Pregnancy Success Rate after IUI by Using Puresperm Density Gradient

Kyumars Safinejad Department of Biology, Broojerd branch, Islamic Azad University, Broojerd, Iran Email: Q_safinejad@yahoo.com

> Leila Yadegar Department of Biology, Payam Noor University, Qom, Iran Email: lyadegar@yahoo.com

Saber Safinejad Department of Human Genetics, Shafagh Pars clinic, Broojerd, Iran Email: md.saber_safinejad@yahoo.com

Abstract—One of the simplest ways of treatment of infertility is the intrauterine insemination (IUI) method, transferring the strengthened sperm into the uterus, as compared to the more developed methods, it is less expensive and it does not need anesthesia.

The aim of this research is to introduce the success of applying the IUI method by using of puresperm density gradient in the couples referring to Qom-Royan infertility center and its comparison with pregnancy rate by using other techniques of IUI method.

The day of ovulation, semen of male partner are processed by passing from puresperm density gradient; and the processed spermatozoa are transferred to the uterus of female partner. From 139 patients underwent IUI method by use of this technique pregnancy was happened in 28 cases. The pregnancy rates were 21.34, 23.33, 8.33 and 12.5 percent in patients who had unexplained infertility, sperm motility, sperm motility-morphology and cervical secretions problems respectively.

Our results showed that, 1) The best results obtained in cases who had unexplained infertility and sperm motility problems, 2) It can be increased, the percentage of success in IUI method by eliminating some couples who don't have the necessary conditions for performing IUI method.

Index Terms—percentage of pregnancy, IUI, infertile couples, pure sperm, Qom-Royan

I. INTRODUCTION

Infertility defined as problem in achieving a pregnancy within one year by regular coital exposure [1]. About 15% of couples are infertile, of which 40% are infertile because of male factor infertility and 40% are because of female factor infertility, and in the remainder, both factors are implicated [2]. Infertility and its treatment are psychologically stressful with virtually hundreds of articles published on this issue.

Intrauterine insemination (IUI) is a popular treatment for infertility around the world. The return from treatment, like most fertility treatments is dependent upon the etiology and the age of the woman. The laboratory techniques and equipment required for IUI are less intensive than that required for in vitro fertilization (IVF). Hence, particularly in countries where there is only modest or no health fund reimbursement for fertility treatment, IUI is usually the first-line choice for assisted conception. Insemination (IUI), in which male sperm are injected into the mother, have been available for a long time and are still widely used. Prescription fertility drugs, which affect a woman's ovulation and fertility, have been used since at least the 1970's.

IUI is a simple first line treatment for infertile couples [3]. This inexpensive treatment, in comparison with other assisted reproductive techniques (ART), has been widely used to treat infertile couples with a variety of indications such as male subfertility, unexplained fertility, cervical mucus hostility and endometriosis related infertility. [4]-[9] Pregnancy rates after IUI differ between studies according to patient selection criteria, the presence of various infertility factors, ovarian stimulation methods, number of cycles performed, different sperm parameters, and preparation technique. IUI can be used with ovarian stimulation, which increases the number of available oocytes at the site of conception, and increase pregnancy rates than IUI alone. [10]-[13] Diagnostic laparoscopy is the standard procedure in diagnosing infertility. For infertile patients where pathologic abnormalities are considered, COH and IUI should be performed after the laparoscopic surgery.

Some couples solve their problem with regular clinical treatments. However, it is untreatable in 48% of them and they need special laboratory techniques called ART. There have been interests in cognitive sciences, neuronal networks, fuzzy theory and statistical neuronal models as the most effective tools in prediction problems.

There are several methods for treatment of infertility, one of which is IUI. In spite of high relative invariant prevalence of infertility, give service to this infertile couple has increased considerably in recent years. In

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contrast to other methods recommended for infertility treatment, the IUI method is simpler, cheaper and it dosen't need anesthesia [14].

