

Analysis of blastocyst morphokinetics assessed by time-lapse technology and its correlation with human IVF treatment outcome

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Received: February 16, 2026 • Revised manuscript received: May 15, 2026 • Accepted: May 18, 2026

Published online: June 11, 2026

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ABSTRACT

Human embryo selection in in vitro fertilisation (IVF) treatments is traditionally based on evaluating cell number, morphology, and fragmentation to select the most viable embryos. However, this static evaluation method has limitations, as it captures only a snapshot of the embryo at a single time point. Recent advancements in time-lapse imaging and artificial intelligence (AI) have improved embryo assessment by enabling dynamic and continuous observation of embryonic development.

This retrospective cohort study aimed to evaluate morphometric and morphokinetic parameters of 102 human embryos cultured during IVF cycles. Embryos were included if they reached the blastocyst stage on Day 5 and were selected for fresh single embryo transfer. Morphological and morphokinetic parameters were assessed using time-lapse technology to compare embryos resulting in clinical pregnancy and those that did not.

Clinical pregnancy was achieved in 47.1% of transfers. Although no significant differences were observed in blastocyst area, diameter, or inner cell mass (ICM) size between embryos that implanted (P+) and those that did not (P–), trends toward larger dimensions were observed in the P+ group. Morphokinetic parameters showed slightly faster developmental kinetics in P+ embryos, although not significantly.

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Morphokinetic scores were calculated using time-lapse embryo evaluation systems (KIDScore D5 and iDAScore, Vitrolife). iDAScore values were significantly higher in implanted embryos. iDAScore, which incorporates AI-based scoring, showed better predictive performance compared with KIDScore in ROC and logistic regression analyses, indicating its potential as a reliable tool for embryo selection. These findings support the use of AI-based morphokinetic analysis for improved embryo selection in IVF.

KEYWORDS

human IVF, embryo morphology, embryo morphokinetics, artificial intelligence, IVF outcome

INTRODUCTION

The primary goal of in vitro fertilisation (IVF) treatment is to select the most viable embryo for transfer, with the aim of maximising the likelihood of implantation and achieving a successful pregnancy. Embryo selection is traditionally based on the timing of cell divisions and morphological evaluation during development, as these parameters are considered key indicators of embryo quality during the cleavage stage and the blastocyst stage.

In most IVF laboratories, blastocyst selection is based on the Gardner grading system introduced by Gardner and Schoolcraft in 1999 [1, 2]. Although the system does not encompass all aspects of blastocyst morphology, it remains the most widely used method worldwide. The grading evaluates blastocyst expansion and the morphological appearance of the inner cell mass (ICM) and trophoctoderm (TE) cells.

Recent studies have examined whether these parameters are predictive of implantation and live birth outcomes. Ahlström et al. [3] reported that larger and more cohesive ICMs were associated with higher implantation rates. Similarly, Almagor et al. [4] demonstrated that the ICM-to-blastocyst diameter ratio correlates with clinical pregnancy outcomes in single blastocyst transfers.

The introduction of time-lapse technology (TLT) into human IVF has revolutionised embryo assessment by enabling continuous and objective monitoring of developmental dynamics. To ensure reproducibility and standardisation of morphokinetic data, the Time-Lapse User Group published comprehensive guidelines defining and harmonising the nomenclature and annotation of key developmental events [5]. These guidelines established reference points for dynamic parameters such as pronuclear fading, cleavage timing, and blastocyst formation, which have since become the basis for time-lapse embryo evaluation worldwide. In the present study, embryo development was annotated according to these established criteria to ensure consistency and comparability of data.

In recent years, computational and artificial intelligence-based models have been developed to assist embryologists in the selection and ranking of embryos for transfer. Among these, the KIDScore D5 model uses predefined morphokinetic and morphological parameters from large datasets of embryos with known implantation outcomes, allowing for a semi-automated estimation of implantation potential [6, 7]. More recently, the iDAScore model has been introduced as a fully automated, artificial intelligence-based system that analyses entire time-lapse sequences using deep learning networks. The latest version of iDAScore has shown improved

predictive performance compared with both previous algorithmic models and the original iDA-Score version, and is now increasingly implemented in IVF laboratories worldwide [8]. These systems assist embryologists in establishing embryo ranking and selection order, contributing to more objective decision-making and reducing inter-observer variability in IVF practice.

