RESEARCH PAPER

The impact of severe hypertriglyceridemia on glycosylated hemoglobin test in type 2 diabetes mellitus

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Abstract

Background: although the glycosylated hemoglobin (HbA1c) test is a valid test for diabetes diagnosis and evaluation of glycemic control, many factors are known to affect the results of the HbA1c test and make it reflects a false image of the glycemic control. One of these factors is hypertriglyceridemia.

Objectives: the study aims to assess the impact of severe hypertriglyceridemia (TG ≥ 1000 mg/dl) on HbA1c test results in type 2 diabetes mellitus.

Method: a retrospective study on two groups of adult patients with type 2 DM; were implemented in Faiha Diabetes, Endocrine and Metabolism Centre (FDEMC) in Basrah–Iraq. A total of 267 patients with type 2 DM and severe hypertriglyceridemia and a total of 313 patients with type 2 DM and normal triglycerides levels were included. Then the mean HbA1c of the patients with severe hypertriglyceridemia was measured in each FBS range and compared to the mean HbA1c of the patients with normal triglycerides levels in the same FBS range to determine the impact of severe hypertriglyceridemia on the readings of HbA1c in the diabetic patients.

Results: the mean HbA1c in both groups was compared in each FBS range and revealed that despite of recording nearly the same mean FBS, HbA1c was higher in patients with severe hypertriglyceridemia by 1.33% in the FBS range 130-149 mg/dl with a statistically significant p-value = 0.003. While it was lower in the FBS range 200-229 mg/dl, 230-259 mg/dl, 260-299 mg/dl, 300-349 mg/dl and 350-399 mg/dl by 1.02%, 0.85%, 1.77%, 2.29% and 2.23%, respectively with a statistically significant p-value that equal to 0.005, 0.034, < 0.001, < 0.001 and 0.039, respectively.

Conclusion: this study reveal that severe hypertriglyceridemia has a significant impact on HbA1c test results. The HbA1c values in diabetic patients with severe hypertriglyceridemia are unpredictable.

Keywords: HbA1c test, type 2 DM, severe hypertriglyceridemia.

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Introduction

Glycosylated hemoglobin (HbA1c) is the percentage of hemoglobin A that has glucose linked to it. While the percentage is generally at a low level, in diabetes mellitus the increased blood glucose causes greater hemoglobin binding, resulting in a higher HbA1c value.[1] Glycosylated hemoglobin (HbA1c) has long been recognized as a valid and reliable test for diabetes diagnosis and evaluating glycemic control in diabetic patients.[2] Its result reflects the mean glucose level during the previous 8 to 12 weeks. It has less pre-analytic and biological variability than other diabetes diagnostic tests,

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and it is more convenient for patients and doctors because no fasting is needed.[3]As a result, clinical practice guidelines and societies have endorsed the HbA1c as the standard for determining whether a patient is in or out of the glycemic control range, as well as for guiding treatment decisions regarding adjustments in the treatment regimen.[4] However, its reliability is compromised in certain medical situations such as anemia, hemoglobinopathies, acute blood loss within three months, blood transfusion within three months, chronic ingestion of alcohol, salicylates, opioids, and high doses of vitamins A and C, chronic kidney disease, chronic liver disease, HIV on antiretroviral therapy, pregnancy, hypersplenism, hypertriglyceridemia and hyperbilirubinemia.[5] Not identifying these factors may influence the decision-making process and adversely affects the patients' health by intensifying the treatment and the risk of adverse events like hypoglycemia or wrongly de-intensify it.[6] There are four most used techniques for measuring HbA1c. They are immunoassay, ion-exchange high-performance liquid chromatography, boronate affinity high-performance liquid chromatography, and enzymatic assays.[7] The small intestine absorbs dietary triglycerides, which are then released into the lymph system before entering the systemic circulation as chylomicrons via the thoracic duct. Some of the triglycerides are removed from the chylomicrons by muscle and adipose tissues, and the remaining chylomicrons are taken up by the liver and processed into cholesterol-rich lipoproteins. Although the small intestine absorbs the majority of the triglycerides in the blood, the liver generates and secretes a little quantity.[8] Many causes can lead to hypertriglyceridemia including: obesity, overeating, diabetes mellitus and insulin resistance, excess alcohol intake, nephrotic syndrome and kidney failure, some types of familial hyperlipidemia like familial combined hyperlipidemia (Type II hyperlipidemia), deficiency of lipoprotein lipase, hypothyroidism, systemic lupus erythematosus and medications like beta-blockers, anti-psychotics, bile acid sequestrants, estrogen, immunosuppressants, protease inhibitors, tamoxifen, glucocorticoids, thiazides and isotretinoin.[9] Hypertriglyceridemia must be diagnosed using fasting serum triglycerides levels. According to Endocrine Society, mild hypertriglyceridemia is defined as triglycerides level of 150-199 mg/dl, moderate hypertriglyceridemia is defined as triglycerides level of 200-999 mg/dl and severe hypertriglyceridemia is defined as triglycerides level of ≥ 1000 mg/dl.[10]

