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"Histological evaluation of the ovary and some vital variables in women with polycystic ovary syndrome"

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Abstract:

Polycystic ovary syndrome (PCOS) is a medical condition that affects the function of the ovaries and hormone production in women. It causes symptoms such as irregular menstruation, increased testosterone secretion, weight gain, excessive hair growth and insulin resistance. The causes are not completely known, but genetic, environmental and lifestyle factors play a role. Possible complications include increased risk of heart disease, diabetes, and high blood pressure. It is diagnosed through laboratory tests, such as examining hormones and white blood cells. Recently, studies are focusing on the level of nesfatin-1 protein in PCOS patients, and the results indicate that its levels differ between patients and healthy people, and it is considered a diagnostic tool that needs extensive study.

This study aims: Evaluating the histological changes in the ovaries of women with polycystic ovary syndrome and some vital variables.

Materials and working methods: 270 blood samples were collected, divided into two groups. The first group included 180 blood samples from women with polycystic ovary syndrome, and the second group included 90 blood samples from healthy women (control samples). The results between the two groups were carefully evaluated, including evaluating the levels of testosterone and follicle-stimulating hormone. As well as evaluating the level of nesfatin-1 and white blood cells between women with polycystic ovary syndrome and healthy women.

Results: Our results showed a relatively high level of nesfatin-1 in polycystic ovary syndrome patients compared to healthy controls. The results showed statistically significant differences at the P≤0.05 level

The levels of testosterone, follicle-stimulating hormone, luteinizing hormone, and progesterone also showed an increase in affected patients, and it also showed that there was a significant difference at the p level ≥0.05.

While the levels of estrogen (E2) and prolactin showed no significant differences at the level of P>0.05.

Women with this syndrome had relatively high white blood cell counts compared to unaffected women.

Conclusions: Our results indicate a decreased level of nesfatin-1, as nesfatin-1 is a metabolic regulatory protein. The results also showed an increase in both follicle-stimulating hormone and testosterone and a slight increase in white blood cells, which leads to an imbalance in the regulation of hormones and the menstrual cycle and a threat to the reproductive process.

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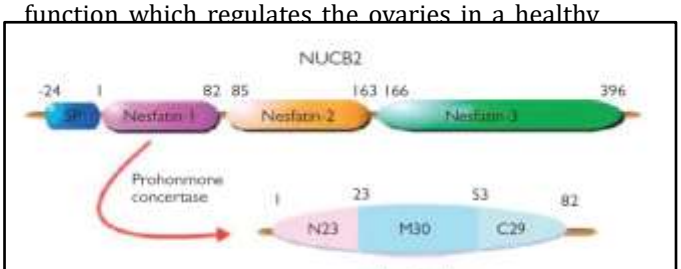
Key words:

Polycystic ovary syndrome (PCOS), nesfatin-1, total testosterone, follicle-stimulating hormone (FSH), luteinizing hormone(LH) , estrogen (E2) , prolactin.