Despite these advancements, there is no universally recommended algorithm that currently integrates morphological and kinetic data for embryo selection, and the predictive value of specific features, such as blastocyst expansion rate or ICM growth dynamics, remains under investigation. Nevertheless, despite the promising performance of AI-assisted embryo evaluation systems, their clinical utility and generalisability remain under active investigation. Differences in laboratory conditions, culture protocols, patient populations, image acquisition systems, and annotation practices may influence model performance across IVF centres. In addition, the interpretability of deep learning-based predictions remains limited, and further multicentre validation studies are required before universal implementation of AI-guided embryo prioritisation strategies can be fully established [9, 10].

The aim of this study was to evaluate morphometric and morphokinetic parameters associated with implantation outcome in human IVF embryos cultured using TLT. We investigated how morphological characteristics of embryos at the expanded blastocyst stage, particularly blastocyst diameter and ICM size, relate to implantation success. Additionally, we examined cleavage kinetics and the morphokinetic scores (KIDScore, iDAScore) provided by a time-lapse embryo monitoring system. We hypothesized that AI-based morphokinetic scoring systems may provide superior predictive performance for implantation outcome compared with conventional morphometric measurements alone.

MATERIALS AND METHODS

Study design

We retrospectively analysed embryo morphology and development from IVF cycles conducted at the Division of Assisted Reproduction, Department of Obstetrics and Gynaecology, Semmelweis University, Budapest, Hungary. Embryos were included in the study if they reached the blastocyst stage on the day of embryo transfer (Fig. 1). Only fresh single blastocyst transfers performed on Day 5 were included in the study. A total of 102 embryo transfers from 102 patients were analysed, with one transfer included per patient. Both conventional IVF and intracytoplasmic sperm injection (ICSI) cycles were included in the analysis. Among the included embryos, 66 were derived from conventional IVF cycles and 36 from ICSI cycles. In most cases, fertilisation was performed using ejaculated sperm provided by the male partner, while donor sperm was used in one case only. Cycles involving testicular sperm extraction (TESE) were not included. Preimplantation genetic testing (PGT) was not used in any of the included cases. Eligibility criteria did not include maternal age, cause of infertility, or the number of previous IVF cycles.

All laboratory procedures were performed using Vitrolife G-series media (Vitrolife, Gothenburg, Sweden). Oocyte incubation and conventional IVF procedures were carried out using G-IVF Plus medium. Embryos were cultured in single-step G-TL medium in the EmbryoScope⁺ incubator using either EmbryoSlide[®] 8-well dishes for individual culture or 16-well group culture dishes, depending on the number of retrieved oocytes, as our working group

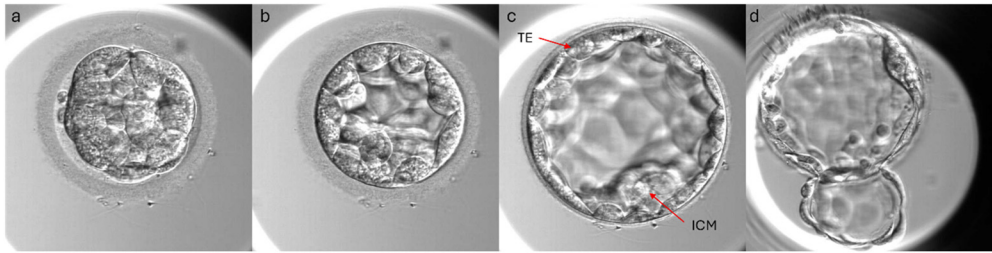


Fig. 1. Blastocyst development. Representative time-lapse images illustrating blastocyst development from early blastocyst formation to the hatching blastocyst stage. Sequential panels show (a) early blastocyst with initial cavity formation, (b) progressing blastocoel expansion, (c) fully expanded blastocyst with visible inner cell mass (ICM), trophoctoderm (TE), and thinning of the zona pellucida, and (d) embryo hatching through the zona pellucida

previously described [11]. This incubator system is equipped with a built-in microscope and a camera that captures images in 11 focal planes every 10 min. Compared with conventional static assessment, time-lapse technology enables continuous, non-invasive monitoring of embryonic development and provides detailed morphokinetic data [12]. Embryo transfer was performed using EmbryoGlue[®] medium according to the manufacturer's recommendations.