Preparation of spermatozoa by use of concentration gradient of pure sperm technique is recently available as an alternative to percoll technique. Sperm samples prepared by pure sperm density gradient resulted in a significant decrease in the percentage of fragmented DNA in spermatozoa as shown by TUNEL assay and improve sperm quality. The IUI method is one of the infertility treatment methods in which the spermatozoa processed and transferred to uterus of female partner directly through certain catheter [15].

In the coital operation partial number of spermatozoa passed through cervical canal and reach to the uterus; but in IUI method many spermatozoa are entered into the uterus, so chance of fertility is increased. In general IUI is useful in below cases: 1) When female partner has cervical secretion problems, 2) When male partner cannot ejaculate in vaginal canal because of psychological problems, 3) When semen fluid of male partner contain anti sperm antibodies [16], 4) When semen retrogrades to bladder instead of depletion through the penis, 5) When the cause of infertility has not been recognized, 6) Before expensive advanced infertility methods such as IVF and ICSI.

The aim of this study is to report of success of IUI method by the use of pure sperm concentration gradient technique in the infertile couples referring to the Qom-Royan infertility center in 2014; and its comparison with the world pregnancy rate with IUI method.

II. MATERIAL AND METHODS

A total of 139 semen samples of patients undergoing IUI therapy were evaluated according to WHO (1999), except for morphology which was evaluated by strict criteria [17].

First in day 1-3 of menstrual cycle, the female partner examined with vaginal ultrasound and administration of fertility drugs such as clomiphene citrate, tamoxifen, HMG, initiated by physician diagnosis, then vaginal ultrasound is used for determining of size of follicles in female partner; and when the prominent follicle reaches 18-20 mm size, hCG injected and about 34-36 hours after the hCG injection, the eggs are retrieved.

On the day of insemination, a semen specimen was collected in a sterile container following 3-5 days of sexual abstinence. After liquefaction at room temperature, initial volume, viscosity, mobility, progression, and sperm count were assessed The semen samples were processed by pure sperm and the motile spermatozoa separated by passing from pure sperm concentration gradient and followed by trial washing and is placed in co2 incubator for one hour to get capacity and been strengthened. Intrauterine insemination was performed using an intrauterine catheter with a 1- or 2-ml syringe, 36_4 hours after HCG injection. The catheter was gently passed through the cervical canal and the sperm suspension slowly pushed into the uterine cavity. Insemination volume ranged from 0.5-2.0 ml. The

catheter was subsequently withdrawn and the patient remained in the supine position for 15 min after insemination.

In couples under IUI treatment infertility causes, include: 1) unexplained causes (when infertility cause is not detected by laboratory tests), 2) problems of motility (when motility of spermatozoa is between 20-40 percent), 3) problems of motility and morphology (when motility and morphology of spermatozoa are 20-40 percent and 10-20 percent respectively), 4) problems of cervical secretions (when anti sperm antibodies are found in cervical secretions which may reduce sperm motility or kill them). This research is a cross-sectional study of descriptive kind and place of this study was in Qom-Royan infertility and Broojerd Shafagh Pars genetics and infertility centers.

III. RESULTS AND DISCUSSION

From the 139 cases, in 19 out 89 (21.34%) with unexplained infertility, 7 out 30 (23.33%) with sperm motility problems, 1 out 12 (8.33%) with sperm motility and morphology problems and 1 out 8 (12.5%) with cervical secretions problems pregnancy occurred (Table I).

Infertility is one of the most significant problems that affect the health of families and societies. Determining the prevalence of infertility is important for evaluating the capacity of fertility in a society.

 TABLE I.
 PREGNANCY RATE IN INFERTILE COUPLES WITH DIFFERENT CAUSES IN THIS STUDY

Infertility causes	Number of females under treatment	Number of pregnant females	Pregnancy rate
Unexplained causes	89	19	21.34
sperm motility problems	30	7	23.33
sperm motility and morphology problems	12	1	8.33
cervical secretions problems	8	1	12.5

Infertility is a condition that individuals have less control on it. Studies have shown that individuals with infertility problems experience psychosocial problems including anxiety, depression, social isolation, loneliness, social stigmatization, sexual dysfunction and low selfesteem. The use of various ways of coping seems to have different impact on women's infertility stress. Nearly 14% of couples have infertility. 15-25% of infertile women have unexplained infertility. The principal methods for treatment unexplained infertility are IUI and IVF. GnRH antagonists and GnRH agonists and Gonadotropins which used in IUI and IVF cycles have different effect on serum prolactin level.

