Provisionally accepted for publication

ORIGINAL ARTICLE

Could elevated serum prolactin positively impact pregnancy outcomes in females undergoing IVF/ICSI?

Serum prolactin vs pregnancy outcomes

Sally **A. El-Attar** [1,*], Mona **M. Shaban** [1], Ahmed **E. Saleh** [2], Mazen **Abdel-Rasheed** [3], Mahmoud **Abdelhameed** [1], Safaa **I. Mahmoud** [1]

[1] Department of Obstetrics and Gynecology, Faculty of Medicine, Cairo University, Cairo, Egypt.

[2] Department of Obstetrics and Gynecology, El-Mokattam Insurance Hospital, Cairo, Egypt.

[3] Department of Reproductive Health and Family Planning, National Research Centre, Cairo, Egypt.

***Corresponding author:** Sally **A. El-Attar**, M.D. Department of Obstetrics and Gynecology, Faculty of Medicine, Cairo University, Al Kasr Al Aini, Cairo, Egypt. Email: dr_sally_elattar@yahoo.com

ORCID: 0009-0003-0257-4972

Doi: 10.36129/jog.2024.195

ABSTRACT

Objective. Optimal reproductive results may need a particular amount of circulating prolactin. Our aim in this study is to assess whether mildly elevated prolactin in females undergoing IVF/ICSI is more effective regarding pregnancy outcomes.

Materials and Methods. A prospective study was conducted on 222 infertile women undergoing IVF/ICSI using the long “gonadotrophin-releasing hormone” (GnRH) agonist protocol. Based on their basal prolactin level, 111 women with normal basal prolactin levels (< 25 ng/ml) were allocated to Group 1, and 111 women with basal prolactin levels of 25 to 50 ng/ml were allocated to Group 2. The characteristics of IVF/ICSI cycles and pregnancy outcomes cycles in both groups were recorded.

Results. As expected, the prolactin level at trigger day showed a significant difference between both groups (64.71 ± 27.74 *vs* 103.24 ± 29.95, p < 0.001). Although there was no significant difference between both groups regarding the number of oocytes retrieved (p = 0.473), the number of mature oocytes (p = 0.281), the number of total embryos (p = 0.224), and the number of transferred embryos (p = 0.420), the clinical and ongoing pregnancy rates significantly increased in group 2 with mildly elevated basal prolactin level (55.34% with p = 0.034 and 46.60% with p = 0.038, respectively)

Conclusions. Mildly elevated blood prolactin levels (25 to 50 ng/ml) were associated with higher clinical and ongoing pregnancy rates.

Key words

IVF/ICSI; clinical pregnancy rate; ongoing pregnancy rate; prolactin level.

INTRODUCTION

According to accumulating investigations, prolactin plays a pleiotropic function in reproduction, growth, metabolism, behavior, immune system, and carcinogenesis [1,2]. The lactotrophs in the anterior pituitary gland are responsible for prolactin hormone secretion. The hypothalamus regulates prolactin secretion via secreting both prolactin-inhibiting factors (PIF), which is predominantly exerted by dopamine, and prolactin-releasing factors (PRF) [3,4].

The major physiological response during the postpartum period is to prepare the breasts for breastfeeding. Prolactin regulates mammary gland growth and development, as well as milk production and secretion. Serum prolactin in non-pregnant women is typically less than 25 ng/L; a level higher than that limit is classified as hyperprolactinemia, provided that the venous sample is taken without undue stress. Hyperprolactinemia causes gonadotropin deficit, resulting in hypogonadotropic hypogonadism [5–7].

Dopamine agonists are frequently prescribed to inhibit prolactin secretion and restore ovulation in infertile women with hyperprolactinemia who want to get pregnant. However, follicle formation in women receiving in-vitro fertilization/intracytoplasmic sperm injection (IVF/ICSI) relies on exogenous gonadotropin intake rather than endogenous hormones. In addition, luteal phase support is mainly guaranteed by exogenous adequate progesterone intake [8,9].

Objectives

The study aims to assess the role of mildly elevated serum prolactin levels (below 50 ng/ml) in improving pregnancy outcomes.

MATERIALS AND METHODS

Study registration, ethical and methodological standards

This study involved 222 infertile women recruited for assisted reproductive technique (ART) at the IVF Unit of the Obstetrics and Gynecology Department, Kasr Al Aini Cairo University Hospital, Egypt, after being approved by the Research Ethics Committee. Before starting, all participants signed an informed written consent.

