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The impact of PRP and ovarian needle puncture on the level of ovarian androgens and the results of ICSI in women with PCOS

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Abstract

PCOS is caused by hyperandrogenism, ovulatory dysfunction, and aberrant GnRH pulsation that results in irregular gonadotropin production. Platelet Rich Plasma (PRP), which is useful for ovarian rejuvenation by enhancing folliculogenesis, is being used more and more for a variety of conditions, including infertility. To assess the impact of combining PRP and ovarian needle puncture on the levels of ovarian androgens (free testosterone and androstenedione), as well as the characteristics of the ICSI cycle and its outcome in women with PCOS. Seventy women from infertile couples participated in this study; they were divided into two groups at random: PRP Group: 35 women who received PRP and ultrasound-guided transvaginal ovarian needle puncture at the same time during the previous ICSI cycle. Patients in the non-PRP group (35 women) did not get PRP and ultrasound-guided transvaginal ovarian needle puncture at the same time during the previous ICSI cycle. CD7–CD8: Under general light anesthesia, an ultrasound-guided transvaginal ovarian needle puncture was performed concurrently with ovarian PRP for the PRP group. After PRP therapy, mean serum free testosterone decreased significantly (from 1.35 ± 0.91 to 0.86 ± 0.57, P < 0.001), although androstenedione levels did not alter significantly. On the day of ova pick-up, there were no appreciable variations in stimulation, oocyte, embryonic traits, or hormone levels between the PRP and non-PRP groups. Although not statistically significant, the PRP group had greater pregnancy rates for both fresh (34.3% vs. 20.0%, P <0.113) and frozen ET (20.0% vs. 14.3%, P <0.292). When administered concurrently to PCOS women prior to an ICSI cycle, ovarian needle puncture and PRP effectively lower basal levels of free testosterone and may increase the likelihood of conception. To elucidate the efficacy of PRP in PCOS, more research is necessary.

Keywords: Ultrasound-guided transvaginal ovarian needle puncture, Platelet-rich plasma, Polycystic ovary syndrome

Introduction

About 6–13% of women who are of reproductive age suffer from polycystic ovarian syndrome (PCOS), which has a genetic component. Globally, up to 70% of women with PCOS go misdiagnosed. The most frequent cause of anovulation and a major contributor to infertility is PCOS. Numerous chronic illnesses that affect both mental and physical health are linked to the syndrome. 1. known as Stein-Leventhal syndrome or hyperandrogenic anovulation2. PCOS is exacerbated by hyperandrogenism, ovulatory dysfunction, insulin resistance, and aberrant GnRH pulsation that results in irregular gonadotropin production. PCOM is brought on by increased androgen production from ovarian dysfunction, which also interferes with follicular growth and ovulation.

A rise in LH production and aberrant negative or positive feedback mechanisms involving progesterone and estrogen are two factors contributing to hyperandrogenism's dysregulation of the pulsatile secretion of GnRH3. Through the activation of IGF-1 via androgen receptors, androgens facilitate the shift of primordial to secondary follicles. Additionally, they promote Graafian follicle development by increasing FSH sensitivity through the creation of more FSH receptors.

For antral follicles to produce oestradiol, androgens such as testosterone and androstenedione are essential. Hormonal levels in PCOS patients show a shift from progestogenic to more estrogenic follicles, which is accompanied by higher expression of HSD17B2, CYP11A, and CYP19A in FF. Additionally, oocyte quality5 is negatively correlated with the mRNA levels of CYP11A, CYP19A, and HSD17b1 in exosomes detected in FF.

According to studies, increased activity and

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expression of steroidogenic enzymes such as CYP17A1, CYP11A1, HSD3B2, SRD5A1, and AKR1C36 cause Theca cells (Cs) in PCOS patients to create excess androgens.

Although ART techniques like IVF/ICSI-ET are frequently used to treat infertility in women with PCOS, they are less safe than for women without PCOS7 due to added risks such cycle cancellation, OHSS, and an increased risk of multiple pregnancies.