Patients and methods

A retrospective study on two groups of adult patients (Age >18 years) with type 2 DM; those with normal triglycerides levels (TG < 150 mg/dl) and those with severe hypertriglyceridemia (TG ≥ 1000 mg/dl) were implemented in Faiha Diabetes, Endocrine and Metabolism Centre (FDEMC) in Basrah–Iraq. Data were collected from the electronic registry of patients who consulted Faiha Diabetes, Endocrine and Metabolism Centre (FDEMC). A total of 267 patients with type 2 DM and severe hypertriglyceridemia were collected from the electronic registry of patients who consulted to Faiha Diabetes, Endocrine and Metabolism Centre (FDEMC) from the 24th of August, 2008 to the 16th of September, 2021. A total of 313 patients with type 2 DM and normal triglycerides levels were collected from the electronic registry of patients who consulted FDEMC from the 18th of July, 2021 to the 16th of September, 2021. For both groups, we recorded patients' age, sex, body

mass index, fasting blood sugar, glycosylated hemoglobin, Fasting triglycerides level, LDL, VLDL, HDL, cholesterol, non-HDL and duration of diabetes. Both groups of patients were subdivided into sub-groups each one with its specific range of FBS. The ranges of FBS in these sub-groups were chosen to ensure good number of patients in each sub-group and nearly the same mean FBS in patients with normal triglycerides levels and those with severe hypertriglyceridemia in each range of FBS. Then the mean HbA1c of patients with severe hypertriglyceridemia was measured in each FBS range and compared to the mean HbA1c of patients with normal triglycerides levels in the same FBS range to determine the impact of severe hypertriglyceridemia on readings of HbA1c test in diabetic patients (type 2). Patients with conditions, other than severe hypertriglyceridemia, that affect or may affect the HbA1c test results (anemia, hemoglobinopathies, acute blood loss within three months, blood transfusion within three months, chronic ingestion of alcohol, salicylates, opioids, and high doses of vitamins A and C, chronic kidney disease, chronic liver disease, HIV on antiretroviral therapy, pregnancy, hypersplenism, hypertriglyceridemia and hyperbilirubinemia)[5] have been excluded from the study. The Continuous variables have been summarized by using descriptive statistics presented as mean and SD. Categorical variables have been summarized by using counts and percentages. This study has considered the two-tailed probability values with ($p \leq 0.05$) to be statistically significant. All statistical calculations were performed with IBM SPSS Statistics 26 for Windows.

Results

Both groups (those with normal triglycerides levels and those with severe hypertriglyceridemia) were divided into sub-groups each one with its specific FBS range. The mean FBS and the mean HbA1c were measured in each sub-group, (Table-1).

Table 1. Variation of HbA1c in normal triglycerides and severe hypertriglyceridemia groups adjusted to FBS

