

Diabetes and pregnancy outcomes in retrograde ejaculation by sperm source and aetiology: an exploratory ICSI study

Haitian Ding

First Affiliated Hospital of Anhui Medical University

Ruijun Liu

Taian City Central Hospital

Sana Atta

First Affiliated Hospital of Anhui Medical University

Yuqin Hu

The 901st Hospital of the Joint Logistics Support Force of PLA

Aylla Qayyum

First Affiliated Hospital of Anhui Medical University

Dongdong Tang

First Affiliated Hospital of Anhui Medical University

Hao Geng

First Affiliated Hospital of Anhui Medical University

Ping Zhou

First Affiliated Hospital of Anhui Medical University

Zhaolian Wei

First Affiliated Hospital of Anhui Medical University

Yuping Xu

First Affiliated Hospital of Anhui Medical University

Feng Ni

The 901st Hospital of the Joint Logistics Support Force of PLA

Yunxia Cao

First Affiliated Hospital of Anhui Medical University

Yingchun Liu

`efy102204@fy.ahmu.edu.cn`

Second Affiliated Hospital of Anhui Medical University

Keywords: retrograde ejaculation, pregnancy outcome, intracytoplasmic sperm injection, embryo transfer, testicular sperm, diabetes mellitus

Posted Date: July 13th, 2026

DOI: <https://doi.org/10.21203/rs.3.rs-10181784/v1>

License: © ⓘ This work is licensed under a Creative Commons Attribution 4.0 International License.
[Read Full License](#)

Additional Declarations: No competing interests reported.

Diabetes and pregnancy outcomes in retrograde ejaculation by sperm source and aetiology: an exploratory ICSI study

Haitian Ding^{a,b,c,h}, Ruijun Liu^{e,h}, Sana Atta^{a,d,h}, Yuqin Hu^f, Aylla Qayyum^{a,d}, Dongdong Tang^{a,b,c}, Hao Geng^{a,b,c}, Ping Zhou^{a,b,c}, Zhaolian Wei^{a,b,c}, Yuping Xu^{a,b,c}, Feng Ni^{f,*}, Yunxia Cao^{a,b,c,*}, Yingchun Liu^{g,*}

^a Reproductive Medicine Center, Department of Obstetrics and Gynecology, the First Affiliated Hospital of Anhui Medical University, No 218 Jixi Road, Hefei 230022, China

^b NHC Key Laboratory of Study on Abnormal Gametes and Reproductive Tract (Anhui Medical University), No 81 Meishan Road, Hefei 230032, China

^c Key Laboratory of Population Health Across Life Cycle (Anhui Medical University), Ministry of Education of the People's Republic of China, No 81 Meishan Road, Hefei 230032, China

^d Anhui Province Key Laboratory of Reproductive Health and Genetics, No 81 Meishan Road, Hefei 230032, China

^e Affiliated Taian City Central Hospital of Qingdao University, Taian 271000, China

^f Reproductive Medicine Center, The 901st Hospital of the Joint Logistics Support Force of PLA, Hefei 230031, Anhui, China.

^g Reproductive Medicine Center, Department of Obstetrics and Gynecology, the Second Affiliated Hospital of Anhui Medical University, Hefei 230601, China

^h These authors contributed equally to this work.

*Corresponding author:

*Feng Ni, Reproductive Medicine Center, The 901st Hospital of the Joint Logistics Support Force of PLA, Hefei 230031, Anhui, China. Email: 20070208nifeng@163.com

*Yunxia Cao, Reproductive Medicine Center, Department of Obstetrics and Gynecology, the First Affiliated Hospital of Anhui Medical University, No 218 Jixi Road, Hefei 230022, China. Email: caoyunxia5972@ahmu.edu.cn

*Yingchun Liu, Reproductive Medicine Center, Department of Obstetrics and Gynecology, the Second Affiliated Hospital of Anhui Medical University, Hefei 230601, China. Email: efy102204@fy.ahmu.edu.cn

Abstract

Background: Retrograde ejaculation is a rare cause of male infertility, and the optimal sperm source for intracytoplasmic sperm injection and the impact of diabetes on assisted reproductive outcomes remain unclear. This retrospective multicentre study included 79 patients with retrograde ejaculation undergoing intracytoplasmic sperm injection–embryo transfer to exploratorily evaluate pregnancy outcomes comparing testicular versus ejaculated sperm and diabetic versus non-diabetic aetiology.

Results: The testicular sperm group showed numerically higher implantation (30.6% vs. 23.7%, $P=0.240$) and clinical pregnancy rates (54.0% vs. 39.4%, $P=0.092$), with a trend toward lower miscarriage (8.8% vs. 28.6%, $P=0.053$) compared to the ejaculated sperm group. The non-diabetic group had significantly higher implantation (33.3% vs. 19.4%, $P=0.020$) and clinical pregnancy rates (54.8% vs. 34.5%, $P=0.020$) than the diabetic group. Diabetes was identified as an independent risk factor for pregnancy failure (odds ratio, 3.35; 95% confidence interval, 1.58–7.11; $P=0.002$). The use of ejaculated sperm showed a non-significant association with higher failure risk (odds ratio, 2.02; $P=0.061$).

Conclusions: In this exploratory analysis, diabetes was identified as an independent predictor of poorer intracytoplasmic sperm injection outcomes in patients with retrograde ejaculation. Although numerical trends favored testicular sperm, no definitive conclusion can be drawn due to the limited sample size. Larger prospective studies are needed to confirm these findings.

Keywords: retrograde ejaculation; pregnancy outcome; intracytoplasmic sperm

injection; embryo transfer; testicular sperm; diabetes mellitus

Introduction

The global incidence of infertility is approximately 8%–12%, half of which is attributed to male factors [1]. Retrograde ejaculation (RE) accounts for 0.3%-2% of male infertility [2]. RE is a disorder in which some or all male semen flows back into the bladder during ejaculation due to the failure to close the bladder neck (Fig 1). Patients often report ejaculation during orgasm without feeling semen discharged through the urethra, or only a small amount [3].

The symptoms of early incomplete RE are not obvious and patients tend to ignore the occurrence of the disease. Full-blown RE can lead to infertility and is the main reason for consultation in couples of reproductive age. Its aetiology includes lesions in the bladder neck anatomy and neuropathy caused by several factors, such as diabetes [4, 5]. RE can be treated using single or combined approaches involving drugs [6] and surgery [7-9] to restore neuromodulation and normal anatomy of the bladder neck.

