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Original Research Article

Ovarian function following intra-ovarian injection of platelet rich plasma in premature ovarian aging

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ABSTRACT

Background: Premature ovarian aging as evidenced by AMH levels less than 0.5 ng/ml is predictive of reduced success in management of infertility. The most effective treatment is in vitro fertilization which is not affordable or acceptable to most of our patients with low socioeconomic background. Intra-ovarian injection of autologous platelet rich plasma (PRP) improves the ovarian reserve markers but it remains to be seen whether it improves the pregnancy rate.

Methods: This was a prospective study of 29 patients with poor ovarian reserve (age between 30-40 years, AMH<0.5 ng/ml), AFC<4 in both ovaries) who received intra ovarian injection of platelet rich plasma. The patients were followed up for one year and data was collected about clinically relevant outcomes like pregnancy.

Results: Four women (13.8%) had pregnancy, of which 3 were spontaneous and one was following ovarian stimulation and intrauterine insemination. All had diagnosed clinical pregnancy, of which two women had live birth.

Conclusions: Given the 13.8% rate of pregnancy within one year we can speculate that PRP injection may be a reasonable alternative to in vitro fertilization in women with diminished ovarian reserve.

Keywords: Diminished ovarian reserve, Platelet rich plasma, In vitro fertilization

INTRODUCTION

Premature ovarian aging or occult primary ovarian insufficiency is a milder form of ovarian insufficiency characterized by abnormally high follicular stimulating hormone (FSH) though below 40 mIU/ml and abnormally low age-specific antimullerian hormone (AMH).¹ The most effective therapy for these infertile women is in vitro fertilization (IVF). IVF may not be successful due to poor quantity and quality of the remaining egg pool. Donor oocytes for IVF in women with diminished ovarian reserve is often not acceptable for ethical, religious or psychological reasons and higher expenses.² Women with diminished ovarian reserve are likely to go a long way to

conceive or have a genetically related offspring. Though of unproven effectiveness and little value, platelet rich plasma (PRP) is often offered to couples under desperate circumstances only to be readily accepted.³

Platelet rich plasma is derived from peripheral blood. The activated alpha granules of platelets release a variety of growth factors which promotes healing and regeneration of tissues.⁴ There have been reports that intra ovarian injection of autologous platelet rich plasma has resulted in spontaneous pregnancy and follicular growth with improved pregnancy outcome in IVF treatment.⁵ Use of PRP may regenerate oocytes and restore fertility in women with ovarian insufficiency.² PRP has 5-10 fold higher

concentration of growth factors than whole blood. Murine ovaries when injected with growth factors appeared to develop mature eggs from oogonial stem cells, according to a research done on Harvard University.⁶ Ovarian rejuvenation with autologous PRP was first introduced by Pantos et al.⁷ Advanced maternal age is associated with poor ovarian reserve. Ovarian function is expected to improve when oogonial stem cells are exposed to activated PRP. The half life of growth factors in PRP is very short, thus limiting the duration of PRP effect. Laparoscopic approach is advantageous when ovaries without growing follicles are difficult to define with ultrasound.

We have women with diminished ovarian reserve who cannot afford in vitro fertilization. We added intraovarian injection of PRP to laparoscopic evaluation of tuboperitoneal factor, offering ovarian stimulation with intrauterine insemination to those with patent tubes. An observational study of these women revealed significant rise in AMH and fall in FSH over the next three months.⁸ A one year follow up study of these women was undertaken to find out more clinically relevant outcomes like change in menstrual cycle and pregnancy.

METHODS

This prospective observational study was carried out in the department of Reproductive Endocrinology and Infertility from July 2019 to December 2021. The study population of interest were 29 infertile women with poor ovarian reserve, age between 30-40 years. Inclusion criteria were AMH<0.5 ng/ml, AFC<4 in both ovaries and exclusion criteria were primary ovarian insufficiency (FSH>25 mIU/ml on two occasions one month apart), previous ovarian surgery or chemotherapy, ongoing anticoagulant use. They were offered laparoscopic evaluation of tuboperitoneal factor followed by ovarian stimulation and intrauterine insemination as they did not afford in vitro fertilization. Intraovarian injection of PRP was given during laparoscopy. Ethical approval from institutional review board was taken. Written informed consent was taken from each participant.

