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Original Research Article

## Comparing day five and day six blastocyst transfers in IVF: the influence of male fertility factors on outcomes

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### ABSTRACT

**Background:** Advances in in vitro fertilization (IVF) technology now permit blastocyst transfers on both day five and day six of embryo development. However, the influence of male fertility factors on the success of these transfers is not thoroughly understood. This study aims to evaluate and compare the outcomes of day five versus day six blastocyst transfers and to explore how male fertility parameters impact IVF success rates.

**Methods:** A prospective study was conducted over 24 months, involving 732 IVF cycles. Participants were categorized into two groups: one receiving day five blastocyst transfers (n=366) and the other receiving day six transfers (n=366). Variables such as male age, smoking habits, obesity, and semen quality were analysed. The primary outcomes-implantation rates, clinical pregnancy rates and live birth rates-were compared using chi-square and Fisher's exact tests.

**Results:** The success rates were notably higher for day five blastocyst transfers, with an implantation rate of 58.9% and a clinical pregnancy rate of 53.5%, compared to day six transfers, which had an implantation rate of 47.5% and a clinical pregnancy rate of 42.9%. Factors such as younger male age (<35 years), non-smoking status, normal weight and good semen quality were associated with better IVF outcomes.

**Conclusions:** Day five blastocyst transfers generally yield higher success rates than day six transfers. Addressing male fertility factors, including age, smoking and semen quality, is essential for optimizing IVF success.

**Keywords:** Assisted reproductive technology, Clinical pregnancy rate, Day 5 blastocyst transfer, Day 6 blastocyst transfer, Embryo viability, IVF outcomes

### INTRODUCTION

In vitro fertilization (IVF) has significantly advanced reproductive medicine, providing new opportunities for couples facing infertility challenges. The option to transfer blastocysts on either day five or day six after fertilization allows for the selection of embryos that are further developed, enhancing the likelihood of successful implantation.<sup>1,2</sup> Day five blastocyst transfers are

commonly preferred due to the advanced stage of embryo development, which often correlates with higher viability. However, some embryos may benefit from an additional day of culture, leading to day six transfers.<sup>3,4</sup> Despite this, the comparative effectiveness of day six versus day five transfers remains a topic of ongoing debate.<sup>5</sup> Male fertility factors, such as age, lifestyle choices (e.g., smoking, obesity) and semen quality, play a crucial role in determining the success of IVF procedures.<sup>6,7</sup> As men age, there is a natural decline in semen quality, including

reduced sperm count and motility and increased DNA fragmentation.<sup>8</sup> This study aims to investigate the outcomes of day five and day six blastocyst transfers and assess the impact of male fertility factors on these outcomes.

## METHODS

This prospective cohort study took place at Ashoka Advanced IVF Center (Raipur and Bhopal) from 2021 to 2023. Participants were selected using simple random sampling to ensure that each eligible couple had an equal probability of being included. This method minimized selection bias and enhanced the representativeness of the sample. A total of 732 IVF cycles were included, with participants divided into day five (n=366) and day six (n=366) blastocyst transfer groups. Couples were selected based on good ovarian reserve, normal uterine anatomy and absence of chronic conditions affecting fertility. Participants underwent either standard IVF or intracytoplasmic sperm injection (ICSI).

Among the total cycles, 490 IVF cycles involved patients with a good prognosis (e.g., tubal block, unexplained infertility), while 242 ICSI cycles were due to severe oligozoospermia (low sperm quality). Embryos were cultured in Medicult/Origio media (Europe). Primary outcomes—implantation, clinical pregnancy and live birth rates—were assessed for both groups. Male fertility factors, including age, smoking status, obesity (BMI) and semen quality, were evaluated using WHO guidelines. Statistical analysis used chi-square and Fisher's exact tests, with significance set at  $p < 0.05$ .

## RESULTS

### Day five vs day six blastocyst transfers

Day five transfers had a significantly higher implantation rate (58.9%) and clinical pregnancy rate (53.5%) compared to day six (implantation rate 47.5%, clinical pregnancy rate 42.9%).

### Male age

Implantation, clinical pregnancy and live birth rates declined with advancing male age. Males <35 years showed higher success rates compared to those >40 years.

### Smoking status

Smokers had lower implantation (37%) and live birth rates (26%) compared to non-smokers (implantation 46%, live birth 35%).

### Obesity

Obese individuals had lower success rates than those with normal BMI, with implantation, clinical pregnancy and live birth rates of 32%, 27% and 22%, respectively.

## Semen quality

Normal semen quality was associated with better outcomes, with implantation rates of 46%, compared to 36% for poor semen quality.

