A Comparative Study between the Levels of Adropin in Iraqi Women with Metabolic Syndrome and Diabetes

¹Riyam Hussein Assaf *   **²Layla Othman Farhan**

1,2 Department of Chemistry, College of Sciences for Women, University of Baghdad, Baghdad, Iraq.

*Corresponding author**: rayam.hussein1205a@csw.uobaghdad.edu.iq**

Abstract

By measuring Adropin, fasting blood glucose (FBG), cholesterol, high-density lipoprotein (HDL), triglycerides (TG), low-density lipoprotein (LDL), and very low-density lipoprotein (VLDL) in the sera of Iraqi patients with MetS and type 2 diabetes mellitus (T2DM), the current study was designed to compare some crucial markers in metabolic syndrome (MetS) sera and diabetic patients (T2DM) with control. Twenty female subjects were divided into three groups: group I=40 with MetS and group II=40 with T2DM, and 40 healthy subjects were employed as a control group. Compared to the control group, Adropin levels in the Mets group and T2DM group decreased significantly ($p < 0.05$). In contrast, none of the patient groups (MetS and T2DM) showed any change compared to themselves. In conclusion, according to the present data, higher levels of FBG, lipid profile, and increased blood pressure (BP) were found in patients with MetS and T2DM. A drop in level could be considered a novel indicator of MetS and T2DM. Compared to T2DM patients, Adropin levels are thought to have a more sensitive diagnostic function than those with MetS and newly diagnosed DM.

Keywords: Adropin, Type 2 diabetes mellitus, Metabolic syndrome, Body mass index, Blood pressure.

1. Introduction

Metabolic syndrome (MetS) has increased more widely and is known to dramatically increase the risk of acquiring diabetes and cardiovascular disease. Obesity, atherogenic dyslipidemia, hypertension, and insulin resistance (IR) are a group of cardiac risk factors that collectively make up the syndrome. In women, the MetS has a few distinctive characteristics. Dietary and lifestyle changes are two of the most important aspects of controlling the MetS [1]. Adropin is a peptide hormone composed of 76 amino acids. It was first characterized as a secreted peptide with residues 1–33 encoding a secretory signal peptide sequence [2]. This protein has the same amino acid sequence in humans, mice, and rats. Recent results [3,4] indicate a role in energy balance and the regulation of glucose and fatty acid metabolism, even if our understanding of the precise physiological effects of this poorly characterized peptide is still developing [5].

The gene encoding this protein, Enho, is mainly expressed in the liver and the central nervous system. Adropin, however, may be a membrane-bound protein that controls cell-cell communication, as has been proposed more recently [6]. Adropin has also been found in several physiological fluids and tissues, including milk, colostrum, liver, kidney, pancreas, small intestine, endothelial cells, brain, cerebellum, liver, kidney, and heart [7,8]. Low levels of Adropin have been linked to a risk of dyslipidemia, IR, and other MetS symptoms [9]. Age and body mass index (BMI) are antagonistic to blood Adropin concentrations [10]. Adropin levels are controlled by dietary macronutrients during feeding circumstances and rise with dietary fat content [11].

Adropin is not necessary for controlling appetite; its actions impact adiposity and help prevent IR, dyslipidemia, and poor glucose tolerance. This study aims to compare Adropin levels in diabetic females with and without MetS and compare them with the control group [12].

2. Materials and Methods

Metabolic syndrome criteria includes:-

1) Triglyceride levels above 150 mg/dL (or specific treatment for this lipid abnormality).

2) decrease in HDL-cholesterol levels (40 mg/dL in men and 50 mg/dL in women) or treatment for this lipid abnormality.

3) High BP; systolic or diastolic readings of 130 or more millimeters mercury; or (having been diagnosed with hypertension and were treated).

4) Higher than normal fasting blood glucose (FBG; 110 mg/dl) or have been given a type II diabetes diagnosis.

