Platelet-rich plasma as an adjuvant in the endometrial preparation of patients with refractory endometrium

Aixa Molina1,2, Jose Sánchez1,2, William Sánchez1,2, Vanessa Vielma1,2

1Unidad de Asistencia Materno Reproductiva (UNAMATER) Mérida. Venezuela
2Instituto Venezolano de Fertilidad Caracas (IVF CARACAS) Caracas D.C. Venezuela

ABSTRACT
Objective: To improve endometrial quality and implantation rates after the administration of platelet-rich plasma in patients with refractory endometrium.

Methods: 19 patients undergoing in vitro fertilization, aged between 33 and 45 years with a history of refractory endometrium, to whom platelet rich plasma was given by infusion with a catheter into the uterine cavity on the tenth day of the hormone replacement therapy, and then 72 hours after the first administration.

Results: Endometrial thicknesses >7mm was reported with the first use; and in all cases, endometrial thickness >9mm were evident after the second administration. The entire study group qualified for Embryo Transfer at 72 hours after the first administration.

Conclusions: Platelet-rich plasma and its biostimulation effects on the endometrial microvasculature seems to be beneficial to patients with refractory endometrium, providing an increase in endometrial receptivity and a consequent increase in implantation rates. As an autologous resource, they are easy to obtain and inexpensive. Thus, we recommend it to be included in the different protocols for endometrial preparation, including those in which a natural cycle is preferred.

Keywords: Platelet-rich plasma, refractory endometrium, endometrial thickness

INTRODUCTION
The refractory endometrium, characterized by atrophy with endometrial interface measurements below 6 mm by ultrasound; and/or the presence of intrauterine adhesions and synechia through hysteroscopy, which is the so-called Asherman syndrome, continues to represent a serious problem for the practitioner who does the endometrial preparation for patients undergoing Assisted Reproductive Techniques (ART). This type of endometrial pattern is still responsible for many embryo transfer (ET) cancellations or implantation failures (Coughlan et al., 2014; Kasius et al., 2014). Refractory endometrium can be idiopathic, congenital, surgical (uterine curettage); caused by inflammatory processes, infections and radiation therapy, among other causes (Revel, 2012). These different etiological agents cause an important lesion on the basal layer of the endometrium, with consequent decrease in its regeneration capacity. Hormone replacement therapy (HRT) with estradiol valerate, has been registered in the endometrial preparation of infertile patients, requiring low and high complexity treatments. However, other drugs have been added to it such as: acetylsalicylic acid (aspirin); Sildenafil, vitamin E, GnRH agonists, HCG, L-arginine, pentoxifylline, and others, to ensure an adequate implantation window and to promote the synchronization between a prepared embryo to implant and a receptive endometrium (Qublan et al., 2008; Takasaki et al., 2010; Coughlan et al.; 2014). Recently, growth factors, mainly G-CSF, stem cells, PRP and bone marrow have been used as alternative treatment modalities; and the extreme resource of uterine transplantation has been tested (Ohl et al., 2002; Del Priore et al., 2013; Barad et al., 2014; Kunicki et al., 2014; Singh et al., 2014; Chang et al., 2015).

Originally, PRP was defined as "the volume of autologous plasma having a platelet concentration above the basal line". It was later defined as a group of soluble and diffusible polypeptide substances able to regulate the growth, differentiation and phenotype of numerous cell types (Vega et al., 2000). PRP is an autologous source of many factors and substances, especially platelet-derived growth factor and transforming growth factor-β. These growth factors released after platelet degranulation are those that provide the mechanisms required to produce the necessary biological synthesis for the tissue regeneration process (Grageda, 2004; Grageda et al., 2005).

A normal blood clot contains 93% red blood cells, 6% platelets and, in some cases, less than 1% of white blood cells. In contrast, a PRP clot contains 94% of platelets, only 5% of red blood cells and 1% of white blood cells. This change in the ratio of cells that do not stimulate regeneration (red blood cells) vis-à-vis those that do stimulate all phases of regeneration (platelets), explains its ability to reinforce tissue regeneration (Anitua et al., 2004; Marx, 2004; Marx & Garg, 2005; Sampson et al., 2008). The primarily biostimulation function of PRP is directly linked to the platelet release of growth factors. These factors have tissue regeneration inducing properties.