In general, semen analysis is basic test for infertile male partner. In semen analysis volume, count, motility and morphology of spermatozoa per ml should be considered [18]. Introduction: Intrauterine insemination is used to treat a variety of conditions, such as diminished sperm quality and 'cervical hostility'. Most agree that a minimum number of motile sperm is necessary for pregnancy after insemination. Very little, however, has been reported whether or not an upper limit exists. Preparation of spermatozoa by use of concentration gradient of pure sperm technique provided motile spermatozoa more than use of other techniques such as simple washing and swim up [19]. The previous studies and results obtained from this study showed that use of pure sperm solution technique for sperm processing for IUI method enhance the chance of pregnancy more than other methods. One of the first treatment drugs for stimulation of ovulation is clomiphene citrate; which is week artificial estrogen that be used as antagonist of estrogen in pharmacological dose for induction of ovulation; but its effects even in cases with good ovulation, have not been recognized precisely. This drug in hypothalamus has anti estrogenic effect; recent studies show that this effect increase GNRH waves in some female partners [20].

The pregnancy rate is high by use of clomiphene citrate; but for some patients with HH (hypogonadotropic hypogonadism), HMG is selected drug. For prevention of backward movement of sperm suspension, insemination is done with 0.3 to 0.5 ml volume. When the size of dominant follicle reaches 18 to 20 mm, ovulation induction with 1000 to 5000 IU of HCG is done. Thereafter, insemination is done within 38 to 40 hours [21].

In general the use of gonadotropin preparations to treat fertility problems has been known since the first half of the 20th century [22]. The IUI methods as compare to other methods are well studied. Male factor infertility is one of general indications for IUI treatment. The Allinson's beliefs that sperm processing before IUI, cause to get capacitation of spermatozoa; and capacitation is completed in female body so fertilization is increased [23]. Previous studies showed that IUI has good results in patients with anti-sperm antibodies; also without induction by HMG success is low even in patients with unexplained infertility [24]. The previous studies showed that defects that untreated by surgery may be treated by IUI method [15].

Intrauterine insemination using the male partner's sperm is commonly performed to overcome factors, as well as to increase the probability of conception in diverse etiologies [7], [11], [15]. Therefore, several semen parameters have been evaluated as predictors of successful outcome with intrauterine insemination. This study also sought to evaluate the effects of sperm characteristics on IUI-related pregnancy and also attempted to determine good parameters to predict pregnancy rate. A wide range success in achieving pregnancy was reported IUI for various indications [11]. IUI is a viable, simple, noninvasive, and inexpensive option, particularly for male-factor infertility patients. The accurate prediction of IUI success is important for many reasons: 1) to appropriately counsel the couple about their chances of conception using the different ARTs available (IUI or IVF with or without ICSI) and help them make the right decision; 2) to avoid conducting a procedure on a couple knowing in advance that its potential benefit is slim; 3) to avoid the economic, emotional, and psychological trauma of repetitive failed IUI cycles; and 4) to save time, which is a crucial factor in infertility management, by directing the couple toward more effective techniques (IVF with or without ICSI), particularly for those with advanced maternal age.

Previous studies and our study showed that male factor infertility with problem of sperm motility and selected couples and unexplained infertile couples are the best cases for infertility treatment with IUI method because enhance chance of fertility considerably [24], [25].

Pregnancy rate per cycle in male factor infertility in our study was 20%. This is more than 8 and 16% with other techniques such as percoll methods.in the literature and it seems that our result is acceptable, and comparison to other studies revealed that we have significantly higher pregnancy rate [26].

One of our aims in this study was to clarify the relationship between semen parameters in semen analysis and pregnancy rate. IUI is the first line treatment for unexplained infertility. However there is some sperm preparation method but none of them are ideal.