Introduction

Polycystic ovary syndrome (PCOS) refers to several endocrine and metabolic disorders that occur in adult women of reproductive age [1]. Although the exact cause of polycystic ovary disease remains unknown, genetic and hormonal factors are believed to play an important role in its appearance [2]. Polycystic ovary syndrome contributes to disruption of women's health and vitality, as it can cause the risk of menstrual problems, type 2 diabetes, other hormonal disorders, and blood pressure, in addition to visible symptoms such as scalp hair loss, body hair growth, and acne[3]. Polycystic ovary syndrome is associated with weight loss problems and insulin resistance. Accurate diagnosis of PCOS depends on radio ultrasound and the characteristic clinical symptoms of menstrual irregularity, hypometabolism, or hyperandrogenism, and what is diagnosed in the reproductive stages of life in women. Patients with PCOS suffer from infertility or symptoms of excess androgens such as testosterone, follicle-stimulating hormone (FSH), and white blood cells[4] This syndrome affects the level of Nesfatin-1 appears for the first time in the form of a peptide consisting of 82 amino acids. Nesfatin-1 is considered a type of protein nucleobindin 2 (NUCB2), which is secreted from the nucleus of the hypothalamus. The number of amino acids in this protein is 396 due to proteolytic processes. The NUCB2 hormone is divided into three parts. Nesfatin-1 consists of an N-terminal part, while Nesfatin-2 and Nesfatin-3 consist of a C-terminal part of the three peptides that make up the NUB2 hormone only. It has been shown that Nesfatin-1 has an effect on regulating appetite and food intake [5]. But the function of nesfatin-2 and nesfatin-3 is not yet known. Nesfatin-1 consists of three parts [5]. The first section, known as N23, begins at the N-terminal end and continues with the 23rd amino acid[6]. The second section known as M30 consists of amino acids 23 to 53. The third section designated C29 includes amino acids 53-82 which is located near the carboxy terminus of M30 and is said to be useful in controlling appetite and food consumption [6]. Figure 1 shows the composition of nesfatin-1 and the structure of the NUB2 protein [5].Since its identification in 2006, the nesfatin-1 protein has been investigated for possible roles in appetite regulation, energy balance, and reproductive function, among other physiological functions. For women of childbearing age, PCOS in hormones is characterized by ovarian cysts and menstrual cycles. Irregular, symptoms include weight gain and hirsutism. (Excessive hair growth) [3]. Although the exact origin of PCOS is unknown, a combination of environmental and genetic factors are thought to play a role. Women with PCOS often exhibit insulin resistance and hormonal imbalances, including high levels of androgens, or male hormones. The potential role of Nesfatin-1 in PCOS has been studied, particularly in light of its association with insulin resistance and metabolic disorders. Nesfatin-1 may be involved in the control of insulin production and glucose metabolism which are often abnormal in individuals with PCOS according to certain research [7]. Furthermore, nesfatin-1 levels have been found to differ between PCOS patients and healthy subjects, although results have been mixed. In addition, studies on the presence of nesfatin-1 in pancreatic B cells found that it increased insulin secretion by activating the release of calcium ions from L-type channels in the cell[8]. Satiety peptides secreted effect on gonadal function which regulates the ovaries in a healthy



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from the Hypothalamus which have been shown to have beneficial effects on energy balance, obesity and glucose metabolism are also known to have an

Figure 1: Structure of nesfatin-1 and structure of the NUCB2 protein.

We studied some hormonal markers, such as Total testosterone, as Total testosterone is released in the adrenal cortex. Patients diagnosed with PCOS have elevated levels of testosterone (LH levels rise as a result, and eggs produce more testosterone as a result) [10] [11] and follicle-stimulating hormone (FSH) PCOS patients have insufficient FSH secretion, prolonged GnRH pulses, and an increase in LH can all lead to ovulation failure, leading to increased estrogen production which has a significant impact on PCOS Ovarian cyst [12]. We also studied white blood cells One of the main characteristics of PCOS with or without obesity is subclinical inflammation, which is characterized by increased levels of C-reactive protein (CRP) and interleukin-6 (IL-6) white blood cells (WBCs) [13].

Methods

This current study was conducted on women of reproductive age with polycystic ovary syndrome, at Samarra General Hospital in the city of Samarra, Salah al-Din Governorate, and they were diagnosed according to the Rotterdam criteria. In this study, some women had a clinical indicator of insulin resistance, and samples were collected from 10/12/2023 until 4/23/2024, when 270 blood samples were collected from women, representing two groups. The first group obtained consisted of 180 blood samples from infected women, in addition to the information contained in the questionnaire. As for polycystic ovary syndrome and the second group, 90 samples from healthy women (control samples) were measured, and the level of the nesfatin-1 protein associated with insulin resistance and metabolism, testosterone, follicle-stimulating hormone, luteinizing hormone, estrogen, prolactin hormone, and blood cells were measured. White blood.

Collect blood samples

A venous blood sample of 5 ml was drawn from each patient under study. Medical syringes with capacity of 5 ml were used, then blood was drawn and the procedure was performed under the necessary sterile conditions.

