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ORIGINAL ARTICLE

Assessment of Serum Level of Asprosin in Obese Children and its Association with Determinants of Metabolic Syndrome

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ABSTRACT

Background: Asprosin, a new adipocytokine, has reportedly been associated with glucose release, dyslipidemia, and insulin resistance (IR). However, the relationship of asprosin with metabolic syndrome (MetS) remains unknown. This study aimed to investigate the correlation between serum asprosin levels and the factors influencing metabolic syndrome. **Patients and Methods:** This is a case control study was conducted on 100 children who attended and were selected from the General Pediatrics Outpatient Clinic, Zagazig University Hospital during the period of the study. Children were divided into two groups; Group1: included 50 obese children and Group 2: included 50 healthy normal-weight children of matched age and sex. **Results:** Regarding to serum asprosin there was a highly significant increase in the obese group compared to and control group. There were significant positive correlations between serum asprosin and weight, body mass index (BMI), waist circumference, systolic blood pressure (SBP), total cholesterol, triglycerides, fasting blood glucose (FBG), homeostasis model assessment (HOMA-IR) and glycosylated hemoglobin (HbA1c). There were significant negative correlations between serum asprosin and height, DBP and fasting insulin level. **Conclusions:** this study suggests a direct correlation between elevated levels of asprosin and key indicators of obesity and metabolic syndrome highlighting its potential role in contributing to the complex web of metabolic dysregulation. **Keywords:** Adipokine, Asprosin; Childhood obesity; Metabolic Syndrome

INTRODUCTION

Asprosin is a hormone that is released by the white adipose tissue which stimulates the release of glucose from the liver into the blood circulation.The white adipose structure is the main resource of asprosin. Asprosin is a cleavage product of the C-terminal of profibrillin which is encoded by the FBN1 gene**.** Asprosin targets many organs including the skeletal muscle, pancreas, liver, and cardiac system. In addition, asprosin stimulates appetite leading to weight gain. It also influences glucose metabolism, cell apoptosis, and insulin resistance. Furthermore, it has been implicated in some medical conditions such as obesity and diabetes **[1].**

Asprosin is a novel diabetogenic adipokine classified as a caudamin hormone protein. This adipokine is released from white adipose tissue during fasting and elicits glucogenic and orexigenic effects. Although white adipose tissue is the dominant source for this multitask adipokine, other tissues also may produce asprosin such as salivary glands, pancreatic B-cells, and cartilage **[2]**.

asprosin, is emerging as a potential mediator of obesity and MetS in children. Asprosin is a hormone protein derived from profibrillin and plays a role in the hypothalamic control of food intake as well as hepatic glucose release. In adults, its serum levels have been correlated with various MetS features, including obesity, hypertrigly- ceridemia, elevated cholesterol levels, T2D, and IR. However, conflicting data still exist regarding its association with these factors in children [3].

Asprosin deficiency is found in patients with **neonatal progeroid syndrome (NPS)**, while excess production of asprosin is detected in the condition of insulin resistance and obesity. It is proposed that asprosin in cooperation with ghrelin is beneficial to cachexia-anorexia, a complex metabolic syndrome occurring in severe burn victims. These findings imply that asprosin plays an essential role in a range of metabolic-related diseases [4].

The correlation between asprosin and pathologies directly associated with MetS such as obesity, insulin impairment, T2DM, and inflammation has been the subject of several studies in the scientific community. Other pathologies were also contemplated and asprosin correlated that are somewhat MetS associated like polycystic ovary syndrome (PCOS) [5].

Thus we aim to investigate the correlation between serum asprosin levels and the key indicators of metabolic syndrome.

METHODS

This case-control study was carried out on 100 children who attended the Pediatrics outpatient clinic, at Zagazig University Hospital during the period of the study.

Participants were selected from the general pediatric outpatient clinic of Zagazig University hospitals.

Sample size: Assuming that mean fasting asprosin concentration in the obese group was 2360 ± 5094 ng/ml and in the normal weight group was 307± 833 Ng/ml, so the sample size was 100 (50 in each group) "calculated using open Epi, with power 80% and cl 95 %

All children's parents or guardians provided written informed consent, and the study was authorized by the Research Ethical Committee of the Faculty of Medicine, Zagazig University (International Review Board) ZU-IRB. The World Medical Association's Code of Ethics (Declaration of Helsinki) was responsible for the work of human studies.