When antegrade ejaculation cannot be re-established, sperm must be retrieved using an alternative technology to facilitate assisted pregnancy [10]. The choice of assisted reproductive therapy is generally based on the condition of the recipient female and the quantity and quality of sperm obtained from the male donor. Intracytoplasmic sperm injection (ICSI) is only marginally affected by sperm parameters, fallopian tube-related factors, and other variables. It has become the leading treatment for infertility caused by male factors such as severe oligoasthenospermia, teratozoospermia, and obstructive azoospermia [11].

Currently, the investigation of pregnancy outcomes in patients with retrograde

ejaculation (RE) undergoing intracytoplasmic sperm injection-embryo transfer (ICSI-ET) is mainly based on insights from case reports or small-sample studies. This exploratory study aimed to evaluate these outcomes in a cohort of patients with RE, with a focus on comparing different sperm sources (testicular vs. ejaculated) and aetiologies (diabetic vs. non-diabetic), while acknowledging the limited sample size and the need for cautious interpretation.

Materials and methods

Ethical approval

This study was conducted in accordance with the ethical standards of the Declaration of Helsinki and ICMJE recommendations. Ethical approval was granted by the Ethics Committee of a university-affiliated hospital in China (reference number: P2020-12-36). Additional approval was obtained from the Ethics Committee of a collaborating hospital through a multicentre ethical review collaboration mechanism in accordance with regional guidelines. All participants provided written informed consent after receiving detailed explanations regarding study objectives, potential risks and benefits, and data usage protocols. Personal identifiers were removed using pseudonymization before analysis, and all clinical data were stored in secure, encrypted databases.

Patients and controls

We retrospectively analysed 79 patients diagnosed with RE who underwent ICSI

between August 2001 and August 2025. Of these, 59 patients were treated at the Reproductive Center of a university-affiliated hospital in a city of China, and 20 patients were enrolled from another collaborating hospital in the same city, as part of a multicentre collaboration. The inclusion criteria were as follows: 1) couples younger than 40 years with a normal chromosomal karyotype; 2) the male partner had no genital abnormalities and agreed to provide a medical history, including general health conditions, surgery, and diabetes, to facilitate a preliminary diagnosis of RE; 3) no abnormalities in the female reproductive system; and 4) normal general examination results before initiating assisted reproduction in both partners. The exclusion criteria were as follows: 1) male with no ejaculation, dry ejaculation, other ejaculation disorders, obstructive azoospermia, and oligoasthenospermia caused by other reasons; 2) abnormal sex hormone levels in both spouses. According to the aetiology of RE, patients were divided into diabetic ($n = 33$) and non-diabetic ($n = 46$) groups. According to the sources of ICSI sperm, patients were divided into a testicular sperm group ($n=39$) and an ejaculated sperm group ($n=44$). A control group of 200 men with oligozoospermia or obstructive azoospermia was also included for comparison (Fig 2). Pregnancy outcomes, including blastocyst formation, embryo implantation, and pregnancy rates, were compared.

Due to national regulations requiring legal marriage for assisted reproductive technology in China, each male patient was paired with his female partner. Therefore, the number of female patients was identical to male patients in all analyses (79 women in the RE group, 33 in the diabetic subgroup, 46 in the non-diabetic subgroup, and 200

women in the control group). No separate female cohort analysis was required.

Diabetes subtype was not formally classified because this was a retrospective study without endocrinologist assessment, autoantibody testing, or C-peptide measurement. Based on medical records showing adult onset and absence of childhood diabetes, type 2 diabetes was considered the most probable diagnosis, but this remains a limitation.

Semen and urine analysis

To diagnose retrograde ejaculation, each patient was instructed to empty the bladder before the examination. He then provided a semen sample by masturbation. Immediately after ejaculation, he was asked to urinate, and the post-ejaculatory urine sample was collected. Both the semen sample (if any) and the urine sample were centrifuged. The resulting pellets were examined microscopically for the presence of spermatozoa, and the urine supernatant was tested for fructose. The presence of spermatozoa and/or fructose in the post-ejaculatory urine, particularly when the antegrade semen sample showed low volume or no sperm, confirmed the diagnosis of RE [12].

ICSI treatment

The male partner underwent a sperm retrieval procedure based on individual circumstances, either involving sperm separation and purification after ejaculation into alkalized urine or surgical retrieval of sperm from the testis or epididymis. The

available sperm were selected for ICSI under a microscope. The woman underwent ovarian stimulation, possibly including a superovulation procedure, depending on her clinical condition. Oocytes were retrieved under transvaginal ultrasound guidance, and ICSI followed by in vitro embryo culture was performed to obtain transferable embryos, which were subsequently transferred [13] (Fig 3). Thirty days after embryo transfer, vaginal B-ultrasound was used to determine the number of intrauterine gestational sacs and the presence or absence of cardiac pulsation. ... embryo implantation rate = number of gestational sacs detected/number of transferred blastocysts $\times$ 100%

Statistical analysis

All continuous data are expressed as mean $\pm$ standard deviation ($\bar{x} \pm S$). T-tests and rank-sum tests were used to compare the data. All count data are expressed as rates (%), and comparisons between count data were made using the chi-square test. SPSS (version 16.0; SPSS, Chicago, IL, USA) was used for statistical analysis; $p < 0.05$ was regarded as statistically significant. Given the limited sample size, particularly after subgroup stratification, all non-significant comparisons should be interpreted as exploratory and hypothesis-generating rather than conclusive.

Results

There were no significant differences in baseline data between groups ($p > 0.05$). There was no significant difference in fertilization rate between the testicular sperm group

and the ejaculated sperm group (64.3% vs. 65.3%, $P=0.707$). The implantation rate in the testicular sperm group (30.6%) was numerically higher than that in the ejaculated sperm group (23.7%); however, the difference was not statistically significant ($P=0.240$). The clinical pregnancy rate was also numerically higher in the testicular sperm group (54.0% vs. 39.4%, $P=0.092$), and the miscarriage rate was numerically lower (8.8% vs. 28.6%, $P=0.053$). None of these differences reached statistical significance, likely due to the limited sample size (Table 1). When comparing aetiologies, the non-diabetic group had significantly higher implantation (33.3% vs. 19.4%, $P=0.020$) and clinical pregnancy rates (54.8% vs. 34.5%, $P=0.020$) than the diabetic group (Table 2). These differences were statistically significant despite the limited sample size.