FBS (Mg/dl)	Patients with normal triglycerides Levels					Patients with severe hypertriglyceridemia					P-value (FBS)	P-value (HbA1c)
	No.	Mean FBS	SD	Mean HbA1c	SD	No.	Mean FBS	SD	Mean HbA1c	SD		
72-129	68	111.6	12.5	7.16	1.47	8	114.0	18.6	8.16	1.95	0.629	0.083
130-149	56	138.8	5.6	7.81	1.28	12	138.0	6.0	9.14	1.66	0.696	0.003
150-169	48	158.6	5.3	8.51	1.24	17	158.7	4.4	8.92	1.60	0.947	0.279
170-199	46	183.02	9.22	9.30	0.95	33	186.4	9.0	9.53	1.53	0.111	0.426
200-229	28	214.6	7.9	10.82	1.31	36	215.5	8.6	9.80	1.47	0.666	0.005
230-259	27	242.07	9.76	11.57	1.07	36	245.78	9.62	10.72	1.81	0.138	0.034
260-299	22	281.0	10.7	12.45	0.78	36	279.8	11.4	10.68	1.57	0.68	<0.001
300-349	14	314.9	16.0	13.53	1.69	45	319.9	14.7	11.24	1.91	0.279	<0.001
350-399	4	373.25	23.36	13.83	2.77	18	371.39	15.26	11.6	1.59	0.379	0.039
400-513	0	-	-	-	-	26	446.58	38.16	12.63	2.2	-	-

P-value measured by independent t-test.

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For the FBS range 72-129 mg/dl, 150-169 mg/dl, and 170-199 mg/dl, the mean FBS was nearly the same for both groups with no significant statistical difference. The mean HbA1c in patients with severe hypertriglyceridemia was higher than the mean HbA1c in patients with normal triglycerides levels by 1%, 0.41% and 0.23%, respectively, but it is not significant statistically. For the FBS range 130-149 mg/dl, the mean FBS was nearly the same for both groups with no significant statistical difference (P-value = 0.696), the mean HbA1c in patients with severe hypertriglyceridemia was higher than the mean HbA1c in patients with normal triglycerides levels by 1.33% with statistically significant p-value = 0.003. For the FBS range 200-229 mg/dl, 230-259 mg/dl, 260-299 mg/dl, 300-349 mg/dl and 350-399 mg/dl, the mean FBS was nearly the same for both groups with no significant statistical difference, the mean HbA1c in patients with severe hypertriglyceridemia was lower than the mean HbA1c in patients with normal triglycerides levels by 1.02%, 0.85%, 1.77%, 2.29% and 2.23%, respectively, with statistically significant p-value equals to 0.005, 0.034, < 0.001, < 0.001, and 0.039 respectively.

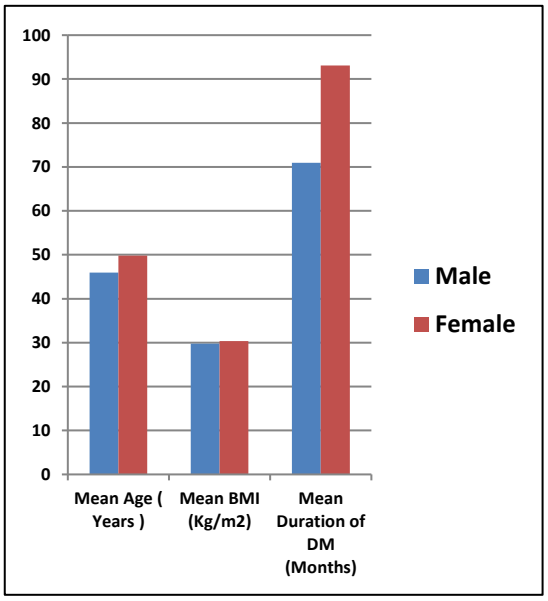

Fig 1. Diabetic patients (type 2) with normal triglycerides levels: Basic characteristics

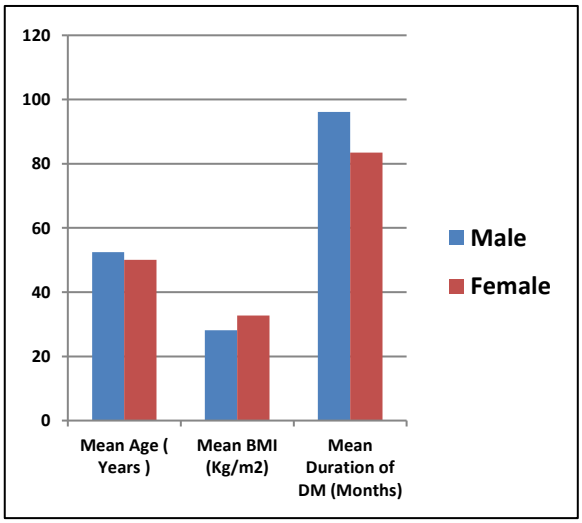

Fig 2. Diabetic patients (type-2) with severe hypertriglyceridemia: Basic characteristics

