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Review

Time-lapse culture with morphokinetic embryo selection improves pregnancy and live birth chances and reduces early pregnancy loss: a meta-analysis

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KEY MESSAGE

A meta-analysis was conducted on five studies with 1637 patients. The analysis showed that the application of time-lapse monitoring together with an embryo-evaluating algorithm was associated with a significantly higher ongoing pregnancy rate, a significantly lower early pregnancy loss and a significantly higher live birth rate.

A B S T R A C T

Embryo evaluation and selection is fundamental in clinical IVF. Time-lapse follow-up of embryo development comprises undisturbed culture and the application of the visual information to support embryo evaluation. A meta-analysis of randomized controlled trials was carried out to study whether time-lapse monitoring with the prospective use of a morphokinetic algorithm for selection of embryos improves overall clinical outcome (pregnancy, early pregnancy loss, stillbirth and live birth rate) compared with embryo selection based on single time-point morphology in IVF cycles. The meta-analysis of five randomized controlled trials (n = 1637) showed that the application of time-lapse monitoring was associated with a significantly higher ongoing clinical pregnancy rate (51.0% versus 39.9%), with a pooled odds ratio of 1.542 ($P < 0.001$), significantly lower early pregnancy loss (15.3% versus 21.3%; OR: 0.662; $P = 0.019$) and a significantly increased live birth rate (44.2% versus 31.3%; OR 1.668; $P = 0.009$). Difference in stillbirth was not significant between groups (4.7% versus 2.4%). Quality of the evidence was moderate to low owing to inconsistencies across the studies. Selective application and variability were also limitations. Although time-lapse is shown to significantly improve overall clinical outcome, further high-quality evidence is needed before universal conclusions can be drawn.

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Introduction

A receptive endometrium, a genetically and metabolically sound embryo and the appropriate synchronization between them are fundamental for achieving a successful ongoing pregnancy and the birth of a healthy baby during the course of IVF for infertility treatment. One of the critical steps limiting success rates in the laboratory phase is the embryo culture and the proper evaluation of the available embryos. It has been generally accepted that, although there are evident correlations, embryo morphology is not always a robust and absolute indicator for implantation potential since sometimes the best looking blastocyst fails to produce pregnancy, or, a morphologically suboptimal embryo can develop into a healthy baby. Finally, on average, only one-third of all cycles result in a pregnancy (Calhaz-Jorge et al., 2016).

Morphological evaluation of the embryos at specific time points has been the method of choice for embryo selection for decades (Cummins et al., 1986; Edwards et al., 1981), although its limitations have later been recognized (Guerif et al., 2007; Racowsky et al., 2009). Morphological evaluation started with the strategy of measuring single features, such as pronuclear size and alignment (Sadowy et al., 1998; Scott et al., 2000; Wright et al., 1990), multinucleation in early cleavage stages (Alikani et al., 2000; Hardy, 1997), blastomere fragmentation (Plachot and Mandelbaum, 1990; reviewed in Munné and Cohen, 1998; Alikani et al., 1999) or blastocyst morphology (Fehilly et al., 1985; Gardner et al., 2000; Hartshorne et al., 1991). Together with classical morphology, timing of cleavages has been also considered to measure the quality of embryos (Johnson and Day, 2000; reviewed in Johnson, 2002). It was shown as early as in the mid-eighties, that embryos with early first and second cleavages can have implantation rates well above 30% (Edwards and Beard, 1999; Edwards et al., 1984). More recent proposals for scoring embryo quality often combine the results of multiple single-point observations (Nagy et al., 2003; Qian et al., 2008; Scott et al., 2007). Consensus guidelines (ALPHA, SART) also propose multiple evaluations; however, they also disclose their limitations in predicting implantation potential (Racowsky et al., 2009; ALPHA, 2011; Hossain et al., 2016). Although multiple observations will increase the robustness of embryo evaluation, it imposes multiple disturbances to the culture environment, possibly stressing the embryo and reducing the embryos' potential to develop and implant. The way to circumvent this 'observational dilemma' is incubation using time-lapse monitoring. This provides information about the development of the embryos in time intervals of 5–10 min, adding up to about 1000 images in each focal plane per embryo during a 5-day culture period compared with the 2–4 static time point observations carried out in normal routine. This imaging procedure alters the basis of embryo evaluation from single discrete time-point observation to continuous observation, changing the timing variable from discrete to continuous. This transition was enabled by the introduction of advanced microscopy for live cell imaging, focusing on the special needs of the human embryo (Cruz et al., 2011; Pribenszky et al., 2010; Wong et al., 2010).