Although the exact mechanism behind transvaginal ovarian needle punctures (UTND) is unknown, it might be similar to laparoscopic ovarian drilling (LOD) or ovarian wedge resection. In women with PCOS, whose ovarian tissues and fluids are normally high in androgens8, UTND quickly reduces intraovarian androgen levels by puncturing the ovary and aspirating follicular fluid. Research indicates that transvaginal ovarian cold drilling can improve the outcomes of in-vitro fertilization in people with PCOS who are difficult to treat. It is less intrusive and less expensive than LOD, but the impact is temporary and fades within six months.

A person's blood is used to make platelet-rich plasma (PRP), which contains a high concentration of platelets that promote healing and coagulation with little chance of immunological rejection. Pure PRP, white blood cell and PRP, pure platelet-rich fibrin, and white blood cell and platelet-rich fibrin are the four categories of platelet concentrates based on the amount of white blood cells and fibrin they contain. In regenerative medicine, PRP is utilized to heal injured tissues. Although its exact mechanism is still unknown, recent research suggests PRP may help the female reproductive system by encouraging endometrial and follicle growth.

PRP is being utilized more and more to treat a variety of conditions, such as infertility, and as an ART supplement for women who have early menopause, premature ovarian insufficiency, or low ovarian reserve. In women with low ovarian reserve, PRP is useful for ovarian rejuvenation because, after four weeks, clinical hormone levels of FSH and AMH returned to normal. Furthermore, PRP may boost ovarian volume and antral follicle count (AFC), increasing the chance of ovulation and possibly subsequent conception.

In ICSI 12, 13, PRP improved endometrial parameters and improved implantation. Intraovarian PRP was found to improve folliculogenesis, decrease follicular atresia and mRNA damage, increase progesterone and estradiol, decrease testosterone, LH, FSH, and androstenedione, increase ovarian antioxidant potential, and boost estrogen receptor expression in a mouse model of PCOS14. It is relevant to take into account the possible effects of PRP-related growth factors on different molecular components within ovaries affected by PCOS15, given the crucial role that growth hormones play in early and late folliculogenesis, the initiation of oocyte growth, cell proliferation, and the inhibition of apoptosis, especially during the later stages of development. Few human research has looked at how intraovarian PRP affects hormonal imbalance or ovulatory dysfunction in PCOS-afflicted patients.

Patients and techniques

The High Institute for Infertility Diagnosis and Assisted Reproductive Technologies at AL Nahrain University in Baghdad, Iraq, is the site of this prospective randomized (comparative) trial. Seventy women from infertile couples who visited the institute's infertility diagnosis clinic participated in the trial, which ran from October 1, 2023, until April 1, 2025. The Local Medical Ethical Committee approved the trial, and each patient provided written consent.

Rotterdam's criteria for PCOS diagnosis and eligibility for intracytoplasmic sperm injection (ICSI) were the inclusion criteria.

1. Hyperandrogism: This condition is clinically diagnosed by either hyperandrogenemia (high androgen test) or cutaneous signs of excess androgen.
2. Dysfunction of ovulation (oligo/anovulation).
3. Ultrasound assessment of polycystic ovarian morphology.

ICSI management was included in this investigation because PCOS-infertile women may have additional variables, such as a tubal etiology or a prolonged period of infertility.

The study included PCOS women from couples with a male factor that indicated ICSI, such as severe

Oligoasthozoospermia, Teratozoospermia, and severe Oligoasthoteratozoospermia (OAT). Couples with infertile PCOS women and husbands who had infertility causes were included in the study. They had to be between the ages of 18 and 40, have a BMI between 18 and 35, and have platelet counts within acceptable ranges (150000–400000) per microliter. Exclusion criteria: illnesses that are psychologically or medically incompatible with pregnancy, Untreated gynecological diseases, such as pelvic infections, ovarian tumors, and uterine causes of infertility, endometriosis, and untreated endocrine disorders Patients with blood problems, those on anticoagulant or antiplatelet medications, and those who were unable to produce Grade 1 embryos.