The diabetic and non-diabetic groups were further stratified into testicular and ejaculated sperm subgroups for comparison. In the diabetic group, fertilization and cleavage rates were numerically higher in the ejaculated sperm subgroup than in the testicular sperm subgroup (71.0% vs. 66.0% and 68.6% vs. 60.1%, respectively), but these differences were not significant ($p>0.05$). The blastocyst formation rate was significantly higher in the testicular sperm subgroup of the diabetic group (38.3% vs. 27.1%, $P=0.004$). In the non-diabetic group, the ejaculated sperm subgroup had a higher blastocyst formation rate (28.8% vs. 28.0%, $P=0.836$) and a high-quality blastocyst rate (21.3% vs. 19.4%, $P=0.488$); however, these differences were not statistically significant. In both the diabetic and non-diabetic subgroups, numerical trends favored the testicular sperm approach for pregnancy outcomes. In the diabetic

group, the testicular sperm subgroup showed numerically higher clinical pregnancy (35.7% vs. 20.0%) and implantation (21.7% vs. 15.0%) rates, and a numerically lower miscarriage rate (10.0% vs. 40.0%). In the non-diabetic group, the testicular sperm subgroup also showed numerically higher clinical pregnancy (65.7% vs. 50.0%) and implantation (35.9% vs. 28.4%) rates, and a numerically lower miscarriage rate (8.7% vs. 21.0%). However, none of these differences in pregnancy outcomes reached statistical significance ($p>0.05$) (Tables 3, 4), likely due to the limited sample size after subgroup stratification.

Compared to the control group, the RE group had significantly lower rates of fertilization (64.9% vs. 80.6%, $P<0.001$), high-quality blastocyst formation (18.8% vs. 29.9%, $P<0.001$), implantation (26.9% vs. 44.2%, $P<0.001$), and clinical pregnancy (45.8% vs. 59.7%, $P=0.007$), indicating that RE itself poses challenges to ICSI success. There was no significant difference in miscarriage rates between the RE group and control group (18.3% vs. 12.0%, $P=0.210$) (Table 5).

Binary logistic regression analysis of clinical pregnancy outcomes identified diabetes as an independent risk factor for failure to achieve pregnancy in ICSI patients with RE (odds ratio [OR], 3.35; 95% confidence interval [CI], 1.58-7.11; $P=0.002$). This association remained significant after adjusting for potential confounders including female age, reinforcing the independence of diabetes as a risk factor. The use of ejaculated sperm was associated with an increased odds of failure (OR, 2.02) but did not reach statistical significance (95% CI, 0.97-4.20 ; $P=0.061$) (Table 6).

Discussion

RE is treated by restoring antegrade ejaculation. Nevertheless, clinical treatments, including drug therapy and surgical treatment, all have drawbacks such as certain limitations and treatment failure or recurrence [14,15]. Therefore, assisted reproductive technology has become the most effective means to solve the fertility needs of patients with RE. RE results in reduced sperm production and poor sperm quality. To reduce the impact of sperm factors on infertility [16], ICSI was used to overcome RE-related male infertility. Despite the feasibility and necessity of ICSI for RE [17], the limited number of reported cases requires further verification in larger cohorts. Given the rarity of RE and the modest sample size available, the present study should be considered exploratory.

Retrieval of sperm from urine has been commonly used in ICSI to overcome RE-related male infertility in previous reports [18,19]. However, it is hypothesized that urine may be toxic to semen [20], as the pH of fresh semen (7.2–8.0) differs from that of urine, and urine may contain bacteria, cell fragments, and ammonia components that could potentially damage sperm [21,22]. Surgical sperm retrieval has therefore emerged as an alternative option [19,23–25]. Testicular sperm extraction (TESE) is widely used for severe oligospermia and non-obstructive azoospermia. Greco et al. [26] reported that testicular sperm used in ICSI produced better pregnancy outcomes than ejaculated sperm, which they attributed to lower sperm DNA fragmentation (SDF) in testicular sperm. It has been proposed that, as sperm pass through the reproductive tract, oxidative stress may increase SDF [27], and high

SDF in ejaculated sperm may lead to reduced embryo implantation and early pregnancy loss [28]. Although we did not measure SDF, oxidative stress, or urine parameters in this study, the numerical trends we observed (higher implantation and clinical pregnancy rates with testicular sperm) are consistent with these hypotheses. We speculate that TESE may avoid potential damage from urine exposure and excessive oxidative stress, potentially resulting in higher-quality sperm. However, this remains speculative and requires direct mechanistic investigation. Moreover, compared to non-invasive retrieval from urine, TESE carries risks of postoperative complications [29,30]. Therefore, our exploratory data suggest that TESE-ICSI-ET may be considered for RE patients, but the choice should be individualized.

Diabetes is common in patients with RE [31,32] and is known to have negative effects on male fertility. Studies have reported abnormal apoptosis of spermatogenic cells in diabetic patients [33], differences in the expression of genes involved in oxidative stress and DNA repair [34], endocrine abnormalities [35], and microvascular lesions [36] that may affect sperm formation and maturation. Some studies have suggested that diabetic patients have higher SDF levels [37], and damaged sperm DNA could potentially affect embryo development [38]. However, we did not directly measure any of these molecular or physiological parameters in our cohort. Our finding that the diabetic group had significantly lower implantation and clinical pregnancy rates (Table 2), and that diabetes was an independent risk factor for pregnancy failure (OR=3.35, 95% CI 1.58–7.11, P=0.002), indicates a strong association. Nevertheless, the underlying mechanisms remain speculative and should

be investigated in future studies. Of note, diabetes was not subtyped in this study due to the lack of endocrinological evaluation and specific biomarkers (autoantibodies, C-peptide); based on adult onset and absence of childhood diabetes, type 2 diabetes is considered the most probable, but this remains unconfirmed.

RE also occurs in the early stages of diabetes [39] and is more likely to be overlooked. Therefore, attention should be paid to symptoms of hyperglycaemia in young and middle-aged men, as well as to the screening and treatment of diabetes, to reduce the risk of RE associated with sperm damage and autonomic neuropathy. According to our analysis, for non-diabetic patients with RE, while there was no significant difference in pregnancy outcome between sperm sources within this subgroup, obtaining testicular sperm surgically appeared to yield better pregnancy outcome trends (clinical pregnancy rate 65.7% vs. 50.0%; implantation rate 35.9% vs. 28.4%; miscarriage rate 8.7% vs. 21.0%). However, this should be further verified by analysing more samples.