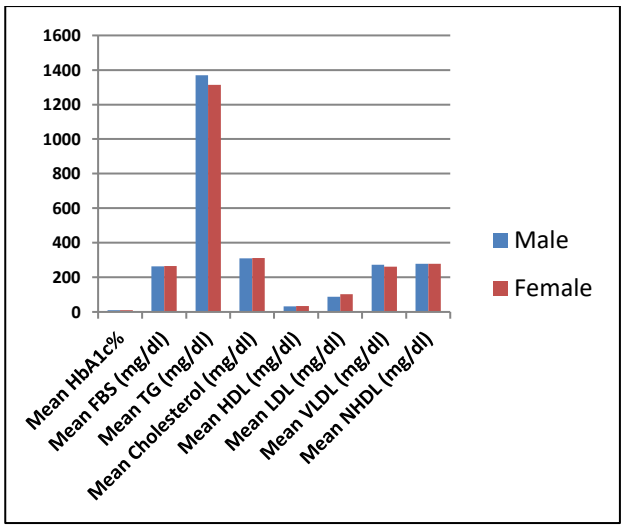

Fig 3. Diabetic patients (type 2) with normal triglycerides levels: Biochemical characteristics

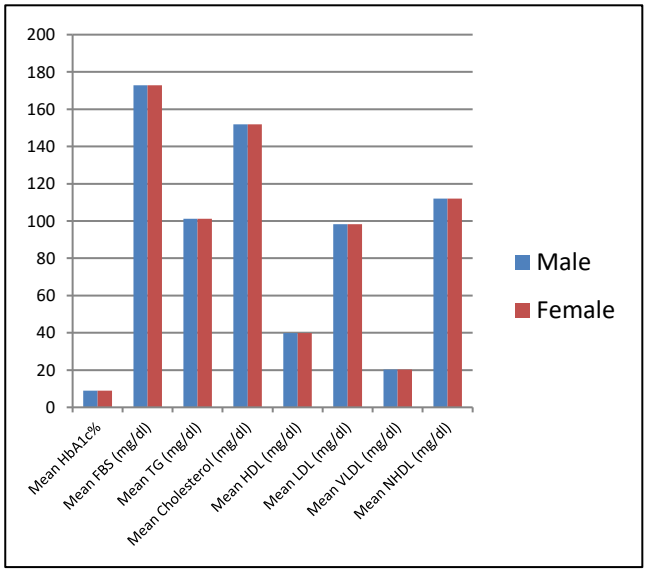

Fig 4. Diabetic patients (type-2) with severe hypertriglyceridemia:Biochemical characteristics

Discussion

There are few studies discussing the effects of hypertriglyceridemia on glycosylated hemoglobin (HbA1c) test. In the current study, it was found that severe hypertriglyceridemia has a significant impact on HbA1c test results in the diabetic patients (type-2). The HbA1c values in the diabetic patients (type-2) with severe hypertriglyceridemia are unpredictable, they may be equal to, higher or lower than the actual values. There was no statistically significant difference between the mean HbA1c of diabetic patients (type-2) with severe hypertriglyceridemia and those with normal triglycerides levels in the FBS range 72-129 mg/dl, 150-169 mg/dl and 170-199 mg/dl. These findings are consistent with a study published on the 12th of February, 2019 by Rene Rodriguez-Gutierrez et al. in which they included 44 patients with hypertriglyceridemia (TG values 378-2986 mg/dl). Most of them had DM (77.3%) and 50% had type 2 DM. They measured the HbA1c values at the beginning of the study and then, after a rapid reduction of triglycerides levels, they measured HbA1c values again. They found that hypertriglyceridemia had no significant impact on HbA1c values.[11] In the FBS range 130-149 mg/dl, the mean HbA1c in the diabetic patients (type 2) with severe hypertriglyceridemia was higher than of those with normal triglycerides levels by 1.33% with a statistically significant p-value. In a study with a similar finding, Falko et al. published a case study of misleading HbA1c increase in a woman with type 2 DM and severe hypertriglyceridemia (23,000 mg/dl). They analyzed these findings further by washing out the patient's samples with saline solution to determine if severe hypertriglyceridemia was interfering with the HbA1c result because HbA1c did not accurately correspond with glycemic control (HbA1c =28.5% and FBS=400mg/dl). They reached the conclusion that HbA1c was falsely elevated when triglycerides levels were above 1750 mg/dl.[12] While the mean HbA1c in the diabetic patients (type 2) with severe hypertriglyceridemia was lower than of those