Autologous PRP preparation

Around 30 ml of autologous venous blood with anti coagulant was centrifuged for 10 minutes at 1000-1200 rpm. The white blood cells and platelets were separated from the red blood cells at the bottom. The supernatant top layer was platelet rich plasma (PRP). The 10 ml PRP aspirated was used for intraovarian injection as early as possible.

Follow up

The women had monthly estimation of ovarian reserve markers for three months. They were offered ovarian stimulation and intrauterine insemination once the serum AMH was >0.6 ng/ml. The women were contacted over cell phone at least one year after they had the intra ovarian

injection of autologous platelet rich plasma. Data was collected about clinically relevant outcomes like change in menstrual function, hypoestrogenic features, spontaneous or assisted pregnancy and pregnancy outcome. Statistical analysis was done with Microsoft excel.

RESULTS

The demographic and clinical characteristics of the study participants are described in Table 1. All the 29 women completed the second follow up with ovarian reserve markers. Three women (10.34%) became pregnant by 3rd month. Another 3 participants were lost to follow up due to COVID 19 pandemic. So 23 women completed the third visit for estimation of ovarian reserve.

Table 1: Demographic and clinical characteristics of the study participants.

Parameters	Percent
Age (years) mean±SD	35.90±3.2
Socio economic status by monthly income	
Lower middle (<20,000 tk)	62.07
Middle (20,000-40,000 tk)	34.48
High >40,000 tk	3.45
Occupation	
Housewife	65.52
Service	34.48
Education	
<10 years	37.93
10-12	20.69
>12	41.38
Habitat	
Urban	62.10
Rural	37.90
Types of infertility	
Primary	68.97
Secondary	31.03
Duration of infertility (years) mean±SD	
	6.80±3
Menstrual phenotype	
Eumenorrhea	89.66
oligomenorrhea	10.35
Early menopause in family	6.90

The mean rise in AMH (ng/ml) was 0.04±0.15 after first month, 0.18±0.21 after second month and 0.20±0.16 after third month. Only one woman had fertility treatment following intra ovarian injection of PRP and conceived. Patients received in addition to PRP treatment medications like DHEA, vitamin D and Coenzyme Q alone or in various combinations.

Four women (13.8%) had pregnancy, of which 3 spontaneous and one following ovarian stimulation and intrauterine insemination. All had diagnosed clinical pregnancy, of which two women had live birth.

Regarding menstrual function, 3 out of 4 women had improvement of menstrual cycle after PRP treatment from oligomenorrhea to eumenorrhea. Other women had regular menstrual cycles before and after PRP treatment. Menopausal symptoms like hot flush was reported by only one woman.

DISCUSSION

The study reveals that pregnancy rate in one year follow up of the women with diminished ovarian reserve was 13.8%. Regarding approximate live birth rates in women with low AMH, it is 8% with FSH/TUI and 12% per cycle start with IVF when the age is <35 years and 6% with FSH/TUI and 10% per cycle start with IVF when age 35-40 years.^{9,10} The threshold for low AMH was 0.4 by La marca and 0.84 by Brodin.^{9,10} So the pregnancy rate achieved by intraovarian PRP injection is comparable to that achieved by in vitro fertilization in women with diminished ovarian reserve.

There were six studies for PRP in diminished ovarian reserve: three case series, two observational studies and one observational non randomized study. Total number of subjects was 469. Improvement of ovarian reserve markers was detected on all studies.¹¹ Melo et al conducted a prospective controlled non-randomized study on 83 women with diminished ovarian reserve (age >38 years, FSH>12 mIU/ml and AMH<0.8 ng/ml).¹² Forty six women were given monthly ovarian intracortical injections for three months and 36 women received no intervention. All patients had subsequent ART with antagonist protocol and were followed up for one year. They reported biochemical pregnancy (26.1% versus 5.4%) and clinical pregnancy (23.9% versus 5.4%) in PRP versus controls. Petryk et al had a study on 38 women 31-45 years of age with low ovarian reserves and at least two unsuccessful attempts to receive their oocytes through IVF.¹³ PRP injections into the ovaries were given as an ultrasound-guided procedure or a laparoscopic-assisted approach. After PRP treatment, women were followed for 12 months. There was restoration of hormone levels in 79.2%. Natural conception occurred in 4 women.