Additionally, in our study, compared to the control group, the RE group had significantly lower fertilisation (64.9% vs. 80.6%, $P<0.001$), high-quality blastocyst formation (18.8% vs. 29.9%, $P<0.001$), implantation (26.9% vs. 44.2%, $P<0.001$), and clinical pregnancy rates (45.8% vs. 59.7%, $P=0.007$), suggesting that RE itself poses challenges to ICSI success, with diabetes as a compounding factor. Our pregnancy outcomes were better than those reported by Nikolettos et al. [17] (all 16 couples used urine sperm), possibly because 39 of our 79 couples underwent TESE. However, these findings must be interpreted in light of several limitations that also

explain why some numerical trends (e.g., higher pregnancy rates with testicular sperm) did not reach statistical significance: the modest sample size (79 RE patients) and further subgroup stratification substantially reduce statistical power; the retrospective design may introduce bias; we did not measure mechanistic factors (e.g., sperm DNA fragmentation, oxidative stress); diabetes was not formally subtyped (type 2 likely but unconfirmed); and the control group may not be ideal. Consequently, all non-significant comparisons should be interpreted as exploratory trends rather than definitive evidence. A multicentre prospective study with a larger sample size is needed to verify our results.

Conclusion

In this exploratory study, RE was associated with poorer ICSI outcomes compared to a control group of other male factor infertility patients. Diabetes was identified as an independent risk factor for pregnancy failure in RE patients undergoing ICSI. These findings suggest that active glycemic management may be important in this population. While numerical trends favored testicular sperm over ejaculated sperm for implantation and clinical pregnancy rates, and showed a trend toward lower miscarriage, none of these differences reached statistical significance, likely due to the limited sample size. Therefore, no definitive recommendation can be made regarding the optimal sperm source. Testicular sperm extraction may be considered as an option, particularly for diabetic patients, but this remains preliminary and requires confirmation in larger prospective studies. Individualized treatment planning based on patient characteristics

and preferences is essential.

Declarations

Ethical approval

This study was conducted in accordance with the ethical standards of the Declaration of Helsinki and ICMJE recommendations. Ethical approval was granted by the Ethics Committee of a university-affiliated hospital in China (reference number: P2020-12-36). Additional approval was obtained from the Ethics Committee of a collaborating hospital through a multicentre ethical review collaboration mechanism in accordance with regional guidelines. All participants provided written informed consent after receiving detailed explanations regarding study objectives, potential risks and benefits, and data usage protocols. Personal identifiers were removed using pseudonymization before analysis, and all clinical data were stored in secure, encrypted databases.

Consent to participate

All participants gave written informed consent to participate in the study.

Consent for publication

All authors consent to the publication of this work and authorize the granting of this license.

Conflict of interest

The authors declare no competing interests.

Funding

This study was supported by the National Natural Science Foundation of China [Grant No. 82471648, 82201803, 82271639, and 82371622], Clinical Translational Medicine Research Program [202204295107020037], Research Funds of Center for Big Data and Population Health of IHM [JKS2023004], Natural Science Foundation of Anhui Province [2208085Y31], Foundation of the Education Department of Anhui Province [2024AH050825 and gxgwfx2022007], Foundation of the Key Laboratory of Anhui Province [RDOGD-2024-02 and RHG-2021-1], University Synergy Innovation Program of Anhui Province [GXXT-2019-044], the Anhui Provincial Higher Education Research Project [2022AH010072] and the 2023 Annual Open Topic of MOE Key Laboratory of Population Health Across Life Cycle [JK20232].

Authors' contributions

Haitian Ding, Ruijun Liu, and Sana Atta contributed equally to this work. Study concept and design were led by Yingchun Liu, Yunxia Cao, and Feng Ni. Data acquisition and clinical patient management were performed by Haitian Ding, Ruijun Liu, Sana Atta, Aylla Qayyum, Yuqin Hu, Dongdong Tang, Hao Geng, Ping Zhou, Zhaolian Wei, and Yuping Xu. Statistical analysis and data interpretation were carried out by Haitian Ding, Ruijun Liu, and Sana Atta. The manuscript was drafted by Haitian

Ding, Ruijun Liu, and Sana Atta, and critically revised for important intellectual content by Yingchun Liu, Yunxia Cao, and Feng Ni. All authors reviewed and approved the final version of the manuscript. Yingchun Liu is the principal corresponding author who takes overall responsibility for the work. Yunxia Cao and Feng Ni serve as co-corresponding authors.

Acknowledgements

The authors thank all study participants, clinical staff, and funding sources for their support.

Data availability

Data will be made available on request.

Figure Captions

Fig 1. Pathophysiology of normal and retrograde ejaculation. In normal ejaculation, the internal sphincter contracts and the external sphincter relaxes, allowing antegrade semen flow. In retrograde ejaculation, the internal sphincter relaxes while the external sphincter contracts, directing sperm into the bladder.

Fig 2. Study flow diagram. 79 RE patients were stratified by aetiology (diabetic, n=33; non-diabetic, n=46) and sperm source (testicular, n=39; ejaculated, n=44). Implantation, clinical pregnancy, and miscarriage rates were compared, with binary logistic regression for risk factors.

Fig 3. Schematic of ICSI-ET procedure for RE patients. Sperm were obtained via TESE or urine separation. Following oocyte retrieval, ICSI was performed, and resulting embryos were cultured and transferred.

Abbreviations:

RE: retrograde ejaculation;

ICSI: intracytoplasmic sperm injection;

ET: embryo transfer;

ICSI-ET: intracytoplasmic sperm injection–embryo transfer;

TESE: testicular sperm extraction;

SDF: sperm DNA fragmentation;

OR: odds ratio;

CI: confidence interval.