In their alpha granules, platelets contain a cocktail of chemical mediators that trigger the onset of the regenerative phase, both through the promotion of angiogenesis and the initiation of cell regeneration by mitotic mediators of mesenchymal cells, which is the "kick off cocktail". Platelet granules’ action is key in the move from the haemostatic to the regenerative phase. Their lysosomes contain many proteolytic enzymes, their dense granules contain prothrombotic factors and their α-granules a high content of pro-regenerative growth factors, among them PDGF and TGF-β. In addition, throughout their formation, platelets have the property of accumulating, by endocytosis, numerous molecules from the medium, among which are lipid mediators, such as sphingosine-1-phosphate (SPP), phosphatidic acid and lysophosphatidate. These substances have an anti-apoptotic effect on endothelial cells, they are involved in the chemotaxis of endothelial cells, and their proliferation and promotion of adhesion bonds form capillary-like structures. Sphingosine appears to be critical to the onset of angiogenesis, since it is the ligand of endothelial differentiation gene (EDG) receptors. Finally, the overexpression of VEGFR2 renders the endothelium sensitive to the production of VEGF (vascular endothelial growth factor) to initiate angiogenesis from endothelial cells (Romagnani et al., 2004; Paiva et al., 2011).

Growth factors are obtained by genetic and recombinant engineering procedures; however, they are expensive and repeated doses are required to achieve an optimal...
therapeutic effect. In contrast, PRP is easy to obtain, it is of low cost and contains a high concentration of growth factors, thus providing an excellent alternative source. Since PRP is an autologous preparation, therefore non-toxic and non-allergenic, it can be used in various medical conditions, as an adjuvant to conventional treatment with mostly satisfactory results. In the case of the endometrial preparation for ART and specifically on the refractory endometrium, not enough studies have been carried out; however, the few reported results demonstrate improvements with its use, in relation to an increase of endometrial thickness, as well as, an increase in implantation rates.

OBJECTIVE
To improve endometrial quality and implantation rates after the administration of platelet-rich plasma in patients with refractory endometrium undergoing in vitro fertilization + embryo transfer treatment in assisted reproduction units (UNAMATER) and (IVF Caracas) in the period from February 2016 to February 2017.

MATERIAL AND METHODS
Type of research
Prospective Design.

Physical environment
This study was carried out in the Maternal Reproductive Assistance Unit (in Spanish UNAMATER) in Merida, Venezuela and at the Venezuelan Institute of Fertility Caracas (in Spanish IVF Caracas) in Caracas D.C, Venezuela.

Study subjects
The study population consisted of 19 infertile patients undergoing High Complexity ART (In Vitro Fertilization), aged between 33 and 45 years with a previous history of refractory endometrium and at least one failed IVF attempt. To these patients, an autologous PRP serum was applied in a quantity of 1 cc by infusion with a catheter into the uterine cavity, on the tenth day of HRT with Estradiol Valerate, and again at 72 hours of the first administration (day 12 of HRT). Endometrial thickness was determined by transvaginal ultrasound.

Time frame
This study was carried out for one (1) year, from February 2016 to February 2017.

Selection criteria

Inclusion criteria
All patients with failed IVF, either performed in our Units of Human Assisted Reproduction or in External Units, who expressed a desire to undergo a new IVF+ET treatment, in whom preliminarily refractory endometrium was detected. During the period from February 2016 to February 2017.

Exclusion criteria
• Patients without unsuccessful IVF attempts.
• Patients with previous IVF but without refractory endometrium.

Resources

Human Resources
Clinicians, Embryologists, Anaesthesiologists, Nurses, Cardiologist, Psychologist, among others.

Institutional Resources
Assisted Human Reproduction Units: Maternal Reproductive Assistance Unit (UNAMATER) in Merida, Venezuela and Venezuelan Institute of Fertility Caracas (IVF Caracas) in Caracas D.C, Venezuela.