Then part of the blood is placed in a tube of EDTA and the other part is placed in a tube containing gel and left at room temperature for 15 minutes in order for the blood to be separated by a centrifuge at a speed of 6000 rpm for 5 minutes in order to obtain serum Then it is withdrawn using a fine pipette and placed in 2 ml Eppendorf tubes, and the sample number, patient name and group number are written on them using an adhesive tape to be stuck on them. It is kept in the refrigerator at -20 °C until testing.

Histological sections

Samples of size $1m^3$ were taken from the ovary that was removed during the surgical operations performed at Samarra Hospital in Salah al-Din Governorate, and 10% formalin was added to the removed organ until the tissue sections were prepared, which were prepared according to the method of Mancroft and Stephen, and the necessary steps for microscopic preparation were examined. Tissue sections were taken using a

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compound light microscope using a 40X lens, and their results were read and interpreted.

Nesfatin-1 concentration

The concentration of nesfatin-1 was estimated using a kit produced by the Chinese company Sun Long using ELISA technology and according to the company's steps and working principle.

Estimation of hormone concentration

The concentration of total testosterone, FSH, luteinizing hormone, estrogen and prolactin was estimated using a kit produced by the Chinese company Monobind Inc using ELISA technology and according to the company's steps and method.

WBC concentration

The white blood cell count was measured using a CBC device from the company SYSMEX-Japan

Statistical Analysis

The current results were analyzed using a t-test at a significance level of P≤ 0.05 using SPSS V20 [14].

Results and discussion

Parameters	Mean ±SD	
	Women control	Patients women
FSH (mU/mL)	7.17±2.68	14.60±4.34*
Testosterone (mIU/mL)	0.466±0.160	1.526±0.506*
LH (mIU/mL)	0.466±0.160	1.526±0.506*
Estrogen (mIU/mL)	90.7±31.2	101.4±34.0 n.s
prolactin (mIU/mL)	10.32±3.37	11.75±4.16 n.s

Table:1 evolution of sex hormones

The asterisk (*) indicates a significant difference at the 0.05 level

n.s (non-significant) represents no significant difference •

The current study showed a significant decrease in the level of nesfatin-1 in women with polycystic ovary syndrome, which reached (75.87±8.70). Compared to the level of nesfatin-1 in healthy women, which was (173.2±85.8) at a significance level of 0.05 As shown in table (1). As for hormonal indicators, the statistical results showed that there were statistically significant differences in the levels of both testosterone and follicle-stimulating hormone (FSH), as shown in Table (2), and the level of white blood cells (WBC) also showed a relative increase in patients with polycystic ovary syndrome compared to For healthy women, as shown in table (3)

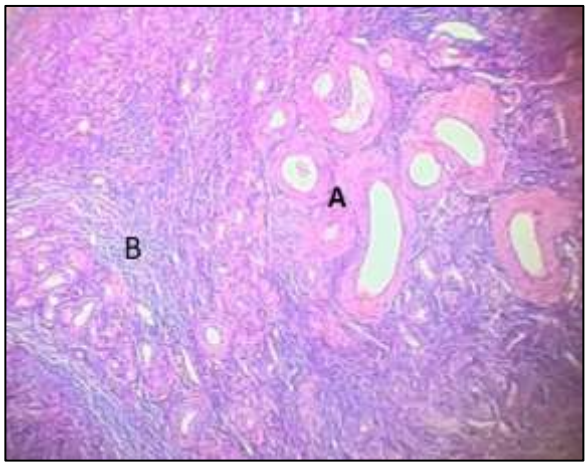
Parameters	Mean ±SD	
	Women control	Patients women
Nesfatin-1 (pg/mL)	69.8±16.0	144.4±39.2*

Table:2 Nesfatin-1 levels in serum of patients and control groups

The asterisk (*) indicates a significant difference at the 0.05 level .

Histological sections

T Tissue sections were taken from polycystic ovary syndrome patients, and the results were as follows: CT images (2-1).



