УДК 618.177-06:616.43/.45:578.834:616.988.7
DOI: 10.35278/2664-0767.1(53).2024.315216

NEURO-ENDOCRINE ASPECTS OF INFERTILITY IN WOMEN WITH LONG-COVID-19

BOICHUK O.G.[1], GOLOVCHAK I.S.[1], KOLOMIICHENKO T.V.[2]
[1]Ivano-Frankivsk National Medical University, Ukraine,
[2]Shupyk National University of Health Care of Ukraine, Kyiv

Currently, the exact mechanisms of the impact of COVID-19 on women's reproductive health remain poorly defined. ***Purpose*** *of the study is to assess the hormonal function of patients with infertility and long-term COVID-19.* ***Materials and methods.*** *The main group consisted of 80 women with infertility who had symptoms of "long-COVID", the comparison group - 40 patients without a history of COVID-19. Hormonal studies of follicle-stimulating hormone (FSH), luteinizing hormone (LH), prolactin, melatonin, estradiol (E2), progesterone (P), free testosterone (Tf), dehydroepiandrosterone (DHEA), cortisol, anti-Mьllerian hormone (AMH) were conducted.* ***Results.*** *Violation of gonadotropic regulation of reproductive function is manifested by an increase in the serum concentration of LH and FSH and an increase in their ratio. A tendency to hyperprolactinemia, a relative decrease in serum melatonin was revealed. A more than 2-fold increase in the E2/P ratio on the 22nd day of the menstrual cycle indicates a tendency to hyperestrogenemia. An increase in the level of serum androgens (Tf and DHEA), which also play a role in female reproduction, was detected. Elevated levels of the stress hormone cortisol can negatively affect the chances of getting pregnant in assisted reproductive technologies programs. A decrease in AMH was observed in patients with infertility and long-term COVID-19, indicating a decrease in their ovarian reserve.* ***Conclusion.*** *The identified neuro-endocrine changes in patients with infertility and long-term COVID-19, due to the direct and indirect effects of the transmitted disease and its consequences, may be a marker of reduced reproductive potential and unsuccessful attempts to use assisted reproductive technologies.*

Key words: *women's reproductive health, long-COVID-19, infertility, pituitary and pineal hormones, steroid hormones*

Currently, the exact mechanisms of the impact of COVID-19 on women's reproductive health remain poorly defined. It is assumed that the causative agent can directly affect the menstrual and reproductive functions of women by affecting the receptors of angiotensin-converting enzyme 2, which is widely expressed in the ovaries and uterus. Specific interactions between the reproductive system and SARS-CoV-2 infection are believed to occur at the ovarian/endometrial level [2].

SARS-CoV-2 infection can affect the "hypothalamus-pituitary-ovary-endometrium" axis, which leads to disorders of a woman's reproductive function. There is a reciprocal relationship between the hypothalamic-pituitary-adrenal axis, which ensures the formation of a response to stress, and the hypothalamic-pituitary-ovarian axis, in which the activation of one axis leads to the suppression of the other. Chronic stress suppresses the production of estrogens [5], so it is necessary to assess the potential impact of COVID-19 on the reproductive system.

The impact of the transferred COVID-19 on the reproductive system of a woman can be mediated by the toxic effects of the drugs used, the duration of hospitalization in the intensive care unit and intensive care unit, and the decompensation of concomitant chronic diseases. It is currently unknown what long-term consequences for women's reproductive health may be associated with the transfer of COVID-19. In this regard, all women who have experienced COVID-19, especially in a severe form, should be assigned to a group of high risk of developing complications and be subjected to more careful dispensary observation within 1 year after inpatient treatment for COVID-19. It is necessary to determine further management tactics: restoration of the menstrual cycle, treatment of infertility, operative treatment of gynecological pathology, selection of contraceptive methods, hormone replacement therapy and other specific therapy according to gynecological indications [7].