Patients were divided into two groups at random: PRP Group: 35 women who received PRP and ultrasound-guided transvaginal ovarian needle puncture at the same time during the previous ICSI cycle. Patients in the non-PRP group (35 women) did not get PRP and ultrasound-guided transvaginal ovarian needle puncture at the same time during the previous ICSI cycle. Every girl should undergo a hormonal test that measures the basal cycle day two within the usual range. Prolactin, AMH, FSH, LH, TSH,

Free testosterone and androstenedione. CD7–CD8: Under general light anesthesia, an ultrasound-guided transvaginal ovarian needle puncture was performed concurrently with ovarian PRP for the PRP group. The entire PRP preparation process was conducted at 24 degrees Celsius in an air-conditioned setting.

In general, there are three phases involved in PRP preparation:

1. Gathering samples Since PRP is made of plasma, making plasma from the patient's blood is the initial step in making PRP. To get 2-4 mL for PRP of both ovaries, the patient had about 20-30 mL of blood drawn via peripheral venipuncture. The most advised anticoagulant is sodium citrate since it improves platelet preservation16, 17.

2. Centrifugation: By centrifuging and separating its various components, platelet-rich plasma (PRP) is made from whole blood, which contains plasma (55%), red blood cells (41%), platelets, and white blood cells (4%). Red blood cells are eliminated during the centrifugation and separation procedure, and plasma with five to ten times higher amounts of growth factors is produced 18. varied centrifuge machines have varied rotor sizes and radii (R). Rotation per minute (rpm) is therefore calculated using relative centrifugal force (g) measurements. The following is the conversion factor from "g" to "rpm": R (rpm) = (1.118 x 10-5) g 2 19. following a 6-minute 1800g centrifugation. Platelet-poor plasma was represented by the upper layer, which was aspirated and disposed of. The lower layer, which corresponded to red blood cells, was disposed of after the buffy coat layer was aspirated and transferred to a different sterile conical tube for an additional centrifugation cycle. Four milliliters of PRP were aspirated into a sterile syringe, and the procedure was repeated a second time without the addition of activators.

3. Storage: Preserving the PRP's quality before using it in a therapeutic setting. In order to preserve the leukocyte concentration and pH of solution19, PRP should ideally be utilized no later than eight hours after centrifugation. Within an hour, PRP was injected into our patients.

About 2 mL of PRP per ovary was injected intraovarianly utilizing ultrasound-guided transvaginal injection while under intravenous sedation or general anesthesia administered by a certified anesthesiologist. The injection was carried out in multifocal areas, and three to four transvaginal punctures per ovary were made using a 22-gauge needle and guide18 in order to diffuse the PRP in the subcortical layers. The patient was discharged on antibiotics and little analgesic medications after a well-tolerated operation.

To begin the ICSI cycle on cycle day two, all women are advised to visit the institute's infertility clinic. For the PRP group, a venipuncture was used to obtain a blood sample for hormone retesting: FreeT and Androstendion.

Protocols for Controlled Ovarian Hyperstimulation: The flexible antagonist protocol type was used for ICSI20 since PCOS affects all women. Daily subcutaneous injections of rFSH (Gonal F, Merk Serono) at a dose ranging from 75 IU to 225 IU per day at a set time were used to initiate stimulation on CD2. As soon as three or more follicles were at least 17–18 mm in size,

With the goal of avoiding OHSS, one of two methods was used to initiate ovulation and the maturation of the final oocytes: A. The Dual Trigger: 0.2 mg of triptorelin (Decapeptyl; Ferring Pharmaceuticals) and recombinant hCG (Ovitrelle®, 250 IU Merk Serono, Switzerland) are subcutaneously injected 35 hours prior to the time for ova pick-up. B. Agonist Trigger for GnRH: A GnRH-a trigger's main clinical advantage is its capacity to cause quick, reversible luteolysis, which lowers the risk of OHSS. For final oocyte maturation, patients were given a single 0.2 mg bolus of GnRH-a (triptorelin; decapeptyl, Ferring). Segmentation of the IVF cycle is the process of freezing all embryos following GnRH-a trigger and transferring them in a subsequent cycle. Women who were at risk for OHSS21 had undergone this treatment.

Every woman in both groups consented to have a blood sample taken in the morning of the ova pick-up day in order to measure the hormones Androstendion Δ4-A and FreeT.

