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The Influence of Insulin Resistance on the Levels of Neuron Specific Enolase in the Pathogenesis of Obesity-Related Complications.

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Abstract

Obesity is a complicated and heterogeneous condition characterized by excessive adipose tissue buildup, which is closely linked to the development of insulin resistance which in turn leads to metabolic and cardiovascular issues like diabetes, dyslipidemia, and inflammation, contributing to the pathophysiology of obesity-related conditions. The purpose of this study was to estimate the levels of Neuron-Specific-Enolase (NSE), Glutathione S Transferase (GST), and HOMOIR in obese individuals with and without type 2 diabetes mellitus(T2DM) as biomarkers for the development of related pathophysiological conditions associated with obesity. The study included a group of 46 non-obese as a control (from both sexes) and 92 obese participants as patients. The patients were categorized into two groups: 42 with T2DM (from both sexes) and 50 without T2DM (from both sexes). Anthropometric parameters including body mass index (BMI), and waist-to-hip ratio (WHR) were assessed. The enzyme linked immunesorbent assay (ELISA) method was used to measure NSE, GST levels, and insulin levels. Furthermore, the lipid profile and FBG levels were manually assessed. Overall, obese patients with and without T2DM had significantly higher BMI, HOMA IR, and NSE levels, as well as a significant decline in GST levels compared to the healthy controls. Regarding gender, there was a significant increase in NSE levels in obese patients without T2DM (both sexes) in contracts to obese with T2DM patients (both sexes). however, there was no significant difference in BMI and GST levels The findings suggest that non-diabetic obese individuals exhibit elevated levels of NSE, a marker exclusive to nerve cells. in addition, increased BMI, HOMOIR, and dyslipidemia all contribute to the development of obesity and type 2 diabetes, along with their associated complications..

1. Introduction:

Obesity has significantly increased globally, with 33% of the world's population currently classified as overweight or obese [1]. Obesity is defined as an excessive buildup of adipose and is a significant medical condition. Abdominal fat is recognized as the major risk for obesity related diseases such as: dyslipidemia, coronary heart disease hypertension, type 2 diabetes mellitus, non-alcoholic fatty liver disease, stroke, etc. Moreover, pro-inflammatory and pro-oxidant conditions are linked to fat accumulation [2]. Many risk factors, both hereditary and environmental, have been connected to the etiopathology of obesity [3]. One of these causes is known as oxidative stress. Changing dietary habits and promoting the accumulation of white adipose tissue can exacerbate obesity and its related complications [4].

The body mass index (BMI) and markers of oxidative stress have a significant direct relationship. Numerous in vitro investigations have demonstrated elevated levels of reactive oxygen species, and oxidative stress promote the prolifera-

tion, differentiation, and growth of adipocytes, while also regulating hunger and satiety responses [5]. Excess adipose tissue, especially visceral fat, is known to increase oxidative stress through various biochemical mechanisms, including the action of NADPH oxidase and mitochondrial dysfunction, which further exacerbates insulin resistance and inflammation [6]. This heightened oxidative state not only contributes to insulin resistance by impairing insulin signaling pathways, but also promotes the development of diabetic complications [7].

In diabetics, persistent hyperglycemia has been linked to the appearance of neurological problems [8]. It is easier to identify and track the neurological consequences of diabetes early when nerve damage is assessed by measuring neuron-derived proteins such neuron-specific enolase (NSE) [9]. Neuron-specific enolase (EC 4.2.1.11; NSE) alternatively referred to as gamma () enolase or enolase-2 (Eno2), is an isozyme of glycolytic enolase that is thought to have multiple functions. NSE is primarily expressed in the cytoplasm of neuroendocrine and neuronal cells.

Apart from its well-known glycolysis role in the cytoplasm, NSE expression variations and changes in cell localization are linked to a number of pathologies, including cancer, autoimmune diseases, inflammation, and infection [10]. It is possible for neuron-specific enolase to be released into the bloodstream due to nerve injury and metabolic disorders [8]. Antioxidant strategies can prevent or treat diabetes by reducing oxidative stress. Fruits and vegetables rich in antioxidants can raise glucose levels, and various diets have shown benefits in diabetes prevention [11]. These constituents may act as antioxidants, enhance insulin sensitivity and secretion, and lower blood cholesterol levels [12].