The severe stress caused by the new infection of COVID-19, and even the state of emergency during the pandemic, can have a significant impact on the reproductive system. There is a reciprocal relationship between the hypothalamic-pituitary-adrenal axis, which ensures the formation of the response to stress, and the hypothalamic-pituitary-ovarian axis, in which the activation of one axis leads to the suppression of the other. Chronic activation of reactions in response to stress suppresses the production of estrogens, which can cause disruption of the menstrual cycle and the appearance of anovulatory cycles [1].

Stress-related disorders of the menstrual cycle are a spectrum of disorders that includes secondary amenorrhea (absence of menstruation for 3 months or more, provided the previous regular menstrual cycle) and a more rare form - primary stressogenic amenorrhea [6].

In addition, there are gender differences in the regulation of the stress response, mainly due to the interaction between the hypothalamic-pituitary-adrenal and hypothalamic-pituitary-gonadal axes [4, 5, 8, 9, 12].

PURPOSE of the study is to evaluate the hormonal function of patients with infertility and long-term COVID-19.

MATERIALS AND RESEARCH METHODS

120 patients who applied to the clinic of reproductive technologies for infertility treatment were included in the comprehensive examination, and they were divided into 2 groups. The main group consisted of 80 women with infertility who showed signs of "long-COVID", the comparison group - 40 patients without a history of COVID-19.

Hormonal studies of follicle-stimulating hormone (FSH), luteinizing hormone (LH), prolactin, melatonin, estradiol (E_2), progesterone (P), free testosterone (T_f), dehydroepiandrosterone (DHEA), cortisol, anti-Mullerian hormone (AMH) were conducted.

The content of hormones in the blood serum was determined by the immunehistochemical method with ECLIA electrochemiluminescence detection and the enzyme immunoassay method (placental lactogen, melatonin). Blood was taken for the determination of hormones on the 3rd day of the menstrual cycle (MC), and for the determination of P and T_f on the 3rd and 22nd days of the MC.

The research was carried out in accordance with the main provisions of GCP ICH and the Declaration of Helsinki, agreed with the ethics committee of the Ivano-Frankivsk National Medical University and the Shupyk National University of Health of Ukraine, Kyiv. All studies were carried out after receiving the patient's informed consent for diagnosis and treatment.

The obtained data were processed by the methods of variational statistics accepted in medicine, using the Student's t-test for numerical indicators after checking for normality of distribution, with a critical significance level of $p<0.05$. The Microsoft Excel statistical analysis package was used.

RESEARCH RESULTS AND THEIR DISCUSSION

According to the data we received, in women with infertility and long-term COVID-19, a violation of the gonadotropic regulation of reproductive function was noted, which was manifested by an increase in the serum concentration of LH and FSH (Table 1), the imbalance of such an increase in these two indicators is evidenced by an increase in the ratio of LH/ FSH up to 1.58±0.07 versus 1.12±0.04 in women without a history of COVID-19 ($p < 0.05$). Such shifts may be a reflection of reduced ovarian reserve.

The increased level of prolactin found in patients after COVID-19 (16.41±0.42 vs. 4.95±0.21 ng/ml, respectively, $p < 0.05$) may be a consequence of the stressful impact of the disease and, in turn, negatively affect female fertility, because hyperprolactinemia suppresses the cyclic activity of the hypothalamic-pituitary-gonadal HPG axis and is considered one of the causes of infertility in men and women [13].

Table 1. Hormones of the pituitary gland and pineal gland on the 3rd day of the cycle in patients with infertility depending on the transferred COVID-19

Indicator	After COVID-19, n=80	No history of COVID-19, n=40
LH, IU/L	8.79±0.42*	4.95±0.21
FSH, IU/L	5.52±0.33	4.42±0.45
LH / FSH	1.58±0.07*	1.12±0.04
Prolactin, ng/ml	16.41±0.42*	11.73±0.31
Melatonin, ng/ml	16.50±0.26*	27.32±0.63

Note. * - the difference in the indicator relative to the group of women without a history of COVID-19 is statistically significant ($p < 0.05$)