Data collection
The data was collected in a form especially designed for this study.

The patients included in this study consented with their signature and that of a witness; an additional written Informed Consent was used for the application of autologous PRP serum.

Statistical analysis of the information
After completing collection, the data was transcribed to a database and statistically analysed using the SPSS software version 20.0. The descriptive statistical results are presented in the form of tables of frequency distribution, figures and measurements.

RESULTS
In the population studied, endometrial thicknesses >7 mm was obtained with the first application of PRP, and in all cases endometrial thicknesses greater than 9 mm became evident after the second application of the autologous preparation. The entire study group qualified for ET in the blastocyst stage.

Of a total of 19 patients with refractory endometrium who underwent IVF+TE during the period between February 2016 and February 2017 and who received two PRP applications as adjuvant in the endometrial preparation, 14 positive pregnancy tests, which corresponded to 73.7%; and 5 presented negative pregnancy tests, which was equivalent to 26.3%. The average age of the patients studied was 39.21±3.22 years (range 33-45 years). When categorizing age values into two age groups, 57.9% (n=11) were grouped around 33-39 years and 42.1% (n=8) aged 40-45 years (Table 1).

| Age   | No. of Patients | %  | % collected |
|-------|----------------|----|-------------|
| 33    | 1              | 5.3| 5.3         |
| 34    | 1              | 5.3| 10.5        |
| 36    | 2              | 10.5| 21.1       |
| 38    | 4              | 21.1| 42.1       |
| 39    | 3              | 15.8| 57.9       |
| 40    | 2              | 10.5| 68.4       |
| 41    | 2              | 10.5| 78.9       |
| 42    | 1              | 5.3 | 84.2       |
| 44    | 2              | 10.5| 94.7       |
| 45    | 1              | 5.3 | 100.0      |
| Total | 19             |     | 100.0       |

With regards to the number of previous IVF attempts, 57.9% (n=11) had had one (1) previous IVF; followed by 31.6% with 2 previous IVF and 10.5% with 3 previous IVF (Table 2). In relation to oocyte origin, 68.4% (n=13) used their own oocyte and 31.6% (n=6) used oocyte donation. (Table 3).
In relation to the number of previous IVF and the result of the pregnancy test a 42.1% (n=8) with a positive test had a history of (1) previous IVF, 15% (n=3) with a negative pregnancy test. A 21.1% (n=2) with positive test presented (2) previous IVF vs. 10.5% (n=2) with negative pregnancy test. Finally, 10.5% (n=2) of the patients with positive pregnancy tests presented a history of (3) IVF previous vs. no case in patients with negative pregnancy tests (Table 8).

Regarding the type of pregnancy related to the number of previous IVF, the highest percentages of 21.4% (n=3) and 14.3% (n=2) of ongoing clinical pregnancies and live births corresponded to patients with one (1) previous IVF, followed by 14.3% (n=2) of live births and ongoing clinical pregnancies in patients with two (2) previous IVF. Biochemical pregnancies with 14.3% (n=2) and anembryonic pregnancy of 7.1% (n=1) in patients with (1) previous IVF. Fetal death at 16 weeks added up to 7.1% (n=1) in patients with three (03) previous IVF. (Table 9).

When referring to patient age in relation to the number of oocytes, the highest percentage was 52.6% (n=10) of own oocytes in the age group between 33-39 years old and 15.8% (n=3) of own oocytes in the group between 40 - 45 years. A 5.3% (n=1) oocyte donation in the age group of 33-40 years vs. 26.3% (n=5) oocyte donation in the age group of 40 - 45 years (Table 10).

When contrasting the age of the patients and the number of previous IVF, 42.1% (n=8) of the patients aged between 33-39 years had previous IVF, 10.5% (n=2) of the same age range group presented 2 previous IVF and 5.3% (n=1) of patients presented 3 previous IVF. Similarly, a 15.8% (n=3) of the patients between 40-45 years had a history of 1 previous IVF, 21.1% (n=4) of this age group presented 2 previous IVF and a 5.3% (n=1) had 3 previous IVF procedures (Table 11).