The information obtained through time-lapse monitoring gives us knowledge about the kinetic and morphologic changes and abnormalities an embryo undergoes *in vitro*. Kinetic events can be precisely timed and the correlation of these timings and intervals to blastocyst formation, implantation, live birth and time to pregnancy were investigated in various publications (Castelló *et al.*, 2016 [review]; Ebner *et al.*, 2016). The time-lapse technique puts a time-stamp on all images; such digitalization paves the way for calculated assessments and therefore less subjectivity.

The introduction of time-lapse imaging systems in clinical human IVF, however, has stirred discussions about how new technologies should be implemented in the daily clinical routine. Many reviews and observational studies have discussed the value of time-lapse monitoring in routine laboratory practice (Freour et al., 2015; Kaser and Racowsky, 2014; Kirkegaard et al., 2012, 2014; Montag et al., 2011; Racowsky et al., 2015; Wong et al., 2013). It is as yet unclear, however, whether the observed benefits come from the undisturbed culture or improved selection based on continuous time-lapse images. In short: what is the weight of these benefits in the added value of time-lapse?

Others have suggested that investing in time-lapse and changing the daily routine would not lead to clinical benefits (Armstrong et al., 2015a; Wong et al., 2014). It has been suggested that the clinical benefits of applying new technologies should be verified and documented by randomized controlled trials before general implementation in routine clinical IVF (Harper et al., 2012). A Cochrane review based on three randomized trials (Kahraman et al., 2013; Kovacs et al., 2013; Rubio et al., 2014) with 994 patients concluded that insufficient evidence was available for the benefit of time-lapse imaging (Armstrong et al., 2015b). More recently, another two clinical randomized controlled trials were published (Goodman et al., 2016; Siristatidis et al., 2015), increasing the number of treatment cycles by more than 60%, adding up to 1637 patients, thus justifying a new meta-analysis on this subject. Moreover, from three of the studies we could obtain also data on live birth, which would be worth investigating.

We define time-lapse as an intervention that essentially comprises undisturbed embryo culture and the consideration of the continuous visual information provided by time-lapse imaging for embryo evaluation and selection. We completed a thorough literature search for relevant randomized controlled trials and performed a meta-analysis to see whether time-laps monitoring (TLM) intervention could change clinical outcome.

Materials and methods

Sources

The investigators conducted a literature search in major electronic databases, including *SCOPUS* (the Elsevier database), *Web of Science*, *MEDLINE/PubMed*, *Cochrane Central Register of Controlled Trials (CENTRAL)*, *Latin American and Caribbean Literature on the Health Sciences database* (*LILACS*), *Excerpta Medica database (EMBASE)* and *Cumulative Index to Nursing and Allied Health Literature (CINAHL)* in January 2016 and repeated the search in February 2017 to double-check and augment the original one.

The search strategy aimed to identify prospective randomized controlled trials that randomized patients to time-lapse based embryo culture and assessment or to conventional embryo assessment in IVF cycles. The time period covered in the search was publications up to February 27, 2017. The following keywords for title, abstract and keywords were used to identify relevant studies: 'embryo' and 'time-lapse'. The results were further screened by using the terms 'RCT' or 'clinical trial' or 'randomized' or 'prospective' and by eliminating non-human related and non-English studies or duplicates.

Further efforts were made to identify all available studies, including searching trial registries (ClinicalTrials.gov, WHO International

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Clinical Trials Registry Platform and EU Clinical trial register), consulting with www.opengrey.eu site to identify relevant projects in available research reports, doctoral dissertations or conference papers. The authors of this review also contacted authors for clarification and new data and searched the reference lists of the identified articles for further relevant studies.