Patients with type 2 diabetes mellitus (T2DM) using insulin as hypoglycemic therapy, acromegaly, and chronic liver and renal disorders were excluded from the study [6].

2.1 Blood sample collection

After 10–12 hours, ten milliliters of venous blood were collected and divided into two aliquots. The samples were centrifuged at 1500 g for 15 minutes after an incubation period of 30 minutes. A part of the acquired serum was employed to estimate the lipid profile. The following analysis used the second serum component to measure fasting serum insulin.

Weight (in kilograms), height (in centimeters), and waist circumference (in centimeters) were assessed for each subject. Body mass index (BMI) was calculated by dividing weight (in kilograms) by the square of height (m^2) or waist-to-hip ratio (WHR). The BMI was calculated by dividing weight (in kilograms) by the square of height (m^2) [13].

In a seated position, the systolic and diastolic blood pressures (SBP, DBP) (measured in mmHg) were taken using a mercury sphygmomanometer. From these observations, the mean arterial pressure (MAP) was computed using the formula below for calculating the average pressure: MAP= [DBP + SBP-DBP)/3] [14].

An ELISA kit was used to estimate the Adropin level (MyBiosource, America).

2.2 Statistical analysis

The median (25th and 75th percentiles) for numerical variables with an abnormal distribution is used to interpret the data. The Shapiro-Wilk test was applied to check if the data were distributed normally. The (Mann-Whitney) test was employed to describe numerical variables that were not regularly distributed. The significance level was set at a p-value of 0.05. moreover, receiver operating characteristic (ROC) was used to estimate the Adropin cut-off value.

3. Results

The median (25th and 75th percentiles) of age and BMI for MetS, T2DM patients, and control are listed in Table 1. Age in patients with MetS was 49.0 (39.75–53.0), in T2DM was 48.50 (51.25–60.0), and in the control group was 46.50 (38.0–52.0). The BMI distribution of the MetS was 31.50 (30.0–35.0), 29.0 (27.0–35.0) in T2DM patients, and 24.0 (24.0–25.0) in the healthy subjects. The data of the WHR distribution of the MetS was 0.94 (0.89–1.01), T2DM patients was 0.98 (0.94–1.03), and the healthy subjects group was (0.83–0.84).

Table 1. The demographic characteristics of patients and control group

Variables	Median		
	MetS (n= 40)	T2DM (n= 40)	Control (n= 40)
Age (Years)	49.0(39.75 -53.0)	48.50(51.25-60.0)[b]	46.50 (38.0 – 52.0)
BMI	31.50 (30.0 – 35.0)[a,c]	29.0(27.0-35.0)[b,c]	24.0 (23.0 – 25.0)
WHR	0.94 (0.89 – 1.01)[a,c]	0.98(0.94-1.03)[c]	0.83 (0.80 – 0.84)

The median (25th and 75th percentiles) of the obtained data were evaluated using the Mann-Whitney test at the 0.05 threshold; there was a significant difference between the two independent means.
a) Show how MetS Group and control differ significantly from each other.
b) Show that the T2DM group and control have a significant difference.
c) Show that the MetS Group and the T2DM Group have important differences.

Table 2 shows the serum levels of serum lipid profiles in MetS, T2DM patients, and controls. No significant difference appeared in cholesterol, T.G., HDL,VLDL, and LDL levels when compared between MetS and T2DM patients and the control group with p>0.05 as shown in **Table 2**. Much evidence has linked obesity with sympathetic overactivity (increase in heart rate, blood pressure, and breathing rate).