Embryo development was continuously monitored using the EmbryoScope+ time-lapse system, and the EmbryoViewer software (Vitrolife) was used for automatic annotation and analysis of key developmental events, including pronuclear fading (tPNf), first mitotic division (t2), subsequent cleavage stages (t4, t8), and the onset of blastocyst formation (tB). Morphological and morphokinetic evaluation was performed by four experienced embryologists according to standardised laboratory protocols. Cleavage timings and morphological characteristics, including ICM and TE grading, were assessed continuously during embryo development and were used for routine clinical embryo prioritisation. The timing of each event was defined by the first frame in which the relevant morphological change became evident [5]. Morphokinetic annotations followed the Vitrolife annotation guidelines, while morphological grading was performed according to the Gardner grading system. During manuscript preparation, all annotations and measurements were retrospectively reviewed and verified by the first author to ensure consistency of the dataset. Automatic annotations provided by the software were reviewed and validated manually to correct potential errors from image segmentation or atypical cleavage patterns. Only embryos with clearly traceable developmental timelines were included in the final analysis. These data were later correlated with clinical implantation outcomes to evaluate potential predictive relationships.

Morphometric and morphological assessment

Time-lapse monitoring allowed continuous visualisation of blastocyst development up to expansion (Fig. 2), enabling accurate identification of the developmental stage used for morphometric assessment.

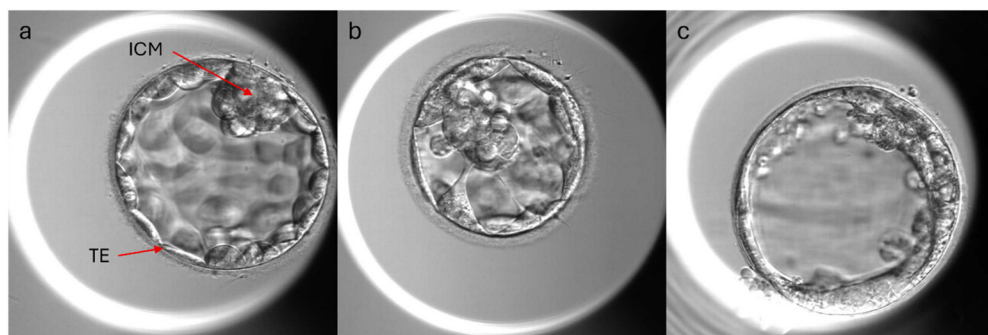


Fig. 2. Morphological grading. ICM was graded A–C based on cell number and compactness (A: tightly packed, numerous cells; B: loosely grouped, several cells; C: very few, loosely aggregated cells), while TE was graded A–C according to epithelial continuity and cell number (A: many cells forming a cohesive epithelium; B: few cells forming a loose epithelium; C: very few large cells). Figures show embryos with grading (a) A/A, (b) B/B and (c) C/C; embryos with grade D ICM or TE were excluded from this study

Morphological grading was performed according to the Gardner and Schoolcraft (1999) system and the Istanbul Consensus criteria, with the 2024 update considered during interpretation [13, 14].

ICM grades

- A: Tightly packed, many cells
- B: Loosely grouped, several cells
- C: Very few, loosely grouped cells
- D: No visible cell or degeneration

TE grades

- A: Many cells forming a cohesive epithelium
- B: Few cells forming a loose epithelium
- C: Very few large cells
- D: Signs of degeneration

Embryos with grade D ICM or TE morphology were excluded from further analysis.

Manual morphometric measurements were performed to quantify blastocyst and inner cell mass (ICM) dimensions. All measurements were taken between 112 and 116 h post-fertilisation, corresponding to the expanded blastocyst stage (Fig. 3). Embryos reaching the expanded blastocyst stage after 116 h post-fertilisation were excluded.

The blastocyst diameter (μm) was measured at its largest cross-section using the straight-line tool, while the ICM diameter (μm) was measured in the focal plane where the ICM borders were most clearly distinguishable. The blastocyst area (μm^2) and ICM area (μm^2) were calculated using the ellipse tool. In cases of irregularly shaped or loosely aggregated ICM structures, measurements were performed using the best-fitting ellipse approximation based on the visible ICM borders.