Endogenous antioxidant enzymes like catalase, SOD, GST, and GPx play a crucial role in oxidative stress defense [13]. glutathione S-transferases (GST) are essential phase II detoxification enzymes. These enzymes are involved in the glutathione-coupling reactions of a broad range of electrophilic substances, allowing for easier metabolism, excretion, and detoxification. For this reason, they are critical for cell defense against a variety of xenobiotics, carcinogens, oxidative stress products, chemotherapy, and environmental contaminants [14].

The main aim of this study was to evaluate the potential impact of Neuron-Specific Enolase (NSE) and Glutathione S-Transferase (GST) on obese individuals as predictive markers for oxidative stress and the emergence of issues associated with obesity.

2. Methods:

2.1 Study Design and Sample Selection:

The study was conducted from October 2023 to December 2023 at the University of Baghdad, College of Science for Women, with sample collecting performed at AL-Kindi Teaching Hospital. A total of 138 subjects were divided into two groups according to the findings of their medical examinations and their body mass index (BMI) 46 non-obese people with BMI less than 25 as a control (27 males, 19 females) and 92 obese participants with BMI more than 25 as patients, the patients also were divided into two groups, the first group included 22 obese males and 20 obese females with T2DM, the second group included 22 males and 28 females suffering from obesity and without T2DM. Inclusion criteria consisted of the following: obese diabetic individuals who were on metformin and sulfonylurea medications, and obese non-diabetic individuals. The exclusion criteria consisted of the following: Individuals with polycystic ovarian syndrome, thyroid conditions, heart disease, diabetes mellitus type 2 with complications, diabetes mellitus type 2 with insulin injection, renal diseases, pregnant women's.

The project was approved by the local ethical committee in the University of Baghdad College of Science for Women. According to the cod number 4166/22 on 26.07.2023.

2.2 Blood Sample Collection and Outcome Measurements:

In the morning, blood samples of 10ml were obtained from fasting patients using gel tubes specifically designed for serum separation. The tubes were then left at room temperature (20–25^{o}C) until clotting occurred. After clot formation, and in order to separate the serum, the blood samples were centrifuged for 10 minutes at 3000 rpm. The serum was then divided into five sections and transferred into plastic tubes. Subsequently, the serum samples were frozen at -20^{o}C until they were ready to undergo biochemical assays, including tests for fasting blood sugar (FBS), lipid profile,neuron-specific enolase, glutathione S-transferase (GST), and insulin levels. ELISA Kit (colud-clone crop) was used to measure neuron-specific enolase, GST, and insulin levels. The colorimetric method was used to measure the lipids profile and blood glucose. Body Mass Index was calculated using the formula (weight / height^2), while waist-hip ratio (waist cm $\div$ hip cm) and the semi-empirical formula were used to determine insulin resistance. The homeostatic model assessment for insulin resistance, or HOMA-IR, is calculated as follows: glucose * insulin / 405 (glucose mg dL^{-1}).

2.3 Statistical Analysis:

Statistical analysis was conducted using SPSS software version 26.0, with normal distribution data presented as mean $\pm$ SE, ANOVA a tool was utilized to establish the contrast among three different variables. ROC analysis was also performed. P value was considered significant when < 0.05 [14].

3. Result:

Table 1 shows the mean value of age in the control group (G1) was (38.39 ± 1.28),in obese with T2DM (G2) was (51.59 ± 1.07) and in the obese without T2DM (G3) was (43.06 ± 1.17). The results were statistically significant. The mean value of BMI (kg m^{-2}) in the control group was (24.37 ± 0.25), in the obese with T2DM was (34.42 ± 0.51), and in the obese without T2DM was (32.93 ± 0.74). The results showed statistical significance between groups G1, G2, and G3, but not between groups G2 and G3. The mean value of the W/H ratio in G1 was (0.91 ± 0.01), and in G2 and G3, it was (0.99 ± 0.01, 0.93 ± 0.007), respectively. We found it to be statistically significant between G1 and G2, as well as between G2 and G3. The lipid parameter results in Table 1 show that the mean values of TC, TG, LDL, and VLDL were significantly higher in obese patients with T2DM compared to controls and obese patients without T2DM ($p < 0.05$). While there was a significant decline in the mean value of HDL-c in obese people with T2DM compared to other study groups,

Table 1 shows a significant ($P < 0.05$) increase in FBG (189.28±13.78 mg dL^{-1}) in obese with T2DM compared to obese without T2DM and control groups (104.14±1.37 mg dL^{-1}, 97.75±1.48 mg dL^{-1}), with no significant difference in FBG in obese without T2DM when compared to control group ($p<0.05$). Obese individuals with and without type 2 diabetes had similar mean insulin values, but both had significantly higher insulin levels than controls (6.61±0.66, 6.34±0.41, 2.57±0.04). All study groups showed a significant difference ($p< 0.05$) in mean HOMA IR values.