Recently, melatonin has been considered not only in the regulation of sleep/wake processes in the body, but also in connection with a number of pathological mechanisms of various diseases, in particular, a large number of studies have demonstrated its immunomodulating, antioxidant, anti-inflammatory and antiviral properties, in particular in relation to SARS-Cov-2 , effects [10]. Numerous studies have revealed the connection between melatonin and female reproductive function [3], in particular, a positive correlation between follicular melatonin levels and markers of ovarian reserve, anti-Мьllerian hormone and initial FSH level has been established [11]. The same authors also found a similar correlation between follicular fluid melatonin levels and in vitro fertilization (IVF) outcomes and oocyte quality. Similarly, [14] found a significant positive correlation between the concentration of melatonin in follicular fluid and the number of antral follicles in women undergoing IVF, which is also consistent with a supportive or protective effect of melatonin on the progression of the ovarian cycle. Therefore, the relative decrease in the serum hormone we found (16.50±0.26 relative to 27.32±0.63 pg/ml) may be, on the one hand, associated with long-term COVID-19, and on the other hand, be a marker of reduced reproductive potential and unsuccessful attempts at IVF.

All stages of cyclical changes in the secretion of steroid hormones in infertile patients after infertile patients took place against the background of an increased level of estrogen and a reduced concentration of progesterone in both the follicular (on the 3rd MC) and the luteal (on the 22nd day) phase of the cycle (Table 2). A more than 2-fold increase in the E2/P ratio on the 22nd day of MC indicates the predominance of estrogen and a tendency to hyperestrogenemia, which was confirmed by colpocytology ("hormonal mirror"). According to the results of colpocytology, an anovulatory cycle of the hyperestrogenic type with luteal phase insufficiency was established in 45 (56.3%) women of the long-COVID-19 group against 14 (35.0%) patients without a history of COVID-19.

Table 2. Steroid hormones and AMH in patients with infertility depending on the experienced COVID-19

Indicator	After COVID-19, n=80	No history of COV-ID-19, n=40
E_2 on the 3rd day of the MC, pg/ml	58.25±2.14	39.47±1.85
P on the 3rd day of the MC, ng/ml	0.98±0.08	1.24±0.10
E_2 on the 22nd day of the MC, pg/ml	86.35±3.12	48.6±2.73
P on the 22nd day of the MC, ng/ml	6.91±0.49	10.37±0.76
E_2/P on the 22nd day of the MC	12.36±0.54	5.34±0.31
T_f, pg/ml	1.73±0.04	1.42±0.06
DHEA, μg/dL	78.85±3.20	65.81±2.30
Cortisol, μg/dL	8.42±0.15	7.35±0.14
AMH, ng/ml	2.16±0.18	3.04±0.16

Note. the difference of all indicators relative to the group of women without a history of COVID-19 is statistically significant ($p < 0.05$)

Some increase in the level of serum androgens (T_f and DHEA), which also play a role in female reproduction, was detected.

The elevated level of the stress hormone cortisol in the examined patients (8.42±0.15 vs. 7.35±0.14 μg/dL, $p < 0.05$), which may be associated with long-term COVID-19, negatively affects the chances of receiving pregnancy in ART programs, because it is known that high levels of cortisol and prolactin negatively affect the secretion of estradiol during ovulation and contribute to the weakening of the ovulatory surge of LH. High cortisol concentrations lead to insensitivity of the pituitary gland to gonadotropin-releasing hormone and the ovaries to luteinizing hormone (LH). The consequence of this is the restriction of the release of LH, follitropin (FSH) and estrogens; thus, when the follicular phase lengthens, the menstrual cycle becomes irregular and long, and the probability of ovulation decreases [13].

A decrease in AMH was observed in patients with infertility and long-term COVID-19 (2.16±0.18 vs. 3.04±0.16, respectively, $p < 0.05$), which indicates a decrease in their ovarian reserve.

So, neuro-endocrine changes were detected in patients with infertility and long-term COVID-19, which can be caused by the direct and indirect effects of the transferred disease and its consequences.