Table 2. The Serum lipid profile in MetS, T2DM, and control

Variables	Median		
	MetS	**T2DM**	**Control**
	(n= 40)	**(n= 40)**	**(n= 40)**
Cholesterol (mg/dL)	203.0(187.0-216.25)a	182.50(151.25-217.50)b	51.30(45.43-142.63)
T.G (mg/dL)	176.50(134.0-191.50)	197.50(129.50-263.0)b	81.90(62.63-90.40)
HDL-C (mg/dL)	40.70 (39.05-43.83)a	22.60(19.30-45.43)	42.0(36.0-46.0)b
LDL-C (mg/dL)	125.60(108.60-142.35)a	109.0(86.0-146.50)b	80.40(71.77-88.05)
VLDL (mg/dL)	35.30(26.85-38.30)a	39.50(25.30-53.0)b	19.80(15.0-22.60)

The median (25th and 75th percentiles) of the obtained data were evaluated using the Mann-Whitney test at the 0.05 threshold; there was a significant difference between the two independent means.

a) Show how MetS Group and control differ significantly from each other.

b) Show that the T2DM Group and Control have a significant difference.

c) Show that the MetS Group and the T2DM Group have important differences.

The cholesterol levels increased significantly in MetS and T2DM groups 203.0 (187.0-216.25) and 182.50 (151.25-217.50) when compared with the control group 51.30 (45.43-142.63), $p > 0.05$. At the same time, there was no significant difference in Cholesterol compared with the two groups of patients. The T.G. and VLDL levels increased significantly in patient groups; MetS, 176.50 (134.0-191.50), 35.30(26.85-38.30) and T2DM, 197.50 (129.50-263.0), 39.50 (25.30–53.0) when compared with the control group 81.90 (62.63-90.40), and 19.80 (15.0-22.60), $p > 0.05$. At the same time, there was no significant difference in Cholesterol when we compared the MetS and T2DM groups [15]. The HDL and LDL levels showed a high significant difference when comparing patient groups; MetS 40.70 (39.05-43.83) and 125.60 (108.60-142.35), T2DM 22.60 (19.30-45.43), 109.0 (86.0-146.50), and control 42.0 (36.0-46.0), and 80.40 (71.77-88.05), as shown in **Table 2**. The high visceral fat accumulation characterized by obesity is a distinct feature of MetS [16]. Patients with MetS have low HDL and high T.G. levels due to the slower clearance of these lipoproteins from the bloodstream [17,18]. Increased FBG levels can cause oxidative stress and inflammation and hasten the development of dyslipidemia brought on by some harmful variables, like air pollution. Elevated FBG levels and dyslipidemia have been linked in numerous studies [13,19,20], and in comparison to the control group, the DBP, SBP, and FBG significantly increased in the MetS and T2DM patient groups ($p < 0.05$, $p < 0.001$). When comparing patient groups (MetS with T2DM), the results revealed a significant difference in diastolic blood pressure, SBP, and FBG ($p < 0.05$, $p < 0.00$). The result showed a substantial decrease in Adropin levels in the MetS and T2DM groups. When compared with the control group, $p < 0.05$, there was no significant difference between the patient groups (MetS, T2DM) and the control group, as shown in **Table 3**.

Table 3. Clinical and biochemical factors of patients and control group

Variables	Median			*p*-value
	MetS	**T2DM**	**Control**	
	(n= 40)	**(n= 40)**	**(n= 40)**	
DBP (mmHg)	(77.34±25.10)a,c	(78.22±22.10)b,c	73.4±15.10	0.05
SBP (mmHg)	(131.03 ±13.05)a,c	(140.03 ±15.05)b,c	122.03 ±17.07	0.05
FBG(mg/dL)	117.50(92.0-118.0)a,c	170.50(142.25-236.50)[b,C]	112.65(96.0-142.63)	0.000
Adropin(pg/mL)	100.0(87.18-111.50)a	97.75(87.70-106.45)b	228.65(167.60-241.18)	N.S

The collected data were analyzed by mean ± stander deviation and median (25th and 75th percentiles) of the obtained data were evaluated using the Mann-Whitney test at the 0.05 threshold; there was a significant difference between the two independent means.

a) Show how MetS group and control differ significantly from each other.

b) Show that the T2DM group and control have a significant difference.

