Journal of Rare Cardiovascular Diseases
ISSN: 2299-3711 (Print) | e-ISSN: 2300-5505 (Online)
www.jrcd.eu
RESEARCH ARTICLE

Evaluation of Serum Levels of Leucine-Rich α-2 Glycoprotein, Fibroblast Growth Factor-19, Spexin, and Copeptin in Obese Males

MOHAMMED NSAIF JASSAM[1] , Omar Thaer Jawad[2]
Department of Biology, College of Education for Pure Sciences, Samarra University

*Corresponding Author

Article History
Received: 15.07.2025
Revised: 31.07.2025
Accepted: 18.08.2025
Published: 29.09.2025

Abstract: ***Background:*** Spexin (SPX), a neuropeptide implicated in appetite regulation and energy metabolism, has been linked to obesity and related metabolic disorders. However, findings remain inconsistent across populations. ***Objective:*** To examine the serum SPX levels of healthy, overweight, and obese adults and to determine the serum SPX levels in relation to anthropometric and biochemical indices of obesity. ***Methods:*** A case-control study was carried out among 90 adults who were equally divided into the healthy, overweight and obese group. As measurements were taken, body mass index (BMI) and waist circumference for the individual were assessed, fasting glucose and lipid profile were measured. Serum SPX levels were measured by using the method of enzyme immunoassays (ELS). ***Results:*** The BMI and waist circumference had a significant difference between groups ($p<0.001$). While numerical differences in levels of SPX were found, no statistically significant differences were found between study groups. Moreover, results of correlation analysis showed no significant correlation with the levels of SPX with the markers of obesity. ***Conclusion:*** Circulating SPX levels did not significantly differ between healthy, overweight, and obese adults in this cohort, nor were they correlated with anthropometric or biochemical parameters. These results indicate that SPX is not a good biomarker of obesity in this population. Further studies to elucidate the role of SPX in human obesity have to be conducted with larger groups of overweight or obese individuals with a broad range of ethnic populations.

Keywords:

INTRODUCTION

According to the World Health Organization (WHO), obesity is abnormal or excessive fat accumulation that may become a detrimental issue contributing to reduced health.

This syndrome is characterized by a body mass index (BMI) of 30 kg/m² or more, a threshold that mainly applies to White populations. Obesity, defined as a BMI of 30 kg/m² or more, affects about 890 million people (13%) worldwide. BMI is a practical, though imperfect, method for determining fat mass, primarily used as a screening tool for obesity and for epidemiological applications (Lingvay et al., 2024a).

A 2021 WHO report stated that more than one billion people suffer from obesity, including 650 million adults, 340 million adolescents, and 39 million children, based on data collected in 2016. The prevalence of overweight and obesity increased globally by 27% in adults and 47% in children between 1980 and 2013 (Avgerinos et al., 2019).

The metabolic consequences of obesity include type 2 diabetes, metabolic dysfunction-associated fatty liver disease, hypercholesterolemia, chronic kidney disease, and cardiovascular diseases linked to metabolic dysfunction. These disorders share the same pathophysiology, as the constant need to store excess calories leads to adipose tissue dysfunction, characterized by elevated circulating fatty acids and increased production of pro-inflammatory cytokines (Zheng et al., 2024).

These abnormalities trigger oxidative stress, mitochondrial and endoplasmic reticulum stress, resulting in beta-cell dysfunction, insulin resistance in multiple organs, endothelial dysfunction, hypercoagulability, and lipid metabolism disturbances, ultimately leading to clinical symptoms (Klein et al., 2022).

Adipose tissue is recognized as a multifunctional entity, essential not only for fat and energy storage but also for endocrine and immune activities (Bou Matar et al., 2025). Adipose tissue functions as an important endocrine organ by secreting numerous physiologically active chemicals, known as adipose-derived secretory factors or adipokines, which exhibit either pro-inflammatory or anti-inflammatory properties. Abnormal synthesis or secretion of these adipokines resulting from adipose tissue dysfunction may contribute to the pathophysiology of obesity-associated problems (Arias et al., 2024).

Certain proteins and biological components serve as markers that help in understanding metabolic changes in obese patients. This has led to the identification of new biomarkers for obesity and related metabolic disorders. Leucine-rich a-2 glycoprotein (LRG1) is a member of the leucine-rich repeat (LRR) superfamily of proteins. It plays a role in several biological processes,

such as inflammation, immune response, and cell adhesion (Camilli et al., 2022). Lysaccharide-Recognition Proteins (LRGP-1 - LRG1) alone or together with other known markers are thought to be a candidate inflammation and obesity biomarker. LRG1 concentrations are positively correlated with obesity and lower plasma concentrations of LRG1 are predictive for weight loss after bariatric surgery and the improvement of metabolic disorders (Pek et al., 2018).

Fibroblast Growth Factor-19 (FGF-19) became an emerging metabolic hormone that has been extensively studied with regard to its potential as a therapeutic agent for the treatment of obesity and metabolic recovery after bariatric surgery, in particular diabetes. This is principally secreted by enterocytes, and acts as an obesity, energy homeostasis, lipid metabolism, and glucose homeostasis metabolic regulator. Fibroblast growth factor (FGF)-19 plays key roles in the regulation of bile acid, decrease in food intake as well as hedonic food preferences in the central nervous system, induction of fatty acid oxidation, insulin-sensitivity, and increase in the energy expenditure (Hosseinzadeh et al., 2020).

Spexin, a 14 amino acid neuropeptide (also called neuropeptide Q) was discovered in 2007. Primarily it is produced by humans in their white adipose tissue (Behrooz et al., 2020). Spexin has the functions of obesity, energy metabolism, appetite control, satiety, glucose and lipid metabolism, fatty acid absorption, cardiovascular system, renal function, endocrine balance, reproduction, and gastrointestinal function (Ma et al., 2018a). Spexin is associated with its decreased expression in adipose tissue in overweight individuals and decreased levels of these circulating concentrations as well in people with obesity (badri et al., 2025).

First described by Holwerda in 1972, Ccopeptin is a 39-amino acid peptide that has a leucine-rich core (Holwerda, 1972). Copeptin is a cleavage product of AVP precursor, which regulates fluid balance and endocrine stress responses, and it has been associated with metabolic disorders (Akbulut et al. 2025). In adults, numerous cross-sectional and prospective studies have demonstrated that copeptin goes together with abdominal obesity, insulin resistance and metabolic syndrome (Shu et al., 2025).