The study found a statistically significant difference ($P < 0.05$) in the levels of serum NSE (ng mL^{-1}) among three groups G1, G2, and G3 (4.06 ± 0.11,6.74 ± 0.44,10.75 ± 0.53 ng mL^{-1}), respectively. Furthermore, there was a statistically significant difference in the levels of serum GST (ng mL^{-1}) between G1, G2, and G3 (7.55 ± 0.23, 3.54 ± 0.13, and 3.80 ± 0.21 ng mL^{-1}), respectively. However, there was no statistically significant difference between G2 and G3 ($P < 0.05$).

Table 2 presents a comparison of anthropometric measurements among patient study groups according to the gander and shows there was a significant increase in the mean ± SE of age for obese males with T2DM (51.86 ± 1.39 years) when compared to obese males without T2DM (42.86 ± 1.75 years) ($p < 0.05$).

Similarly, there was a significant increase in the mean ± SE of age for obese females with T2DM(51.30 ± 1.70 years) than obese females without T2DM(43.21 ± 1.61 years). Also, the mean ± SE of BMI for obese males with T2DM and obese without T2DM participants was (33.59 ± 0.55 and 31.44 ± 0.57 kg kg m^{-2}), respectively, and for obese females with T2DM and obese without T2DM participants, it was (35.34 ± 0.85 and 34.11 ± 1.21 kg kg m^{-2}), respectively. The BMI mean value did not significantly differ in study groups. Moreover, the mean ± SE waist-hip ratio of obese males with T2DM was significantly higher than that of obese males without T2DM (1.02 ± 0.024, 0.96±0.008, respectively) ($p < 0.05$). Furthermore, in female participants, the mean ± SE WHR of obese with T2DM was higher than that of obese without T2DM (0.97 ± 0.01, 0.911 ± 0.008, respectively).

The lipid parameters, which are displayed in Table 3, indicate that there was no significant difference in the mean ± SE of Cho, TG, LDL, HDL, and VLDL in obese males and females with T2DM and obese individuals without T2DM groups)$p < 0.05$).

Table 4 shows the mean ± SE of FBG, insulin, and HOMOIR between obese patient groups. The mean value of FBG was significantly increased in obese males and females with T2DM (202.67 ± 22.87, 174.55 ± 14.23 mg dL^{-1}) in comparison to obese males and females without T2DM. (101.93 ± 1.91,105.88 ± 1.90 mg dL^{-1}) at P value (≤ 0.05). While there were no statistical differences in the mean ± SE of the insulin hormone in all study groups, the results also found statistical differences in the mean ± SE of HOMA IR between obese females with T2DM and obese females without T2DM (3.37 ± 0.55, 1.66 ± 0.174, respectively) ($p \leq 0.05$). However, there were no statistical differences in the mean ± SE of HOMA IR between obese males with T2DM and obese males without T2DM (3.01 ± 0.68, 1.58 ± 0.125), respectively.

Table 5 presents the mean ± SE values of NSE and GST (ng mL^{-1}) for obese patient groups. It was shown that there is a significant increase in the mean value of the NSE in obese males without T2DM (10.92 ± 1.00 ng mL^{-1}) compared with obese males without T2DM (7.43 ± 0.72 ng mL^{-1}) Similarly, the mean ± SE of NES in obese females without T2DM was (10.62 ± 0.54 ng mL^{-1}) significantly higher than that in obese females with T2DM (5.99 ± 0.46 ng mL^{-1}). while there was no significant difference in the mean ± SE of GST between obese males with T2DM (3.37 ± 0.14 ng mL^{-1}) and obese males with T2DM (3.77 ± 0.22 ng mL^{-1}) patients at p-value ($p < 0.05$) and between obese female with T2DM (3.72 ± 0.23 ng mL^{-1}) and obese females without T2DM (3.83 ± 0.35 ng mL^{-1}). As shown in Table 6 and Figure 1, the result of the ROC analysis of NSE is the area under the curve = 0.837, the cut-off value for NSE ¿ 79.790. The sensitivity was estimated for NSE at 84% and specificity at 26.2% in obese patients.