References:

- [1] Ombelet, W., et al., Infertility and the provision of infertility medical services in developing countries. *Human reproduction update*, 2008. 14(6): p. 605-621. <https://doi.org/10.1093/humupd/dmn042>
- [2] Yavetz, H., et al., Retrograde ejaculation. *Human reproduction* (Oxford, England), 1994. 9(3): p. 381-386. <https://doi.org/10.1093/oxfordjournals.humrep.a138513>
- [3] Thomas, A.J.J., Ejaculatory dysfunction. *Fertility and sterility*, 1983. 39(4): p. 445-454. [https://doi.org/10.1016/s0015-0282\(16\)46929-3](https://doi.org/10.1016/s0015-0282(16)46929-3)
- [4] Parnham, A. and E.C. Serefoglu, Retrograde ejaculation, painful ejaculation and hematospermia. *Translational andrology and urology*, 2016. 5(4): p. 592-601. <https://doi.org/10.21037/tau.2016.06.05>
- [5] Sandler, B., Idiopathic retrograde ejaculation. *Fertility and sterility*, 1979. 32(4): p. 474-475. [https://doi.org/10.1016/s0015-0282\(16\)44308-6](https://doi.org/10.1016/s0015-0282(16)44308-6)
- [6] Narayan, P., P.H. Lange and E.E. Fraley, Ejaculation and fertility after extended retroperitoneal lymph node dissection for testicular cancer. *The Journal of urology*, 1982. 127(4): p. 685-688. [https://doi.org/10.1016/s0022-5347\(17\)53996-8](https://doi.org/10.1016/s0022-5347(17)53996-8)
- [7] Abrahams, J.I., et al., The surgical correction of retrograde ejaculation. *The Journal of urology*, 1975. 114(6): p. 888-890. [https://doi.org/10.1016/s0022-5347\(17\)67166-0](https://doi.org/10.1016/s0022-5347(17)67166-0)
- [8] Kurbatov, D., et al., Correction of Retrograde Ejaculation in Patients with Diabetes Mellitus Using Endourethral Collagen Injection: Preliminary Results. *The journal of sexual medicine*, 2015. 12(11): p. 2126-2129. <https://doi.org/10.1111/jsm.13024>
- [9] Reynolds, J.C., et al., Bladder neck collagen injection restores antegrade ejaculation after bladder neck surgery. *The Journal of urology*, 1998. 159(4): p. 1303-1303.

- [10] Jefferys, A., D. Siassakos and P. Wardle, The management of retrograde ejaculation: a systematic review and update. *Fertility and sterility*, 2012. 97(2): p. 306-312. <https://doi.org/10.1016/j.fertnstert.2011.11.019>
- [11] Nijs, M. and W. Ombelet, Intracytoplasmic sperm injection in assisted reproductive technology: an evaluation. *Human fertility (Cambridge, England)*, 2000. 3(3): p. 221-225.
- [12] Colpi, G., et al., EAU guidelines on ejaculatory dysfunction. *European urology*, 2004. 46(5): p. 555-558. <https://doi.org/10.1080/1464727002000199021>
- [13] Mansour, R., Intracytoplasmic sperm injection: a state of the art technique. *Human reproduction update*, 1998. 4(1): p. 43-56. <https://doi.org/10.1093/humupd/4.1.43>
- [14] Arafa, M. and O. El Tabie, Medical treatment of retrograde ejaculation in diabetic patients: a hope for spontaneous pregnancy. *The journal of sexual medicine*, 2008. 5(1): p. 194-198. <https://doi.org/10.1111/j.1743-6109.2007.00456.x>
- [15] Middleton, R.G. and R.L. Urry, The Young-Dees operation for the correction of retrograde ejaculation. *The Journal of urology*, 1986. 136(6): p. 1208-1209. [https://doi.org/10.1016/s0022-5347\(17\)45284-0](https://doi.org/10.1016/s0022-5347(17)45284-0)
- [16] Palermo, G.D., et al., Intracytoplasmic sperm injection: state of the art in humans. *Reproduction (Cambridge, England)*, 2017. 154(6): p. F93-F110. <https://doi.org/10.1530/REP-17-0374>
- [17] Nikolettos, N., et al., The outcome of intracytoplasmic sperm injection in patients with retrograde ejaculation. *Human reproduction (Oxford, England)*, 1999. 14(9): p. 2293-2296. <https://doi.org/10.1093/humrep/14.9.2293>

- [18] HOTCHKISS, R.S., A.B. PINTO and S. KLEEGMAN, Artificial insemination with semen recovered from the bladder. *Fertility and sterility*, 1954. 6(1): p. 37-42. [https://doi.org/10.1016/s0015-0282\(16\)31863-5](https://doi.org/10.1016/s0015-0282(16)31863-5)
- [19] Rosenlund, B., et al., In-vitro fertilization and intracytoplasmic sperm injection in the treatment of infertility after testicular cancer. *Human reproduction* (Oxford, England), 1998. 13(2): p. 414-418. <https://doi.org/10.1093/humrep/13.2.414>
- [20] Aust, T.R., et al., Development and in vitro testing of a new method of urine preparation for retrograde ejaculation; the Liverpool solution. *Fertility and sterility*, 2008. 89(4): p. 885-891. <https://doi.org/10.1016/j.fertnstert.2007.04.042>
- [21] Auroux, M.R., et al., Is the sperm bacterial ratio a determining factor in impairment of sperm motility: an in-vitro study in man with *Escherichia coli*. *International journal of andrology*, 1991. 14(4): p. 264-270. <https://doi.org/10.1111/j.1365-2605.1991.tb01091.x>
- [22] Kim, S.C. and H.W. Kim, Effects of nitrogenous components of urine on sperm motility: an in vitro study. *International journal of andrology*, 1998. 21(1): p. 29-33. <https://doi.org/10.1046/j.1365-2605.1998.00083.x>
- [23] Craft, I.L., et al., Factors influencing the outcome of in-vitro fertilization with percutaneous aspirated epididymal spermatozoa and intracytoplasmic sperm injection in azoospermic men. *Human reproduction* (Oxford, England), 1995. 10(7): p. 1791-1794. <https://doi.org/10.1093/oxfordjournals.humrep.a136175>
- [24] Goldstein, M. and C. Tanrikut, Microsurgical management of male infertility. *Nature clinical practice. Urology*, 2006. 3(7): p. 381-391.

<https://doi.org/10.1038/ncpuro0524>

[25] Kanto, S., et al., Fresh testicular sperm retrieved from men with spinal cord injury retains equal fecundity to that from men with obstructive azoospermia via intracytoplasmic sperm injection. *Fertility and sterility*, 2009. 92(4): p. 1333-1336.