The study is to measure serum levels of LRG1, FGF-19, Spexin and Copeptin in obese males and to compare them with non-obese individuals and whether they could be used as biomarkers exceeding in the diagnosis or evaluating the severity of obesity related metabolic therapy.

MATERIAL AND METHODS

Study Design

This study had a total of 90 male participants who were divided in three groups:
30 healthy participants (control group),
30 obese people (people diagnosed with morbid obesity by a specialist physician and according to the measure of height and weight to calculate the BMI),
30 overweight individuals.

Blood samples were obtained from outpatients attending clinics and laboratories, the age of the subjects varied between 14 and 70 years of age. The time frame of the study ran from November 5, 2024 through January 2nd of 2025.

Collection of Blood Samples

A total of 5 ml of blood from the veins of both the patients and the healthy individuals were drawn through the use of sterile medical syringes. The blood was transferred into gel tubes that were not anticoagulant treated and was allowed to clot for one hour at room temperature. Serum was then separated by centrifugation (x g, 1000 minutes).

The serum was then spotted into four Eppendorf tubes (0.5 ml of serum in each Eppendorf), then frozen at -20 degC until analysis time. Each tube was only used once in order to prevent freezing and unfreezing of the samples.

Assessment of Body Mass Index (BMI)

Height and weight measurement was done with stadiometer (m) and a sensitive balance (kg). The BMI was based on the following formula:

BMI = \frac{\text{Weight (in kg)(\text{weight in Kilograms to Augmented SI units)}}{\text{Height (in m)}^2(\text{height in meters to Augmented SI units)

Biochemical Analysis

Measurement of the Concentration of Fibroblast Growth Factor-19 (FGF-19)
The serum level of FGF-19 was determined by using a particular immunoassay as recommended by the manufacturer.

Evaluation of the Spexin Concentration
The serum level of Spexin was measured using a validated immuno assay method.

Evaluation of Copeptin Level
The serum concentration of Copeptin was determined with the help of a standardised enzyme convertase immunodoassay (ELS).

RESULTS AND OBSERVATIONS:

The Body Mass Index (BMI) of the three groups was significant. The mean value obtained as a BMI in the healthy group was 21.333 +- 0.326 (95% confidence interval; CI, 20.666 and 22.001). The mean BMI in the group of overweight is

27.233 +- 0.294 (95% CI: 26.631-27.835), the mean BMI in the group of obese is 37.733 +- 0.705 (95% CI: 36.292-39.174).

One-way ANOVA analysis revealed a highly significant overall difference among the groups (**F = 300.215, P < 0.001**). According to Duncan's multiple range test, the mean rankings were **a, b, and c** for obesity, overweight, and healthy groups, respectively, indicating statistically significant differences between each pair of groups at a significance level of 0.05. Furthermore, the observed differences exceeded the Least Significant Difference (LSD) threshold of approximately **1.35**, confirming that these differences were real rather than random.

Similarly, Waist Circumference (WC) demonstrated a progressive upward trend across the groups, with highly significant statistical differences. The mean WC in the healthy group was **80.567 ± 1.353** (95% CI: **77.799–83.335**), in the overweight group **93.567 ± 0.681** (95% CI: **92.174–94.959**), and in the obese group **122.067 ± 1.434** (95% CI: **119.133–125.000**). ANOVA analysis yielded **F = 310.575, P < 0.001**, confirming strong statistical significance.

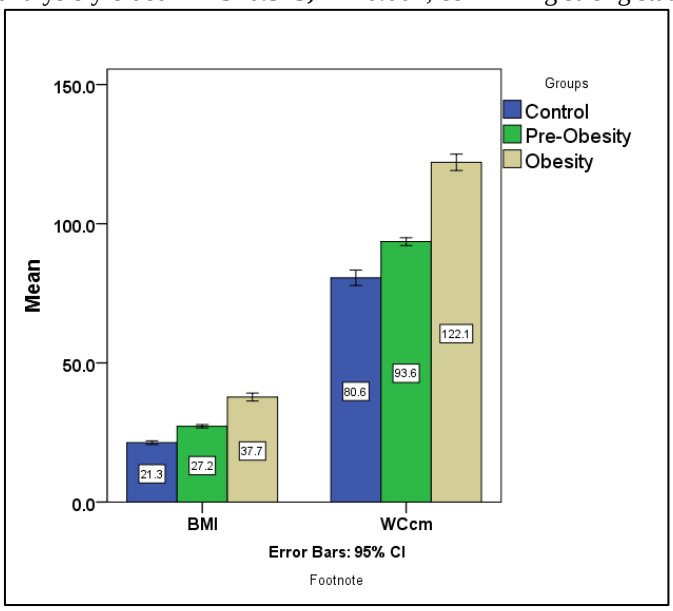

Figure 4-1. Body Mass Index (BMI) and Waist Circumference (WC) among the Three Study Groups

LRG1 values showed highly significant differences among the three groups. The mean value in the healthy group was 24.070 ± 0.524 with a 95% confidence interval of 22.998 – 25.142. The overweight group recorded a mean of 28.608 ± 0.270 with a confidence interval of 28.056 – 29.161, while the obesity group showed a mean of 36.981 ± 0.233 with a confidence interval of 36.504 – 37.458.

One-way ANOVA revealed a highly significant overall difference (F = 320.010, P < 0.001). According to Duncan's test, the order was: a = obesity, b = overweight, and c = healthy. All pairwise differences exceeded the LSD value at 5% (LSD ≈ 1.03), confirming that LRG1 levels were highest in the obesity group, followed by overweight, and then healthy individuals. This is illustrated in Table (4-3) and Figure (4-2).

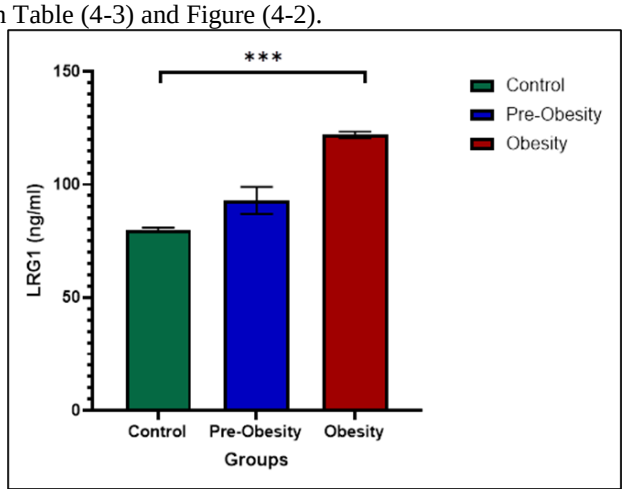

**Figure 4-2. Leucine-rich α-2 Glycoprotein 1 (LRG1) among the study groups, *=p<0.001

The measurable values of FGF19 showed a clear and significant inverse relationship with obesity status. The highest mean FGF19 levels were observed in the healthy group (148.544 ± 4.723, 95% CI: 138.884 – 158.203), followed by the overweight group (101.566 ± 8.180, 95% CI: 84.837 – 118.296), and then the obesity group (47.686 ± 4.746, 95% CI: 37.978 – 57.393).