4. Discussion:

Obesity is a complex condition influenced by numerous factors, including environmental, genetic, aging, and alterations in the gut microbiome Environmental factors such as diet, lifestyle, and exposure to toxic chemicals interact with

Table 1. Grain size distribution and the sediments texture of the three studied sites.

Groups Parameters	Control Group No. (46)	Diabetes Mellitus with Obesity Group No. (42)	Obesity Group No. (50)	P-value
Age (year)	38.39 ± 1.28^{a} (35)	51.59 ± 1.07^{c} (52)	43.06 ± 1.17^{b} (41.50)	0.0001 **
BMI (kg m^{-2})	24.37 ± 0.25^{a} (24.65)	34.42 ± 0.51^{b} (33.61)	32.93 ± 0.74^{b} (31.42)	0.0001 **
W/H ratio	0.91 ± 0.01^{a} (0.91)	0.99 ± 0.01^{b} (0.97)	0.93 ± 0.007^{a} (0.93)	0.0001**
TC (mg dL^{-1})	146.33 ± 4.61^{a} (148.44)	184.21 ± 9.30^{b} (173)	173.33 ± 5.34^{b} (171.34)	0.0001**
TG (mg dL^{-1})	111.44 ± 4.51^{a} (113.80)	196.85 ± 20.03^{b} (161.50)	154.28 ± 11.42^{ab} (122.70)	0.0001**
HDL-C (mg dL^{-1})	44.04 ± 0.93^{b} (42.51)	39.57 ± 1.34^{a} (40)	43.63 ± 1.20^{b} (41.44)	0.016**
LDL-C (mg dL^{-1})	80.70 ± 4.61^{a} (80.41)	106.39 ± 8.08^{b} (95)	98.84 ± 4.65^{ab} (96.94)	0.008**
VLDL-C (mg dL^{-1})	22.39 ± 0.89^{a} (22.84)	39.18 ± 0.89^{b} (22.84)	30.85 ± 2.28^{ab} (24.54)	0.0001**
FBG (mg dL^{-1})	97.75 ± 1.48^{a} (97.91)	189.28 ± 13.78^{b} (171.50)	104.14 ± 1.37^{a} (104.30)	0.0001**
Insulin (μU/mL)	2.57 ± 0.04^{a} (2.58)	6.61 ± 0.66^{b} (5.56)	6.34 ± 0.41^{b} (5.29)	0.0001**
HOMA IR	0.62 ± 0.01^{a} (0.63)	3.18 ± 0.44^{c} (2.40)	1.63 ± 0.11^{b} (1.35)	0.0001**
NES (ng mL^{-1})	4.06 ± 0.11^{a} (4.11)	6.74 ± 0.44^{b} (5.63)	10.75 ± 0.53^{c} (9.94)	0.0001**
GST (ng mL^{-1})	7.55 ± 0.23^{b} (7.18)	3.54 ± 0.13^{a} (3.59)	3.80 ± 0.21^{a} (3.49)	0.0001**

Data were presented as Mean $\pm$ SE (Median)
**Significant difference between means using ANOVA -test at 0.01 level.
-Significant variants are denoted by different small letters.
-Non-significant variations are denoted by identical small letters.

genetic predispositions and significantly impact the risk and development of obesity [15]. The present study demonstrated in Table 1 that age, BMI, and WHR were significantly higher in obese patients with and without T2DM in comparison to the control group. These results are confirmed by Vaishnavi et al.[16] who found that diabetic obese subjects had significantly higher values for BMI and WHR compared to non-diabetic subjects. Also, the study groups showed statistically significant differences in lipid profile parameters. Dyslipidemia and diabetes were found to be significantly related in the study; diabetic patients have lower levels of HDL and higher levels of LDL. this result in line with Biadgo et al., [17] demonstrated that type 2 diabetes mellitus patients have lower levels of high-density lipoprotein and higher levels of total cholesterol, low-density lipoprotein, and triacylglycerol. Individuals who have dyslipidemia and type 2 diabetes remain highly susceptible to cardiovascular disease (CVD)[18].

Furthermore, the results indicate there are significant differences in the mean of HOMA IR between study groups, with a significant increase in obese patients with and without T2DM, this is further supported by findings that show a positive correlation between HOMA-IR and body mass index (BMI), as well as waist circumference (WC), which are both indicators of obesity [19]. The result presented in Table 1 indicates a significant increase in the level of NSE in the serum of obese patients with and without T2DM in comparison to control, this indicates the role of oxidative stress in the progression of disease related to obesity. These are in

Table 2. Comparisons of anthropometric parameters for males and females in obese with T2DM and obese without T2DM groups.