<https://doi.org/10.1016/j.fertnstert.2008.07.1776>

[26] Greco, E., et al., Efficient treatment of infertility due to sperm DNA damage by ICSI with testicular spermatozoa. *Human reproduction* (Oxford, England), 2005. 20(1): p. 226-230. <https://doi.org/10.1093/humrep/deh590>

[27] Muratori, M., et al., Investigation on the Origin of Sperm DNA Fragmentation: Role of Apoptosis, Immaturity and Oxidative Stress. *Molecular medicine* (Cambridge, Mass.), 2015. 21(1): p. 109-122. <https://doi.org/10.2119/molmed.2014.00158>

[28] Esteves, S.C., M. Roque and N. Garrido, Use of testicular sperm for intracytoplasmic sperm injection in men with high sperm DNA fragmentation: a SWOT analysis. *Asian journal of andrology*, 2018. 20(1): p. 1-8.

https://doi.org/10.4103/aja.aja_7_17

[29] Esteves, S.C., et al., Reproductive potential of men with obstructive azoospermia undergoing percutaneous sperm retrieval and intracytoplasmic sperm injection according to the cause of obstruction. *The Journal of urology*, 2013. 189(1): p. 232-237.

<https://doi.org/10.1016/j.juro.2012.08.084>

[30] Schlegel, P.N. and L.M. Su, Physiological consequences of testicular sperm extraction. *Human reproduction* (Oxford, England), 1997. 12(8): p. 1688-1692.

<https://doi.org/10.1093/humrep/12.8.1688>

- [31] Fedder, J., et al., Retrograde ejaculation and sexual dysfunction in men with diabetes mellitus: a prospective, controlled study. *Andrology*, 2013. 1(4): p. 602-606. <https://doi.org/10.1111/j.2047-2927.2013.00083.x>
- [32] Gaunay, G., H.M. Nagler and D.S. Stember, Reproductive sequelae of diabetes in male patients. *Endocrinology and metabolism clinics of North America*, 2013. 42(4): p. 899-914.
- [33] Guneli, E., et al., Effect of melatonin on testicular damage in streptozotocin-induced diabetes rats. *European surgical research. Europäische chirurgische Forschung. Recherches chirurgicales europeennes*, 2008. 40(4): p. 354-360. <https://doi.org/10.1159/000118032>
- [34] Mallidis, C., et al., The influence of type 1 diabetes mellitus on spermatogenic gene expression. *Fertility and sterility*, 2009. 92(6): p. 2085-2087. <https://doi.org/10.1016/j.fertnstert.2009.06.006>
- [35] Schoeller, E.L., S. Schon and K.H. Moley, The effects of type 1 diabetes on the hypothalamic, pituitary and testes axis. *Cell and tissue research*, 2012. 349(3): p. 839-847. <https://doi.org/10.1007/s00441-012-1387-7>
- [36] Padrón, R.S., et al., Semen analyses in adolescent diabetic patients. *Acta diabetologica latina*, 1984. 21(2): p. 115-121. <https://doi.org/10.1007/BF02591100>
- [37] Agbaje, I.M., et al., Insulin dependant diabetes mellitus: implications for male reproductive function. *Human reproduction (Oxford, England)*, 2007. 22(7): p. 1871-1877. <https://doi.org/10.1093/humrep/dem077>
- [38] Mulholland, J., et al., Male diabetes mellitus and assisted reproduction treatment

outcome. Reproductive biomedicine online, 2011. 22(2): p. 215-219.

<https://doi.org/10.1016/j.rbmo.2010.10.005>

[39] Kam, J., V.H. Tsang and V. Chalasani, Retrograde Ejaculation: A Rare Presenting Symptom of Type 1 Diabetes Mellitus. Urology case reports, 2016. 10: p. 9-10.

<https://doi.org/10.1016/j.eucr.2016.10.008>

Table 1. Comparison of basic data, blastocyst formation and pregnancy outcome between the testicular sperm group and the ejaculated sperm group

	Testicular sperm Group	Ejaculated sperm Group	<i>p</i> -value
No. of patients	39	44	-
Mean male age (years)	31.8±5.0	32.7±6.7	0.487
Mean female age (years)	29.2±5.2	30.6±4.7	0.204
Infertility age (years)	3.9±2.8	3.3±2.0	0.271
No. of ICSI cycles	48	56	/
Fertilisation rate (count, %)	436/678 (64.3)	437/669 (65.3)	0.707
Cleavage rate (count, %)	410/678 (60.5)	366/669 (54.7)	0.032
Blastocyst formation rate (count, %)	221/678 (32.6)	185/669 (27.6)	0.049
High quality blastocyst formation rate (count, %)	128/678 (18.9)	125/669 (18.7)	0.922
No. of thawing cycles	63	71	/
Mean number of embryos transferred	1.7±0.6	1.7±0.6	/
Implantation rate (count, %)	34/111 (30.6)	28/118 (23.7)	0.240
Clinical pregnancy rate (count, %) per thawing cycle	34/63 (54.0)	28/71 (39.4)	0.092
Miscarriage rate (count, %)	3/34 (8.8)	8/28 (28.6)	0.053

Note: Due to legal marriage requirements for ART in China, the number of female patients equals the number of male patients in each group (testicular sperm group: 39 women; ejaculated sperm group: 44 women).

Table 2. Comparison of basic data, blastocyst formation and pregnancy outcome between the diabetic group and the non-diabetic group

	DM Group	N-DM Group	<i>p</i>-value
No. of patients	33	46	-
Mean male age (years)	32.2±4.6	32.0±6.9	0.865
Mean female age (years)	30.8±4.4	28.9±4.1	0.052
Infertility age (years)	3.7±3.2	3.2±1.8	0.335
No. of ICSI cycles	44	60	/
Fertilisation rate (count, %)	368/565 (65.1)	505/781 (64.7)	0.877
Cleavage rate (count, %)	321/565 (56.8)	455/781 (58.3)	0.578
Blastocyst formation rate (count, %)	184/565 (32.6)	222/781 (28.4)	0.087
High quality blastocyst formation rate (count, %)	94/565 (16.6)	159/781 (20.4)	0.069
No. of thawing cycles	58	73	/
Mean number of embryos transferred	1.6±0.6	1.7±0.6	/
Implantation rate (count, %)	20/103 (19.4)	40/120 (33.3)	0.020
Clinical pregnancy rate (count, %) per thawing cycle	20/58 (34.5)	40/73 (54.8)	0.020
Miscarriage rate (count, %)	5/20 (25.0)	6/40 (15.0)	0.498

Note: The number of female patients equals the number of male patients in each group (DM group: 33 women; N-DM group: 46 women).