Regarding the age of the patient in relation to the result of the Pregnancy Test, 42.1% (n=8) of the patients between 33-39 years of age had a positive test compared to 15.8% (n=3) in this age group that had negative pregnancy tests. The 31.6% (n=6) of patients between 40 - 45 years of age had a positive test versus 10.5% (n=2) of negative pregnancy tests in the same age group (Table 12).

Finally, when correlating the age of the patients and type of pregnancy, we found that the highest percentages in both age groups corresponded to ongoing clinical pregnancies and live births, with a 14.3% (n=2) and a 21.4% (n=3) in the age group between 33-39 years. 21.4% (n=3) a 14.3% (n=2) respectively, for the age group 40-45 years. Biochemical gestation in the same percentage 7.1% (n=1) for both age groups. A 7.1% (n=1) anembryonic pregnancy and a fetal death of 7.1% (n=1) occurred only in the 33-39 age group (Table 13).

**DISCUSSION**

The main objective of this study was to improve endometrial quality and implantation rates after the administration of platelet-rich plasma in patients with refractory endometrium undergoing in vitro fertilization + embryo transfer treatment. In this sense, the detection performed for one year, showed improvements regarding endometrial thickness in the population studied, after the use of the autologous PRP. Regarding the implantation rates expressed in positive pregnancy tests, 73.7% (n=14) presented very satisfactory results, matching the findings obtained in the few previous serial clinical studies on the matter.

The main advantage of this detection would be the availability of an autologous resource of low cost, which offers obvious benefits regarding the development of a receptive endometrium that favours embryo implantation.
Table 5. Previous in-vitro fertilization attempts vs. Type of oocyte

| Previous in-vitro fertilization attempts | Type of oocyte | Total |
|----------------------------------------|----------------|-------|
|                                        | Own | %     | Donated | %     | No. | %     |
| 1                                      | 9   | 47.4  | 2       | 10.5  | 11  | 57.9  |
| 2                                      | 3   | 15.8  | 3       | 15.8  | 6   | 31.6  |
| 3                                      | 1   | 5.3   | 1       | 5.3   | 2   | 10.5  |
| Total                                  | 13  | 68.4  | 6       | 31.6  | 19  | 100.0 |

Table 6. Type of Oocytes vs. Pregnancy test

| Type of Oocytes | Pregnancy test | Total |
|----------------|----------------|-------|
|                | Positive | Negative |       |       |
|                | No. | %     | No. | %     | No. | %     |
| Own            | 8   | 42.1  | 5   | 26.3  | 13  | 68.4  |
| Donated        | 6   | 31.6  | 0   | 0.0   | 6   | 31.6  |
| Total          | 14  | 73.7  | 5   | 26.3  | 19  | 100.0 |

Table 7. Type of oocytes vs. type of pregnancy

| Type of pregnancy | Type of oocytes | Total |
|-------------------|----------------|-------|
|                   | Own | %     | Donated | %     | No. | %     |
| Ongoing pregnancies | 3   | 21.4  | 2       | 14.3  | 5   | 35.7  |
| Live births        | 2   | 14.3  | 3       | 21.4  | 5   | 35.7  |
| Biochemical pregnancies | 1   | 7.1   | 1       | 7.1   | 2   | 14.3  |
| Anembryonic pregnancy | 1   | 7.1   | 0       | 0.0   | 1   | 7.1   |
| Fetal death (16 weeks) | 1   | 7.1   | 0       | 0.0   | 1   | 7.1   |
| Total              | 8   | 57.1  | 6       | 42.9  | 14  | 100.0 |

Table 8. Previous in-vitro fertilization attempts vs. Pregnancy test

| Previous in-vitro fertilization attempts | Pregnancy test | Total |
|----------------------------------------|----------------|-------|
|                                        | Positive | Negative |       |
|                                        | No. | %     | No. | %     | No. | %     |
| 1                                      | 8   | 42.1  | 3   | 15.8  | 11  | 57.9  |
| 2                                      | 4   | 21.1  | 2   | 10.5  | 6   | 31.6  |
| 3                                      | 2   | 10.5  | 0   | 0.0   | 2   | 10.5  |
| Total                                  | 14  | 73.7  | 5   | 26.3  | 19  | 100.0 |