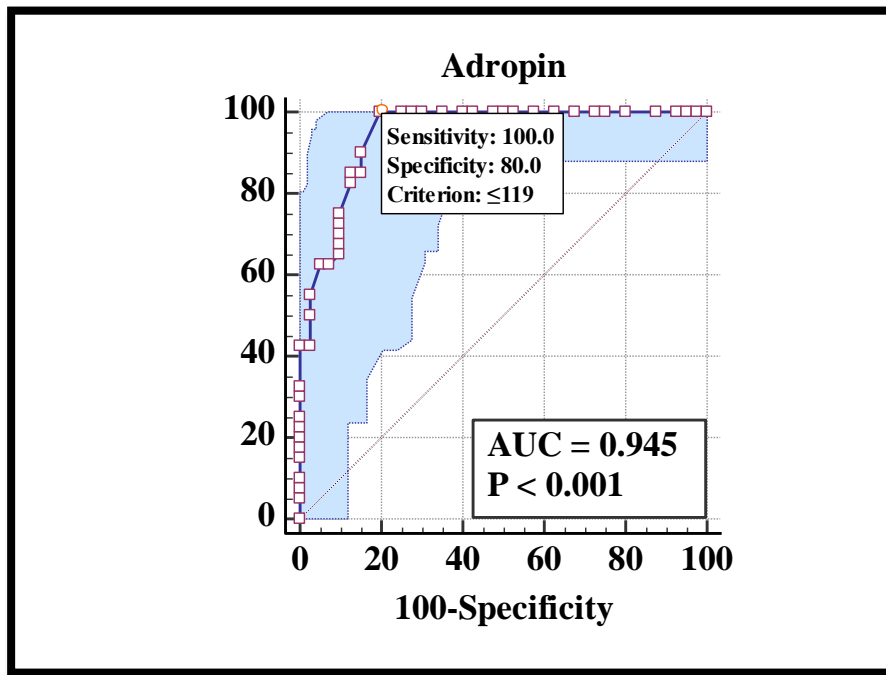c) Show that the MetS group and the T2DM group have important differences.

Figure 1. The ROC curve analysis of serum Adropin concentration in MetS patients against healthy subjects

Table 4. Adropin AUC and validity in distinguishing between MetS and healthy subject

Variable	AUC	*P*-Value	cutoff value	Sensitivity	Specificity	Accuracy	(+ve) predictive value	(-ve) predictive value
Adropin	0.945	0.001	119	100.0	80.0	0.800	83.0	100.0

4. Discussion

Much evidence has linked obesity with sympathetic overactivity (increased heart rate, blood pressure, and breathing rate). Numerous studies have connected sympathetic overactivity and obesity (increased heart rate, blood pressure, and breathing rate) [21].

As shown below, the ROC was carried out twice, once for MetS patients from healthy subjects and once for T2DM patients from healthy subjects [22]. Using ROC curve analysis, the ability of serum Adropin levels to identify MetS patients from healthy individuals was examined **Table 4** and **Figure 1**. The ROC curve was significantly lower than the diagnostic test for MetS, but it had higher validity (high sensitivity (100.0) and specificity (80.0). The area under the curve (AUC) of the receiver operating characteristic curve for the existence of a MetS diagnosis was 119 (p < 0.001), which was the optimal level of correct MetS prediction [23].

Diabetes and low blood Adropin levels trigger atherosclerosis; hence, their interaction may result in more severe atherosclerosis. Additional research is required to determine how Adropin, diabetes, and atherosclerosis are related causally [24,25,26]. Adropin participates in several biological processes and is controlled by various nutrients, including lipids and carbohydrates [27].

Adropin treatment can increase glucose oxidation and decrease fatty acid oxidation in skeletal muscle cells [28]. A drop in can lessen IR and delay the onset of obesity by improving lipid catabolism, which has preventive effects in T2DM. Inhibiting the development of 3T3-L1 preadipocytes into mature adipocytes, which reduces fat accumulation and inflammation, is how Adropin controls the expression of peroxisome proliferator-activated receptors-γ. Another study found that patients with moderate and severe obstructive sleep apnea had decreased plasma levels of Adropin and higher levels of inflammation markers like tumor necrosis factor and interleukin-6 compared to healthy people [29,30].