ANOVA analysis revealed statistically significant differences across the groups (F = 68.382, P = 0.001). According to Duncan's test, the order was: a = healthy, b = overweight, and c = obese. Since FGF19 values were < LSD at 5% (LSD ≈ 17.17), this indicates significant differences as individuals transition from the healthy group to the obese group. These findings are illustrated in Table (4-4) and Figure (4-3).

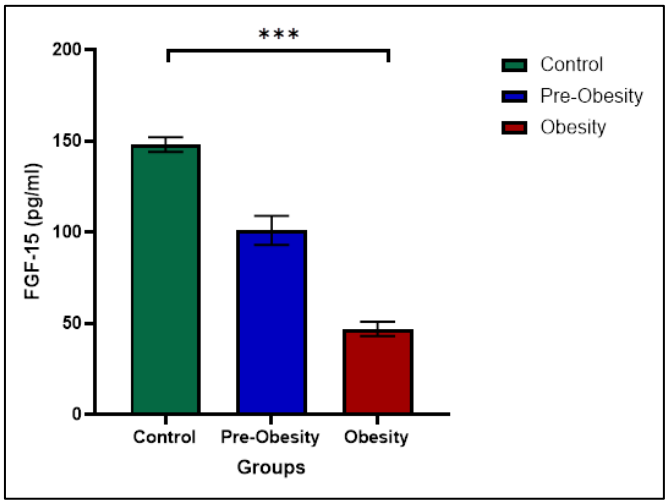

Figure 4-3. Fibroblast Growth Factor 19 (FGF19) among the study groups, p < 0.001

The mean values differed numerically between the groups but without overall statistical significance. The mean in the healthy group was 372.007 ± 54.696 with a 95% CI of 260.141–483.873; in the overweight group it was 280.386 ± 41.019 with a 95% CI of 196.493–364.280; and in the obese group it was 266.581 ± 41.286 with a 95% CI of 182.141–351.020. ANOVA showed F = 1.544 with P = 0.219, indicating no overall significant difference. Duncan's letters were identical (a) for all three groups, and the pairwise differences did not exceed the approximate LSD value at 5% (LSD ≈ 129.77), supporting the absence of statistical significance at the level of pairwise comparisons. This is shown in Table (4-5) and Figure (4-4).

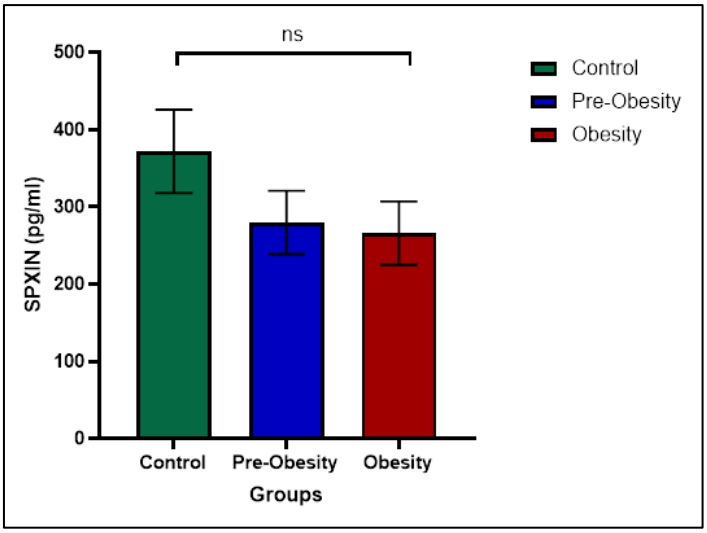

Figure 4-4. Spexin (Spxin) levels among the three study groups, ns = p > 0.05

The results showed comparable Copeptin levels among the groups without statistical significance. Healthy individuals recorded 303.228 ± 23.107 with a 95% CI of 255.969 – 350.487, the overweight group 322.732 ± 16.563 with a 95% CI of 288.857 – 356.608, and the obese group 308.646 ± 22.121 with a 95% CI of 263.403 – 353.889.

ANOVA analysis yielded F = 0.234 with P = 0.792, indicating no significant differences. Duncan's test assigned identical letters (a) to all groups, and pairwise differences did not exceed the LSD value at 5% (LSD ≈ 58.53), confirming the absence of significant differences in Copeptin levels among the groups.

As shown in Table (4-6) and Figure (4-5).

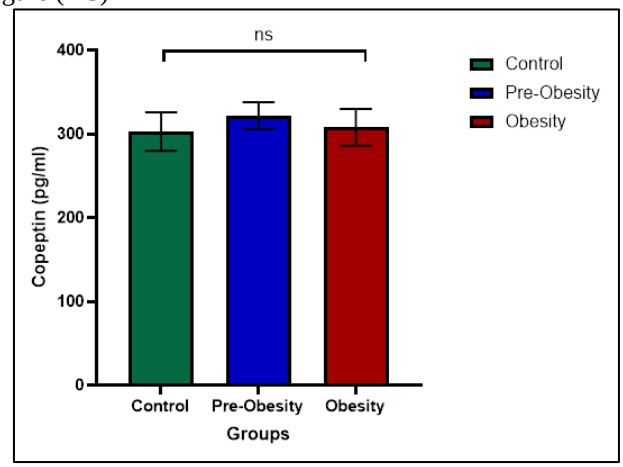

Figure 4-5. Copeptin levels among study groups, ns = p > 0.05

Based on ANOVA analysis and Duncan's post-hoc comparisons, LRG1 showed a clear upward trend from healthy to overweight and then obese subjects, while FGF19 displayed the opposite pattern, with a gradual decrease across the groups. Both differences were highly significant (P < 0.001). In contrast, Spexin and Copeptin did not show significant differences among the groups (P = 0.219 and 0.792, respectively).