Autologous PRP injection of ovaries were done in similar study populations like poor responder, primary ovarian insufficiency, peri menopausal as well as menopausal women wishing pregnancy.¹⁴

Ovarian rejuvenation with autologous PRP was first introduced by Pantos et al.⁷ The case series reported on two women with premature ovarian failure (POF) aged 40 and 27 years, respectively, and one menopausal woman aged 46 years. Patients achieved pregnancy through natural conception within 2–6 months following PRP treatment.

Cakiroglu et al had a study on 311 women, age 24-40 years with primary ovarian insufficiency diagnosed according to ESHRE criteria having intraovarian PRP injection.¹⁵ One

to two cycles after the PRP procedure 23 women (7.4%) conceived spontaneously. Case series of 30 women with primary ovarian insufficiency who received intra ovarian injection of PRP: 3/30 women had spontaneous pregnancy and live birth. Restoration of menstruation was observed in approximately half of women.⁵ Perfect injection of PRP is possible because of laparoscopy.¹¹

Farimani et al did a retrospective study on 96 women with poor ovarian response who were grouped according to POSIEDON criteria and received intraovarian PRP prior to assisted reproductive techniques (ART).¹⁶ Group 4 was most prevalent (58.3%). There was significant increase in total oocyte number and MII oocytes. Of 96 cases 14 (14.6%) had clinical pregnancy following ART. Case series of 30 women with poor ovarian response according to Bologna criteria receiving intraovarian PRP and having ICSI thereafter reported that clinical pregnancy happened in 14 women followed by 12 live births.⁵

The women with primary ovarian insufficiency have spontaneous pregnancy and live birth. A recent review of two randomized controlled trials, two observational studies and one interventional study report the occurrence of pregnancies naturally or with different treatments (hormonal replacement therapy, IVF, in-vitro maturation and stem cell therapy as a rate of 2.2-14.2% in these women.¹⁷ Mean age in patients who achieved a pregnancy was 30 years, highlighting that oocyte quality in these patients is likely unaffected. No treatment has thus far shown its superiority in improving fertility in women with POI. Women with relatively higher ovarian reserve who has not received any ovarian stimulation has a potential to conceive without PRP stimulation. The spontaneous conceptions are difficult to attribute to intervention with PRP.³

AMH is a sensitive and reliable ovarian reserve. AMH predicts oocytes yield as well as poor or excessive ovarian response in IVF, but do not predict pregnancy after OS/TUI for unexplained infertility or do not predict spontaneous pregnancy in infertile women. Reproductive potential is poorly predicted by AMH only. This is because ovarian reserve testing like AMH or AFC test only quantity but not quality of remaining oocytes pool.¹⁸ Ovarian reserve tests may be false positive. Diminished ovarian reserve diagnosed by low AMH is not equivalent to inability to conceive. Age is a much stronger predictor of pregnancy than ovarian reserve.¹⁸ An analysis of 5,000 fresh autologous IVF cycles of patients with extremely low levels of AMH (<0.16 ng/ml) found that live birth rate was 9.5% per cycle start (20.5% per transfer).¹⁸ Younger women have larger intra individual variation in AMH levels; 17 out of 22 women under 38 years show a variation of more than 0.5 ng/ml in AMH concentration.¹⁹

There is insufficient evidence by comparative studies in favor of effectiveness of PRP. Most studies are small, before- after studies without clinically relevant endpoints

like pregnancy and live birth. Some domestic and international congress has negative reports on ovarian rejuvenation by PRP injection. Publication of positive reports only allows us to suspect publication bias.³

The PRP treatment may have some risks yet to experience. We advice PRP for infertile women but the treatment may not be always on the best interest of the couple. As responsible professionals we should counsel the couples that PRP is still experimental. Unbiased explanation of possible results will prevent development of undue expectations.³

Limitations

Limitations of the study include small sample size. Selection of study population from a single centre limits external validity.

CONCLUSION

Given the 13.8% rate of pregnancy within one year we can speculate that PRP injection may be a reasonable alternative to in vitro fertilization in the women with diminished ovarian reserve.