Table 3. Comparison of basic data, blastocyst formation and pregnancy outcome between the testicular sperm group and the ejaculated sperm group in the diabetes group

	Testicular sperm Group	Ejaculated sperm Group	<i>p</i>-value
No. of patients	16	19	-
Mean male age (years)	32.7±4.5	32.7±4.6	1.000
Mean female age (years)	30.3±4.9	31.6±3.9	0.398
Infertility age (years)	5.0±3.4	3.1±2.6	0.078
No. of ICSI cycles	20	23	/
Fertilisation rate (count, %)	179/271 (66.0)	189/295 (64.0)	0.621
Cleavage rate (count, %)	163/271 (60.1)	158/295 (53.7)	0.114
Blastocyst formation rate (count, %)	104/271 (38.3)	80/295 (27.1)	0.004
High quality blastocyst formation rate (count, %)	49/271 (18.1)	45/295 (15.2)	0.367
No. of thawing cycles	28	30	/
Mean number of embryos transferred	1.7±0.7	1.6±0.6	/
Implantation rate (count, %)	10/46 (21.7)	6/52 (15.0)	0.173
Clinical pregnancy rate (count, %) per thawing cycle	10/28 (35.7)	6/30 (20.0)	0.181
Miscarriage rate (count, %)	1/10 (10.0)	4/10 (40.0)	0.304

Note: In the diabetic group, the testicular sperm subgroup included 16 female patients and the ejaculated sperm subgroup included 19 female patients.

Table 4. Comparison of basic data, blastocyst formation and pregnancy outcome between the testicular sperm group and the ejaculated sperm group in the non-diabetic group

	Testicular sperm Group	Ejaculated sperm Group	p-value
No. of patients	23	25	-
Mean male age (years)	31.5±4.9	32.7±7.0	0.531
Mean female age (years)	28.3±5.2	29.8±5.2	0.322
Infertility age (years)	3.2±1.9	3.3±1.5	0.842
No. of ICSI cycles	28	32	/
Fertilisation rate (count, %)	257/407 (63.1)	248/374 (66.3)	0.353
Cleavage rate (count, %)	247/407 (60.7)	208/374 (55.6)	0.150
Blastocyst formation rate (count, %)	117/407 (28.8)	105/374 (28.0)	0.836
High quality blastocyst formation rate (count, %)	79/407 (19.4)	80/374 (21.3)	0.488
No. of thawing cycles	35	38	/
Mean number of embryos transferred	1.6±0.6	1.6±0.6	/
Implantation rate (count, %)	23/64 (35.9)	19/67 (28.4)	0.353
Clinical pregnancy rate (count, %) per thawing cycle	23/35 (65.7)	19/38 (50.0)	0.175
Miscarriage rate (count, %)	2/23 (8.7)	4/19 (21.0)	0.384

Note: In the non-diabetic group, the testicular sperm subgroup included 23 female patients and the ejaculated sperm subgroup included 25 female patients.

Table 5. Comparison of basic data, blastocyst formation and pregnancy outcome between the retrograde ejaculation group and the control group

	RE Group	Control Group	p-value
No. of patients	79	200	-
Mean male age (years)	32.1±5.9	29.0±3.8	<0.001
Mean female age (years)	29.7±4.9	30.3±4.4	0.345
Infertility age (years)	3.5±2.5	3.9±2.6	0.236
No. of ICSI cycles	104	218	/
Fertilisation rate (count, %)	873/1346 (64.9)	2250/3165 (80.6)	<0.001
Cleavage rate (count, %)	776/1346 (57.7)	2138/3165 (67.6)	<0.001
Blastocyst formation rate (count, %)	406/1346 (30.2)	1182/3165 (37.4)	<0.001
High quality blastocyst formation rate (count, %)	253/1346 (18.8)	947/3165 (29.9)	<0.001
No. of thawing cycles	131	308	/
Mean number of embryos transferred	1.6±0.6	1.8±0.5	/
Implantation rate (count, %)	60/223 (26.9)	248/561 (44.2)	<0.001
Clinical pregnancy rate (count, %) per thawing cycle	60/131 (45.8)	184/308 (59.7)	0.007
Miscarriage rate (count, %)	11/60 (18.3)	22/184 (12.0)	0.210

Note: The RE group included 79 female patients and the control group included 200 female patients.

Table 6. Binary logistic regression analysis of clinical pregnancy outcomes for retrograde ejaculation

	B	S.E.	Wald	P	OR	95% CI
DM	1.209	0.384	9.90	0.002	3.35	1.58 - 7.11
Ejaculated sperm	0.701	0.374	3.51	0.061	2.02	0.97 - 4.20