The oocytes were collected by the ART specialist 34–36 hours after triggering, right before follicular rupture. Oocytes were extracted from patients under the supervision of transvaginal ultrasound after they were admitted to the operating room fasting. Usually, the entire process took 20 to 30 minutes. Patients were then treated with analgesics, antibiotics, and proge-sterone22 to support the luteal phase. Fresh embryo transfers were performed on women who got dual trigger, and frozen embryo transfers were performed on women receiving GnRH agonist trigger in both the PRP and groups using the same technique.

The number of embryos that were chosen for transfer was based on the patient's age, the rank of the attempt, the clinical history, and the embryonic quality. Only two embryos were transplanted during the initial try in women under 35. We transplanted three embryos to ladies above the age of 35. About two weeks following embryo transfer, a pregnancy test was performed to ensure successful implantation.

Taking samples: Serum sampling: A. Group: Free testosterone, androstendion. On the day of ova collection and Cycle CD2:

B. PRP Group: Free T, Androstenedione: Prior to PRP sampling, on CD2 of the ICSI cycle, following PRP sampling, and on the day of ova collection: The study's Androstendion and FreeT measurement kits were the quantitative in vitro diagnostic measurement of androstenedione in serum was performed using the Demeditec Androstenedione ELISA DE3265 Kits from Germany and the Free Testosterone Demeditec Testosterone free ELISA DE2924 Kits from Germany.

Findings

Demographics: There were 35 patients in the PRP group and 35 in the non-PRP group in this study. Table 4.1 displays a summary of the demographic traits of both groups. The mean age, mean body mass index, mean duration of infertility, proportions of patients by type of infertility, and proportions of patients by etiology of infertility did not differ significantly ($p > 0.05$).

Table 1: Comparing the demographic traits of the PRP group to the non-PRP group

Characteristic	PRP group 35 cases	Non-PRP group 35 cases	*P*
Age (years			
Mean ±StDe.	28.71 ±5.36	29.43 ±5.54	0.585
Min.-Max.	18 -39	19 -39	N
BMI (kg/m^2)			
Mean ±StDe.	27.72 ±3.73	28.31 ±3.16	0.480
Min.-Max.	21 -33.7	21 -35	N
Duration of Infertility (years)			
Mean ±StDe.	6.49 ±4.01	7.74 ±3.76	0.181
Min.-Max.	1 -20	2 -18	N
Type of Infertility			
Primary	29 (82.9 %)	26 (74.3 %)	0.382
Secondary	6 (17.1 %)	9 (25.7 %)	N

Cause of Infertility			
Male Factor	17 (48.6 %)	13 (37.1 %)	0.488 N
Female Factor	4 (11.4 %)	3 (8.6 %)	
Dual Factor	14 (40.0 %)	19 (54.3 %)	

StDe.: standard deviation; BMI: body mass index; N: not significant

Hormonal serum levels: Comparison of mean serum hormonal levels at baseline between Non PRP group and PRP group is shown in table 4.2 in which we reported lack of significant variations in mean levels of FSH, LH, TSH, prolactin, estradiol and AMH (p> 0.05).

Table.2: Comparison of mean serum hormonal levels at baseline

Characteristic	**PRP group**	**Non-PRP group**	***P***
	35 cases	**35 cases**	
CD2 FSH (mIU/ ml)			
Mean ±StDe.	5.65 ±1.08	5.49 ±1.60	0.633 N
Min.-Max.	2.8 -7.3	2.88 -11.2	
CD2 LH (mIU/ ml)			
Mean ±StDe.	5.11 ±1.92	5.88 ±2.11	0.115 N
Min.-Max.	1.6 -8.7	1.9 -12.9	
TSH (pg/ml)			
Mean ±StDe.	1.89 ±0.58	1.98 ±0.60	0.536 N
Min.-Max.	0.56 -3	0.63 -2.7	
Prolactin (mIU/ ml)			
Mean ±StDe.	19.64 ±5.02	17.46 ±6.65	0.125 N
Min.-Max.	7 -29	3.2 -32	
E2 (pg/ml)			
Mean ±StDe.	30.50 ±11.09	33.89 ±10.21	0.187 N
Min.-Max.	12 -51.2	10 -52	
AMH (ng/ml)			
Mean ±StDe.	4.53 ±1.69	4.70 ±1.41	0.641 N
Min.-Max.	1.6 -9.2	2.87 -9.78	

E2: estradiol; AMH: anti-mullerian hormone; FSH: follicle stimulating hormone; LH: luteinizing hormone; TSH: thyroid stimulating hormone; CD2: cycle day 2; N: not significant

Table 2 compares the baseline mean serum levels of free testosterone and androstenedione in the PRP group and the non-PRP group. Prior to treatment, the mean levels of free testosterone in the study group and the non-PRP group did not differ significantly (p = 0.379). There was no significant change in the meaning of androstenedione between the PRP and non-PRP groups (p = 0.088).