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with normal triglycerides levels in the FBS range 200-229 mg/dl, 230-259 mg/dl, 260-299 mg/dl, 300-349 mg/dl and 350-399 mg/dl by 1.02%, 0.85%, 1.77%, 2.29% and 2.23%, respectively, with a statistically significant p-value. As matched to these findings, Garrib et al. reported a case of falsely low HbA1c. They detected a diabetic patient with an HbA1c of 4.8%, blood glucose monitoring of 200-325 mg/dl, and triglycerides level of 2388 mg/dl. After washing out the samples with saline solution, they re-evaluated the test. It showed a more accurate HbA1c for the glycemic control of the patient than in the prior trial (HbA1c=12.2%). Furthermore, they evaluated the HbA1c and triglycerides of 98 patients with diabetes and found that when triglycerides levels were >1320 mg/dl, HbA1c increased significantly in the washed samples (P=0.0001).[13]

Limitations of the study:

The small sample size may affect the results due to its impact on the statistical analysis. Also The retrospective nature of the study may not show the complete picture of the impact of severe hypertriglyceridemia on the readings of HbA1c. and Post-prandial glucose was not measured in this study and the comparison of HbA1c between the diabetic patients (type-2) with severe hypertriglyceridemia and those with normal triglycerides levels adjusted to the values of FBS alone.

Conclusion & recommendation, In the setting of elevated triglycerides, doctors face uncertainty when assessing HbA1c readings. Our results suggest that severe hypertriglyceridemia has a significant impact on HbA1c test results. The HbA1c values in diabetic patients (type-2) with severe hypertriglyceridemia are unpredictable. They may be equal to, higher or lower than the actual values. Then we recommend that when assessing glycemic control of patients with type 2 DM, doctors should pay attention to the patients' triglycerides levels as severe hypertriglyceridemia may lead to falsely higher or lower HbA1c readings and hence gives a wrong estimation of the patients' glycemic control leading eventually to improper decisions regarding treatment regimes. Also doctors should rely more on repeated FBS and RBS readings instead of HbA1c tests to assess the control of diabetes in diabetic patients (Type-2) with severe hypertriglyceridemia.

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تأثير إرتفاع الدهون الثلاثية الشديد على اختبار الهيموغلوبين الغليكوزيلاتى فى داء السكرى النوع الثانى