These patterns, consistent with the progressive increase in BMI and WC across the groups, suggest that LRG1 and FGF19 represent more sensitive biomarkers of obesity status and could be incorporated into discrimination or risk prediction models. Meanwhile, Spexin and Copeptin did not provide clear discriminative value within this sample. Confirmation of these findings in larger cohorts with better adjustment for confounding factors is recommended.

The biomarkers are summarized in Figure (4-6).

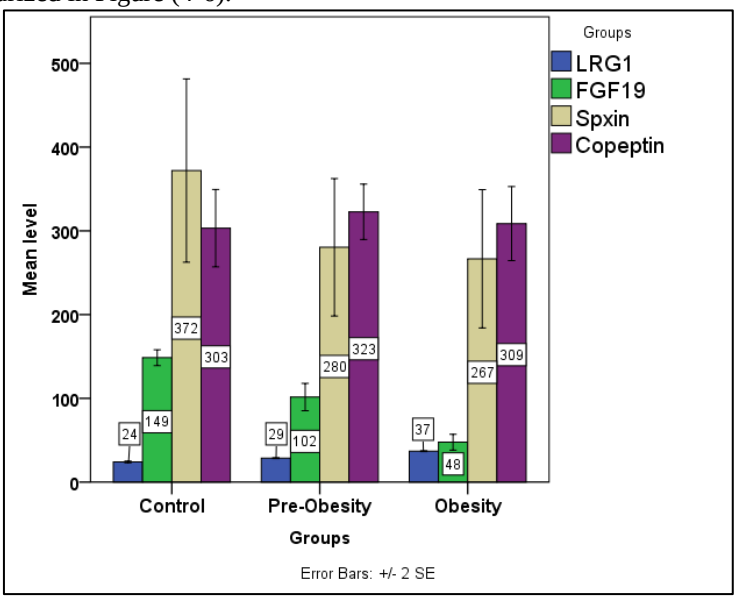

Figure 4-6. Analysis of differences in the biomarkers

DISCUSSION

The progressive increase in both Body Mass Index (BMI) and Waist Circumference (WC) across the study groups highlights the reliability of anthropometric indicators as primary markers of fat accumulation. The significant differences among the groups are consistent with the common scientific view that both indices rise gradually when moving from normal weight to overweight and then to obesity. This observation has been documented in geographically proximate and diverse settings, reinforcing the robustness of BMI and WC as diagnostic tools for classifying individuals according to obesity status (Chinedu et al., 2013).

Despite the widespread use of BMI, it remains limited by its inability to distinguish between fat-free mass and fat mass. Nevertheless, many studies across different populations confirm a strong association between BMI and metabolic complications, validating its role as a practical epidemiological measure. The findings of this study support this perspective: the clear elevation in BMI among obese individuals aligns with the reported increase in metabolic risk in similar studies, while the intermediate values in the overweight group reflect a transitional state in which fat accumulation begins to exert pathological effects. This supports the threshold-based classification proposed by earlier research (Sweatt et al., 2024; Cancela-Carral et al., 2023).

In contrast, WC is increasingly recognized as a more sensitive indicator of central obesity and visceral fat accumulation compared to BMI. The present study's results, which revealed higher WC values in obese individuals, are consistent with evidence highlighting abdominal obesity as a more potent determinant of cardiometabolic risk than total body mass. Investigations in populations with similar ethnic and lifestyle characteristics have shown WC to outperform BMI in predicting insulin resistance, dyslipidemia, and cardiovascular morbidity. Accordingly, our interpretation of the results suggests that WC does not merely complement BMI but may provide a more precise clinical signal when visceral obesity predominates (National Academies of Sciences & Callahan, 2023; Lee et al., 2022).

Other studies show the opposite in people with normal BMI but increased WC as these individuals have been labelled as being normal weight obese. This underlies the need of measuring two indices at the same time, in order to avoid misclassification. Our results were consistent and significant for each measure across all the groups and provided evidence that they; combined were useful in describing the burden of fat. Furthermore, the reliability of this finding in a setting other than in which the data were collected adds more confidence in the use of these as an early screening instrument for at-risk persons (Ross et al., 2020; Lukacs et al., 2019).

Taken as a whole, the study results reported here provide the impetus to reinforce mostly trend when it comes to gradual increase of BMI and WC from non-obesity to obesity as reported in the literature. They corroborate the pertinence of the use of these clinical and epidemiological indicators that are being used in series to screen for the high-risk ones. In parallel, it uncovers the necessity to balance anthropometric indexes with biochemical biomarkers to get improved capacity to differentiate obesity phenotypes with various health risks (Janssen et al., 2004; Singh et al., 2022). The research showed that there was a scientifically significant stepwise increase in the amount of the Leucine-rich a-2 Glycoprotein 1 or LRG1 in light of the healthy people until the overweight and then obese without missing data between all pairwise comparisons.

This pattern indicates a strong association between LRG1 and obesity burden, reflecting the physiopathological load of obesity at the systemic level. This elevation aligns with the characterization of LRG1 as an acute-phase glycoprotein associated with systemic inflammation. Obesity represents a state of low-grade chronic inflammation driven by immune cell infiltration into adipose tissue, secretion of pro-inflammatory cytokines, and metabolic stress. LRG1 expression has been observed to increase in inflammatory contexts, contributing to neutrophil activation, angiogenesis, and vascular remodeling. Accordingly, fat accumulation would be expected to correlate with higher LRG1, a finding supported by the current study (He et al., 2021).

Population-based investigations across diverse ethnic and metabolic backgrounds report the same pattern of association: LRG1 levels rise progressively with increasing weight, central obesity, and insulin resistance. LRG1 is also linked to components of the metabolic syndrome, including fasting blood glucose, dyslipidemia, and hypertension, underscoring its value as an integrative marker that reflects multiple pathways of dysfunction (Alhammad et al., 2022).

By contrast, smaller-scale reports show a less clear picture: in limited samples or distinct disease profiles, the rise in LRG1 appeared milder, or associations weakened after adjustment for confounders such as age, sex, and comorbidities. This variation is often attributed to differences in population structure, disease severity, and measurement methods. Nevertheless, the strength and consistency of the current study's findings—showing significant differences even in overweight compared with healthy individuals—demonstrate that LRG1 changes begin early along the obesity continuum and are not confined to severe cases (Dritsoula et al., 2024).