Patient and public involvement

The inclusion criteria were (a) women of reproductive age (20-45 years old), (b) undergoing ART either for primary or secondary infertility, (c) had either anovulation (except polycystic ovarian syndrome [PCOS] or hyperandrogenism causes), tubal factors or male factors, and (d) their basal prolactin level either normal or mildly elevated below 50 ng/ml. Women were excluded if they had uncontrolled medical disorders, previous pituitary lesions, abnormal

thyroid functions, anovulation due to PCOS or Hyperandrogenism, basal prolactin over 50 ng/ml, recurrent implantation failure, or any uterine abnormalities.

Data collection

Eligible patients had their hormonal profile on the second day of the menstrual cycle preceding induction. Serum prolactin level was measured two times. The first sample was the basal level of prolactin that was done with the hormonal profile on the second day of the menstrual cycle preceding induction and marked as (P0). The second sample was taken on the day of the trigger and marked as (P1). All samples were withdrawn at a specific morning time, after 8 hours of fasting, to avoid serum prolactin level fluctuation.

We used the long "gonadotrophin-releasing hormone" (GnRH) agonist protocol for all patients. GnRH agonist injections (Triptofem. 1mg/ml; Triptorelin Acetate, Marckyrl) were started on the 21st day of the cycle (mid-luteal). On the second day of the next menstrual cycle, patients started gonadotropin intake (Fostimon; UroFollitropin, IBSA) at a personalized dose modified depending on the patient's ovarian response. Folliculometry was obtained till the mature ovum (18:22 mm), and then an intramuscular dose of 10,000 IU of recombinant human chorionic gonadotropin (Choriomon; HCG, IBSA) was given to trigger the final oocyte.

Following that, oocyte retrieval was done using ultrasound-guided transvaginal aspiration 36 hours following the HCG trigger. Embryo growth was monitored daily until the cleavage-stage embryos were transferred fresh (Day 3). Intramuscular injection of 100 mg of (Prontogest; Progesterone, IBSA) was given daily for 16 days for luteal phase support. Pregnancy was tested in blood 11 days after embryo transfer. The pregnancy is considered positive clinically when there is positive β-hCG and an intrauterine gestational sac with a positive fetal pulse at six weeks of gestation.

The primary outcome was the clinical and ongoing pregnancy rates, while the secondary outcomes were the number of retrieved and mature oocytes, as well as embryo quantity and quality.

Sample Size:

Using "PS Power and Sample Size Calculations" software version 3.0.11 for MS Windows, sample size calculation was based on comparing the pregnancy rate between women undergoing IVF/ICSI cycles and have serum prolactin levels < 25 ng/ml and those with serum prolactin level from 25 – 50 ng/ml, using the Chi-square test with α-error level at 0.05, power at 80%. According to a previous investigation, the pregnancy rate among women with serum prolactin levels <30 ng/ml was 25%, while it was 42.7% in women with serum prolactin levels from 30-60 ng/ml. Accordingly, the minimum optimum sample size was 111 IVF/ICSI cycles in each group.

Statistical analysis

Data was analyzed with the "Statistical Package for Social Sciences" (SPSS) version 25 for MS Windows. Categorical variables were presented as frequency and percentages, while quantitative variables were presented as mean ± standard deviation with median and range. The Chi-square test tested the statistically significant differences between the two groups for

qualitative variables and the unpaired t-test/Mann-Whitney test for quantitative variables. A P-value < 0.05 was considered significant.

RESULTS

Following the CONSORT guidelines, we recruited 111 infertile women with basal prolactin levels below 25 ng/ml (assigned to Group 1) and 111 infertile women with basal prolactin levels of 25 to 50 ng/ml without giving dopaminergic agents (assigned to Group 2). After ovulation induction with a long GnRH agonist protocol to undergo an IVF/ICSI cycle, 12 women from group 1 and 8 women from group 2 had failure of fertilization **(Figure 1)**.

The patients' characteristics showed no significant difference between the two groups regarding age (P=0.125), BMI (P=0.366), infertility duration (P=0.130), and type of infertility (P=0.177). The pre-cycle laboratory investigations in both groups also revealed no significant difference regarding the serum level of these hormones: FSH (P=0.807), LH (P=0.268), basal E2 (P=0.888), AMH (P=0.146), and TSH (P=0.365). Only the basal prolactin level showed a significant difference between both groups, as expected (12.47 ± 5.19 vs. 30.35 ± 4.55, p-value <0.001), as it was the base of patient allocation in the two groups (**Table 1**).