المقدمة :على الرغم من أن اختبار الهيموغلوبين الغليكوزيلاتى ((HbA1c هو اختبار صالح لتشخيص مرض السكرى وتقييم السيطرة على نسبة السكر فى الدم، فمن المعروف أن العديد من العوامل تؤثر على نتائج اختبار الهيموغلوبين الغليكوزيلاتى ((HbA1c وتجعله يعكس صورة خاطئة للتحكم فى نسبة السكر فى الدم. أحد هذه العوامل هو ارتفاع الدهون الثلاثية فى الدم.
الغرض من الدراسة : تقييم تأثير إرتفاع الدهون الثلاثية فى الدم الشديد (الدهون الثلاثية ≤ ١٠٠٠ مغ/ديسيلتر) على نتائج اختبار الهيموغلوبين الغليكوزيلاتى (HbA1c) فى مرضى السكرى (النوع الثانى).
المرضى والاساليب :دراسة مقطعية إسترجاعية على مجموعتين من المرضى البالغين المصابين بالسكرى (النوع الثانى)، تم ادراج المرضى الذين لديهم مستوى طبيعى من الدهون الثلاثية (الدهون الثلاثية 150 <مغ/ ديسيلتر) وأولئک الذين يعانون من ارتفاع شديد للدهون الثلاثية فى الدم (الدهون الثلاثية ≤ ١٠٠٠ مغ / ديسيلتر) فى مركز الفيحاء التخصصى للسكرى والغدد الصم والايض فى البصرة-العراق. تم جمع ٢٦٧ مريضا يعانون من السكرى (النوع الثانى) وارتفاع الدهون الثلاثية فى الدم الشديد و٣١٣ مريضا يعانون من السكرى (النوع الثانى) مع مستوى دهون ثلاثية طبيعى. تم تقسيم كلا المجموعتين من المرضى إلى مجموعات فرعية كل مجموعة مع مدى محدد من مستوى سكر الدم الصائم .(FBS) ثم تم قياس متوسط الهيموغلوبين الغليكوزيلاتى HbA1c)) للمرضى الذين يعانون من ارتفاع شديد للدهون الثلاثية فى الدم لكل مدى من مستوى سكر الدم الصائم(FBS)، ومقارنته بمتوسط الهيموغلوبين الغليكوزيلاتى HbA1c)) للمرضى الذين لديهم مستوى دهون ثلاثية طبيعى فى نفس المدى من مستوى سكر الدم الصائم (FBS) لتحديد تأثير ارتفاع الدهون الثلاثية الشديد فى الدم على قراءات الهيموغلوبين الغليكوزيلاتى HbA1c)) فى مرضى السكرى (النوع الثاني)
النتائج: تمت مقارنة متوسط الهيموغلوبين الغليكوزيلاتى (HbA1c) فى كلتا المجموعتين (أولئک الذين لديهم مستوى دهون ثلاثية طبيعى وأولئک الذين يعانون من ارتفاع شديد فى الدهون الثلاثية) فى كل مدى من مستوى سكر الدم الصائم .(FBS) وكشف أنه على الرغم من نفس متوسط سكر الدم الصائم (FBS) تقريبًا ، كان الهيموغلوبين الغليكوزيلاتى HbA1c)) أعلى فى المرضى الذين لديهم ارتفاع شديد فى الدهون الثلاثية بنسبة ١,٣٣٪ فى مدى سكر الدم الصائم ١٣٠-١٤٩ مغ/ديسيلتر مع قيمة P ذات أهمية إحصائية = ٠,٠٠٣ ، بينما كانت أقل فى مدى سكر الدم الصائم ٢٠٠-٢٢٩ مغ/ديسيلتر، ٢٣٠-٢٥٩ مغ/ديسيلتر، ٢٦٠-٢٩٩ مغ/ديسيلتر، ٣٠٠-٣٤٩ مغ/ديسيلتر و ٣٥٠-٣٩٩ مغ/ ديسيلتر بمقدار ١,٠٢٪، ٠,٨٥٪، ١,٧٧٪، ٢,٢٩٪ و ٢,٢٣٪ على التوالى مع قيمة P ذات أهمية إحصائية تساوى ٠,٠٠٥، ٠,٠٣٤، ٠,٠٠١>، ٠,٠٠١ <و ٠,٠٣٩ على التوالى. لم يكن هناک فرق معتد به إحصائياً بين متوسط الهيموغلوبين الغليكوزيلاتى (HbA1c) لكلا المجموعتين فى مدى سكر الدم الصائم ٧٢-١٢٩ مغ/ديسيلتر، ١٥٠-١٦٩ مغ/ ديسيلتر و١٧٠-١٩٩ مغ/ ديسيلتر. بالإضافة الى ذلك، فإن الانحراف المعيارى للهيموغلوبين الغليكوزيلاتى(HbA1c) فى مجموعة ارتفاع الدهون الثلاثية الشديد كان بصورة عامة أعلى من الانحراف المعيارى للهيموغلوبين الغليكوزيلاتى(HbA1c) فى مجموعة مستوى الدهون الثلاثية الطبيعى مما يشير إلى أن البيانات منتشرة بشكل أكبر مع تباين أعلى فى مستوى الهيموغلوبين الغليكوزيلاتى (HbA1c)فى مجموعة ارتفاع الدهون الثلاثية الشديد
الاستنتاج: كشفت هذه الدراسة أن ارتفاع الدهون الثلاثية الشديد لديه تأثير هام على نتائج اختبار الهيموغلوبين الغليكوزيلاتى (HbA1c). إن قيمة الهيموغلوبين الغليكوزيلاتى (HbA1c) فى مرضى السكرى (النوع الثانى) المصابين بارتفاع الدهون الثلاثية الشديد لا يمكن التنبؤ بها. قد تكون مساوية أو أعلى أو أقل من القيمة الحقيقية.
الكلمات المفتاحية: اختبار HbA1c، النوع ٢ DM، ارتفاع شحوم الدم الشديد