Inclusion criteria: Males and females obese children aged from 5-10 years old. Obesity was defined as BMI for age > +2SD based on the WHO growth reference for children.

Exclusion criteria: Children with any systemic disease. Children with infection. Children with physical and mental disability. Children receiving hypoglycemic or lipid lowering drugs.

Randomization: Patients were randomized by computer-generated randomization table into 2 equal groups.

The obese group included 50 obese children (BMI for age >+2SD) based on the WHO growth reference for children [6]. Control group included 50 healthy normal-weight children age and sex matched with obese group one.

All patients were subjected to the following:

Full history taking including personal, complaint, present, past, perinatal, developmental, dietetic, vaccination, and family history.

General examination: general appearance and vital signs (body temperature, pulse rate, respiratory rate, and blood pressure).

Anthropometric measurements

Height was measured by the same examiner using a wall-mounted Harpender 5 stadiometer (Haltain Limited, Crmych, Dyfeed, United Kingdom).

Weight was determined using a calibrated scale (Seca, Hamburg, Germany). BMI was calculated as weight (kg)/ height (m2).

Waist circumference was measured by Wrapping a flexible measuring tape around the natural waist (in between the lowest rib and the top of the hip bone), the umbilicus (belly button), or at the narrowest point of the midsection.

All body systems were examined.

Laboratory tests: Complete Blood Count (CBC), Fasting lipid profile, Fasting Blood Glucose, fasting insulin and hemoglobin A1c (HbA1C), and Homeostatic model assessment-IR (HOMA-IR).

Assessment of serum level of asprosin in obese children and healthy weight children was done.

Measurement of asprosin

Serum asprosin was measured using a commercial human asprosin ELISA Kit (Catalog No: E15190h, Wuhan EIAab Science Co. Ltd., China) according to the manufacturer's instructions. In brief, 100 µL serum covered with the plate sealer was incubated at 37°C for 2 h. First, the liquid was then removed, and 100 µL of detection reagent A was added and incubated at 37°C for 1 h. Second, the sample was washed 3 times, and 100 µL of detection reagent B was added. After 1-h incubation, the sample was washed 5 times, and 90 µL of substrate solution was added. Of note, 50 µL of stop solution was then added, and optical density of 450 nm was determined by automated microplate reader (PerkinElmer, Inc., Waltham, MA, USA). The acquired data were calculated by CurveExpert 1.4 (Hyams D.G., Starkville, MS, USA). The intra-assay coefficient of variation (CV) was ≤6.5%, and the inter-assay CV was ≤9.8%.

Statistical Analysis:

SPSS v28 (IBM Inc., Chicago, IL, USA) was used to conduct the statistical analysis. The quantitative data were provided as mean and standard deviation (SD) and compared between the two groups using an unpaired Student's t-test. When appropriate, qualitative variables were provided as frequency and percentage (%) and were evaluated using the Chi-square test or Fisher's exact test. A two-tailed P value of 0.05 was judged statistically significant. Pearson correlation was used to determine the degree of correlation between two quantitative variables.

RESULTS

Table 1; showed that anthropometric measurements, weight, height, BMI, and waist circumference were significantly higher in the obese group when compared to the control group (P<0.001). Weight z score, height z score, and BMI z score were significantly higher in the obese group compared to the control group (P<0.001).

Table 2; Triglycerides and total cholesterol there was a highly significant increase in obese group compared to the control group (P<0.001). Regarding the assessment of fasting insulin, FBG, HOMA-IR and HBA1C there was a highly significant increase in the obese group compared to the control group (P<0.001). Regarding to serum asprosin there was a highly significant increase in obese group compared to control group (P<0.001). Regarding to Hb, HDL and LDL there was insignificant difference between both groups.

Table 3; showed that there were significant positive correlations between serum asprosin and weight, BMI, waist circumference, SBP, total cholesterol, triglycerides, FBG, HOMA-IR and HBA1C. There were significant negative correlations between serum asprosin and height, DBP and fasting insulin level.