Chang et al. (2015), in a clinical series of 5 patients with endometrial factor associated with ET cancellation, performed PRP in addition to conventional HRT, achieving in all cases endometrium >7 mm, the total was submitted to ET with subsequent positive pregnancy tests in all patients. These results agree with the results obtained in our study. Garcia-Velasco et al. (2016) performed an extensive literature review on the management of refractory endometrium. The review included studies with conventional hormone preparations (estradiol valerate), as well as various drugs including acetylsalicylic acid (aspirin), sildenafil, vitamin E, GnRH agonists, HCG, L-arginine, pentoxifylline among others; as well as autologous preparations as growth factors, mainly G-CSF, stem cells, PRP and bone marrow. Finally, they concluded that despite the vast array of resources available today, it is still not easy to provide a pragmatic evidence-based approach that guides the clinician on how to improve refractory endometrium. In this sense, and in relation to the results of this study, it is considered important to complement them with the formal design of test protocols of the test versus test type, as well as, comparative studies to establish the efficacy between different drugs and preparations, besides the inclusion of several years of data collection.
Table 9. Previous in-vitro fertilization attempts vs Type of pregnancy

| Type of pregnancy       | Previous in-vitro fertilization attempts | Total |
|-------------------------|------------------------------------------|-------|
|                         | 1 No. | 2 No. | 3 No. | %    | No. | %    |
| Ongoing pregnancies    | 3     | 2     | 0     | 0.0  | 5   | 35.7 |
| Live births            | 2     | 2     | 1     | 7.1  | 5   | 35.7 |
| Biochemical pregnancies| 2     | 0     | 0     | 0.0  | 2   | 14.3 |
| Anembryonic pregnancy  | 1     | 0     | 0     | 0.0  | 1   | 7.1  |
| Fetal death (16 weeks) | 0     | 0     | 1     | 7.1  | 1   | 7.1  |
| Total                  | 8     | 4     | 2     | 14.3 | 14  | 100.0|

Table 10. Patients age vs. Type of oocytes

| Age     | Own Type of oocytes | Donated Type of oocytes | Total |
|---------|---------------------|-------------------------|-------|
|         | No. | %   | No. | %   | No. | %   |
| 33-39   | 10  | 52.6| 1   | 5.3 | 11  | 57.9|
| 40-45   | 3   | 15.8| 5   | 26.3| 8   | 42.1|
| Total   | 13  | 68.4| 6   | 31.6| 19  | 100.0|

Table 11. Patient age vs Previous in-vitro fertilization attempts

| IVF | Age     | 33-39 | 40-45 | Total |
|-----|---------|-------|-------|-------|
|     | No. | %   | No. | %   | No. | %   |
| 1   | 8   | 42.1| 3   | 15.8| 11  | 57.9|
| 2   | 2   | 10.5| 4   | 21.1| 6   | 31.6|
| 3   | 1   | 5.3 | 1   | 5.3 | 2   | 10.5|
| Total | 11  | 57.9| 8   | 42.1| 19  | 100.0|

Table 12. Patient age vs. Pregnancy test

| Test   | Age     | 33-39 | 40-45 | Total |
|--------|---------|-------|-------|-------|
|        | No. | %   | No. | %   | No. | %   |
| Positive| 8   | 42.1| 6   | 31.6| 14  | 73.7|
| Negative| 3   | 15.8| 2   | 10.5| 5   | 26.3|
| Total  | 11  | 57.9| 8   | 42.1| 19  | 100.0|

CONCLUSIONS AND RECOMMENDATIONS

PRP administration as an adjuvant in the endometrial preparation of patients with refractory endometrium, demonstrated a good sensitivity for increasing endometrial thickness, as well as, to obtain positive pregnancy tests, clinical pregnancies and live births. The use of the PRP preparation and its biostimulation effects on the endometrium microvascular endothelium seems to offer benefits to refractory endometrium, providing an increase in endometrial receptivity and a consequent increase in implantation rates. Being an autologous resource, therefore harmless to the patient, easy to obtain and of very low cost, it is recommended to be included in the different protocols for endometrial preparation in assisted reproduction techniques, even in those where the natural cycle is preferred, that is, without the requirement of hormone replacement therapy.