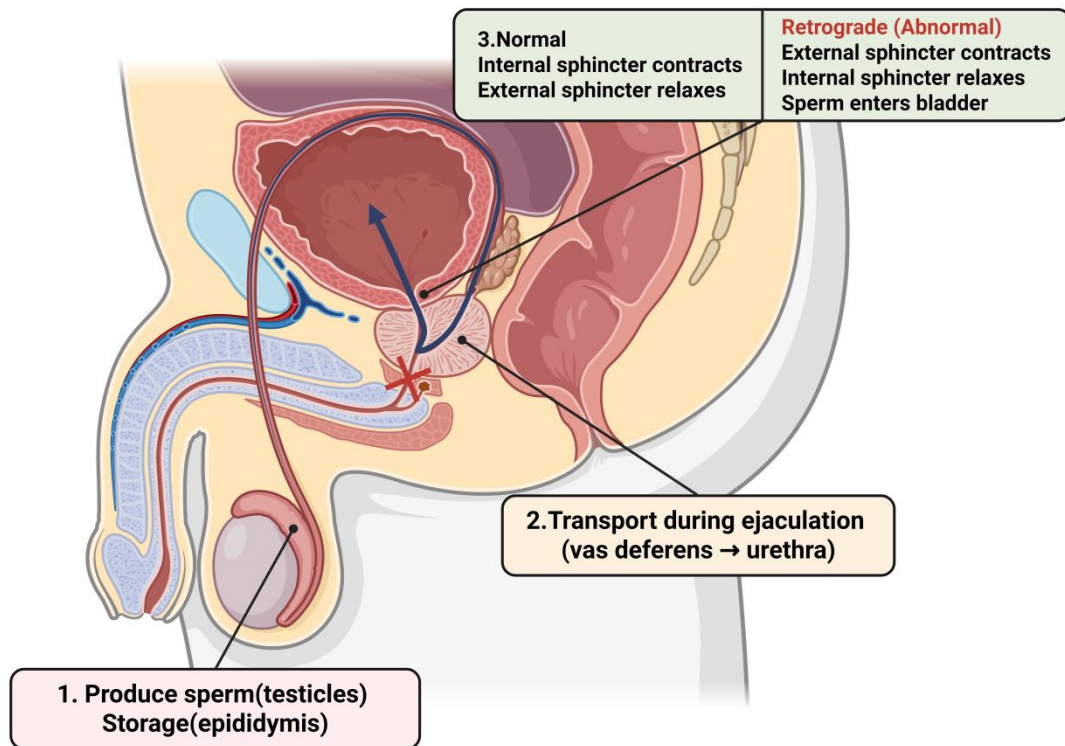


Fig 1. Pathophysiology of normal and retrograde ejaculation.

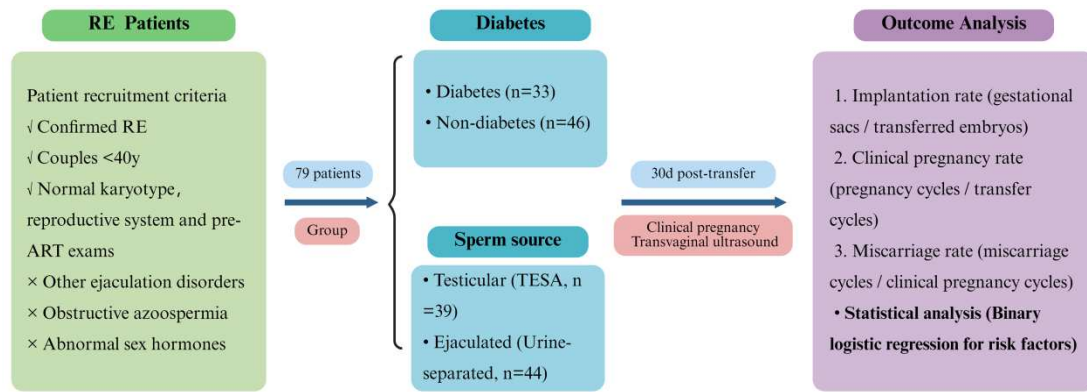


Fig 2. Study flow diagram.

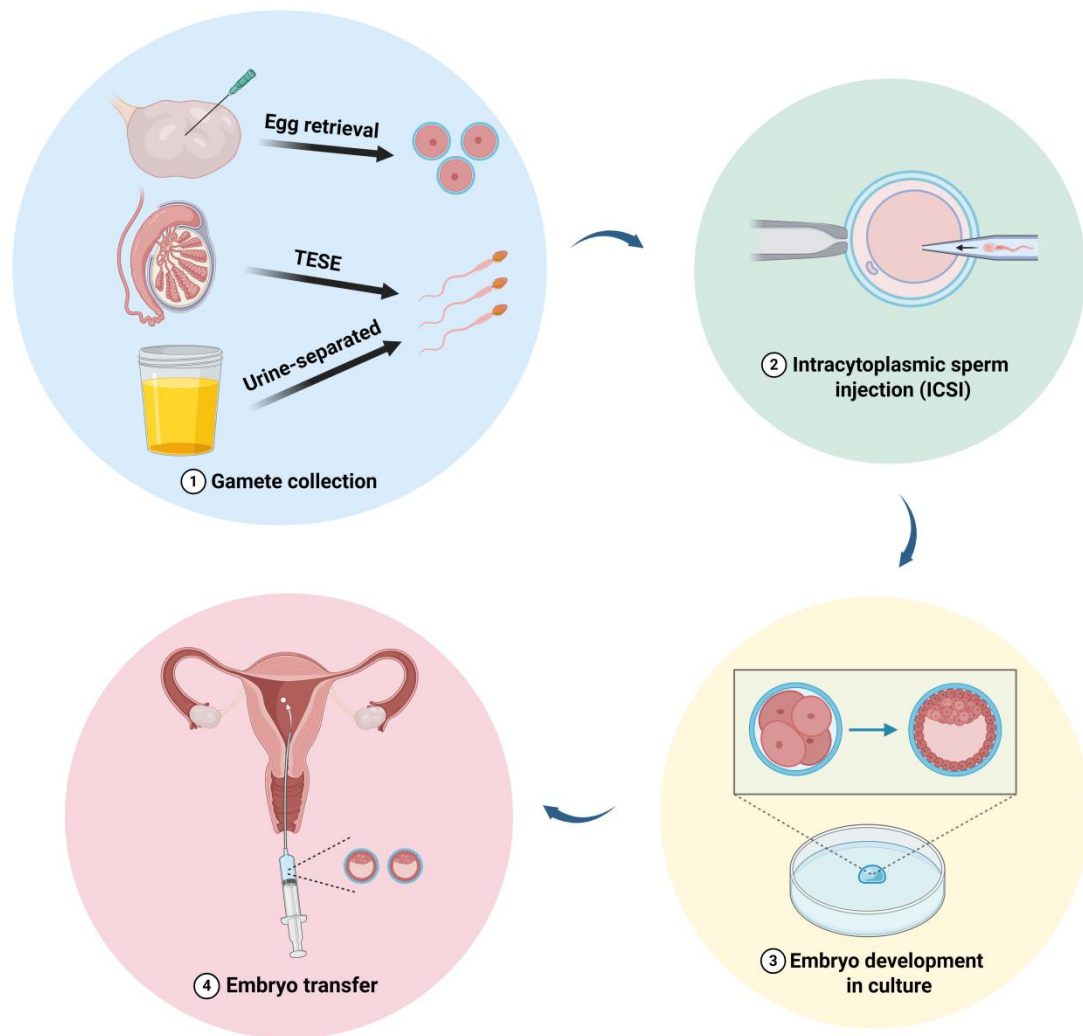


Fig 3. Schematic of ICSI-ET procedure for RE patients.