Recent evidence suggests a mechanistic role for LRG1 beyond that of a passive marker: it may participate in vascular remodeling and promote endothelial dysfunction, common in obesity. Its interaction with TGF-β signaling pathways further implies an active role in the progression to cardiometabolic risk. If confirmed, LRG1 could be positioned both as a diagnostic biomarker and as a potential therapeutic target (Mertlitz et al., 2025).

From an applied perspective, the present study's findings highlight LRG1's ability to sensitively differentiate between healthy, overweight, and obese individuals, adding value to traditional anthropometric indicators. Its stepwise rise across groups suggests utility for early detection of subtle metabolic disturbances not captured by BMI and WC alone. Its

statistical performance supports incorporation into multivariable models to improve prediction of obesity-related diseases (Hoeh et al., 2025).

Synthesizing the evidence, the results align with the prevailing literature that classifies LRG1 as a robust and biologically relevant marker of obesity and its metabolic consequences, with the added dimension of detecting consistent changes as early as the overweight stage. This positions LRG1 as a strong candidate for inclusion in clinical and research frameworks aimed at improving early detection and risk stratification (Choi et al., 2022).

Other studies also highlight a clear stepwise pattern from healthy through overweight to obese individuals, reflecting LRG1's alignment with fat-driven metabolic disturbances. This integrates with mechanistic and clinical evidence supporting its role as a sensitive marker of metabolic dysfunction and possibly as a mediator in some of its pathways (He et al., 2021).

Recent experimental findings reveal a causal contribution of LRG1 to obesity-associated fatty liver and insulin resistance: its elevation promotes de novo lipogenesis, inhibits fatty acid oxidation, and downregulates insulin receptor signaling components such as IRS-1 and IRS-2. This results in fundamental impairments in lipid metabolism, hepatic insulin action, and increased risk (Cao et al., 2025).

Parallel observational studies in adolescents underline LRG1's value as an early marker of metabolic disturbance: higher levels were associated with increased odds of overweight/obesity and with inflammatory and adipokine markers including hs-CRP, leptin, chemerin, IL-6, and IL-10, suggesting it mirrors the emerging inflammatory milieu in the early stages of obesity (Pek et al., 2018).

Animal models reveal a context-dependent dual role: while expression may decrease in dysfunctional adipose tissue, it remains stable or elevated in the liver, where it may exert anti-inflammatory or adaptive effects to preserve tissue balance. This complexity underscores the need to interpret LRG1 changes within a framework that considers tissue specificity and disease stage (Camilli et al., 2022).

The importance of LRG1 extends beyond metabolic disorders, encompassing immune regulation, fibrosis, and responses to biologic therapies in inflammatory diseases—strengthening its status as a multifaceted biomarker and potential therapeutic target. Preclinical models have shown that blocking its function can attenuate disease progression, supporting its validity as a targetable pathway (Carvalho et al., 2024).

Fibroblast Growth Factor 15 (FGF15) is an enterohepatic hormone of ileal origin induced by bile acids. It signals via the FGFR4 receptor complex and the β-Klotho cofactor to regulate bile acid synthesis in the liver, in addition to glucose and lipid metabolism. Physiologically, FGF15 secretion follows circadian and postprandial rhythms, usually peaking within 90–120 minutes after food intake when the intestinal FXR receptor is activated by bile acids (Carvalho et al., 2024). These pulses inhibit the CYP7A1 enzyme and modulate hepatic substrate circulation. This mechanism explains why fasting FGF15 concentrations are relatively low, why postprandial values display clearer gradients, and why obesity is often associated with impaired FXR signaling and altered bile acid substrates, resulting in reduced FGF15. Accordingly, the observed stepwise decrease from healthy subjects to overweight and obese individuals is biologically consistent and aligns with known physiology (Ursic-Bedoya et al., 2022).

A broad range of epidemiological observations indicate an inverse relationship between FGF15 and markers of fat accumulation; low fasting or postprandial levels are associated with higher BMI and WC, as well as with insulin resistance and impaired glucose regulation. The gradient in this study's data reflects this relationship, indicating that a decline in FGF15 is part of the hormonal signature accompanying the transition from metabolic health to dysfunction (Lan et al., 2017).

Interventional studies support this interpretation; following weight loss, especially after metabolic surgery, postprandial FGF15 responses rise significantly, in parallel with improved bile acid kinetics, insulin sensitivity, and glycemic control. Long-term follow-up shows these changes are more pronounced after surgical interventions compared to lifestyle modification, reflecting a remodeling of the gut–liver axis (Guo et al., 2022).

Therapeutic approaches targeting the FXR–FGF15 axis reinforce this concept; FXR activation enhances FGF15 secretion, while impaired FXR signaling suppresses it. Pharmacological FGF15 analogs have shown positive effects on hepatic fat and fibrosis markers in metabolic liver diseases, supporting the notion that restoring FGF15 signaling may correct metabolic disturbances rather than merely reflect them (Bozadjieva et al., 2018).

Nevertheless, methodological considerations frame these findings; sampling conditions such as fasting vs. postprandial state, timing, and meal composition all affect FGF15 levels. Moreover, assay variability can complicate cross-study comparisons. Additionally, individual differences in bile acid pools and receptor sensitivity may alter absolute values while preserving the inverse relationship with fat accumulation. These factors account for limited discrepancies without undermining the overall picture (Li et al., 2024).

Mechanistically, the reduction of FGF15 in obesity can be attributed to impaired FXR activation and disrupted gut–liver signaling, which relieve inhibition of CYP7A1, alter hepatic lipid handling, and promote steatosis, insulin resistance, and systemic inflammation. FGF15 functionally complements FGF21; while FGF15 is more closely linked to glucose and bile acid regulation, FGF21 is tied to hepatic lipid accumulation physiology. Together, these hormones provide a broader perspective on endocrine dysregulation in obesity, with several studies supporting this view (Liu et al., 2025).

Overall, the progressive decline in FGF15 across healthy, overweight, and obese individuals aligns with its physiological role and with clinical observations showing that improved metabolic health is accompanied by restored FGF15 signaling. These findings support the view that FGF15 acts as both a biomarker and a mechanistic node in obesity-related disorders, and that combining it with anthropometric and inflammatory markers may enhance risk stratification, especially at the borderline between normal and early obesity stages where conventional measures are less accurate (Antonellis et al., 2019).