Further studies are recommended in terms of the number of population under study and the time frame evaluated, as well as the approach of comparative studies between drugs and autologous preparations, to envisage effective therapeutic alternatives for refractory endometrium. Despite having the latest resources, uterine transplantation, something unimaginable a while ago, arises today in response to fertility preservation as far as uterine factor is concerned. It is necessary to establish preliminary protocols with the use of a wide range of therapeutic alternatives, within which the autologous preparations are becoming increasingly important today based on the positive results obtained.
Table 13. Patient age vs. Type of pregnancy

| Type                      | Age   | Total |
|---------------------------|-------|-------|
|                           | 33-39 | 40-45 |       |
|                           | No.   | %     | No.   | %     | No.   | %     |
| Ongoing pregnancies       | 2     | 14.3  | 3     | 21.4  | 5     | 35.7  |
| Live births               | 3     | 21.4  | 2     | 14.3  | 5     | 35.7  |
| Biochemical pregnancies   | 1     | 7.1   | 1     | 7.1   | 2     | 14.3  |
| Anembryonic pregnancy     | 1     | 7.1   | 0     | 0.0   | 1     | 7.1   |
| Fetal death (16 weeks)    | 1     | 7.1   | 0     | 0.0   | 1     | 7.1   |
| Total                     | 8     | 57.1  | 6     | 42.9  | 14    | 100.0 |

CONFLICT OF INTEREST
No conflict of interest has been declared.

Corresponding author:
Aixa Maria Molina
Unidad de Asistencia Materno Reproductiva (UNAMATER)
Instituto Venezolano de Fertilidad Caracas (IVF CARACAS)
Venezuela.
E-mail: aixaaries@hotmail.com

REFERENCES
Anitua E, Andia I, Ardanza B, Nurden P, Nurden AT. Autologous platelets as a source of proteins for healing and tissue regeneration. Thromb Haemost. 2004;91:4-15. PMID: 14691563 DOI: 10.1160/TH03-07-0440

Barad DH, Yu Y, Kushner VA, Shohat-Tal A, Lazzaroni E, Lee HJ, Gleicher N. A randomized clinical trial of endometrial perfusion with granulocyte colony-stimulating factor in in vitro fertilization cycles: impact on endometrial thickness and clinical pregnancy rates. Fertil Steril. 2014;101:710-5. PMID: 24424357 DOI: 10.1016/j.fertnstert.2013.12.016

Chang Y, Li J, Chen Y, Wei L, Yang X, Shi Y, Liang X. Autologous platelet-rich plasma promotes endometrial growth and improves pregnancy outcome during in vitro fertilization. Int J Clin Exp Med. 2015;8:1286-90.

Coughlan C, Ledger W, Wang Q, Liu F, Demirol A, Gurgan T, Cutting R, Ong K, Sallam H, Li TC. Recurrent implantation failure: definition and management. Reprod Biomed Online. 2014;28:14-38. PMID: 24269084 DOI: 10.1016/j.rbmo.2013.08.011

Del Priore G, Saso S, Meslin EM, Tzakis A, Brännström M, Clarke A, Vianna R, Sawyer R, Smith JR. Uterine transplantation—a real possibility? The Indianapolis consensus. Hum Reprod. 2013;28:288-91. 2320299210.1093/humrep/des406

Garcia-Velasco JA, Acevedo B, Alvarez C, Alvarez M, Bellver J, Fontes J, Landeras J, Manau D, Martinez F, Muñoz E, Robles A, Rodriguez-Tabernero L. Strategies to manage refractory endometrium: state of the art in 2016. Reprod Biomed Online. 2016;32:474-89. PMID: 26947451 DOI: 10.1016/j.rbmo.2016.02.001

Grageda E. Platelet-rich plasma and bone graft materials: a review and a standardized research protocol. Implant Dent. 2004;13:301-9. PMID: 15591991 DOI: 10.1097/01.id.0000148558.91063.06