Parameters	Diabetes Mellitus with Obesity Male Group No. (22)	Diabetes Mellitus with Obesity Female Group No. (20)	Obesity Male Group No. (22)	Obesity Female Group No. (28)	P-value
Age (year)	51.86 ± 1.39^{b} (52)	51.30 ± 1.70^{b} (51.50)	42.86 ± 1.75^{a} (41)	43.21 ± 1.61^{a} (42)	0.0001**
BMI (kg m^{-2})	33.59 ± 0.55^{ab} (33.25)	35.34 ± 0.85^{b} (34.57)	31.44 ± 0.57^{a} (31.02)	34.11 ± 1.21^{ab} (32.24)	0.037*
W/H ratio	1.02 ± 0.024^{c} (0.99)	0.97 ± 0.01^{bc} (0.96)	0.96 ± 0.008^{b} (0.96)	0.911 ± 0.008^{a} (0.91)	0.0001**

Data were presented as Mean ± SE (Median)
**Significant difference between means using ANOVA -test at 0.01 level.
*Significant difference between means using ANOVA -test at 0.05 level.
-Significant variants are denoted by different small letters.
-Non-significant variations are denoted by identical small letters.

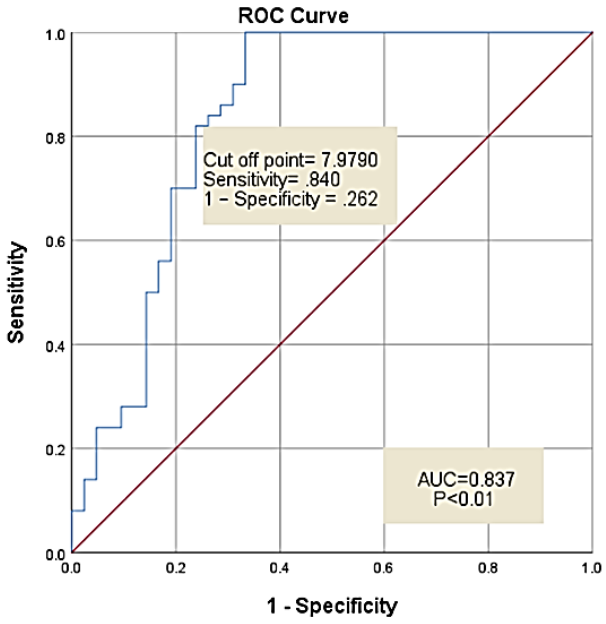

Figure 1. Analysis of the ROC curve for NSE in obese patients.

line with previous research conducted by Roh HT et al., [20] show an increase level of NSE in obese individuals before training. And in line with another study by Majeed et al., [21] showed that, in comparison to healthy individuals, diabetic patients had slightly higher levels of neuron-specific enolase . It has been shown that obesity causes an imbalance between pro-oxidants and oxidants, which increases ROS and causes oxidative stress [22]. According to Abbas et al. [23] , oxidative stress leads to DNA denaturation and apoptosis, which in turn causes neurodegenerative diseases, diabetes, cancer, and cardiac problems. Specifically, brain has higher concentrations of oxygen and unsaturated fatty acids than other organs, but it also exhibits lower levels of antioxidant enzyme activity, which increases the likelihood of neurodegenerative illnesses developing because at-risk neurons may undergo apoptosis [24]. In addition, according to [25] 19.4% of patients with diabetes had abnormal NSE concentrations. The nervous system experiences oxidative stress when exposed to ischemic/hypoxic and hyperglycemic conditions over an extended period [7]]. In diabetics, Persistent hyperglycemia, ischemia, or hypoxia can cause oxidative stress and inflammation of the nervous system, which raises the risk of peripheral neuropathy [26].

To improve peripheral nerve tissue survival and repair, enolase expression is also upregulated in nerve tissue at the same time [27]. Neuron-specific enolase is a glycolytic enzyme that catalyzes the conversion of 2-phosphoglycerate into phosphoenolpyruvate. According to preliminary research, NSE, which can be measured in cerebrospinal fluid and blood, maybe a useful biomarker for determining the degree of neuronal damage and the likelihood of brain injury. Adult brains' gray matter has been shown to contain higher amounts of

Table 3. Levels of Lipid Profile in males and females obese with T2DM and obese without T2DM groups.