The results of this study are in clear agreement with previous research that reported a significant reduction in FGF15 concentrations in obese individuals compared to those with normal weight. Another study supports this observation and suggests that impaired FGF15 secretion reflects defective FXR signaling linked to bile acids, contributing to metabolic dysfunction. This also aligns with evidence that reduced FGF15 is associated with insulin resistance, hepatic steatosis, and adverse lipid profiles, underscoring its role as an integrative marker of metabolic health (Carbone et al., 2025).
Conversely, limited reports have described weaker associations or non-significant differences after adjustment for confounding factors such as age, diet composition, and circadian variation. These discrepancies are likely due to methodological issues, population heterogeneity, and design differences (Carvalho et al., 2024b).

Spexin is a peptide involved in the regulation of appetite, energy expenditure, and lipid metabolism. In this study, although numerical differences in means were observed among healthy, overweight, and obese groups, overall analyses did not reveal significant differences. Duncan's test showed identical group letters, supporting the absence of meaningful pairwise differences. Accordingly, Spexin does not appear, within this study sample, to be a marker capable of differentiating between states of fat accumulation (Behrooz et al., 2020).

This finding is consistent with observational reports that noted high interindividual variability in circulating Spexin levels, which may obscure group-level differences in cross-sectional studies. Other studies have confirmed that circulating Spexin is influenced by multiple confounding factors, including sex, age, diet, and circadian rhythm—factors that may weaken its discriminative value if not methodologically controlled. Parallel works have also reported no clear differences in Spexin between obese and non-obese individuals, limiting its robustness as a standalone obesity marker (Lin et al., 2018; Behrooz et al., 2020).

Conversely, other investigations have reported opposing results, finding lower Spexin concentrations in obese subjects compared with those of normal weight, and linking reduced Spexin to insulin resistance and metabolic dysregulation. These findings suggest a biological relevance even if differences are not consistently observed across all research settings and populations (Khadir et al., 2020; Zhang et al., 2021).
Part of this inconsistency can be explained by differences in population structures, sample sizes, assay kits, and comorbidities such as metabolic syndrome or diabetes. Moreover, sampling timing (fasting vs. postprandial) and circadian rhythm may shift values across individuals and groups. Reports in special contexts such as pregnancy have also documented inconsistent patterns, underscoring the need for tighter control of confounding variables (Behrasi et al., 2024; Behrooz et al., 2020)..

The results of this study partially align with works that found no clear differences in Spexin across obesity categories, while conflicting with other evidence that clearly linked reduced Spexin to obesity. Consequently, Spexin remains an inconsistent marker in the literature, with its value shaped by contextual and methodological determinants. Scientifically, its greater utility may lie in incorporation within multi-marker models alongside anthropometric measures and inflammatory/metabolic biomarkers, rather than as a standalone marker. This conclusion highlights the need for longitudinal and provocative studies (meal tests or exercise protocols), with standardized assay kits and adjustment for sex, age, and circadian rhythm, to clarify whether Spexin can be included in predictive models or serve as a complementary biomarker alongside established indicators (Khadir et al., 2020; Behrooz et al., 2020).

Copeptin is linked to stress-response functions, fluid balance regulation, and the cardiometabolic pathway. In this study, Copeptin values were comparable among healthy, overweight, and obese groups, with no significant differences observed in overall analyses or pairwise comparisons. The identical letters assigned by Duncan's test further supported the absence of meaningful variation. This pattern indicates that, within this sample, Copeptin does not possess discriminative ability for obesity status (Mu et al., 2022).

This finding is consistent with reports that did not identify a clear association between Copeptin and fat

accumulation indices, attributing this to the fact that Copeptin is more strongly influenced by acute physiological stress, hydration status, and renal function than by fat mass itself. Other studies have also suggested that changes in Copeptin are not consistently linked to obesity per se, but rather emerge more clearly in the presence of metabolic syndrome, diabetes, or cardiometabolic complications (Rojas-Humpire et al., 2022; Shu et al., 2025).

CONCLUSION

This study demonstrated that serum spexin concentrations were not significantly altered across healthy, overweight, and obese individuals and showed no meaningful correlation with obesity-related anthropometric or biochemical markers. While SPX has been proposed as a potential regulator of appetite and metabolism, our findings indicate limited diagnostic or predictive value for obesity in this population. Larger multicenter studies are needed to validate these results and to better elucidate the physiological role of SPX in obesity and metabolic disorders.