The sperm count and the total motile sperms count after processing both had significant relationship with pregnancy rate.

Campana [24] and Brasch [25] showed significant relationship between total motile sperms and pregnancy rate. Also, the pregnancy rate with total motile sperms greater than 20×10^6 was higher than those with less than 20×10^6 total motile sperms. We recommend the semen processing methods with higher rate of sperm preservation and higher rate of motility improvement to be used.

IV. CONCLUSION

The pregnancy rate with IUI method can be increased by use of pure sperm concentration gradient technique and eliminating some couples who don't have the necessary conditions for performing of IUI method. The probability of pregnancy increases by the following factors: higher sperm count and total motile sperms in processed sample, repetitive treatment cycles, and lower duration of infertility. Hence, improvement in sperm processing methods that yields to increased sperm count and total motile sperm count, cause higher pregnancy rate with IUI.

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REFERENCES

- P. S. Emlmar, *High Yield Obstertics and Gynecology*, Lippincott Williams & Wilkins, 2001, pp. 131-135.
- [2] F. N. Shamma and A. H. DeCherney, "Infertility: A Historical Perspective," in *Infertility: Evaluation and Treatment*, W. R. Keye, R. J. Chang, R. W. Rebar, M. R. Soules, Eds. Philadelphia, Pa: WB Saunders, 1995, pp. 3-7.
- [3] Y. Zhao, N. Vlahos, D. Wyncott, C. Petrella, J. Garcia, H. Zacur, and E. E. Wallach, "Impact of semen characteristics on the success of intrauterine insemination," *J Assist Reprod Genet*, vol. 21, no. 5, pp. 143-148, 2004.
- [4] B. J. Van Voorhis, M. Barnett, A. E. Sparks, C. H. Syrop, G. Rosenthal, and J. Dawson, "Effect of the total motile sperm count on the efficacy and costeffectiveness of intrauterine insemination and *in vitro* fertilization," *Fertil Steril*, vol. 75, no. 4, pp. 661-668, 2001.
- [5] R. P. Dickey, R. Pyrzak, P. Y. Lu, S. N. Taylor, and P. H. Rye, "Comparison of the sperm quality necessary for successful intrauterine insemination with World Health Organization threshold values for normal sperm," *Fertil Steril*, vol. 71, no. 4, pp. 684-689, 1999.
- [6] R. P. Dickey, S. N. Taylor, P. Y. Lu, B. M. Sartor, P. H. Rye, and R. Pyrzak, "Effect of diagnosis, age, sperm quality, and number of preovulatory follicles on the outcome of multiple cycles of clomiphene citrate-intrauterine insemination," *Fertil Steril*, vol. 78, no. 5, pp. 1088-1095, 2002.
- [7] A. J. Goverde, J. McDonnell, J. P. Vermeiden, R. Schats, F. F. Rutten, and J. Schoemaker, "Intrauterine insemination or *in vitro* fertilisation in idiopathic subfertility and male subfertility: A randomised trial and cost-effectiveness analysis," *Lancet*, vol. 355, no. 9197, pp. 13-18, 2000.
- [8] M. Zafar, T. Jameel, and K. N. Abdullah, "Impact of intrauterine insemination as first line treatment of subfertility," *J Pak Med Assoc.*, vol. 57, no. 3, pp. 133-136, 2007.
- [9] S. Bhattacharya and M. Hall, "Cost-effective treatment of couples with subfertility," *Lancet*, vol. 355, no. 9197, p. 2, 2000.
 [10] B. J. Cohlen, P. Vandekerckhove, E. R. Velde, and J. D. Habbema,
- [10] B. J. Cohlen, P. Vandekerckhove, E. R. Velde, and J. D. Habbema, "Timed intercourse versus intrauterine insemination with or without ovarian hyperstimulation for subfertility in men," *Cochrane Database Syst Rev.*, vol. 2, no. 2, p. CD000360, 2000.
- [11] D. S. Guzick, S. A. Carson, C. Coutifaris, *et al.*, "Efficacy of superovulation and intrauterine insemination in the treatment of infertility," *N Engl J Med.*, vol. 340, no. 3, pp. 177-183, 1999.
- [12] J. L. Hwang, L. W. Huang, B. C. Hsieh, *et al.*, "Ovarian stimulation by clomiphene citrate and hMG in combination with cetrorelix acetate for ICSI cycles," *Hum Reprod*, vol. 18, no. 1, pp. 45-49, 2003.
- [13] R. Sikandar, S. Virk, S. Lakhani, H. Sahab, and J. Rizvi, "Intrauterine insemination with controlled ovarian hyperstimulation in the treatment of subfertility," *J Coll Physicians Surg Pak*, vol. 15, no. 12, pp. 782-785, 2005.
- [14] A. R. Martiniez, R. E. Bernardus, et al., "Basic question on in trauterne inseminations and update," Obstet Gyncol Surv, vol. 48, pp. 811-828, 1993.
- [15] G. M. D. Sher, D. M. Virginia, R. N. M. N. Jean, *In vitro fertilization*, M. A., 1998, pp. 146-150.
- [16] T. F. Kruger and Coetzee, "The role of sperm morphology in assisted reproduction," *Hum. Reprod. Update*, vol. 5, pp. 172-178, 1999.
- [17] J. F. Griffin and S. R. Ojeda, *Text Book of Enocrine Physiology*, New York: Oxford University Press, 1996, pp. 234-245.
- [18] I. G. Dwimn, P. Meniru, and R. Brinsden, Hand Book of IUI, Craft, 1997, pp. 4-19.
- [19] G. K, Beruram, Basic Clinical Pharamacology. M. C. Graw-Hill, 2001, pp. 699-701.
- [20] G. Prietl, H. van der Ven, and D. Krebs, "Homologous intrauterine in semination," in *Manual on Assisted Reproduction*, T. Rabe, K. Diedrich, B. Runnebaum, Eds. Berlin: Springer-Verlag, 1997, pp. 178-203.
- [21] M, F. Ludwig, R. E. Diedrich, *et al.*, "Ovarian stimulation from basic science to clinical application," *Repord Biomed on Line*, vol. 5, no. Suppl. 1, pp. 73-86, 2002.