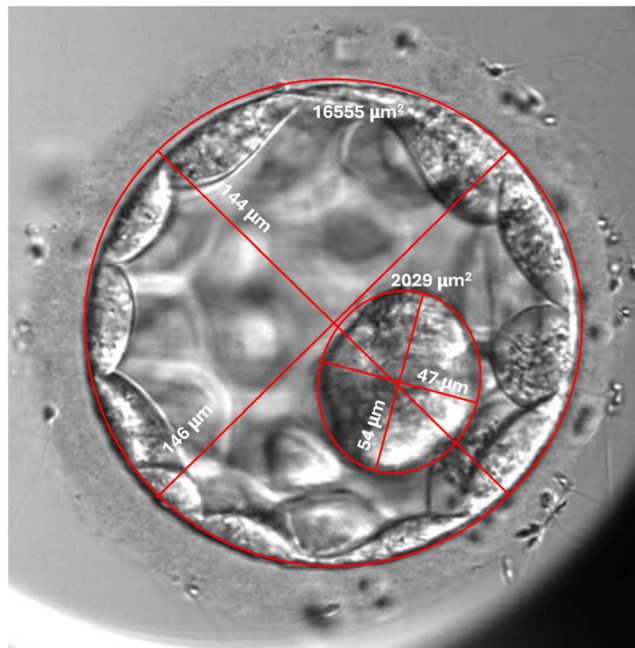


Fig. 3. Measuring the embryo. Area and diameter of the blastocyst and ICM at the largest cross-section measured using ellipses and straight lines fitted to the embryo

From these data, two ratios were calculated to normalise ICM size relative to overall embryo dimensions: ICM-to-blastocyst diameter ratio and ICM-to-blastocyst area ratio. We used these ratios to compare the relative contribution of the ICM to total blastocyst size between embryos that resulted in clinical pregnancy (P+) and those that did not (P-).

Embryos reaching the expanded blastocyst stage but showing insufficiently visible ICM structures or incomplete visualization within the field of view were excluded from morphometric analysis because reliable measurements could not be obtained.

Morphokinetic scores

Two embryo evaluation systems were used to estimate implantation potential based on morphokinetic and morphological parameters.

KIDScore D5 algorithm (Vitrolife) assigns a numerical score ranging from 1.0 to 9.9, which reflects the likelihood of implantation. The score is calculated using annotated developmental timings (including pronuclear number, cleavage events, and time to blastocyst formation) together with morphological parameters of the ICM and TE.

iDAScore system is based on artificial intelligence and evaluates entire time-lapse image sequences without manual annotation. The software automatically generates a morphokinetic score ranging from 1.0 to 9.9, which correlates positively with implantation potential.

In the present study, KIDScore D5 v3.1 and iDAScore v1.0 models (Vitrolife) were used, as these versions were clinically implemented in our laboratory during the study period. For each embryo, both the KIDScore and iDAScore values were recorded at the time of evaluation. These morphokinetic scores were subsequently compared between embryos that resulted in clinical pregnancy (P+) and those that did not (P–).

Pregnancy outcome

Biochemical pregnancy was defined as a positive serum β -hCG test (>10 IU/L) performed 10–14 days after embryo transfer. Clinical pregnancy was confirmed by the presence of a gestational sac on transvaginal ultrasound examination approximately 4–6 weeks after embryo transfer.

Live birth was defined as the delivery of a live newborn after ≥ 24 weeks of gestation. Spontaneous abortion was defined as pregnancy loss before 24 weeks of gestation without medical induction.

Statistical analysis

Data were analysed using Microsoft[®] Excel (v16.65) and Statistica (Cloud Software Inc., v14.0.1.25 and 14.2.0.18) software. Continuous variables are presented as mean $\pm$ standard deviation (SD). Group comparisons between implanted (P+) and non-implanted (P–) embryos were performed using two-sample Student's *t*-tests (two-tailed), with statistical significance set at $P < 0.05$.

Receiver operating characteristic (ROC) curve analysis and area under the curve (AUC) calculations were performed to evaluate the predictive performance of iDAScore and KIDScore for clinical pregnancy outcome. Effect sizes for group differences were estimated using Cohen's *d*. Variance differences between groups were assessed using Levene's test.