Table 1: Anthropometric measurements of the studied groups

		Group 1 (n=50)	Group 2 (n=50)	P value
Weight (Kg)	**Mean ± SD**	34.88±4.6	23.58±4.47	**<0.001***
	Range	26-42	16-36	
Weight z score	**Mean ± SD**	2.57 ±1.6	-0.26 ±.5	**<0.001***
	Range	1.9-3.7	-0.1-1.5	
Height (cm)	**Mean ± SD**	116.22±12.98	104.78±6	**<0.001***
	Range	94-140	90-115	
Height z score	**Mean ± SD**	0.25±0.5	-0.09 ± 0.3	**<0.001***
	Range	-0.3-2	-0.5-1.9	
BMI (Kg/m^2)	**Mean± SD**	32.18±3.06	17.7±3.08	**<0.001***
	Range	26.7-41	13.3-17.6	
BMI z score	**Mean± SD**	3.2±1.78	0.15±0.39	**<0.001***
	Range	2.2-3.9	-0.6-1.9	
Waist circumference (cm)	**Mean± SD**	98.38±6.79	62.2±6.83	**<0.001***
	Range	80-109	50-78	

BMI: body mass index, *: statistically highly significant as P value <0.001

Table 2: Laboratory investigations of the studied groups

		Group 1 (n=50)	Group 2 (n=50)	P value
Hb (g/dL)	**Mean ± SD**	12 ± 1.16	11.7 ± 1.21	0.145
	Range	8.7 - 14	8.7 - 13.3	
Total cholesterol (mg/dL)	**Mean ± SD**	181.5 ± 26.8	164.6 ± 38.39	**<0.001***
	Range	140 – 240	94 - 241	
Triglycerides (mg/dL)	**Mean ± SD**	159.5 ± 37.08	83.8 ± 23.72	**<0.001***
	Range	100 - 240	44 - 137	
HDL (mg/dL)	**Mean ± SD**	48 ± 10.59	52.8 ± 13.67	0.053
	Range	33 - 80	36 - 80	
LDL (mg/dL)	**Mean ± SD**	123.5 ± 18.65	124.4 ± 16.81	0.809
	Range	89 - 160	98 - 156	
Fasting insulin (µIU/mL)	**Mean ± SD**	14.4 ± 3.61	3.1 ± 1.01	**<0.001***
	Range	5.2 – 20	1.7 - 5.2	
FBG (mg/dL)	**Mean ± SD**	156.6 ± 16.37	88.5 ± 11.46	**<0.001***
	Range	122 - 195	67 - 110	
HOMA-IR	**Mean ± SD**	5.5 ± 0.98	1.8 ± 0.33	**<0.001***
	Range	2.1 - 8.9	1.3 - 2.7	

		Group 1 (n=50)	Group 2 (n=50)	P value
HBA1C (%)	**Mean ± SD**	5.6 ± 0.26	5.1 ± 0.25	**<0.001***
	Range	5.1 - 6	4.7 - 5.5	
Serum asprosin (ng/ml)	**Mean ± SD**	119.2 ± 16.29	65.2 ± 16.96	**<0.001***
	Range	87 - 161	42 - 99	

Hb: hemoglobin, HDL: high density lipoprotein, LDL: low density lipoprotein, FBG: fasting blood glucose, HOMA-IR: homeostatic model assessment of insulin resistance, *: statistically significant as P value <0.05

Table 3: Correlation between serum asprosin and different parameters

	Serum asprosin (ng/ml)	
	r	**p**
Age (years)	0.008	0.936
Weight (Kg)	0.650	**<0.001***
Height (cm)	-0.390	**<0.001***
BMI (Kg/m^2)	0.749	**<0.001***
Waist circumference (cm)	0.807	**<0.001***
Heart rate (beats/min)	0.021	0.836
SBP (mmHg)	0.397	**<0.001***
DBP (mmHg)	-0.398	**<0.001***
RR (breath/min)	-0.021	0.837
Temperature (º c)	-0.155	0.123
Hb (g/dL)	0.074	0.467
Total cholesterol (mg/dL)	0.270	**0.007***
Triglycerides (mg/dL)	0.689	**<0.001***
HDL (mg/dL)	-0.143	0.157
LDL (mg/dL)	0.041	0.684
Fasting insulin (µIU/mL)	-0.788	**<0.001***
FBG (mg/dL)	0.753	**<0.001***
HOMA-IR	0.783	**<0.001***
HBA1C (%)	0.556	**<0.001***