The characteristics of IVF/ICSI cycles are shown in **Table 2.** There is no significant difference between the two groups regarding the gonadotropin dose used for induction (P=0.2) or the antral follicle count found during folliculometry (P=0.308). Also, the prolactin level at trigger day showed a significant difference between the two groups, as expected (64.71 ± 27.74 vs. 103.24 ± 29.95, p-value <0.001). Although there was no significant difference between the two groups regarding the number of oocytes retrieved (P=0.473), the number of mature oocytes (M2) (P=0.281), the number of total embryos (P=0.224), and the number of transferred embryos (P=0.420), we found that group 2 with mildly elevated basal prolactin level had significantly higher clinical pregnancy rate (55.34% vs. 40.4%, P=0.034) and significantly higher ongoing pregnancy rate (46.6% vs. 32.32%, P=0.038) compared to group 1 with normal basal prolactin level.

DISCUSSION

Main findings

Hyperprolactinemia has been recognized as a contributing factor to amenorrhea and infertility. However, optimal reproductive results may require a specific amount of serum prolactin. During the menstrual cycle, changes in prolactin levels were observed, indicating that prolactin plays a role in several processes of reproduction. During controlled ovarian stimulation (COS) for IVF/ICSI, a specific dynamic of serum prolactin levels, defined by temporary hyperprolactinemia, has a notable importance and influence on fertility outcome [1].

The current study aimed to assess the effect of increased basal prolactin level (between 25-50 ng/ml) on clinical and ongoing pregnancy rates. The prolactin level after HCG trigger was significantly higher in females with mildly elevated basal prolactin levels compared to females with normal basal prolactin levels. The incidence of clinical and ongoing pregnancy rates were significantly higher among females with mildly elevated basal prolactin levels when compared with females with normal basal prolactin levels.

Interpretation and comparison with other literature

Supporting our finding, in a study about fertility treatment outcomes, Wyse et al. (2021) found that prolactin was the only predictor for fertilization rate in a study of 22 normal-weight females and 22 overweight females, with an AUC of 0.76 (sensitivity of 0.77, specificity of 0.78) [10].

Zhang et al. (2020) conducted a large retrospective study on 3009 patients with basal prolactin levels <50 ng/mL undergoing IVF/ICSI cycles. They showed that baseline prolactin levels greater than 30 ng/mL were associated with a higher clinical pregnancy rate, which was consistent with our findings. They found that patients with favorable pregnancy outcomes had substantially higher prolactin levels at all measurement sites than those with negative results. They suggested that the favorable influence of prolactin on pregnancy outcomes may be attributable to the promotion of oogenesis and embryo development, as well as the enhancement of luteal support [9].

On the contrary, Trikoilis et al. (2022) studied 109 infertile nulliparous women undergoing their first cycle of IVF/ICSI therapy in order to assess the state of anxiety, measure the levels of stress biomarkers, such as prolactin, and determine how they related to the results of IVF/ICSI. They found that serum prolactin levels did not differ between the negative and positive pregnancy test groups [11].

In addition, Hassan (2020) conducted a study on 53 women undergoing IVF/ICSI cycles and divided them into two groups: PCOS women and non-PCOS women. He found that women with high blood prolactin levels had a reduced pregnancy rate in both PCOS and non-PCOS groups. He concluded that women with PCOS are more likely to have hyperprolactinemia than normal ovulating females. Elevated serum prolactin levels impair implantation and reduce the likelihood of pregnancy following ICSI [12].

Iancu et al. (2022) conducted their study on 544 patients with serum prolactin levels of 25.07±23.7 ng/ml and found no correlation between serum prolactin levels and oocyte or embryo count [13]. However, a later review article by Iancu et al. (2023) suggested the role of mildly elevated prolactin levels in reproductive biology with different mechanisms. Oocyte competency is linked to elevated prolactin levels in follicles. Prolactin may also have a role in the development and survival of the corpus luteum, endometrial receptivity, blastocyst implantation, and survival of sperms with weak motility [1].

Endometrial cells were found to have prolactin receptors. Endometrial prolactin production maintains endometrial receptivity and has been demonstrated to offer an optimum environment for the implanting blastocyst transplanted during IVF cycles when circulating prolactin levels are normal [14,15].