REFERENCES

1. Alhammad, R., Abu-Farha, M., Hammad, M. M., Thanaraj, T. A., Channanath, A., Alam-Eldin, N., Al-Sabah, R., Shaban, L., Alduraywish, A., Al-Mulla, F., Rahman, A., & Abubaker, J. (2022a). Increased LRG1 Levels in Overweight and Obese Adolescents and Its Association with Obesity Markers, Including Leptin, Chemerin, and High Sensitivity C-Reactive Protein. International Journal of Molecular Sciences, 23(15), 8564. https://doi.org/10.3390/IJMS2315856
2. Behrasi, F., Khayat, S., Karajibani, M., Fanaei, H., & Montazerifar, F. (2024). Association between spexin, nutritional status of pregnant women with excessive gestational weight gain, and newborn weight: A cross-sectional study. Clinical Nutrition Open Science, 55, 79–90. https://doi.org/10.1016/J.NUTOS.2024.03.005
3. Behrooz, M., Vaghef-Mehrabany, E., Maleki, V., Pourmoradian, S., Fathifar, Z., & Ostadrahimi, A. (2020a). Spexin status in relation to obesity and its related comorbidities: a systematic review. Journal of Diabetes and Metabolic Disorders, 19(2), 1943. https://doi.org/10.1007/S40200-020-00636-8
4. Behrooz, M., Vaghef-Mehrabany, E., Maleki, V., Pourmoradian, S., Fathifar, Z., & Ostadrahimi, A. (2020b). Spexin status in relation to obesity and its related comorbidities: a systematic review. Journal of Diabetes and Metabolic Disorders, 19(2), 1943. https://doi.org/10.1007/S40200-020-00636-8
5. Bozadjieva, N., Heppner, K. M., & Seeley, R. J. (2018). Targeting FXR and FGF19 to Treat Metabolic Diseases—Lessons Learned From Bariatric Surgery. Diabetes, 67(9), 1720. https://doi.org/10.2337/DBI17-0007
6. Camilli, C., Hoeh, A. E., De Rossi, G., Moss, S. E., & Greenwood, J. (2022). LRG1: an emerging player in disease pathogenesis. Journal of Biomedical Science, 29(1), 6. https://doi.org/10.1186/S12929-022-00790-6
7. Cancela-Carral, J. M., Bezerra, P., Lopez-Rodriguez, A., & Silva, B. (2023). Degree of association between the body mass index (BMI), waist-hip ratio (WHR), waist-height ratio (WHtR), body adiposity index (BAI) and conicity index (CI) in physically active older adults. Clinical Nutrition ESPEN, 58, 335–341. https://doi.org/10.1016/J.CLNESP.2023.10.007
8. Cao, X., Wang, N., Yang, M., & Zhang, C. (2025). Lipid Accumulation and Insulin Resistance: Bridging Metabolic Dysfunction-Associated Fatty Liver Disease and Chronic Kidney Disease. International Journal of Molecular Sciences 2025, Vol. 26, Page 6962, 26(14), 6962. https://doi.org/10.3390/IJMS26146962
9. Carbone, F., Després, J. P., Ioannidis, J. P. A., Neeland, I. J., Garruti, G., Busetto, L., Liberale, L., Ministrini, S., Vilahur, G., Schindler, T. H., Macedo, M. P., Di Ciaula, A., Krawczyk, M., Geier, A., Baffy, G., Faienza, M. F., Farella, I., Santoro, N., Frühbeck, G., … Montecucco, F. (2025). Bridging the gap in obesity research: A consensus statement from the European Society for Clinical Investigation. European Journal of Clinical Investigation, 55(8), e70059. https://doi.org/10.1111/ECI.70059
10. Carvalho, M. B. de, Jorge, G. M. C. P., Zanardo, L. W., Hamada, L. M., Izabel, L. dos S., Santoro, S., & Magdalon, J. (2024a). The role of FGF19 in metabolic regulation: insights from preclinical models to clinical trials. American Journal of Physiology - Endocrinology and Metabolism, 327(3), E279–E289. https://doi.org/10.1152/AJPENDO.00156.2024,
11. Carvalho, M. B. de, Jorge, G. M. C. P., Zanardo, L. W., Hamada, L. M., Izabel, L. dos S., Santoro, S., & Magdalon, J. (2024b). The role of FGF19 in metabolic regulation: insights from preclinical models to clinical trials. American Journal of Physiology - Endocrinology and Metabolism, 327(3), E279–E289. https://doi.org/10.1152/AJPENDO.00156.2024,
12. Carvalho, M. B. de, Jorge, G. M. C. P., Zanardo, L. W., Hamada, L. M., Izabel, L. dos S., Santoro, S., & Magdalon, J. (2024c). The role of FGF19 in metabolic regulation: insights from preclinical models to clinical trials. American Journal of Physiology - Endocrinology and Metabolism, 327(3), E279–E289. https://doi.org/10.1152/AJPENDO.00156.2024/ASSET/IMAGES/LARGE/AJPENDO.00156.2024_F001.JPEG
13. Chinedu, S. N., Ogunlana, O. O., Azuh, D. E., Iweala, E. E. J., Afolabi, I. S., Uhuegbu, C. C., Idachaba, M. E., & Osamor, V. C. (2013).

Correlation Between Body Mass Index and Waist Circumference in Nigerian Adults: Implication as Indicators of Health Status. Journal of Public Health Research, 2(2), e16. https://doi.org/10.4081/JPHR.2013.E16