R: coefficient correlation, BMI: body mass index, SBP: systolic blood pressure, DBP: diastolic blood pressure, RR: respiratory rate, Hb: hemoglobin, HDL: high-density lipoprotein, LDL: low-density lipoprotein, FBG: fasting blood glucose, HOMA-IR: homeostatic model assessment of insulin resistance, *: statistically significant as P value <0.05

DISCUSSION

It has been reported that asprosin is an orexigenic hormone and stimulates the increase in food consumption and body weight gain. Secreted asprosin reaches the liver and triggers glucose release in response

to low dietary glucose. In response to insulin, the liver stocks excess glucose in glycogen form followed by the meal. In fasting, the liver is induced to break down the stored glycogen and secrete glucose, as well as synthesize new glucose. The secreted. Research & Reviews in Health Sciences 55 glucose into the circulation so that the brain and other organs that utilize from the glucose can maintain their normal function [3].

The weight, height, BMI, and waist circumference were substantially higher in the obese group compared to the control group (P<0.001). In particular, the weight z score, height z score and BMI z score were found to be significantly elevated in obese group to the control group.

A marked divergence in dietetic history emerged between the two groups, demonstrating statistical significance (P<0.001). The control group exhibited a history of a balanced diet among all patients, while the obese group demonstrated a history of unhealthy dietary habits. Furthermore, a higher prevalence of family history related to Hypertension and dyslipidemia was observed in the obese group in comparison to the control group. Physical activity assessment revealed moderate physical activity for all cases in the control group, while the obese group showcased predominantly low physical activity.

Laboratory investigations, encompassing hemoglobin showed no statistically significant differences between the two groups. About the lipid profile, triglyceride levels were significantly elevated in the obese group compared to the control group (P<0.001). Conversely, total cholesterol, high-density lipoprotein, and low-density lipoprotein presented no substantial differences between the two groups.

Assessment of blood sugar levels between the studied groups revealed that patients within the obese group exhibited significantly higher levels of FBG, Fasting insulin, HOMA-IR, and HbA1c in comparison to control group (P<0.001).

Similarly, Sünnetçi Silistre & Hatipoğl [7] involved the participation of three distinct cohorts of children, namely 54 who were classified as overweight, 44 who were categorized as obese, and 60 falling within the normal-weight range. The study found a statistically significant difference among the groups in terms of insulin, HOMA-IR, insulin sensitivity index, and HbA1c.

Our results showed an elevation of serum asprosin levels within obese group in contrast to the control group, with this difference being statistically highly significant (P<0.001).

These results came by those reported by Sünnetçi Silistre & Hatipoğl [7] who demonstrated a statistically significant elevation in asprosin levels among obese children as opposed to those with normal weight (P = 0.009). Furthermore, the multiple regression analysis identified asprosin as a predictive factor for obesity.

Similarly, Mirr et al [8] encompassed a cohort comprising 50 non-diabetic individuals with obesity and an equivalent number of healthy volunteers. Notably, the investigation unveiled markedly elevated levels of asprosin concentrations among the obese patient group, with a statistically significant difference evident ($p < 0.001$).

The study by Moradi et al [9] was conducted on a sample size consisting of 35 normal-weight children and an equal number of children with obesity. The findings demonstrated a statistically significant elevation in circulating asprosin levels among obese children in comparison to the control subjects.

These results came under a previous study by Hong et al [10] that showed an evident elevation in serum asprosin levels in patients diagnosed with metabolic syndrome in comparison to the healthy control group. Additionally, a discernible ascending pattern was observed in serum asprosin levels as the number of metabolic components increased within the population.

In the investigations conducted by Liu et al [11] findings indicated heightened levels of asprosin in obese children. They observed a significant increase in circulating asprosin levels among obese children with non-alcoholic fatty liver disease (NAFLD) in contrast to their obese counterparts without NAFLD. However, no discernible variation in asprosin concentrations was observed between obese children without NAFLD and their lean counterparts.