Grageda E, Lozada JL, Boyne PJ, Caplanis N, McMillan PJ. Bone formation in the maxillary sinus by using platelet-rich plasma: an experimental study in sheep. J Oral Implantol. 2005;31:2-17. PMID: 15751383 DOI: 10.1563/0-692.1

Kasius A, Smit JG, Torrance HL, Eijkemans MJ, Mol BW, Opmeer BC, Broekmans FJ. Endometrial thickness and pregnancy rates after IVF: a systematic review and meta-analysis. Hum Reprod Update. 2014;20:530-41. PMID: 24664156 DOI: 10.1093/humupd/dmu011

Kunicki M, Łukaszuk K, Wociatkiew-Potocka I, Liss J, Kulwickowska P, Szczypańska J. Evaluation of granulocyte colony-stimulating factor effects on treatment-resistant thin endometrium in women undergoing in vitro fertilization. Biomed Res Int. 2014;2014:913235. PMID: 24693540 DOI: 10.1155/2014/913235

Marx RE. Platelet-rich plasma: evidence to support its use. J Oral Maxillofac Surg. 2004,62:489-96. PMID: 15085519 DOI: 10.1016/j.joms.2003.12.003

Marx RE, Garg AK. Dental and craniofacial applications of platelet-rich plasma. Surrey: Quintessence Publishing Company; 2005.

Ohl J, Lefebvre-Maunoury C, Wittemer C, Nisand G, Laurent MC, Hoffmann P. Nitric oxide donors for patients undergoing IVF. A prospective, double-blind, randomized, placebo-controlled trial. Hum Reprod. 2002;17:2615-20. PMID: 12351537 DOI: 10.1093/humrep/der027

Paiva P, Hannan NJ, Hincks C, Meehan KL, Pruyser E, Dimitriadis E, Salamonsen LA. Human chorionic gonadotrophin regulates FGF2 and other cytokines produced by human endometrial epithelial cells, providing a mechanism for enhancing endometrial receptivity. Hum Reprod. 2011;26:1153-62. PMID: 21345913 DOI: 10.1093/humrep/der027

Qublan H, Aminar Z, Al-Qudah M, Diab F, Nawasreh M, Malkawi S, Balawneh M. Luteal phase support with GnRH-a improves implantation and pregnancy rates in IVF cycles with endometrium of <or=7 mm on day of egg retrieval. Hum Fertil (Camb). 2008;11:43-7. DOI: 10.1080/14647270701704768

Revel A. Defective endometrial receptivity. Fertil Steril. 2012;97:1028-32. PMID: 22542142 DOI: 10.1016/j.fertnstert.2012.03.039
Romagnani P, Lasagni L, Anunziato F, Serio M, Romagnani S. CXC chemokines: the regulatory link between inflammation and angiogenesis. Trends Immunol. 2004;25:201-9. PMID: 15039047 DOI: 10.1016/j.it.2004.02.006

Sampson S, Gerhardt M, Mandelbaum B. Platelet rich plasma injection grafts for musculoskeletal injuries: a review. Curr Rev Musculoskelet Med. 2008;1:165-74. PMID: 19468902 DOI: 10.1007/s12178-008-9032-5

Singh N, Mohanty S, Seth T, Shankar M, Bhaskaran S, Dharmendra S. Autologous stem cell transplantation in refractory Asherman's syndrome: A novel cell based therapy. J Hum Reprod Sci. 2014;7:93-8. PMID: 25191021 DOI: 10.4103/0974-1208.138864

Takasaki A, Tamura H, Miwa I, Taketani T, Shimamura K, Sugino N. Endometrial growth and uterine blood flow: a pilot study for improving endometrial thickness in the patients with a thin endometrium. Fertil Steril. 2010;93:1851-8. PMID: 19200982 DOI: 10.1016/j.fertnstert.2008.12.062

Vega JA, García-Suárez O, Martínez-Almagro A. Cartílago Articular y Factores de Crecimiento (primera parte). Mapfre Med. 2000;11:212-25.