Exploratory binary logistic regression analysis was additionally performed to evaluate whether iDAScore and KIDScore remained associated with clinical pregnancy outcome after adjustment for maternal age.

RESULTS

In this study, we analysed the development and morphology of blastocyst stage embryos. During the study period, 213 fresh Day 5 single embryo transfers were performed. Among these, 166 transferred embryos reached the expanded blastocyst stage. In 107 cases, both the blastocyst and ICM could be reliably measured using time-lapse imaging. Five additional cases were excluded because of technical limitations affecting morphokinetic score extraction, resulting in a final study cohort of 102 embryos. Each embryo transfer included in the analysis originated from a different patient, and no patient contributed more than one treatment cycle to the study cohort.

The mean maternal age at the time of embryo transfer was 35 ± 4.4 years (mean $\pm$ SD). Maternal age did not differ significantly between the implanted (P+) and non-implanted (P–) groups (34.8 ± 4.3 vs. 34.9 ± 4.4 years, respectively; $P = 0.949$).

Of the 102 embryo transfers included in the final study, 58 (56.9%) resulted in biochemical pregnancy, and 48 (47.1%) progressed to clinical pregnancy. Nine clinical pregnancies ended in spontaneous or induced abortion, while 39 pregnancies resulted in live birth, corresponding to a live birth rate of 38.2% per embryo transfer. Two twin pregnancies occurred (2% of all transfers; 4.2% of clinical pregnancies), both resulting in the delivery of two healthy neonates.

Morphometric measurements of all embryos are summarised in Table 1. Blastocyst areas ranged from 2,220 to 42,616 μm^2 (mean $\pm$ SD = $23,572 \pm 7,662$), while ICM areas ranged from 1,282 to 6,072 μm^2 ($2,968 \pm 892$). Blastocyst diameters ranged from 125 to 223 μm (157 ± 20.4), and ICM diameters ranged from 27 to 89 μm (58 ± 14.8). The mean ICM-to-blastocyst area and diameter ratios were 0.133 ± 0.06 and 0.38 ± 0.12 , respectively. KIDScore values ranged from 3.4 to 9.8 (7.3 ± 1.5), whereas iDAScore values ranged from 5.9 to 9.9 (8.95 ± 0.7).

Comparison between embryos resulting in clinical pregnancy (P+) and those that did not (P–) revealed no statistically significant differences in blastocyst or ICM areas, diameters, or their ratios (Table 2). Mean values of blastocyst area, ICM area, and blastocyst diameter were higher in implanted embryos compared with non-implanted embryos.

Morphokinetic parameters of implanted and non-implanted embryos and the morphokinetic scores estimating the probability of implantation are summarised in Table 3. Developmental timing parameters, including pronuclear fading (tPNf), cleavage to the 2-cell (t2), 4-cell (t4), 8-cell (t8) stages, as well as time to blastocyst formation (tB), were slightly shorter in implanted embryos compared with non-implanted embryos; however, none of these differences reached statistical significance. Correspondingly, only small effect sizes were observed for morphokinetic timing parameters overall.

KIDScore values were slightly higher in implanted embryos, but the difference was not statistically significant (7.5 ± 1.5 vs. 7.2 ± 1.6 ; $P = 0.229$; Cohen’s $d = 0.24$). In contrast,

Table 1. Morphometric parameters of all embryos (N = 102)

Parameter (N = 102)	Min. – Max.	Mean $\pm$ SD
Blastocyst area (μm^2)	2,220 – 42,616	$23,572 \pm 7,662$
ICM area (μm^2)	1,282 – 6,072	$2,968 \pm 892$
ICM/blastocyst area ratio	0.037–0.31	0.133 ± 0.06
Blastocyst diameter (μm)	125–223	157 ± 20.4
ICM diameter (μm)	27–89	58 ± 14.8
ICM/blastocyst diameter ratio	0.15–0.66	0.38 ± 0.12
KIDScore	3.4–9.8	7.3 ± 1.5
iDAScore	5.9–9.9	8.95 ± 0.7

Summary of morphometric parameters and morphokinetic scores measured in all embryos included in the study. Data are presented as minimum, maximum, and mean $\pm$ standard deviation (SD).