The AUC refers to the area under the curve. While the negative predictive value and positive predictive value, respectively. The ROC curve analysis was used to see how the serum Adropin level could show the difference between patients with T2DM and healthy subjects **Table 5** and **Figure 2**. The ROC curve was significantly lower than the diagnostic test for T2DM, indicating greater validity; high sensitivity (95.0) and specificity (87.5). The AUC of the ROC curve for the presence of a diagnosis determined the optimal level of correct T2DM prediction to be 0.956 (*p*= 0.001).

5. Conclusion

In conclusion, according to the present data, higher levels of FBG, lipid profile, and increased BP were found in patients with MetS and T2DM. Adropin level could be considered a novel indicator of MetS and T2DM. Compared to T2DM patients, it is thought that Adropin levels have a more sensitive diagnostic function than those with MetS and newly diagnosed DM.

Ethical Clearance:

This study was approved by the Scientific Committee in the College of Sciences for Women/ University of Baghdad, and verbal consent was obtained from each participant enrolled.

References

1. Schneider, J.G.; Tompkins, C.; Blumenthal, R.S.; Mora, S. The metabolic syndrome in women. *Cardiol. Rev.* **2006**, *14*(*6*), 286–291. https://doi: 10.1097/01.crd.0000233757.15181.67.

2. Kumar, K.G.; Trevaskis, J.L.; Lam, D.D.; Sutton, G.M.; Koza, R.A.; Chouljenko, V.N.; Kousoulas, K.G.; Rogers, P.M.; Kesterson, R.A.; Thearle, M. Identification of Adropin as a secreted factor linking dietary macronutrient intake with energy homeostasis and lipid metabolism. *Cell Metab.* **2008**, *8*(*6*), 468–481. https://doi: 10.1016/j.cmet.2008.10.011.

3. Gao, S.; McMillan, R.P.; Jacas, J.; Zhu, Q.; Li, X.; Kumar, G.K.; Casals, N.; Hegardt, F.G.; Robbins, P.D.; Lopaschuk, G.D. Regulation of substrate oxidation preferences in muscle by the peptide hormone Adropin. *Diabetes* **2014**, *63*(*10*), 3242–3252. https://doi: 10.2337/db14-0388.

4. Hamad, H.T; Mohammed, Y.K. Effect of Clopidogrel on the levels of Adropin and Adiponectin in serum of female Albino Rats. *International Research Journal of Pharmacy and Medical Sciences (IRJPMS)*, **2023**, *6*(*6*),67-71. http://irjpms.com/wp-content/uploads/2023/10/IRJPMS-V6N6P115Y23.pdf.

5. Reddy, V.S.; Reddy, E.P.; Thangappazham, B.; Varshney, S.; Das, V.L.; Munikumar, M. Adropin levels and its associations as a fat-burning hormone in patients with polycystic ovary syndrome: A correlational meta-analysis. Gynecol Endocrinol., **2021**, *37*(*10*), 879-884. https://doi: 10.1080/09513590.2021.1950136.

6. Khan, S.;A.; Ram, N.; Masood, M.Q. Patterns of abnormal glucose metabolism in acromegaly and impact of treatment modalities on glucose metabolism. *Cureus* **2021**, *13*(*3*), e13852. https://doi: 10.7759/cureus.13852.

7. Butler, A.A.; Tam, C.S.; Stanhope, K.L.; Wolfe, B.M.; Ali, M.R.; O'Keeffe, M.; St-Onge, M.-P.; Ravussin, E.; Havel, P.J. Low circulating adropin concentrations with obesity and aging correlate with risk factors for metabolic disease and increase after gastric bypass surgery in humans. *J. Clin. Endocrinol. Metab.* **2012**, *97*(*10*), 3783–3791. https://doi: 10.1210/jc.2012-2194.