(A) A section of an ovary affected by polycystic ovary syndrome: the ovarian cysts appear surrounded by

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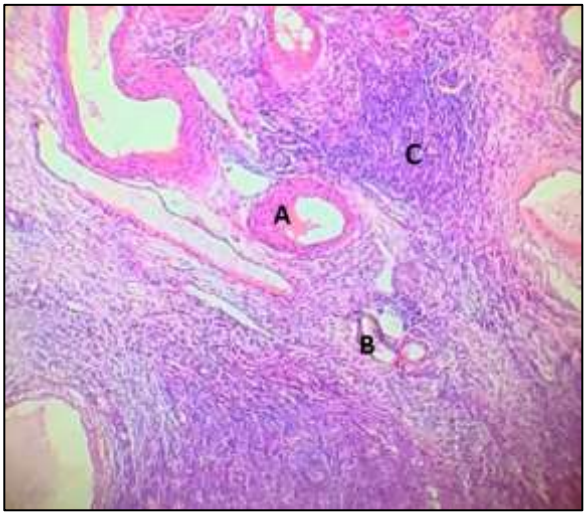
Parameters	Mean ±SD	
	Women control	Patients women
White blood cells (10^9/L)	8.04±2.16	9.20±2.58*

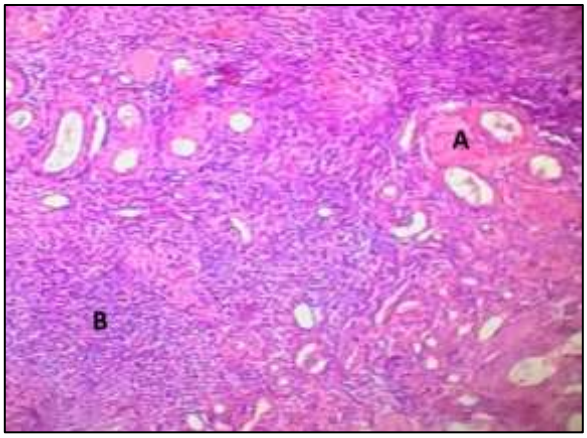
Table:3 White blood cells levels in serum of patients and control groups.

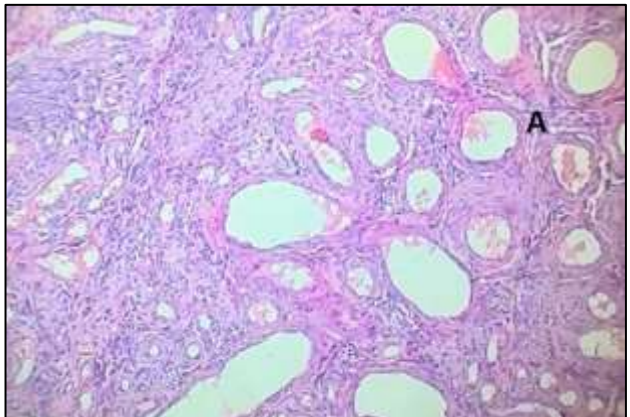
The asterisk (*) indicates a significant difference at the 0.05 level

(B)A section of an ovary affected by polycystic ovary syndrome: the ovarian cysts appear surrounded by bundles of dense colloidal fibroids extending to the extent of the ovarian cortex (A), infiltration of massive numbers of white blood cells (B), stained with hematoxylin & eosin, x40 magnification.



(C) A section of an ovary affected by polycystic ovary syndrome: ovarian cysts appear in different sizes, ranging from large to medium, containing follicular fluid and surrounded by bundles of dense colloidal fibrils (A), tortuous blood vessels devoid of blood and surrounded by fibrils, infiltration of massive numbers of white blood cells (C) Stained with hematoxylin & eosin at magnification x40.





