(82)90349-0.
  23. Muller WA, Faloona GR, Unger RH. Hyperglucagonemia in diabetic ketoacidosis. Its prevalence and significance. *Am J Med.* 1973;54(1):52-7. doi: 10.1016/0002-9343(73)90083-1.
  24. Dumonteil E, Magnan C, Ritz-Laser B, Meda P, Dussoix P, Gilbert M, et al. Insulin, but not glucose lowering corrects the hyperglucagonemia and increased proglucagon messenger ribonucleic acid levels observed in insulinopenic diabetes. *Endocrinology.* 1998;139(11):4540-6. doi: 10.1210/endo.139.11.6294.
  25. Kawamori D, Kurpad AJ, Hu J, Liew CW, Shih JL, Ford EL, et al. Insulin signaling in alpha cells modulates glucagon secretion in vivo. *Cell Metab.* 2009;9(4):350-61. doi: 10.1016/j.cmet.2009.02.007.
  26. D'Alessio, D. The role of dysregulated glucagon secretion in type 2 diabetes. *Diabetes Obes Metab.* 2011;13(1):126-32. doi: 10.1111/j.1463-1326.2011.01449.x.
  27. Hussain MA, Song WJ, Wolfe A. There is kisspeptin – and then there is kisspeptin. *Trends Endocrinol Metab.* 2015;26(10):564-72. doi: 10.1016/j.tem.2015.07.008.
  28. Hauge-Evans AC, Richardson CC, Milne HM, Christie MR, Persaud SJ, Jones PM. A role for kisspeptin in islet function. *Diabetologia.* 2006;49(9):2131-5. doi: 10.1007/s00125-006-0343-z.
  29. Silvestre RA, Egido EM, Hernandez R, Marco J. Kisspeptin-13 inhibits insulin secretion without affecting glucagon or somatostatin release: study in the perfused rat pancreas. *J Endocrinol.* 2008;196(2):283-90. doi: 10.1677/JOE-07-0454.
  30. Vikman J, Ahren B. Inhibitory effect of kisspeptins on insulin secretion from isolated Mouse islets. *Diabetes Obes Metab.* 2009;11(4):197-201. doi: 10.1111/j.1463-1326.2009.01116.x.
  31. Ryan EA. Hormones and insulin resistance during pregnancy. *Lancet.* 2003;362(9398):1777-8. doi: 10.1016/S0140-6736(03)14942-2.
  32. Cetković A, Miljic D, Ljubić A, Patterson M, Ghatei M, Stamenković J, et al. Plasma kisspeptin levels in pregnancies with diabetes and hypertensive disease as a potential marker of placental dysfunction and adverse perinatal outcome. *Endocrine Research.* 2012;37(2):78-88. doi: 10.3109/07435800.2011.639319.