Table 2. Morphometric parameters in implanted (P+) and non-implanted (P–) embryos

Morphological measurements	P+ (N = 48)	P– (N = 54)	p value	Cohen’s d
Blastocyst area (μm^2)	$24,349 \pm 7,090$	$22,881 \pm 8,141$	0.337	0.19
ICM area (μm^2)	$3,069 \pm 977$	$2,878 \pm 806$	0.290	0.21
ICM/blastocyst area ratio	0.13 ± 0.06	0.13 ± 0.06	0.933	0.00
Blastocyst diameter (μm)	159 ± 20.9	154 ± 18.7	0.184	0.25
ICM diameter (μm)	58 ± 15.9	57 ± 13.9	0.573	0.07
ICM/blastocyst diameter ratio	0.38 ± 0.12	0.38 ± 0.11	0.997	0.00

Comparison of morphometric parameters between implanted (P+) and non-implanted (P–) embryos. Data are presented as mean $\pm$ SD. Effect sizes were estimated using Cohen’s d.

Table 3. Kinetic parameters and morphokinetic scores of embryos that resulted in clinical pregnancy (P+) and those that did not (P–)

Morphokinetic parameters	P+ (N = 48)	P– (N = 54)	p value	Cohen’s d
tPNf (hour)	22.6 ± 2.8	23.0 ± 2.8	0.501	–0.13
t2 (hour)	25.3 ± 2.9	25.6 ± 2.9	0.540	–0.12
t4 (hour)	37.1 ± 4.0	37.7 ± 4.6	0.485	–0.14
t8 (hour)	55.6 ± 7.1	56.6 ± 7.9	0.505	–0.13
tB (hour)	101.6 ± 5.2	102.4 ± 5.3	0.444	–0.15
KIDScore	7.5 ± 1.5	7.2 ± 1.6	0.229	0.24
iDAScore	9.1 ± 0.4	8.8 ± 0.8	0.013	0.50

Comparison of key developmental time points and morphokinetic scores between implanted (P+) and non-implanted embryos (P–). Data are presented as mean ± SD. Effect sizes were estimated using Cohen’s d.

iDAScore values were significantly higher in implanted embryos compared with non-implanted embryos (9.1 ± 0.4 vs. 8.8 ± 0.8 ; $P = 0.013$), corresponding to a moderate effect size (Cohen’s $d = 0.50$).

In addition to higher mean values, implanted embryos demonstrated significantly lower variability in iDAScore values. Variance analysis using Levene’s test confirmed a significantly narrower score distribution among implanted embryos ($P = 0.0012$), indicating greater consistency of iDAScore predictions in embryos resulting in clinical pregnancy (Fig. 4).

Receiver operating characteristic (ROC) curve analysis demonstrated slightly superior predictive performance of iDAScore compared with KIDScore for clinical pregnancy outcome. The area under the curve (AUC) was 0.606 for iDAScore and 0.563 for KIDScore.

Exploratory binary logistic regression analysis was performed to evaluate whether morphokinetic scoring systems remained associated with clinical pregnancy outcome after adjustment for maternal age. In the model including iDAScore and maternal age, iDAScore remained a significant independent predictor of clinical pregnancy outcome (odds ratio [OR] = 2.41, 95% confidence interval [CI]: 1.15–5.06; $P = 0.020$), whereas maternal age was not significant ($P = 0.938$). In contrast, in the model including KIDScore and maternal age, KIDScore did not remain a significant predictor of implantation outcome (OR = 1.17, 95% CI: 0.91–1.52; $P = 0.226$).