8. Hamad, M.S.; Sarhat, E.R.; Sarhat, T.R.; ABASS, K.S. Impact of serum Adropin and Irisin in Iraqi patients with congestive heart failure. *PJMH S* **2021**, *15*(*2*), 497–499. https://pjmhsonline.com/2021/feb/497.pdf.

9. Yu, H.; Zhao, P.; Wu, M.; Liu, J.; Yin, W. Serum Adropin levels are decreased in patients with acute myocardial infarction. *Regul. Pept.*, **2014**, *190*, 46–49. https://doi: 10.1016/j.regpep.2014.04.001.

10. Mustaf, L.A. Adropin levels in the serum of obese type 2 diabetic patients and their relationship to oxidative stress. *African J. Adv. Pure Appl. Sci.*, **2023**, *2(2)*,131–136. https://aaasjournals.com/index.php/ajapas/article/view/343.

11. Jasaszwili, M.; Billert, M.; Strowski, M.Z.; Nowak, K.W.; Skrzypski, M. Adropin as a fat-burning hormone with multiple functions—Review of a decade of research. *Molecules*, **2020**, *25(3)*, 549. https://doi.org/10.3390/molecules25030549.

12. Elgohary, M.; El Maghawry, M.A.; Mostafa, D.; Abd el-Sattar, E.M.; El Tahir, F. The association between serum Adropin level and metabolic complications of type 2 diabetes mellitus. *Egypt. J. Hosp. Med., 2023, 93*, 7323–7328. https://doi.org/10.1186%2Fs13098-022-00796-y.

13. Farhan, L.O. Study of partially purification AST activity in sera of Iraqi patients with diabetic nephropathy. *Atherosclerosis*, **2014**, *14(9)*, 12. https://www.tsijournals.com/articles/study-of-partially-purification-ast-activity-in-sera-of-iraqi-patients-with-diabetic-nephropathy.pdf.

14. DeMers, D.; Wachs, D. Physiology, mean arterial pressure. StatPearls Publishing, **2021**. https://www.ncbi.nlm.nih.gov/books/NBK538226.

15. Huang, J.-K.; Lee, H.-C. Emerging evidence of pathological roles of very-low-density lipoprotein (VLDL). *Int. J. Mol. Sci.*, **2022**, *23(8)*, 4300. https://doi: 10.3390/ijms23084300.

16. Bailey, A.; Mohiuddin, S.S. Biochemistry, high density lipoprotein. StatPearls Publishing, **2022**. https://www.ncbi.nlm.nih.gov/books/NBK549802.

17. Haile, K.; Haile, A.; Timerga, A. Predictors of lipid profile abnormalities among patients with metabolic syndrome in Southwest Ethiopia: A cross-sectional study. *Vasc. Health Risk Manag.***, 2021**, *17*, 461. https://doi.org/10.2147%2FVHRM.S319161.

18. Abed, B.A.; Al-AAraji, S.B.; Salman, I.N. Estimation of Apelin levels in Iraqi patients with type II diabetic peripheral neuropathy, B*aghdad Science Journal*, **2023**, *20(5)*, 1684-1691. https://doi.org/10.21123/bsj.2023.7566.

19. Shi, J.; He, L.; Yu, D.; Ju, L.; Guo, Q.; Piao, W.; Xu, X.; Zhao, L.; Yuan, X.; Cao, Q. Prevalence and correlates of metabolic syndrome and its components in Chinese children and adolescents aged 7–17: The China National Nutrition and Health Survey of Children and Lactating Mothers from 2016–2017. *Nutrients* **2022**, *14(16)*, 3348. https://doi.org/10.3390%2Fnu14163348.

20. Khaleel, F.; N-Oda, N.; Abed, B.A. Disturbance of arginase activity and nitric oxide levels in Iraqi type 2 diabetes mellitus. *Baghdad Sci. J.*, **2018**, *15(2)*,189-191. https://doi.org/10.21123/bsj.2018.15.2.0189.