Table 3: Comparison of mean serum free testosterone and androstenedione levels at baseline between Non PRP group and PRP group

Characteristic	**PRP group**	**Non-PRP group**	***P***
	35 cases	**35 cases**	
Free Testosterone (pg/ml)			
Mean ±StDe.	1.35 ±0.91	1.61 ±1.50	0.379 N
Min.-Max.	0.12 -4.15	0.09 -7.65	
Androstenedione (ng/ml)			
Mean ±StDe.	1.88 ±0.98	2.30 ±1.04	0.088 N
Min.-Max.	0.32 -4.50	0.77 -5.9	

StDe.: standard deviation; N: not significant

Comparison of mean serum free testosterone and androstenedione levels before and after PRP is shown in table 4.4. After PRP, there was significant reduction in free testosterone level (p = 0.001), as shown in Table 3, but no significant alteration in mean serum androstenedione (p = 0.330), as shown in Table 3.

Table 4: Comparison of mean serum free testosterone and androstenedione levels before and after PRP

Characteristic	PRP group Before	PRP group After	*P*
	35 cases	35 cases	
Free Testosterone (pg/ml)			
Mean ±StDe.	1.35 ±0.91	0.86 ±0.57	0.001 S
Min.-Max.	0.12 -4.15	0.05 -1.9	
Androstenedione (ng/ml)			
Mean ±StDe.	1.88 ±0.98	2.05 ±1.01	0.330 N
Min.-Max.	0.32 -4.50	0.15 -4.98	

StDe.: standard deviation; N: not significant; S: significant

Comparison of ICSI Cycle stimulation characteristics: Stimulation characteristics are shown in Table 4 and comparison revealed no significant differences in mean dose of FSH, mean number of GnRH Antagonist Ampoules, mean day of rigger, mean E2 at trigger day, and trigger type (p > 0.05).

Table 5: ICSI Cycle stimulation characteristics

Characteristic	PRP group	Non PRP group	*P*
	35 cases	35 cases	
Dose of FSH			
Mean ±StDe.	1726.40 ±449.55	1727.10 ±488.86	0.995 N
Min.-Max.	1150 -3000	1050 -3100	
Number of GnRH Antagonist Ampoules			
Mean ±StDe.	3.46 ±0.78	3.26 ±0.66	0.250 N
Min.-Max.	2 -5	2 -5	
Trigger day			
Mean ±StDe.	10.63 ±0.91	10.57 ±0.65	0.764 N
Min.-Max.	9 -13	9 -12	
E_2 at trigger			
Mean ±StDe.	2443.00 ±860.16	2429.20 ±949.92	0.949 N
Min.-Max.	1255 -4360	1008 -6680	
Trigger type			
Dual trigger	20 (57.1 %)	17 (48.6 %)	0.473 N
Agonist Trigger	15 (42.9 %)	18 (51.4 %)	

N: not significant

Comparison of Oocytes Characteristics: Oocyte characteristics are shown in (Table 6).