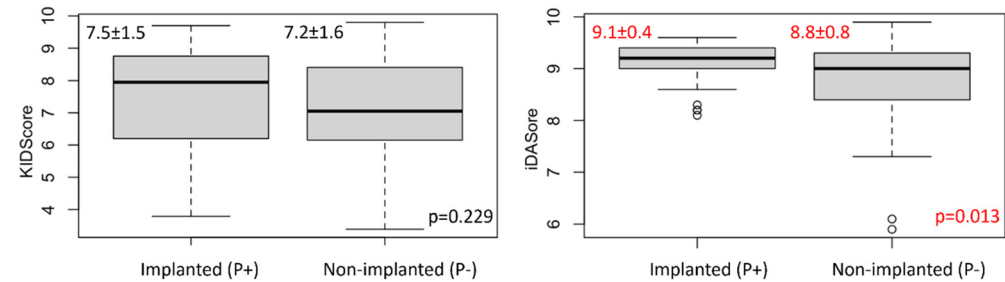


Fig. 4. Distribution of KIDScore and iDAScore values for implanted (P+) and non-implanted embryos (P–). Box plots show the median, interquartile range, minimum and maximum values for each group. Mean ± SD values and p values are indicated within the panels. While KIDScore values did not differ significantly between groups ($p = 0.229$), implanted embryos demonstrated significantly higher iDAScore values ($p = 0.013$) and a narrower score distribution compared with non-implanted embryos

DISCUSSION

Infertility is an increasing global concern, making it essential to continuously work towards improving the effectiveness of IVF treatments. One important aspect is the improvement of embryo culture and evaluation methods. In this study, we aimed to identify novel parameters for embryo prioritisation. We analysed morphometric and morphokinetic parameters of human embryos cultured using TLT and compared those that resulted in clinical pregnancy (P+) with those that did not (P−). No significant differences were observed between the two groups in terms of blastocyst or ICM areas, diameters, or their ratios. In an early study, Richter et al. [15] showed a strong correlation between ICM size and shape and embryo viability. They found that embryos with an ICM larger than $4,500\ \mu\text{m}^2$ had twice as high implantation rate than embryos with a smaller ICM, however, in this older study they did not use TLT. Almagor et al. [4] investigated the ratio of ICM/blastocyst diameters and found that a higher ratio indicates a significantly higher likelihood of implantation. Despite a trend towards larger blastocyst and ICM dimensions in implanted embryos, our findings did not confirm earlier reports suggesting that these features are strong predictors of implantation outcomes. In our study, the lack of confirmation may be attributable to differences in sample size, culture conditions, or the use of more advanced and objective time-lapse monitoring systems.

Recent observations indicate that subcellular morphological structures, such as cytoplasmic strings and trophoctoderm cell architecture, may play an important role in early human embryogenesis and may serve as additional parameters for embryo assessment [16]. Future integration of such features into non-invasive AI-based evaluation systems may improve embryo prioritisation strategies.

In our study, morphokinetic parameters, including the time of pronuclear fading and subsequent cleavage stages, showed slightly faster progression in implanted embryos, although these differences did not reach statistical significance. Desai et al. [17] also found that implanted embryos showed significantly faster development for several parameters; however, their analysis included a substantially larger dataset, highlighting the need for studies with higher statistical power to confirm subtle trends.

Our analysis included both IVF and ICSI cycles, as no significant advantage of intracytoplasmic sperm injection over conventional IVF has been reported in women of advanced maternal age or with a low oocyte number [18]. Because fertilisation timing differs between conventional IVF and ICSI, morphokinetic timings may not be directly comparable between the two insemination methods. In the present study, developmental timings were not normalised to pronuclear fading or other reference points, which represents a limitation of the analysis and may have contributed to the lack of significant differences observed for morphokinetic parameters.

In addition to conventional morphological assessment, two morphokinetic scoring systems were evaluated. KIDScore D5 is a morphokinetic prediction model that combines annotated developmental timings and morphological grading, whereas iDAScore is an artificial intelligence-based system that analyses full time-lapse sequences using deep-learning algorithms to provide an automated implantation probability score. Previous multicentre studies demonstrated that iDAScore outperforms traditional morphokinetic models, including KIDScore D5, in predicting implantation and live birth outcomes [8].

In our dataset, KIDScore values were marginally higher for embryos resulting in clinical pregnancy, but this difference did not reach statistical significance. Conversely, iDAScore values were significantly higher in the P+ group, confirming previous findings on its predictive robustness. Moreover, the dispersion of iDAScore values was narrower among implanted embryos, indicating lower variability and suggesting that higher implantation potential is associated with more consistent developmental dynamics.

Additional statistical analyses further supported the predictive relevance of AI-based embryo assessment. ROC curve analysis demonstrated higher predictive performance for iDAScore compared with KIDScore, while exploratory logistic regression analysis showed that iDAScore remained significantly associated with clinical pregnancy outcome even after adjustment for maternal age. Although the observed predictive performance remained moderate, these findings support the potential clinical utility of AI-assisted embryo prioritisation systems.