- [22] G. Sher, D. Marriage, and J. Stoess, *In Vitro Fertilization*, Facts on Fileinc, 1998, pp. 146-149.
- [23] A. Campana, D. Sakkas, A. Stalberg, and P. G. Bianchi, "Intrauterine inseminiation: Evaluation of the results recording to the womens age, sperm quality, total sperm count per insemination and life table analysis," *Hum Reprod*, vol. 11, no. 4, pp. 732-760, 1996.
- [24] J. H. Chek, A. Bollendrof, and M. Z|accardo, "Insemination and male factor withot superovulation," *Arch Andiol*, vol. 35, no. 2, pp. 135-141.
- [25] P. J. Turek, "Male Infertility," in *Smith's General Urology*, E. A. Tanagho, J. W. McAninch, Eds. 15th ed. NY: Lang Medical Books/McGraw-Hill, 2000, pp. 759-763.
- [26] J. G. Brasch, R. Rawlins, S. Tarchala, and E. Radwanska, "The relationship between total motile sperm count and the success of intrauterine insemination," *Fertil Steril*, vol. 62, pp. 150-154, 1994.



Kyumars Safinejad was born in Asadabad, Iran on 21 September, 1970. His academic background is as follows, B.Sc: microbiology, Qom University, Iran (1992-1996); M.Sc: genetics, Tehran University, Iran (1997-1999) and Ph.D: molecular genetics, Shiraz University, Iran (2007-2011). From 2000 to 2003 he has been a member of genetics and embryology department of Royan institute. Since 2003, he has been a professor at the