Groups Parameters	Diabetes Mellitus with Obesity Male Group No. (22)	Diabetes Mellitus with Obesity Female Group No. (20)	Obesity Male Group No. (22)	Obesity Female Group No. (28)	P-value
TC (mg dL^{-1})	173.27 ± 11.97^{a} (159.50)	196.25 ± 14.24^{a} (196.50)	170.84 ± 7.44^{a} (166.03)	175.29 ± 7.63^{a} (172.90)	0.327
TG (mg dL^{-1})	214.13 ± 33.34^{a} (162.50)	177.85 ± 20.77^{a} (149)	153.73 ± 17.04^{a} (124.65)	154.71 ± 15.67^{a} (121.75)	0.190
HDL-C (mg dL^{-1})	36.95 ± 1.55^{a} (38)	42.45 ± 2.11^{ab} (44)	41.46 ± 1.69^{ab} (40.55)	45.34 ± 1.63^{b} (44.45)	0.008**
LDL-C (mg dL^{-1})	92.04 ± 9.08^{a} (77)	122.17 ± 13.05^{a} (129)	98.63 ± 6.71^{a} (93.24)	99.00 ± 6.52^{a} (98.66)	0.115
VLDL-C (mg dL^{-1})	43.10 ± 6.72^{a} (32.70)	34.88 ± 4.07^{a} (29.80)	30.74 ± 3.40^{a} (24.93)	30.94 ± 3.13^{a} (24.35)	0.177

Data were presented as Mean $\pm$ SE (Median)
**Significant difference between means using ANOVA -test at 0.01 level.
-Significant variants are denoted by different small letters.
-Non-significant variations are denoted by identical small letters.

NSE, whilst the white matter has been reported to contain lower concentrations of NSE [28].

Moreover, the result indicates there is a significant decline in the level of GST in obese patients with and without T2DM when compared to the control group. The results are in line with Mohammed N A et al., [29] which demonstrated that the level of GST decreased in obese patients compared with healthy individuals. Another study shows The homeostatic model assessment for insulin resistance (HOMA-IR), a common measure in T2DM and obesity studies, also showed a correlation with GST activity, where lower GST activity was associated with higher HOMA-IR values, indicating increased insulin resistance [30]. Glutathione S-transferase, a group of antioxidant enzymes, comprises several genes and is essential in the metabolism of electrophilic compounds that cause diseases. Its function includes protecting cells from oxidative harm and monitoring cellular activation mechanisms, thus enhancing cellular health and resistance to stressors.

In this study, considering males and females, the results demonstrated in Table 2, show that age waist and WHR were highly significant in obese diabetic patients (males, females) than obese non-diabetic patients (male , female) this agrees with Wanderley et al. [31] which showed that the age range is significantly higher in diabetic patients. And in line with another study by Joshi et al. [32] also showed that the range of WC and WHR was higher in diabetic participants both male and female than in non-diabetic individuals.

Diabetes prevalence is more strongly correlated with waist circumference and the waist to hip ratio. One possible explanation is that the buildup of abdominal fat or abnormal fat in diabetes may be better reflected by WC and WHtR [33]. The relatively low expandability of the SAT in particular individuals is most likely connected to the excess lipid accumulation in the VAT, which is also a significant source of proinflammatory mediators and free fatty acids (FFAs), both of which are crucial for the onset of IR [34].

The results indicated in Table 3 that there were no significant differences in the mean of Cho, TG, LDL, HDL, and VLDL between obese diabetic patients (males, females) and obese non-diabetic patients (males, females) but the level of TG and VLDL were more than normal range in obese diabetic patients group this disagrees with Sultan et al. [35] Research indicated that obese diabetic males tend to exhibit more pronounced dyslipidemia, characterized by elevated levels of total cholesterol, triglycerides (TG), and low-density lipoprotein (LDL), alongside lower levels of high-density lipoprotein (HDL) compared to their non-diabetic peers.

The hepatic overproduction of VLDL, a common feature in DM, is closely related to insulin resistance and contributes to elevated triglyceride levels and the formation of small, dense LDL particles, which are more atherogenic [36]. The current study demonstrated, Table 4, that there was a significant difference in the level of FBG between obese diabetics (males, females) and non-diabetics, and statistical differences in HOMA IR between obese females diabetics and non-diabetics at a P value (≤ 0.05) While there was no significant difference in the level of insulin hormone in all study groups. This is in line with Gharibeh et al. [37] which showed statistical differences in the mean value of FBG and HOMA IR between obese with DM(males, females) and obese without DM (males, females) and no difference in the

Table 4. Levels of FBS and Insulin hormone and (HOMA IR) in males and females obese with T2DM and obese without T2DM groups.

