External validation of a time-lapse prediction model

Thomas Fréour, Pharm.D., Ph.D.,[a,b] Nicolas Le Fleuter, Pharm.D., M.Sc.,[a,c] Jenna Lammers, M.Sc.,[a,b] Carole Splingart, Pharm.D., M.Sc.,[a] Arnaud Reignier, M.Sc.,[a,b] and Paul Barrière, M.D., Ph.D.[a,b]

[a] Service de Médecine et Biologie du Développement et de la Reproduction, CHU de Nantes, Nantes; [b] INSERM UMR1064, Nantes; and [c] Centre d'AMP Procréalis, La Roche-sur-Yon, France

Objective: To study the performance of a previously published implantation prediction model based on morphokinetics in a different setting, in an unselected population and with various embryo transfer strategies.
Design: Retrospective monocentric study.
Setting: University-based assisted reproduction technology (ART) center.
Patient(s): 450 unselected couples undergoing intracytoplasmic sperm injection (ICSI) cycle with embryo culture in the EmbryoScope (Unisense Fertilitech), corresponding to 528 embryos with known implantation.
Intervention(s): None.
Main Outcome Measure(s): Implantation rates (IR) in embryo categories defined by the model in the overall population and in subgroups according to the day of embryo transfer.
Result(s): The distribution of IR among detailed morphokinetic categories in the overall population and in subgroups according to the day of embryo transfer was more heterogeneous than expected according to the published model. The distribution corresponded better to the original when a simplified version of the model was used, although it worked better in the cleavage-stage group than in the blastocyst-stage group.
Conclusion(s): This study was unsuccessful in replicating the sensitivity of the previously published model for predicting implantation rate of embryos ranked according to morphokinetic categories. Further work is required to assess the utility of the model for embryo selection. Each team using time-lapse technology should build a center-specific prediction model based on its own data and transfer policy. (Fertil Steril® 2015;103:917–22. ©2015 by American Society for Reproductive Medicine.)
Key Words: External validation, implantation, time-lapse, prediction model

Discuss: You can discuss this article with its authors and with other ASRM members at http://fertstertforum.com/freort-time-lapse-prediction-model/

Use your smartphone to scan this QR code and connect to the discussion forum for this article now.*

* Download a free QR code scanner by searching for "QR scanner" in your smartphone's app store or app marketplace.

Despite a large variation in success rates across countries, in vitro fertilization (IVF) still suffers from a relatively low clinical pregnancy rate in some countries and a high multiple pregnancy rate (1). This relatively limited performance of assisted reproductive technology (ART) can be partly attributed to suboptimal embryo culture conditions and imperfect embryo quality assessment methods. Time-lapse monitoring using built-in cameras inside the incubator offers the potential of avoiding disturbance of culture conditions while giving access to a huge amount of data, thanks to continuous monitoring of dynamic embryo development (2). Despite the increasing number of time-lapse-based studies available in the literature, relatively few have aimed at evaluating clinical outcomes after embryo culture and selection with time-lapse monitoring (3, 4).

Given the important amount of data generated by time-lapse devices, multivariable prediction models can be developed to facilitate and improve embryo selection and finally lead to better clinical outcomes. Although many prediction models have been published in the field of reproductive medicine, few have undergone external validation. For example, few predictive models of pregnancy in IVF cycles based on patients' baseline characteristics have been validated externally (5–7). When considering specifically embryo quality assessment in IVF cycles, several predictive models based on conventional morphology have been published (8–13), but to our knowledge none of these models have undergone external validation, except in one very recently published study where the validation was conducted in a different data set but the same setting (14). As it is of utmost

Received August 19, 2014; revised December 12, 2014; accepted December 15, 2014; published online January 23, 2015.
T.F. has nothing to disclose. N.L.F. has nothing to disclose. J.L. has nothing to disclose. C.S. has nothing to disclose. A.R. has nothing to disclose. P.B. has nothing to disclose.
Reprint requests: Thomas Fréour, Pharm.D., Ph.D., Service de médecine et biologie du développement et de la reproduction, CHU de Nantes, 38 boulevard Jean Monnet, 44093 Nantes, France (E-mail: thomas.freour@chu-nantes.fr).

Fertility and Sterility® Vol. 103, No. 4, April 2015 0015-0282/$36.00

http://dx.doi.org/10.1016/j.fertnstert.2014.12.111

importance to evaluate the model's performance in a data set other than the one used to develop the model (i.e., external validation) (15), the implementation of most of these nonvalidated embryo-ranking models in clinical routine practice should be questioned.

In 2011, Meseguer et al. (16) published the first hierarchical model using early morphokinetic parameters. Since then, few investigators have published other prediction models as clinically usable (17). Even if the investigators themselves performed a retrospective analysis of the model's performance in a large data set originating from the Instituto Valenciano de Infertilidad clinics' network (18), only one totally independent external validation has been reported up to now (19). Unfortunately, this work was presented only as an oral communication, which has prevented detailed analyses of the methods and results used in the study. However, the investigators' conclusion was quite straightforward, stating that the model was ineffective when applied to their independent data set. This was also highlighted in a recent commentary by Kirkegaard et al. (20), confirming that a time-lapse prediction model (17) could have limited performance in an external setting, with limited sensitivity and specificity.