Department of Genetic, College of Basic science, Islamic Azad University, Boroujerd Branch, Boroujerd, Iran. He is fluent in infertility treatment, genetic methods and genetic conselling. He recently published a number of articles that include: 1) Kyumars Safi nejad, Nejat Mahdieh, Parvin Mahdipour, Leila Yadegar, Morteza Atri and Gholamreza Javadi, "Sister chromatid exchange in peripheral blood lymphocytesas a possible breast cancer risk biomarker: A study of Iranian patients with breast cancer," Egypt. J. Med. Hum. Genet., vol. 10, no. 1, May, 2009. 2) Kyumars Safinejad, Mojtaba Darbouy, Sayed Mahdi Kalantar, Sirus Zeinali, Reza Mirfakhraie, Leila Yadegar, and Masoud Houshmand, "The prevalence of common CFTR mutations in Iranian infertile men with non-CAVD obstructive azoospermia by using ARMS PCR techniques," J Assist Reprod Genet., vol. 28, no. 11, pp. 1087-1090, Nov. 2011. 3) Kyumars Safinejad, Leila Yadegar, Mojtaba Darbouy, Mohammad Javad Soltani banavandi, Saber Safinejad, and Mohsen Razani, "Common CFTR Polymorphisms in Infertile Men With Non CAVD Obstructive Azoospermia," J. Basic. Appl. Sci. Res., vol. 3, no. 9, pp. 228-232, 2013. 4) Kyumars Safinejad, Leila Yadegar, Massoud Houshmand, Reza Mirfakhraie, Esmaeel Mohammadi Pargoo, "Y chromosome Microdeletions in Infertile Men with Sever Oligozoospermia," J. Basic. Appl. Sci. Res., vol. 3, no. 2, pp. 786-791, 2013. His research interests are infertility & genetics, abridge.



Leila Yadegar was born in Broojerd Iran on 7 December, 1975. Her academic background is as follows, B.Sc: biology, Ahvaz Shahid Chamran University, Iran (1995-1999); M.Sc: zoology, Tehran University, Iran (2004-2007), Since 2005, she has been a master at the Department of Biology, Payam Noor University, Qom, Iran.

She recently published a number of articles including: 1) Kyumars Safinejad, Nejat Mahdieh, Parvin Mahdipour, Leila Yadegar,

Morteza Atri, and Gholamreza Javadi, "Sister chromatid exchange in peripheral blood lymphocytesas a possible breast cancer risk biomarker: A study of Iranian patients with breast cancer," *Egypt. J. Med. Hum. Genet.*, vol. 10, no. 1, May, 2009. 2) Kyumars Safinejad, Mojtaba Darbouy, Sayed Mahdi Kalantar, Sirus Zeinali, Reza Mirfakhraie, Leila Yadegar, and Masoud Houshmand, "The prevalence of common CFTR mutations in Iranian infertile men with non-CAVD obstructive azoospermia by using ARMS PCR techniques," *J Assist Reprod Genet.*, vol. 28, no. 11, pp. 1087-1090, 2011. 3) Kyumars Safinejad, Leila Yadegar, Mojtaba Darbouy, Mohammad Javad Soltani Banavandi, Saber Safinejad, and Mohsen Razani, "Common CFTR polymorphisms in infertile men with non CAVD obstructive Azoospermia," *J. Basic. Appl. Sci. Res.*, vol. 3, no. 9, pp. 228-232, 2013. 4) Kyumars Safinejad, Leila Yadegar, Massoud Houshmand, Reza Mirfakhraie, Esmaeel Mohammadi Pargoo, "Y chromosome Microdeletions in Infertile Men with Severe Oligozoospermia," *J. Basic. Appl. Sci. Res.*, vol. 3, no. 2, pp. 786-791, 2013.



Saber Safinejad was born in Qom Iran on 29 October, 1992. His academic background is as follows, Doctor of Veterinary Medicine (D.V.M), Tehran University, Iran, Since 2013, he has been an employee at the Department of Human Genetics, Shafagh Pars Clinic, Broojerd, Iran. He recently published articles including: Kyumars Safinejad, Leila Yadegar, Mojtaba Darbouy, Mohammad Javad Soltani

banavandi, Saber Safinejad, and Mohsen Razani, "Common CFTR polymorphisms in infertile men with non CAVD obstructive Azoospermia", J. Basic. Appl. Sci. Res., vol. 3, no. 9, pp. 228-232, 2013.