Parameters	Diabetes Mellitus with Obesity Male Group No. (22)	Diabetes Mellitus with Obesity Female Group No. (20)	Obesity Male Group No. (22)	Obesity Female Group No. (28)	P-value
FBS (mg dL^{-1})	202.67 ± 22.87^{b} (177)	174.55 ± 14.23^{b} (166.50)	101.93 ± 1.91^{a} (101.35)	105.88 ± 1.90^{a} (107.3)	0.0001**
Insulin (μU mL^{-1})	5.59 ± 0.83^{a} (4.50)	7.73 ± 1.0^{a} (7.91)	6.29 ± 0.48^{a} (5.66)	6.38 ± 0.63^{a} (4.83)	0.276
HOMA IR	3.01 ± 0.68^{ab} (1.8)	3.37 ± 0.55^{b} (3.05)	1.58 ± 0.125^{a} (1.48)	1.66 ± 0.174^{a} (1.35)	0.005**

Data were presented as Mean $\pm$ SE (Median)
**Significant difference between means using ANOVA -test at 0.01 level.
-Significant variants are denoted by different small letters.
-Non-significant variations are denoted by identical small letters.

Table 5. Levels of NSE and GST levels in males and females obese with T2DM and obese without T2DM groups.

Groups Parameters	Diabetes Mellitus with Obesity Male Group No. (22)	Diabetes Mellitus with Obesity Female Group No. (20)	Obesity Male Group No. (22)	Obesity Female Group No. (28)	P-value
NES (ng mL^{-1})	7.43 ± 0.72^{a} (5.68)	5.99 ± 0.46^{a} (5.27)	10.92 ± 1.00^{b} (10.04)	10.62 ± 0.54^{b} (9.75)	0.0001**
GST (ng mL^{-1})	3.37 ± 0.14^{a} (3.13)	3.72 ± 0.23^{a} (3.79)	3.77 ± 0.22^{a} (3.57)	3.83 ± 0.35^{a} (3.42)	0.628

Data were presented as Mean $\pm$ SE (Median)
**Significant difference between means using ANOVA -test at 0.01 level.
-Significant variants are denoted by different small letters.
-Non-significant variations are denoted by identical small letters.

level of insulin. Long-term, low-grade systemic and local inflammation that leads to insulin resistance-associated diabetes mellitus may be exacerbated by obesity.

Moreover, insulin resistance and hyperinsulinemia may contribute to the development of obesity [38]. Also, this study demonstrated significantly increased NSE levels in (males, females) obese non-diabetics patients when compared to (males, females) obese diabetics patients at P value (≤ 0.05) as shown in Table 5, the elevated NSE levels observed in obese non-diabetic patients compared to their diabetic equals may be influenced by the therapeutic regimens undergone by the latter group (diabetics patients), as indicated by certain research studies. Interventions such as metformin treatment have been shown to modulate oxidative stress levels, inflammation, and DNA damage [39],[40].

Additionally, another study indicated that elevated BMI is linked to elevated NSE serum levels [41]. Furthermore, the study showed that there was no significant difference in the level of GST in all study groups as shown in Table 4; the result disagrees with Pasupathi et al. [42] which showed the activity of antioxidants was significantly decreased in obese diabetic patients when compared to healthy control subjects (obese people). The detractive impact caused by obesity on the body's antioxidant defenses can be explained by it is effective in lowering some antioxidant enzyme levels or altering their activities [43], [44]. The study uses the ROC curve NSE to distinguish between obese individuals with and without type 2 diabetes, revealing a favorable association between NSE and type 2 diabetes and obesity, with an AUC of 0.837 as shown in Table 6 and Figure 1.

5. Conclusions:

The study findings suggest that elevated levels of neuron-specific enolase (NSE), a specific marker for neuronal cells, in obese non-diabetic individuals, indicate the involvement of insulin resistance in inducing oxidative stress, which contributes

Table 6. ROC curve analysis of test variable for NSE in obese patient groups.

Test Result Variable(s)	Area	cut off value	sensitivity Sig.[b]	Specificity	Std. Error[a]	Asymptotic	Asymptotic Confidence Interval	95%
							Lower Bound	Upper Bound
NSE	0.837	7.979	84%	26.2%	0.046	0.000	0.746	0.927

a. Under the nonparametric assumption

b. Null hypothesis: true area = 0.5

to the pathogenesis of obesity-related complications, particularly neurological disorders. Given its established role as a diagnostic indicator for neurological diseases, the disparity in NSE levels between obese diabetic and obese non-diabetic individuals is believed to be influenced by the therapeutic interventions administered to the diabetic group, which are hypothesized to modulate oxidative stress levels. Additionally, these findings underscore the significant role of GSTs in obesity.