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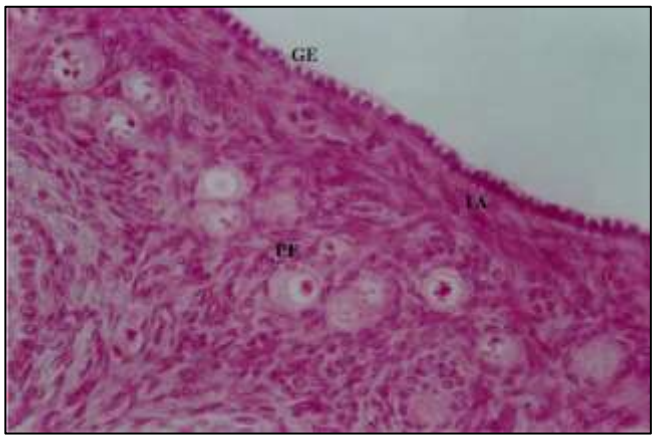
(D) A section of an ovary affected by polycystic ovary syndrome: Ovarian cysts appear in different sizes, ranging from large to medium, and some of them contain follicular fluid and are surrounded by bundles of dense colloidal fibroids. The cysts appear in the form of a group or cluster within the ovarian cortex area (A), stained with hematoxylin & eosin. X40 zoom power.



(F) Structure of the ovary The cortical region is surrounded by a simple epithelium classically named germinative epithelium (GE). Underlying the ovarian surface epithelium is a connective tissue layer, the tunica albuginea (TA). Groups of primordial follicles (PF) are present in the ovarian stroma. Col HE stain x40 [15].

DISCUSSION

In our study, we showed that nesfatin-1 levels were higher in patients with polycystic ovary syndrome compared to healthy subjects. Nesfatin-1 is also closely associated with BMI and insulin resistance. The reason for this increase may be related to hormonal changes and metabolism, as it is a peptide that has a role in regulating appetite and weight. Nesfatin-1 is often associated with insulin resistance, which can lead to an increase in its level as part of the body's response to enhance glucose utilization and energy regulation. Our results are also consistent with a study. Ademoglu and others, who pointed out the high level of nesfatin-1 in blood serum, explained that high nesfatin levels may play an important role in polycystic ovary syndrome [16].

The appearance of acne and hirsutism is the result of an increase in the level of testosterone in our results, which is consistent with the current results and the results of (Abdel Azim et al.) showed an increase in total and free levels. Testosterone in patients with polycystic ovary syndrome [17]. Our study showed that the level of FSH increased relatively in patients with polycystic ovary syndrome, in contrast to what was reported by Shahid and others who said that the level of FSH decreased in patients with polycystic ovary syndrome. Syndrome. Compared to those who do not suffer from this syndrome. [18] Our study showed that there are no significant differences in estrogen and prolactin. Our results are consistent with the study of (Li,A et al) who indicated that there are no significant differences in the level of estrogen. Also, our results are not consistent with what was reported by (Yang et al.,) as they indicated that there was a significant decrease in Prolactin hormone level in patients with polycystic ovary syndrome compared to healthy women [19][20]

Our results also showed an increase in the level of white blood cells in patients with polycystic ovary syndrome. These results contradict what was reported by the scientist (Al-Khobardi NA et al.), where the levels of blood components were measured in patients with polycystic ovary

syndrome, and there were no statistically significant differences, which calls for conducting an intensive study. conducted on this. The issue is acute[21]. The results of histological sections showed infiltration of white blood cells, and this is considered evidence of an inflammatory response, that is, the presence of an immune reaction from the organ. As for fibrosis, it is the irregular accumulation of fibers around the cysts after losing their functional effectiveness. Our results also agreed with those of the scientist (Parte et al) and others, whose histological sections showed the inflammatory effect of white blood cells, as well as the presence of necrosis and damage in ovarian tissue in mice. The results also indicated the presence of numerous follicular cysts in the ovaries and increased levels of testosterone in the serum of mice with polycystic ovary syndrome [22].

Recommendations

Women must pay attention to their dietary pattern to avoid infection with polycystic egg syndrome, They must also undergo periodic hormonal examinations to help in early diagnosis of the disease and early examination of the female reproductive system if the infection is diagnosed. Exercise is also considered an important feature to avoid infection with this syndrome.

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