Table 6: Oocyte characteristics

Characteristic	PRP group	Non PRP group	*P*
	35 cases	35 cases	
Number of Follicles in Ova pick up			
Mean ±StDe.	23.89 ±8.36	24.97 ± 9.97	0.623 N
Min.-Max.	9 -44	11 -58	
Number of Oocytes in Ova pick up			
Mean ±StDe.	17.86 ±7.39	17.43 ±8.01	0.817 N
Min.-Max.	4 -34	8 -51	

Mature (MII) Oocytes			
Mean ±StDe.	13.09 ±6.07	12.86 ±6.98	0.884 N
Min.-Max.	1 -25	3 -40	
Immature metaphase I (MI) oocytes			
Mean ±StDe.	0.94 ±1.16	0.94 ±1.11	1.000 N
Min.-Max.	0 -4	0 -4	
Germinal vesicle (GV) oocyte			
Mean ±StDe.	2.26 ±1.95	2.23 ±2.12	0.953 N
Min.-Max.	0 -7	0 -8	
Abnormal Oocytes			
Mean ±StDe.	1.51 ±2.51	1.06 ±1.43	0.353 N
Min.-Max.	0 -12	0 -5	
Follicle to oocyte ratio			
Mean ±StDe.	74.55 ±16.13	69.86 ±10.06	0.149 N
Min.-Max.	17.4 -93.3	43.8 -87.9	
Maturation index			
Mean ±StDe.	73.28 ±16.81	72.79 ±18.49	0.909 N
Min.-Max.	7.1 -100	18.8 -100	

N: not important

The mean number of oocytes, mean number of follicles, mean number of mature oocytes (MII), mean number of immature oocytes (MI), mean number of germinal vesicle oocytes, mean number of aberrant oocytes, follicle to oocyte ratio, and maturation index did not differ significantly ($p > 0.05$).

Comparison of the embryonic traits of the PRP and non-PRP groups:

The following was discovered when the features of the embryos in the PRP and non-PRP groups were compared: According to Table 6, there was no discernible variation in the number of 2PN oocytes, fertilization rate, mean number of grades 1, 2, 3, and 4 embryos, number of frozen embryos, number of transferred embryos, and kind of embryo transfer ($p > 0.05$).

Table 7: Comparison of embryo characteristics between Non PRP group and PRP group

Characteristic	**PRP group**	**Non-PRP group**	***P***
	35 cases	**35 cases**	
Number of 2PN			
Mean ±StDe.	9.77 ±5.32	10.77 ±6.25	0.474 N
Min.-Max.	1 -19	2 -35	
Fertilization rate			
Mean ±StDe.	75.31 ±19.00	81.25 ±17.48	0.178 N
Min.-Max.	15.8 -100	28.6 -100	
Number of Grade 1 Embryos			
Mean ±StDe.	6.29 ±4.19	6.89 ±4.10	0.547 N
Min.-Max.	1 -15	2 -23	
Number of Grsde 2 Embryos			
Mean ±StDe.	1.44 ±1.60	1.86 ±1.88	0.327 N
Min.-Max.	0 -8	0 -8	
Number of Grade 3 Embryos			
Mean ±StDe.	0.60 ±0.95	0.97 ±1.32	0.180 N
Min.-Max.	0 -4	0 -6	
Number of Grade 4 Embryos			
Mean ±StDe.	1.57 ±1.94	1.09 ±1.54	0.251 N
Min.-Max.	0 -7	0 -6	
Number of Frozen Embryos			
Mean ±StDe.	7.38 ±3.62	5.39 ±5.96	0.154 N

Min.-Max.	1 -19	0 -31	
Number of TE			
Mean ±StDe.	2.26 ±0.51	2.43 ±0.50	0.159 N
Min.-Max.	1 -3	1 -3	
Type of Embryo Transfer			
Fresh Embryo Transfer	18 (51.4 %)	17 (48.6 %)	0.811 N
Frozen Embryo Transfer	17 (48.6 %)	18 (51.4 %)	

N: not important

Table 8 compares the levels of serum free testosterone and androstenedione in the PRP group and the non-PRP group on the day of ovary pickup. Mean free testosterone and mean androstenedione did not differ significantly between the PRP and non-PRP groups ($p > 0.05$).

Table 8: Serum free testosterone and androstenedione levels on the day of Ova pick-up compared between the PRP and non-PRP groups

Characteristic	**PRP group** **35 cases**	**Non-PRP group** **35 cases**	***P***
Free Test. at ova pickup(pg/ml)			
Mean ±StDe.	1.03 ±1.16	1.62 ±1.45	0.063 N
Min.-Max.	0.01 -5.57	0.10 -6.50	
Androstenedione at ova pickup(ng/ml)			
Mean ±StDe.	3.07 ±1.80	2.56 ±1.22	0.169 N
Min.-Max.	1.37 -10.20	0.89 -6.92	

N: not significant

Comparison of positive pregnancy rate between Non PRP group and PRP group is shown in table 9. The differences were not significant ($p > 0.05$).