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Data Availability Statement: All of the data supporting the findings of the presented study are available from corresponding author on request.

Declarations:
Conflict of interest: The authors declare that they have no conflict of interest.

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تأثير مقاومة الأنسولين على مستويات إنزيم الإينولاز الخاص بالخلايا العصبية المرتبطة في تطور مضاعفات السمنة

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الخلاصة

السمنة هي حالة معقدة وغير متجانسة تتميز بتراكم الأنسجة الدهنية المفرط، ترتبط ارتباطًا وثيقًا بتطور مقاومة الأنسولين مما يؤدي إلى مضاعفات ايضية مثل مرض السكري، وخلل شحوم الدم، والالتهابات، والتي تلعب جميعها دورا في في الفسيولوجيا المرضية للحالات المرتبطة بالسمنة. كان الغرض من هذه الدراسة تقييم مستويات الإينولاز الخاص بالخلايا العصبية و الجلوتاثيوناسترانسفيراز ومقاومة الانسولين في مرضى السمنة المصابين،وغير المصابين بداء السكري كمؤشرات حيوية لتطور الحالات الفيزيولوجية المرضية ذات الصلة بالسمنة. شملت الدراسة مجموعة مكونة من 46 من الأفراد الأصحاء(من كلا الجنسين) و 92 مشاركًا يعانون من السمنة المفرطة كمرضى، وتم تصنيف المرضى إلى مجموعتين: 42 مصابًا بـداء السكري (من كلا الجنسين) و 50 غير مصابين بداء السكري (من كلا الجنسين). تم تقييم معاملات القياسات البشرية بما في ذلك مؤشر كتلة الجسم ، ونسبة الخصر إلى الورك. تم قياس مستويات انزيم الإينولاز الخاص بالخلايا العصبية ، الجلوتاثيوناسترانسفيراز إلى جانب هرمون الأنسولين، باستخدام تقنية الاليزا، علاوة على ذلك تم تحديد مستوى الدهون وسكر الصائم باستخدام الطريقة الانزيمية. بشكل عام كان لدى المرضى الذين يعانون من السمنة المفرطة المصابين وغير المصابين بداء السكري ارتفاع ملحوظ في مؤشر كتلة الجسم ومقاومة الانسولين و مستويات الإينولاز الخاص بالخلايا العصبية بالإضافة إلى انخفاض كبير في مستويات الجلوتاثيوناسترانسفيراز على العكس من الأصحاء. فيما يتعلق بالجنس، هناك زيادة ملحوظة في مستويات الإينولاز الخاص بالخلايا العصبية لدى المرضى الذين يعانون من السمنة المفرطة غير المصابين بداء السكري (كلا الجنسين) مقارنة بالمرضى الذين يعانون من السمنة المفرطة المصابين بداء السكري (كلا الجنسين). بينما، لا يوجد فرق كبير في مؤشر كتلة الجسم و مستويات الجلوتاثيوناسترانسفيراز. تشير النتائج إلى أن الأفراد المصابين بالسمنة غير المصابين بداء السكري يظهرون مستويات مرتفعة من الإينولاز الخاص بالخلايا العصبية ، وهو علامة حصرية للخلايا العصبية، بالإضافة إلى ذلك، فإن زيادة مؤشر كتلة الجسم ومقاومة الانسولين وخلل شحوم الدم كلها تساهم في تطور مرض السكري من النوع الثاني والسمنة، فضلا عن المضاعفات المرتبطة بها.

الكلمات الدالة: مؤشر كتلة الجسم ، الجلوتاثيوناسترانسفيراز، الإينولاز الخاص بالخلايا العصبية، السمنة، الإجهاد التأكسدي.

التمويل: لايوجد.

بيان توفر البيانات: جميع البيانات الداعمة لنتائج الدراسة المقدمة يمكن طلبها من المؤلف المسؤول.

اقرارات:

تضارب المصالح: يقر المؤلفون أنه ليس لديهم تضارب في المصالح.

الموافقة الأخلاقية: لم يتم نشر المخطوطة أو تقديمها لمجلة أخرى، كما أنها ليست قيد المراجعة.