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Conflict of interest: None declared

Ethical approval: The study was approved by the Institutional Ethics Committee

REFERENCES

- Barad DH, Albertini DF, Gleicher N. Endocrine effects of intra ovarian injection of platelet rich plasma in women with premature ovarian aging. *Fertil Steril.* 2020;e89:210-6.
- Park H, Ulin M, Cetin E. Ovarian Rejuvenation using platelet rich plasma: a promising option for women in early menopause to have a baby. *Reproductive Sciences.* 2020;12:21-4.
- Urman B, Boza A, Balaban B. Platelet rich plasma another add-on treatment getting out of hand? How can clinicians preserve the best interest of their patients? *Hum Reprod.* 2019;34(11):2099-103.
- Sills SC, Wood HS. Autologous activated platelet rich plasma injection into adult human ovary tissue: molecular mechanism, analysis and discussion of reproductive response. *Bioscience Reports.* 2019;BSR20190805.
- Vo TKC, Tanaka Y, Kawamura K. Ovarian rejuvenation using autologous platelet rich plasma. *Endocrine.* 2021;1:15-27.
- Dawood AS, Salem HA. Current clinical applications of platelet rich plasma in various gynecological disorders: An appraisal of theory and practice. *Club Exp Reprod Med.* 2018;45(2):67-74.
- Pantos K, Simopoulou M, Pantou A, Rapani A, Tsioulou P, Nitsos N, et al. A Case Series on Natural Conceptions Resulting in Ongoing Pregnancies in Menopausal and Prematurely Menopausal Women Following Platelet-Rich Plasma Treatment. *Cell Transplantation.* 2019;28(9-10):1333-40.
- Uddin MJ, Banu J, Ishrat S, Sultana S, Munira S, Shume MM, et al. Effect of autologous platelet rich plasma on anti mullerian hormonr and antral follicle count in subfertile women with poor ovarian reserve. *Int J reprod Contracept Obstet Gynecol.* 2022;1(2):292-8.
- La Marca A, Nelson SM, Siginolfi G, Manno M, Baraldi E, Roli L, et al. Anti-mullerian hormone based prediction model for a live birth in assisted reproduction. *Reprod BioMed Online.* 2011;22(4):341-49.
- Brodin T, Hadziosmanovic N, Burglund L, Olovsson M, Holte J. Antimullerian hormone levels are strongly associated with live birth rates after assisted reproduction. *J Clin Endocrinol Metab.* 2013;98(3):1107-14.
- Elnashar AB. Intra ovarian platelet rich plasma: current status. *Middle East Fertility Society Journal* 2020;1021;26:30.
- Melo P, Navarro C, Jones C, Coward K, Coleman L. The use of autologous platelet rich plasma (PRP) versus no intervention in women with low ovarian reserve undergoing fertility treatment: a non-randomized interventional study. *J Assist Reprod Genetics.* 2020;37:855-63.
- Petryk N, Petryk M. Ovarian Rejuvenation Through Platelet-Rich Autologous Plasma (PRP)—a Chance to Have a Baby Without Donor Eggs, Improving the Life Quality of Women Suffering from Early Menopause Without Synthetic Hormonal Treatment. *Reproductive Sciences.* 2020;27:1975-82.
- Sfakianoudis K, Simopoulou M, Grigoriadis S, Pontou A, Tsiolou P, Maziotis E, et al. Reactivating ovarian function through autologous platelet rich plasma intra ovarian infusion: pilot data on premature ovarian insufficiency, premenopausal,menopausal and poor responder women. *J Club med.* 2020;9:1809.
- Cakiroglu Y, Saltik A, Yuceturk A, Karaosmanoglu O, Kopuk SY, Scott RT, et al. Effects of intraovarian injection of autologous platelet rich plasma on ovarian reserve and ivf outcome parameters in women with primary ovarian insufficiency. *Aging.* 2020;12(11):11211.
- Farimani M, Nazari M, Mohammadi S, Aliabad RA. Evaluation of intra-ovarian platelet-rich plasma administration on oocytes-dependent variables in patients with poor ovarian response: A retrospective study according to the POSEIDON criteria. *Reproductive Biology and Endocrinology.* 2021;19:137.
- Fraison E, Crawford G, Casper G, Harris V, Ledger W. Pregnancy following diagnosis of premature ovarian insufficiency: a systematic review. *Reprod Biomed Online.* 2019;39(3):467.
- Practice Committee of American Society of Reproductive Medicine. Testing and interpreting

measures of ovarian reserve: a committee opinion.
Fertil Steril. 2020;114(6):1151-6.

19. Overveek A, Broekmans FJ, Hehenkamp WJ. Intracycle fluctuations of anti-Mullerian hormone in normal women with a regular cycle: a re- analysis. Reprod Biomed Online. 2012.

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