Table 9: Comparison of positive pregnancy rate between Non PRP group and PRP group

Characteristic	**PRP group**	**Non-PRP group**	**P**
Not Pregnant	16 (45.7 %)	23 (65.7 %)	Reference
Pregnant after fresh ET	12 (34.3 %)	7 (20.0 %)	0.113 N
Pregnant after frozen ET	7 (20.0 %)	5 (14.3 %)	0.292 N

Talk about

Few research has examined the effects of PRP on PCOS women to date, but comparatively more have described the less intrusive interference to the ovary caused by cold ultrasound-guided transvaginal ovarian needle puncture. By doing a small number of needle punctures and injecting PRP into the ovary under the guidance of transvaginal ultrasound, which measures ovarian androgens and ICSI cycle parameters, we examined the impact of using a comparatively less invasive approach to PCO. The use of GnRH triggers and cycle segmentation to lower the risk of ovarian hyperstimulation syndrome (OHSS) are two safer strategies for women with PCOS that have been made possible by advancements in ovarian stimulation protocols and ARTs.

The PRP and non-PRP groups in the current study did not differ significantly in terms of demographics, mean serum hormonal levels at baseline, comparison of mean serum free testosterone and androstenedione levels at baseline, stimulation characteristics, oocyte characteristics, embryo characteristics, or serum free testosterone and androstenedione levels on the day of ova pick-up. When mean serum free testosterone levels were compared before and after PRP, there was no significant difference in androstenedione levels following the research interference, but free testosterone levels significantly decreased after PRP (1.35 ±0.91, 0.86 ±0.57, $P < 0.001$ as shown in Table 3).

The predictive importance of baseline testosterone levels on the result of IVF cycles23 pregnancy is still up for discussion. While dehydroepiandrosterone

(DHEA) was not found to be excessive or diagnostic for PCOS, Abdelazim et al.'s 2020 study of women with PCOS reported that elevated levels of testosterone (both total and free) and androstenedione were markers of excess ovarian androgens. In comparison to controls, the PCOS group had considerably higher levels of androstenedione and testosterone (both free and total testosterone)24,38.

The number of big follicles seen on the day after HCG injection and ovarian response can both be accurately predicted by basal testosterone (T) levels. Additionally, baseline T levels could be helpful in determining how much FSH is needed. Lower T levels may be linked to a potentially reduced ovarian response in the overall female population25. Yang W. blamed the HA PCOS group in 2018 for having a significantly higher number of retrieved oocytes and a significantly lower consumption of total gonadotropin, while also having a significantly higher abortion rate and a significantly lower live birth rate when compared to the control and non-HA PCOS groups. There was no significant difference in the amount of available embryos between the HA PCOS group and the other group.

Wang et al. (2024) examined the Free Androgen Index and found that FAI had no clinical prognostic value for reproductive outcomes in women with PCOS undergoing FET27 because the cutoff threshold of hyperandrogenism has been varied in the studies. AMH is elevated by hyperandrogenaemia (HA), which inhibits follicle growth and ovulation. Additionally, it decreases follicular fluid IGF-II, which is otherwise associated with bigger follicles and elevated levels of estradiol28, 37.

Instead of decreasing negative feedback and promoting increased production of GnRH and LH29, it is expected that a rise in androgen levels will strengthen negative feedback mechanisms, leading to decreased secretion of GnRH and LH. Research argued that Δ4-A levels can be a good indicator of ovarian stimulation response, outperforming FSH and comparable to AMH and AFC. Therefore, although further research is required, baseline Δ4-A may be as good as current leading markers at predicting the results of IVF stimulation. Throughout the menstrual cycle, adrenal androgen levels stay constant, therefore changes are caused by the ovary's synthesis of Δ4-A.