Limitations

The main limiting point in our study is that we targeted infertile patients due to certain causes. We excluded infertile women with anovulation due to PCOS or Hyperandrogenism. In addition, patient recruitment was based on their basal prolactin level. We only targeted women with basal prolactin less than 50 ng/ml. Therefore, we did not study the effect of increased prolactin levels above 50 ng/ml on pregnancy outcomes.

CONCLUSIONS

The number of mature oocytes and embryos among females having IVF/ICSI was not shown to be correlated with serum prolactin levels; however, a little increase in serum prolactin levels (25 to 50 ng/ml) was linked to a greater incidence of clinical and ongoing pregnancy rates.

COMPLIANCE WITH ETHICAL STANDARDS

Authors contribution

MS, SM designed and supervised the study. SE, AS, MA conducted the study and analyzed the data. MA analyzed the data. All authors wrote the draft manuscript and approved the final manuscript.

Funding

This research received no specific grant from any funding agency.

Disclosure of Interests

None of the authors has financial or other conflicts of interest.

Ethical approval

The study protocol was approved by the Research Ethics Committee with reference number (MS-173-2022). All methods were carried out in accordance with relevant guidelines and regulations.

Informed consent

All participants gave their consent after being informed of the study's objective and design, and they were given the option to leave the study at any time.

Data sharing

The data that support the findings of this study are available from Kasr El-Ainy Hospital, but restrictions apply to the availability of these data, which were used under license for the current study, and so are not publicly available. Data are, however, available from the authors upon reasonable request and with the permission of Kasr El-Ainy Hospital.

REFERENCES

[1] Iancu ME, Albu AI, Albu DN. Prolactin Relationship with Fertility and In Vitro Fertilization Outcomes—A Review of the Literature. Pharmaceuticals. 2023;16:122. doi:10.3390/ph16010122

[2] Bernard V, Young J, Binart N. Prolactin - a pleiotropic factor in health and disease. Nat Rev Endocrinol. 2019;15:356–365. doi:10.1038/s41574-019-0194-6

[3] Zhao G-K, Zheng Y, Guo H-X, Wang H-Q, Ji Z-H, Wang T, et al. TRH Regulates the Synthesis and Secretion of Prolactin in Rats with Adenohypophysis through the Differential Expression of miR-126a-5p. Int J Mol Sci. 2022;23:15914. doi:10.3390/ijms232415914

[4] Sadiq NM, Tadi P. Physiology, Pituitary Hormones. StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2024 [cited 2024 Apr 7]. Available from: http://www.ncbi.nlm.nih.gov/books/NBK557556/.

[5] Thapa S, Bhusal K. Hyperprolactinemia. StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2024 [cited 2024 Apr 7]. Available from: http://www.ncbi.nlm.nih.gov/books/NBK537331/.

[6] Vilar L, Vilar CF, Lyra R, Freitas M da C. Pitfalls in the Diagnostic Evaluation of Hyperprolactinemia. Neuroendocrinology. 2019;109:7–19. doi:10.1159/000499694

[7] Karayazi Atıcı Ö, Govindrajan N, Lopetegui-González I, Shemanko CS. Prolactin: A hormone with diverse functions from mammary gland development to cancer metastasis. Semin Cell Dev Biol. 2021;114:159–170. doi:10.1016/j.semcdb.2020.10.005

[8] Mele D, Caprio F, D'Eufemia MD, Schiattarella A, Labriola D, Schettino MT, et al. In vitro fertilization and psychological stress: new insight about different routes of progesterone administration. Ital J Gynaecol Obstet. 2020;32:119. doi:10.36129/jog.32.02.04

[9] Zhang D, Yuan X, Zhen J, Sun Z, Deng C, Yu Q. Mildly Higher Serum Prolactin Levels Are Directly Proportional to Cumulative Pregnancy Outcomes in in-vitro Fertilization/Intracytoplasmic Sperm Injection Cycles. Front Endocrinol. 2020;11:584. doi:10.3389/fendo.2020.00584

[10] Wyse BA, Weizman NF, Defer M, Montbriand J, Szaraz P, Librach C. The follicular fluid adipocytokine milieu could serve as a prediction tool for fertility treatment outcomes. Reprod Biomed Online. 2021;43:738–746. doi:10.1016/j.rbmo.2021.07.001

[[11] Trikoilis N, Mavromatidis G, Tzafetas M, Deligeoroglou E, Tzafetta M, Loufopoulos A, et al. The association of in vitro fertilization/intracytoplasmic sperm injection results with anxiety levels and stress biomarkers: An observational, case-control study. J Gynecol Obstet Hum Reprod. 2022;51:102254. doi:10.1016/j.jogoh.2021.102254

[12] Hassan MF. The frequency of elevated prolactin level in polycystic ovary syndrome women (PCOS) and its effect on pregnancy rate. Glob J Public Health Med. 2020;2:109–117.