14. Choi, C. H. J., Barr, W., Zaman, S., Model, C., Park, A., Koenen, M., Lin, Z., Szwed, S. K., Marchildon, F., Crane, A., Carroll, T. S., Molina, H., & Cohen, P. (2022). LRG1 is an adipokine that promotes insulin sensitivity and suppresses inflammation. ELife, 11. https://doi.org/10.7554/ELIFE.81559,
15. Dritsoula, A., Camilli, C., Moss, S. E., & Greenwood, J. (2024). The disruptive role of LRG1 on the vasculature and perivascular microenvironment. Frontiers in Cardiovascular Medicine, 11, 1386177. https://doi.org/10.3389/FCVM.2024.1386177
16. Guo, J.-Y. ;, Chen, H.-H. ;, Lee, W.-J. ;, Chen, S.-C. ;, Lee, S.-D. ;, Chen, C.-Y., Guo, J.-Y., Chen, H.-H., Lee, W.-J., Chen, S.-C., Lee, S.-D., & Chen, C.-Y. (2022). Fibroblast Growth Factor 19 and Fibroblast Growth Factor 21 Regulation in Obese Diabetics, and Non-Alcoholic Fatty Liver Disease after Gastric Bypass. Nutrients 2022, Vol. 14, Page 645, 14(3), 645. https://doi.org/10.3390/NU14030645
17. He, S., Ryu, J., Liu, J., Luo, H., Lv, Y., Langlais, P. R., Wen, J., Dong, F., Sun, Z., Xia, W., Lynch, J. L., Duggirala, R., Nicholson, B. J., Zang, M., Shi, Y., Zhang, F., Liu, F., Bai, J., & Dong, L. Q. (2021a). LRG1 is an adipokine that mediates obesity-induced hepatosteatosis and insulin resistance. The Journal of Clinical Investigation, 131(24), e148545. https://doi.org/10.1172/JCI148545
18. He, S., Ryu, J., Liu, J., Luo, H., Lv, Y., Langlais, P. R., Wen, J., Dong, F., Sun, Z., Xia, W., Lynch, J. L., Duggirala, R., Nicholson, B. J., Zang, M., Shi, Y., Zhang, F., Liu, F., Bai, J., & Dong, L. Q. (2021b). LRG1 is an adipokine that mediates obesity-induced hepatosteatosis and insulin resistance. The Journal of Clinical Investigation, 131(24), e148545. https://doi.org/10.1172/JCI148545
19. Hoeh, A. E., Chang, J. H., Mueller, R. S., Basche, M., Fantin, A., Sepetis, A., De Rossi, G., Dritsoula, A., Ali, R. R., Turowski, P., Moss, S. E., & Greenwood, J. (2025). LRG1 Alters Pericyte Phenotype and Compromises Vascular Maturation. Cells, 14(8), 593. https://doi.org/10.3390/CELLS14080593/S1
20. Janssen, I., Katzmarzyk, P. T., & Ross, R. (2004). Waist circumference and not body mass index explains obesity-related health risk. The American Journal of Clinical Nutrition, 79(3), 379–384. https://doi.org/10.1093/AJCN/79.3.379
21. Shakir, B. K., Al-Salman, I. M., & Al-Attabi, M. S. (2024). Bioremediation of pesticide Glyphosate (Ground-UP SL) and Remove Cd, Cr elements from polluted aquatic media by using Chlorococcum humicola, Chlorella vulgaris algae. Al-Turath Journal for Biology, 1(01), 8-19.
22. Lan, T., Morgan, D. A., Rahmouni, K., Sonoda, J., Fu, X., Burgess, S. C., Holland, W. L., Kliewer, S. A., & Mangelsdorf, D. J. (2017). FGF19, FGF21, and an FGFR1/β-Klotho-Activating Antibody Act on the Nervous System to Regulate Body Weight and Glycemia. Cell Metabolism, 26(5), 709-718.e3. https://doi.org/10.1016/J.CMET.2017.09.005
23. Lee, S., Kim, Y., & Han, M. (2022). Influence of Waist Circumference Measurement Site on Visceral Fat and Metabolic Risk in Youth. Journal of Obesity & Metabolic Syndrome, 31(4), 296. https://doi.org/10.7570/JOMES22046
24. Li, X., Lu, W., Kharitonenkov, A., & Luo, Y. (2024). Targeting the FGF19–FGFR4 pathway for cholestatic, metabolic, and cancerous diseases. Journal of Internal Medicine, 295(3), 292–312. https://doi.org/10.1111/JOIM.13767,
25. Lin, C. Y., Huang, T., Zhao, L., Zhong, L. L. D., Lam, W. C., Fan, B. M., & Bian, Z. X. (2018). Circulating Spexin Levels Negatively Correlate With Age, BMI, Fasting Glucose, and Triglycerides in Healthy Adult Women. Journal of the Endocrine Society, 2(5), 409. https://doi.org/10.1210/JS.2018-00020
26. Liu, A., Huang, M., Xi, Y., Deng, X., & Xu, K. (2025). Orchestration of Gut–Liver-Associated Transcription Factors in MAFLD: From Cross-Organ Interactions to Therapeutic Innovation. Biomedicines 2025, Vol. 13, Page 1422, 13(6), 1422. https://doi.org/10.3390/BIOMEDICINES13061422
27. Lukács, A., Horváth, E., Máté, Z., Szabó, A., Virág, K., Papp, M., Sándor, J., Ádány, R., & Paulik, E. (2019). Abdominal obesity increases metabolic risk factors in non-obese adults: A Hungarian cross-sectional study. BMC Public Health, 19(1), 1–8. https://doi.org/10.1186/S12889-019-7839-1/TABLES/3
28. Mertlitz, S., Riesner, K., Kalupa, M., Uhlig, N., Cordes, S., Verlaat, L., Jamali, M., Li, N., Mohamed, H. M. E. R., Bullinger, L., Moss, S., Greenwood, J., Jatzlau, J., Knaus, P., Vallecillo-Garcia, P., & Penack, O. (2025). Leucine-rich α-2 glycoprotein 1 (LRG1) during inflammatory complications after allogeneic stem cell transplantation and CAR-T cell therapy. Journal for Immunotherapy of Cancer, 13(3), e009372. https://doi.org/10.1136/JITC-2024-009372
29. Mu, D., Cheng, J., Qiu, L., & Cheng, X. (2022). Copeptin as a Diagnostic and Prognostic Biomarker in Cardiovascular Diseases. Frontiers in Cardiovascular Medicine, 9, 901990. https://doi.org/10.3389/FCVM.2022.901990/XML
30. National Academies of Sciences, E. and M. H. and M. D. F. and N. B. R. on O. S., & Callahan, E. A. (2023). The Science, Strengths, and Limitations of Body Mass Index. Translating Knowledge of

Foundational Drivers of Obesity into Practice. https://doi.org/10.17226/26942

31. Pek, S. L. T., Cheng, A. K. S., Lin, M. X., Wong, M. S., Chan, E. Z. L., Moh, A. M. C., Sum, C. F., Lim, S. C., & Tavintharan, S. (2018). Association of circulating proinflammatory marker, leucine-rich-α2-glycoprotein (LRG1), following metabolic/bariatric surgery. Diabetes/Metabolism Research and Reviews, 34(7). https://doi.org/10.1002/DMRR.3029,
32. Rojas-Humpire, R., Soriano-Moreno, D. R., Galindo-Yllu, B., & Zafra-Tanaka, J. H. (2022). Association between Copeptin and Metabolic Syndrome: A Systematic Review. Journal of Nutrition and Metabolism, 2022, 5237903. https://doi.org/10.1155/2022/5237903
33. Ross, R., Neeland, I. J., Yamashita, S., Shai, I., Seidell, J., Magni, P., Santos, R. D., Arsenault, B., Cuevas, A., Hu, F. B., Griffin, B. A., Zambon, A., Barter, P., Fruchart, J. C., Eckel, R. H., Matsuzawa, Y., & Després, J. P. (2020a). Waist circumference as a vital sign in clinical practice: a Consensus Statement from the IAS and ICCR Working Group on Visceral Obesity. Nature Reviews. Endocrinology, 16(3), 177. https://doi.org/10.1038/S41574-019-0310-7
34. Shu, X., Cai, F., Li, W., & Shen, H. (2025). Copeptin as a diagnostic and prognostic biomarker in pediatric diseases. Clinical Chemistry and Laboratory Medicine, 63(3), 483–498. https://doi.org/10.1515/CCLM-2024-0839/ASSET/GRAPHIC/J_CCLM-2024-0839_FIG_001.JPG
35. Singh, P., Covassin, N., Marlatt, K., Gadde, K. M., & Heymsfield, S. B. (2022). Obesity, Body Composition, and Sex Hormones: Implications for Cardiovascular Risk. Comprehensive Physiology, 12(1), 2949–2993. https://doi.org/10.1002/J.2040-4603.2022.TB00200.X
36. Th, S., & Sh, I. (2024). IL_33 as a Modulator of Neuroinflammation in Alzheimer's disease: An ELISA Approach. Al-Turath Medical Journal, 1(1), 1-10.
37. Zhang, L., Li, G., She, Y., & Zhang, Z. (2021). Low levels of spexin and adiponectin may predict insulin resistance in patients with non-alcoholic fatty liver. Practical Laboratory Medicine, 24, e00207. https://doi.org/10.1016/J.PLABM.2021.E00207