**Table 1: Summary of success rates for day 5 vs day 6 transfers.**

Parameter	Day 5 transfer (%)	Day 6 transfer (%)
<b>Implantation rate</b>	58.9	47.5
<b>Clinical pregnancy rate</b>	53.5	42.9
<b>Live birth rate</b>	49.33	38.2

**Table 2: Implantation rates by male age.**

Male age (in Years)	Implantation rate (%)	Live birth rate (%)
< 35	64	68.5
35-40	50.4	58.2
> 40	38.6	45.1

**Table 3: Influence of smoking on IVF outcomes.**

Smoking status	Implantation rate (%)	Live birth rate (%)
<b>Non-Smoker</b>	46	35
<b>Smoker</b>	37	26

**Table 4: Impact of obesity on IVF success.**

BMI category	Implantation rate (%)	Live birth rate (%)
<b>Normal (&lt;25)</b>	49.2	56.3
<b>Overweight (25-30)</b>	42.7	38.5
<b>Obese (&gt;30)</b>	32	22

**Table 5: Impact of semen quality on IVF outcomes.**

Semen quality	Implantation rate (%)	Live birth rate (%)
<b>Normal semen</b>	46	52.7
<b>Poor semen quality</b>	36	31.5

## DISCUSSION

This study's results indicate that day five blastocyst transfers generally result in higher success rates compared to day six transfers, a trend supported by existing research. The advantage of day five transfers is largely attributed to the embryos' advanced developmental stage, which aligns more closely with the optimal timing for uterine receptivity. Embryos at this stage typically exhibit a higher potential for implantation due to more advanced cellular development and better alignment with the endometrial environment. In contrast, day six transfers might be less

successful due to potential developmental delays that can hinder the embryo's ability to implant effectively.<sup>1,2</sup>

While day six blastocyst transfers can still result in successful pregnancies, they are associated with lower overall embryo viability. The additional culture time may expose embryos to environmental stresses that could impair their development and implantation capabilities. Hence, when feasible, clinicians should prefer day five transfers, as they not only improve success rates but also minimize the risk of multiple pregnancies by reducing the number of embryos transferred.<sup>3,4</sup>

The study underscores the significant role of male fertility factors in IVF outcomes. Male age is a critical determinant, with older men experiencing lower implantation and live birth rates. This decline is likely due to age-related decreases in sperm quality, including reduced motility, lower sperm count and increased DNA fragmentation. This aligns with previous research linking advanced paternal age to higher risks of genetic anomalies and adverse pregnancy outcomes.<sup>5,6</sup>

Lifestyle factors, particularly smoking and obesity, also substantially impact IVF success. Smoking has been linked to diminished semen quality, which affects sperm motility, concentration and increases oxidative stress. Similarly, obesity is associated with hormonal imbalances that negatively influence spermatogenesis, including lower testosterone levels and elevated estrogen. These findings suggest that interventions aimed at improving lifestyle factors could potentially enhance IVF outcomes, especially for male patients affected by smoking or obesity.<sup>7,8</sup>

Furthermore, semen quality emerged as a key factor in determining IVF success. Men with normal semen quality had significantly higher implantation and live birth rates compared to those with compromised semen parameters. This supports earlier findings that highlight the importance of semen analysis in predicting IVF success and emphasizes the need for effective management of poor semen quality, which is often linked to higher rates of genetic abnormalities and reduced fertilization success.<sup>9,10</sup>

This study has several limitations. First, the sample was limited to a single geographic region, potentially restricting the generalizability of the findings. Second, while male fertility factors were evaluated, other potential confounders, such as female age and endometrial receptivity, were not extensively analyzed. Third, the study did not account for variations in laboratory protocols or embryo grading, which may influence outcomes. Lastly, the observational design precludes establishing causal relationships, emphasizing the need for randomized controlled trials to validate these findings further.

## CONCLUSION

This study highlights the superior clinical outcomes associated with day five blastocyst transfers compared to day six transfers in IVF cycles. The success rate of day five transfers (58.5%) was significantly higher than that of day six transfers (42.3%), emphasizing the viability of faster-developing embryos. The findings further demonstrate that transferring fewer embryos on day five reduces the likelihood of multiple pregnancies while maintaining high implantation success. The use of simple random sampling ensured unbiased participant selection, strengthening the validity of the results. These insights underscore the importance of precise embryo transfer timing and the selection of advanced embryo culture systems, such as Medicult/Origio media, to optimize clinical outcomes. Future research should focus on identifying factors influencing embryo development speed to refine IVF protocols further.

The findings reinforce the preference for day five blastocyst transfers to maximize IVF success rates. For patients undergoing IVF, especially those with male partners experiencing age-related fertility issues, focusing on lifestyle modifications and improving semen quality can be crucial. Developing personalized treatment plans that address these factors could enhance overall IVF outcomes and reduce the risk of multiple pregnancies. Incorporating regular evaluations and targeted interventions for male fertility parameters into IVF protocols is recommended to optimize success.

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