Δ4-A may be involved in controlling follicular growth, promoting development past the pre-antral stage, avoiding follicular atresia, and improving the follicular response to stimulation, according to evidence30,31. According to a study, the number of chosen follicles in PCOS cases is better predicted by the early follicular phase elevation in androstenedione than by estradiol. More follicles and an increased risk of an overactive ovarian response are linked to a larger or longer increase in androstenedione levels32,39.

Comparable rates of cleavage, fertility, available embryos, and high-quality embryos were found between the PCOS group and the control group33, suggesting that documented polycystic ovarian morphology does not negatively impact oocyte or embryo quality in IVF/ICSI cycles. PRP auto-located groups had lower serum levels of FSH, LH, testosterone, and androstenedione, according to a 2019 study on animal models by Anvari, S.S., G. Holamreza Dehgan, and Razi, M. Their initial findings indicated that auto-locating PRP fairly ameliorates PCOS-induced pathogenesis. According to the researchers, PRP can help with hormonal imbalance and decrease androgen oversynthesis.

In the end, it may improve the ovulation ratio. Given these results and the high concentrations of many growth factors in PRP, auto-location of this factor may be a novel approach for PCOS patients15. The substantial drop in free testosterone levels following PRP administration to the ovary is in line with the findings of this investigation.

To raise awareness of the possible impacts of intraovarian PRP treatment on folliculogenesis in the context of PCOS, a case report was written.

According to that article, intraovarian PRP treatment resulted in follicular growth and an ovulatory event in an amenorrheic woman with PCOS. The unusually increased blood T level14 also significantly decreased. By simulating a tiny needle drilling 14,34, the paper foxed on an additional mechanism of the potential mechanical effect of inserting the needle into the ovary. Because the ovarian stromal and follicular tissues of women with PCOS produce significantly more androgen, and because their

follicular fluid has a high concentration of androgen8, ovarian puncture and aspiration of follicular fluid through UTND reduces intraovarian androgen and other steroids quickly and directly.

According to Table 8, the pregnancy rate was higher in the PRP group than in the non-PRP group, but the difference was not statistically significant: pregnant after fresh ET 12 (34.3%,) 7(20.0%) P< 0.113) and pregnant after frozen ET 7 (20.0 %), 5 (14.3 %) P< 0.292. The improvement in the embryo euploidy rate after intraovarian PRP application35 may be the cause of this finding. PRP's growth factors may have a local paracrine action that enhances meiotic abnormalities in human oocytes, hence increasing euploidy rates. Large trials18 are still needed to determine whether PRP increases live birth rates and decreases miscarriage rates.

According to an experimental study on the impact of intraovarian PRP injection on ovarian morphology and insulin resistance (PCOS) rats, PRP significantly reduced serum insulin and glucose levels when compared to the PCOS group. Additionally, the insulin resistance index significantly decreased in the PRP-treated groups. Simultaneously, histological investigation showed that a single PRP dosage significantly reduced the number of cystic follicles36 and significantly increased the number of healthy follicles and corpus luteum, improving the ovarian pathological status.

Conclusions

When administered concurrently to PCOS women prior to the ICSI cycle, ovarian needle puncture and PRP effectively lower basal levels of free testosterone and may increase the likelihood of conception. To elucidate the efficacy of PRP in PCOS, more research is necessary.

Abbreviation

PRP	Platelet Rich Plasma
Δ4-A	Androstenedione
OHSS	Ovarian hyperstimulation syndrome
ICSI	Intracytoplasmic Sperm Injection
UTND	Trans-vaginal Ovarian Needle Punctures.

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Author contributions

Rawaa Saad Abunyla, Lubna Amer Al-Anbari, Manal Taha Al-Obaidi conceived, designed the research and apply the study management protocols , Abunyla R. S. Abunyla performed the data analysis and wrote the manuscript and Al-Anbari L. M., Al-Obaidi M. T. conceived reviewed and edited the manuscript.

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Data availability

Data will be made available to the editors of the journal for review or query upon request.

Declarations

The study was approved by the Local Medical Ethical Committee in the High Institute for Infertility Diagnosis and Assisted Reproductive Technologies in AL Nahrain University

Consent to participate

A written consent was obtained from all participants

Consent to publish

Not applicable.

Attestation statement Data regarding any of the subjects in the study has not been previously published unless specified.

Competing interests

The authors declare no competing interests

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