Violation of gonadotropic regulation of reproductive function is manifested by an increase in the serum concentration of LH and FSH, the imbalance of such an increase in these two indicators is evidenced by an increase in their ratio, which may be a reflection of a reduced ovarian reserve. A tendency to hyperprolactinemia was revealed, which is considered one of the factors of infertility.

The detected relative decrease in serum melatonin can be, on the one hand, associated with long-term COVID-19, and on the other hand, be a marker of reduced reproductive potential and unsuccessful attempts at IVF.

A more than 2-fold increase in the E2/P ratio on the 22nd day of MC indicates the predominance of estrogen and a tendency to hyperestrogenemia. Some increase in the level of serum androgens (T_f and DHEA), which also play a role in female reproduction, was detected. Elevated levels of the stress hormone cortisol in examined patients can negatively affect the chances of getting pregnant in ART programs.

A decrease in AMH was observed in patients with infertility and long-term COVID-19, indicating a decrease in their ovarian reserve.

REFERENCES/ЛІТЕРАТУРА

1. Fourman LT, Fazeli PK. Neuroendocrine causes of amenorrhea — an update. The Journal of Clinical Endocrinology and Metabolism. 2015;100(3):812-824. https://doi.org/10.1210/jc.2014-3344 28.
2. Li R, Yin T, Fang F, Li Q, et al. Potential risks of SARS-CoV-2 infection on reproductive health. Reproductive Biomedicine Online. 2020;41(1):89-95. https://doi.org/10.1016/j.rbmo.2020.04.018. 27
3. Olcese JM. Melatonin and Female Reproduction: An Expanding Universe. Front Endocrinol (Lausanne). 2020 Mar 6;11:85. doi: 10.3389/fendo.2020.00085. PMID: 32210911; PMCID: PMC7067698.
4. Onyango MA, Regan A. Sexual and gender-based violence during COVID-19: lessons from Ebola. The Conversation. 2020. https://theconversation.com/sexual-and-gender-based-violenceduring-covid-19-lessons-from-ebola-137541 36.
5. Oyola MG, Handa RJ. Hypothalamic-pituitary- adrenal and hypothalamic-pituitary-gonadal axes: sex differences in regulation of stress responsivity. Stress-the International Journal on the Biology of Stress. 2017;20(5):476-494. https://doi.org/10.1080/10253890.2017.1369523 33
6. Palm-Fischbacher S, Ehlert J. Dispositional resilience as a moderator of the relations- hip between chronic stress and irregular menstrual cycle. Journal of Psychosomatic Obstetrics and Gynaecology. 2014;35(2):42-50. https://doi.org/10.3109/0167482X.2014.912209 29.
7. Phelan N, Behan LA, Owens L. The Impact of the COVID-19 Pandemic on Women's Reproductive Health. Front Endocrinol (Lausanne). 2021 Mar 22;12:642755. doi: 10.3389/fendo.2021.642755. PMID: 33841334; PMCID: PMC8030584.
8. Schneider D, Harknett K, McLanahan S. Intimate partner violence in the great recession. Demography. 2016;53(2):471-505. https://doi.org/10.1007/s13524-016-0462-1 34.
9. Schumacher JA, Coffey SF, Norris FH, Tracy M, Clements K, Galea S. Intimate partner violence and Hurricane Katrina: predictors and associated mental health outcomes. Violence and Victims. 2010;25(5):588-603. https://doi.org/10.1891/0886-6708.25.5.588 35.
10. Souissi A, Dergaa I, Romdhani M, Ghram A, Irandoust K, Chamari K, et al. Can melatonin reduce the severity of post-COVID-19 syndrome?. EXCLI J.