On the other hand, two other studies, Corica et al [12] and Long et al [13] reported diminished levels of asprosin in obese children. Long postulated that these discrepancies might be attributed to the degree of obesity as a primary determinant. It was proposed that in cases of milder obesity, compensatory mechanisms result in reduced asprosin concentrations to counterbalance metabolic disturbances. When compensatory mechanisms prove inadequate, a subsequent escalation in both body mass and asprosin concentration ensues, further accentuating insulin resistance. This implies a dynamic association between asprosin and body mass that is contingent upon the severity of obesity and its accompanying metabolic perturbations. Nonetheless, achieving clarity on these disparities necessitates additional extensive research endeavors.

Regarding metabolic syndrome indicators, the first notable indicator is fasting hyperglycemia, with 39 out of 50 obese children (78%) demonstrating levels above 110 according to the international diabetes federation diagnostic criteria. Regarding HDL levels, 16 out of the 50 subjects (32%) exhibited HDL levels below 40. Examining triglyceride levels, 28 out of 50 obese children (56%) displayed levels above 150.

In terms of blood pressure, 31 out of 50 subjects (62%) demonstrated values exceeding the 90th centile. All 50 subjects

(100%) presented with central obesity.

Furthermore, our results showed a significant positive correlation between serum asprosin levels and parameters including weight, BMI, waist circumference, SBP, triglycerides, FBG, HOMA-IR, and HbA1c. Conversely, a significant negative correlation was observed between serum asprosin levels and height, DBP, and fasting insulin levels.

These results accord with those of Mirr et al [8] who showed that asprosin concentrations exhibited positive correlations with BMI ($p < 0.001$, $r = 0.8$ in females and $r = 0.8$ in males), waist circumference ($p < 0.001$, $r = 0.73$ in females and $r = 0.81$ in males), and all assessed indicators of insulin resistance. Notably, the most robust correlation emerged for the TyG-BMI relationship ($p < 0.001$, $r = 0.78$ in females and $r = 0.81$ in males). It was further noted that circulating asprosin levels exhibited higher values among females ($p < 0.001$). A positive correlation between circulating asprosin and BMI was noted in both obese and non-obese individuals, with the strength of this correlation being more pronounced within the obese group.

In line with our results, Moradi et al [11] observed that obesity was correlated with notable changes in Metrnl and asprosin levels. Consequently, these adipokines emerge as prospective and promising therapeutic targets for addressing the complexities of obesity and its interconnected metabolic disruptions.

Similarly, Hong et al[10] showed that serum asprosin concentrations exhibited positive associations with several parameters, including BMI, waist circumference, FPG, FIns, HOMA-IR, TG, MCP-1, and IL-6. Conversely, a negative correlation was observed with HDL-C.

Further, Hong et al[10] revealed that serum asprosin was found to be an independent factor positively associated with the occurrence of both metabolic syndrome and insulin resistance. This correlation remained statistically significant even after meticulous adjustment for various covariates.

Dyslipidemia and hyperglycemia represent hallmark pathological states within the context of metabolic syndrome, significantly influencing the disease pathogenesis. Our findings demonstrate substantial correlations between serum asprosin levels and parameters including TG, HDL-C, and FBG. These observations lead us to postulate that asprosin may furnish a molecular linkage bridging glucose-lipid metabolism and metabolic syndrome. While inferring direct causal relationships between asprosin and these parameters warrants caution, the potential of therapeutic targeting of asprosin to mitigate metabolic disorders in individuals with metabolic syndrome remains a topic of interest[14].

Limitations

This study is not without its inherent limitations. Primarily, the sample size of our study might not fully represent the diversity of the population, and the findings might not be generalizable to all demographic groups.

Including a larger and more diverse sample would enhance the external validity of the results. Further, participants were recruited from a specific clinic, potentially introducing selection bias. This could affect the representativeness of the sample and the generalizability of the findings to broader populations.

In addition, we included children with age groups ranging from 5 to 10 years old, and could not get a certain diagnosis of metabolic syndrome at that age based on the international diabetes federation diagnostic criteria. Further, participants were recruited from a specific clinic, potentially introducing selection bias. This could affect the representativeness of the sample and the generalizability of the findings to broader populations.

Conclusion: this study suggests a direct correlation between elevated levels of asprosin and key indicators of obesity and metabolic syndrome highlighting its potential role in contributing to the complex web of metabolic dysregulation.

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