[13] Iancu M, Albu A, Albu D. The association between serum prolactin levels and outcomes of in vitro fertilization. Hum Reprod. 2022;37:deac107-646. doi:10.1093/humrep/deac107.646

[14] Auriemma RS, Del Vecchio G, Scairati R, Pirchio R, Liccardi A, Verde N, et al. The Interplay Between Prolactin and Reproductive System: Focus on Uterine Pathophysiology. Front Endocrinol. 2020;11:594370. doi:10.3389/fendo.2020.594370

[15] Pérez-López FR, López-Baena MT, Pérez-Roncero GR. Prolactin secretion in women: narrative review. Gynecol Pelvic Med. 2021;4:20. doi:10.21037/gpm-21-4

Table 1: Patients' characteristics and pre-cycle laboratory investigations.

	Group 1 (n=111)	**Group 2 (n=111)**	**P-value**
Age (years)	30.71 ± 5.42 30 (19 - 43)	29.59 ± 5.43 29 (20 - 42)	0.125
BMI (kg/m^2)	29.48 ± 4.39 29.1 (20.3 - 38.9)	30.03 ± 4.68 29.6 (19.5 - 43.4)	0.366
Duration of infertility (years)	5.61 ± 3.48 5 (1 - 18)	4.95 ± 2.94 5 (1 - 15)	0.130
Type of infertility - Primary - Secondary	 56 (50.45%) 55 (49.55%)	 66 (59.46%) 45 (40.54%)	0.177
FSH	7.02 ± 2.29 6.59 (1.8 - 13.6)	6.94 ± 2.80 6.4 (3.2 - 27.5)	0.807
LH	5.21 ± 2.22 5.05 (0.44 - 14.3)	5.55 ± 2.34 5.2 (1.55 - 14.3)	0.268
E2	54.07 ± 24.30 49 (15.7 - 137)	54.60 ± 31.42 46.2 (10.2 - 198.2)	0.888
AMH	2.34 ± 1.22 2.1 (0.5 - 9.3)	2.11 ± 1.04 1.9 (0.01 - 5.2)	0.146
TSH	2.04 ± 0.89 2 (0.53 - 4.7)	1.94 ± 0.78 1.9 (0.6 - 4.06)	0.365
Basal Prolactin level (P0)	12.47 ± 5.19 12 (3 - 28.3)	30.35 ± 4.55 29.2 (9.8 - 42)	<0.001*

Table 2: Characteristics and outcomes of IVF/ICSI cycles.

	Group 1	**Group 2**	**P-value**
Prolactin level at trigger day(P1)	64.71 ± 27.74 62.7 (13 - 144)	103.24 ± 29.95 103 (19.5 - 164)	< 0.001*
Gonadotropins dose (IU)	267.57 ± 61.20 225 (150 - 450)	277.50 ± 53.32 300 (150 - 450)	0.200
Antral follicular count	12.85 ± 4.02 12 (6 - 25)	12.31 ± 3.87 12 (3 - 20)	0.308
Number of oocytes retrieved	10.42 ± 5.29 10 (2 - 27)	9.95 ± 4.56 9 (2 - 25)	0.473
Number of mature oocytes (M2)	6.31 ± 3.79 6 (0 - 20)	5.80 ± 3.15 5.5 (0 - 16)	0.281
Failed fertilization rate	12/111 (10.81%)	8/111 (7.21%)	0.348
Number of total embryos	4.38 ± 2.28 4 (1 - 10)	3.99 ± 2.32 3 (1 - 14)	0.224
Number of transferred embryos	3.02 ± 0.82 3 (1 - 4)	2.93 ± 0.71 3 (1 - 4)	0.420
Clinical pregnancy rate	40/99 (40.40%)	57/103 (55.34%)	0.034*
Ongoing pregnancy rate	32/99 (32.32%)	48/103 (46.60%)	0.038*

Figure 1: Flowchart of the study.