[Internet]. 2023 Feb. 2 [cited 2024 Sep. 25];22:173-87. Available from: https://www.excli.de/excli/article/view/5864

11. Tong J, Sheng S, Sun Y, Li H, Li WP, Zhang C, Chen ZJ. Melatonin levels in follicular fluid as markers for IVF outcomes and predicting ovarian reserve. Reproduction. 2017 Apr;153(4):443-451. doi: 10.1530/REP-16-0641. Epub 2017 Jan 6. PMID: 28062641. 53
12. Van Gelder N, Peterman A, Potts A, O'Donnell M, Thompson K, Shah N, Oertelt-Prigione S; Gender and COVID-19 working group. COVID-19: Reducing the risk of infection might increase the risk of intimate partner violence. The Lancet. E Clinical Medicine. 2020;21:100348. https://doi.org/10.1016/j.eclinm.2020.100348 38.
13. Wdowiak A, Raczkiewicz D, Janczyk P, Bojar I, Makara-Studzińska M, Wdowiak-Filip A. Interactions of Cortisol and Prolactin with Other Selected Menstrual Cycle Hormones Affecting the Chances of Conception in Infertile Women. Int J Environ Res Public Health. 2020 Oct 16;17(20):7537. doi: 10.3390/ijerph17207537. PMID: 33081268; PMCID: PMC7588978.
14. Zheng M, Tong J, Li WP, Chen ZJ, Zhang C. Melatonin concentration in follicular fluid is correlated with antral follicle count (AFC) and in vitro fertilization (IVF) outcomes in women undergoing assisted reproductive technology (ART) procedures. Gynecol Endocrinol. 2018 May;34(5):446-450. doi: 10.1080/09513590.2017.1409713. Epub 2017 Nov 29. PMID: 29185361

РЕЗЮМЕ

НЕЙРО-ЕНДОКРИННІ АСПЕКТИ НЕПЛІДДЯ У ЖІНОК З ЛОНГ-COVID-19

БОЙЧУК О. Г. [1], *ГОЛОВЧАК І. С.* [1], *КОЛОМІЙЧЕНКО Т.В.* [2]

[1]Івано-Франківський національний медичний університет, м. Івано-Франківськ,

[2]Національний університет охорони здоров'я України імені П.Л. Шупика, Київ

В даний час недостатньо визначеними лишаються точні механізми впливу COVID-19 на репродуктивне здоров'я жінок. **Мета дослідження** - оцінити гормональну функцію пацієнток з безпліддям та лонг-COVID-19. **Матеріали та методи.** Основну групу склали 80 жінок з безпліддям, у яких спостерігались ознаки «лонг-COVID», групу порівняння - 40 пацієнток без COVID-19 в анамнезі. Проводили гормональні дослідження фолікулостимулюючого гормону (ФСГ), лютеїнізуючого гормону (ЛГ), пролактину, мелатоніну, естрадіолу (E_2), прогестерону (П), тестостерону вільного (Тв), дегідроепіандростерону (ДГЕА), кортизолу, антимюлерового гормону (АМГ). **Результати.** Порушення гонадотропної регуляції репродуктивної функції проявляється зростанням рівня сироваткової концентрації ЛГ і ФСГ та підвищення їх співвідношення. Виявлена тенденція до гіперпролактинемії, відносне зниження сироваткового мелатоніну. Зростання більш ніж у 2 рази коефіцієнту E_2/П на 22-й день менструального циклу вказує на тенденцію до гіперестрогенемії. Виявлено підвищення рівня сироваткових андрогенів (Тв та ДГЕА), які також відіграють роль в жіночій репродукції. Підвищений рівень гормону стресу кортизолу може негативно позначитись на шансах отримати вагітність у програмах ДРТ. У пацієнток з безпліддям та лонг-COVID-19 спостерігалось зниження АМГ, що вказує на зменшення у них оваріального резерву. **Висновок.** Виявлені нейро-ендокринні зміни у пацієнток з непліддям та лонг-COVID-19, обумовлені прямим та непрямим впливом перенесеного захворювання та його наслідками, можуть бути маркером зниження репродуктивного потенціалу і невдалих спроб застосування допоміжних репродуктивних технологій.

Ключові слова: репродуктивне здоров'я жінок, лонг-COVID-19, безпліддя, гормони гіпофізу та епіфізу, стероїдні гормони.