For our part, we chose to perform this external validation of Meseguer's prediction model on an unselected population, comprising various days of embryo transfer—that is, cleavage stage on days 2, 3, or 4 or blastocyst stage on days 5 or 6—to evaluate its utility in real life daily conditions. Indeed, both the population included and the day of embryo transfer have an obvious major impact on IVF outcome and subsequently on the model's performance.

Thus, we evaluated the performance of the previously published prediction model by Meseguer et al. (16) in an unselected population. We evaluated its accuracy in predicting implantation according to the day of embryo transfer—the cleavage or blastocyst stage.

MATERIALS AND METHODS

This retrospective observational study was conducted on all intracytoplasmic sperm injection (ICSI) cycles performed with the EmbryoScope (Unisense Fertilitech) between February 2011 and December 2013 in the IVF unit of the University Hospital of Nantes in Nantes, France. All the patients gave written informed consent for the procedures, for digital recording, and for the use of data related to their medical history. Our procedures were approved by the institutional review board.

All patients undergoing ICSI cycles were eligible for embryo culture in the EmbryoScope without any exclusion criteria, provided that free space was available in the EmbryoScope at the time of ICSI. All patients underwent controlled ovarian stimulation with an antagonist protocol. The gonadotropin starting dose was chosen according to the woman's age, ovarian reserve, and previous IVF cycles. The antagonist was started on stimulation day 6, and cycle monitoring consisted of hormone assays and ultrasonographic scans; ovulation was triggered with recombinant human chorionic gonadotropin (hCG) when at least three follicles reached 17 mm in diameter. Oocyte retrieval was performed 34 to 36 hours later.

Cumulus cells surrounding the oocyte were removed 2 hours after ovum pickup by a short treatment with hyaluronidase (SynVitro Hyadase; Origio), and microinjection of all oocytes was performed 30 minutes later. Immature oocytes were cultured for a few additional hours before being injected. All injected oocytes were then immediately placed in individual microwells within a specific culture dish (EmbryoSlide; Unisense Fertilitech). Each microwell was filled with 25 μL of culture medium. These dishes were then loaded into the EmbryoScope, a tri-gas incubator with built-in microscope that allows time-lapse monitoring of early embryo development. Embryo culture was performed at 37°C under controlled atmosphere with low oxygen pressure (5% O_2, 6% CO_2). Vitrolife sequential medium was used for embryo culture, with embryos being cultured in G1-plus medium from days 0–3 and then in G2-plus medium from day 3 onward.

Each embryo was investigated by detailed time-lapse analysis measuring the exact timing of the developmental events in hours after ICSI procedure, as described by Meseguer et al. (16). The terms t2, t3, t4, t5, t6, t7, and t8 are, respectively, used for exact timings of appearance of embryos with 2, 3, 4, 5, 6, 7, and 8 well-defined blastomeres.

The duration of the cellular cycle between each cleavage was also considered, with s2 corresponding to the synchronicity of the second cellular cycle (i.e., the duration in the three-cell stage) and cc2 corresponding to the second cellular cycle (i.e., the time from the two-cell to four-cell stage). Cleavage-stage embryo(s) or single blastocyst transfer was chosen according to medical history, previous IVF attempts, and early embryo development. When the embryo transfer was performed on day 2, 3, or 4, embryos were selected according to their morphology first, and then according to kinetic analysis (early cleavage, absence of direct cleavage from zygote to three-cell embryo, absence of multinucleation on day 2). Younger women (under 32 years) undergoing their first or second IVF cycle were generally counseled to undergo a single-blastocyst transfer (day 5 or 6). Only blastocysts with grade A or B trophectoderm could be chosen for transfer or vitrification. In cases of early embryo transfer (day 2, 3, or 4), a single- or double-embryo transfer was decided conjointly by medical staff and the couple.

Supernumerary embryos were systematically cultured up to day 5 or 6 and were vitrified (RapidVit Blast; Vitrolife) at the blastocyst stage if they reached the required quality criteria (same as for transfer). A pregnancy test was performed 11 or 12 days after embryo transfer; if the test was positive, a clinical pregnancy was confirmed ultrasonographically 4 to 5 weeks later by detection of gestational sac and fetal heart activity. Implantation rate (IR) was defined as the number of gestational sacs with fetal heart activity observed at ultrasound divided by the number of embryos transferred.

To avoid any interpretation bias due to partial implantations in cycles where only one embryo out of the two transferred had implanted, only embryos with known implantation were included in the analysis. That is, we included cycles without implantation (0%) or cycles with 100% implantation (transfer of one embryo resulting in one gestational sac with fetal heart activity, or transfer of two embryos resulting in two gestational sacs with two fetal heart activities).