This is consistent with the growing understanding that AI systems recognise complex visual and kinetic patterns beyond human perception. Certain morphological abnormalities, such as the presence of giant oocytes or excessive perivitelline space, can be readily identified in static images and have been associated with compromised developmental potential [19]. Similarly, features such as cytoplasmic strings or irregular cytoplasmic structures may contribute to the visual patterns detected by deep learning models [20]. The recent release of large annotated human blastocyst image datasets further facilitates the training and benchmarking of these AI systems, underscoring the transformative role of data-driven embryo assessment in reproductive medicine. Because deep learning models evaluate entire image sequences rather than predefined manually annotated parameters, they may capture subtle developmental characteristics that are not readily identifiable during routine embryological evaluation.

Overall, our findings suggest that while morphometric and morphokinetic parameters remain valuable for descriptive embryo evaluation, their independent predictive strength is limited in smaller cohorts. In contrast, AI-based assessment models such as iDAScore provide a more holistic and reproducible approach, capable of integrating complex temporal and morphological data. Rather than supporting absolute embryo selection, advanced morphokinetic assessment may contribute to more effective embryo prioritisation strategies, potentially reducing the time-to-pregnancy by facilitating earlier transfer of embryos with higher implantation potential.

Nevertheless, while embryo prioritisation may improve pregnancy rates per transfer and reduce time-to-pregnancy, strict selection strategies should be interpreted cautiously, as improvements in cumulative live birth rates across all embryos within a treatment cycle are not always guaranteed [21].

Although AI-based systems demonstrate promising predictive performance, their decision-making processes remain only partially interpretable. Current deep learning models can identify complex image-based patterns associated with implantation outcome; however, the specific biological features underlying these predictions are not yet fully understood. Therefore, continued investigation of embryonic developmental parameters remains important both for improving model interpretability and for advancing our understanding of early human embryogenesis. In addition, despite the growing role of AI-assisted systems in reproductive medicine, expert embryologist oversight remains essential for clinical decision-making.

In conclusion, conventional morphometric and morphokinetic parameters showed limited predictive value for clinical pregnancy outcome in the present cohort. In contrast, the AI-based

iDAScore system demonstrated significantly higher predictive performance and lower variability among implanted embryos, supporting its potential utility in embryo prioritisation during IVF treatment. Nevertheless, further large-scale prospective studies are required to validate these findings and to improve the interpretability of AI-assisted embryo evaluation systems.

STUDY LIMITATIONS

Limitations of the present study should be acknowledged. First, this was a retrospective single-centre study with a relatively limited sample size, which may have reduced statistical power for detecting subtle morphokinetic differences between groups. Second, both IVF and ICSI embryos were included without normalisation of developmental timings to fertilisation-related reference points. Third, only transferred embryos reaching the expanded blastocyst stage were analysed, which may introduce selection bias. Finally, the present analysis was performed using iDAScore v1.0, while newer versions of the algorithm have demonstrated improved predictive performance in more recent studies.

Author contributions: AZ, AN, and PF were responsible for the study design and conceptualization. AZ and FP performed the data analysis and interpreted the results. Laboratory IVF procedures were carried out by clinical embryologists AN, JK, DB, and FP. Clinical management, including ovarian stimulation, oocyte retrieval and embryo transfer, was performed by gynaecologist ÉBB, AA, AM, and JU. AZ drafted the manuscript and prepared the figures. PF and JU supervised the study and revised the manuscript. All authors contributed to the critical revision of the article for important intellectual content and approved the final version for submission.

Competing interests: The authors have no financial or proprietary interests in any material discussed in this article.

Ethics statement: Ethical approval was granted by the Institutional Review Board (IRB) of the NNGYK Ethics Committee (Ref: NNGYK/06334-5/2025). As this was a retrospective analysis of de-identified morphokinetic data obtained during routine IVF care, the requirement for informed consent was waived by the committee.

Data availability statement: The data supporting the findings of this study are available from the corresponding author upon reasonable request.

ACKNOWLEDGEMENTS

The authors would like to thank the nursing and administrative staff for their dedication to patient care and assistance with the data collection process.

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