21. Krishnamoorthy, Y.; Rajaa, S.; Murali, S.; Sahoo, J.; Kar, S.S. Association between anthropometric risk factors and metabolic syndrome among adults in India: A systematic review and meta-analysis of observational studies. *Prev. Chronic Dis.*, **2022**, *19*, E24. https://doi: 10.5888/pcd19.210231.

22. Vladu, I.M.; Forțofoiu, M.; Clenciu, D.; Forțofoiu, M.-C.; Pădureanu, R.; Radu, L.; Cojan, Ștefăniță T. Țenea; Rădulescu, P.M.; Pădureanu, V. Insulin Resistance Quantified by the Value of HOMA-IR and Cardiovascular Risk in Patients with Type 2 Diabetes. *Exp. Ther. Med.* **2022**, *23(1)*,73. https://doi: 10.3892/etm.2021.10996. E.

23. Xia, S.-J.; Gao, B.-Z.; Wang, S.-H.; Guttery, D.S.; Li, C.-D.; Zhang, Y.-D. Modeling of diagnosis for metabolic syndrome by integrating symptoms into physiochemical indexes. *Biomed. Pharmacother.*, **2021**, *137*, 111367. https://doi: 10.1016/j.biopha.2021.111367..

24. Grigorescu, E.-D.; Lăcătuşu, C.-M.; Creţu, I.; Floria, M.; Onofriescu, A.; Ceasovschih, A.; Mihai, B.-M.; Şorodoc, L. Self-reported satisfaction to treatment, quality of life and general health of type 2 diabetes patients with inadequate glycemic control from North-Eastern Romania. *Int. J. Environ. Res., Public Health* **2021**, *18*(*6*),3249. https://doi: 10.3390/ijerph18063249.

25. Farhan, L.O.; Taha, E.M.; Farhan, A.M. A case control study to determine macrophage migration inhibitor, and N-Telopeptides of type I bone collagen levels in the sera of osteoporosis patients. *Baghdad Sci. J.*, **2022**, *19*(*4*), 848-854. https://doi.org/10.21123/bsj.2022.19.4.0848.

26. Wei, W.; Liu, H.; Qiu, X.; Zhang, J.; Huang, J.; Chen, H.; Qiu, S.; Lin, R.; Li, S.; Tu, M. The association between serum adropin and carotid atherosclerosis in patients with type 2 diabetes mellitus: A cross-sectional study. *Diabetol. Metab. Syndr.*, **2022**, *14*, 1–8. https://doi.org/10.1186%2Fs13098-022-00796-y.

27. Zhang, H.; Chen, N. Adropin as an indicator of T2DM and its complications. *Food Sci. Hum. Wellness*, **2022**, *11*(*6*), 1455-1463. https://doi.org/10.1016/j.fshw.2022.06.00228.

28. Gao, S.; McMillan, R.P.; Zhu, Q.; Lopaschuk, G.D.; Hulver, M.W.; Butler, A.A. Therapeutic effects of Adropin on glucose tolerance and substrate utilization in diet-induced obese mice with insulin resistance. *Mol. Metab.*, **2015**, *4*(*4*):310-24. https://doi: 10.1016/j.molmet.2015.01.005.

29. Ali, I.I.; D'Souza, C.; Singh, J.; Adeghate, E. Adropin's role in energy homeostasis and metabolic disorders. *Int. J. Mol. Sci.*, **2022**, *23*(*15*):8318. https://doi: 10.3390/ijms23158318.

30. Layla, O.F.; Baydaa, A.A.; Aufaira, S.N.; Wesen, A.M. Total antioxidant capacity and malondialdehyde as a markers of oxidative stress in women with diabetic and disorder hormones. *Adv. Environ. Biol.*, **2016**, *10*(*5*), 172-176. https://doi.org/10.1155%2F2013%2F150693.