Definitions, data, end-points and statistics

Time-lapse is defined as an intervention that essentially comprises undisturbed culture of embryos and the prospective use of the visual information provided by time-lapse monitoring for embryo evaluation.

Eligible studies were screened for number of patients treated. The analysis was conducted using the data from patients randomized (intention to treat). Pregnancy rate was defined as rise in beta-HCG. Ongoing pregnancy rate was defined as presence of gestational sac or fetal heartbeat detected by ultrasound observed between weeks 5 and 16. For early pregnancy loss patients with positive beta-HCG that did not proceed to show gestational sac or did not proceed to fetal heart beat were evaluated. Stillbirth was defined as fetal death after 20 weeks of pregnancy, whereas live birth was considered where the baby was born alive.

The risk of bias was assessed in each of the studies, including randomization, blinding, incomplete outcome data and reporting.

Results are presented as pooled odds ratio with the random effect model applied. The analyses were conducted using MedCalc® 17.4.4 (MedCalc Software, Belgium); $P < 0.05$ was considered statistically significant.

The meta-analysis follows MOOSE and PRISMA guidelines (Stroup et al., 2000; Moher et al, 2009).

Results

Studies, participants and risks of bias

Our database searches and title reviews resulted in 372 publications of interest. After excluding duplicates and non-relevant studies, based on information in the abstracts, 16 studies were further analysed. Seven studies were identified; however, two of those were excluded (Park et al., 2015; Wu et al., 2016) as, in those studies, the visual information from the time-lapse follow-up was disregarded and, because of inclusion of poor-prognosis patients, no embryo selection was possible (Wu et al., 2016).

Finally, five studies (Goodman et al., 2016; Kahraman et al., 2013; Kovacs et al., 2017; Rubio et al., 2014; Siristatidis et al., 2015) involving 1637 randomized patients were included. Study characteristics are described in **Table 1**. Risks of bias were analysed and are described in detail in **Table 2**. Risks of biases are presented in **Table 3**.

Synthesis

The analysis of five studies, including 1637 randomized patients, showed an increase of the ongoing pregnancy rate from 39.9 % to 51.0 % by using time-lapse for continuous embryo assessment compared with the conventional approach at single fixed time points (OR

Table 1 – Characteristics of the studies included in the meta-analysis.

Author	Methods	Participants	Intervention	Outcomes
Kahraman et al., 2013	Single-centre RCT; randomization (1:1) at retrieval; ES at 5% O_2; fresh eSET on D5	Patients under 35 years, first, second, good responders (n = 76/64)	Selection: D5 morphology and Meseguer et al. (2011) hierarchical model for TLM versus D5 morphology in control group;	Embryo development; biochemical pregnancy; clinical pregnancy (w5); miscarriage.
Rubio et al., 2014	Multicentre RCT; randomization (1:1) the day before retrieval; ES at 21% O_2; fresh + frozen eSET, DET on D3 or D5	Patients under 38 years, first, second cycle, autologous or egg donation (n = 856 /843)	Selection: D3 or D5 morphology and Meseguer et al. (2011) hierarchical model for TLM versus D3 or D5 morphology in control group.	Ongoing pregnancy (w12); embryo development; implantation rate; biochemical pregnancy; early pregnancy loss.
Siristatidis et al., 2015	Single-centre RCT: randomization (3:7) after retrieval; PV at 21% O2; fresh eSET, DET, TET on D2 or D3	Patients under 42 years, primary or secondary subfertility (n = 244/ 239)	Selection: according to Meseguer et al. (2011) hierarchical model and Ciray et al., 2014 for TLM versus D2 or D3 morphology in control group.	Clinical pregnancy (w7); ongoing pregnancy (FHB, w12); live birth rates (>w20).
Goodman et al., 2016	Single centre RCT; randomization (1:1) at retrieval; ES at 5.5% O2; eSET or DET on D3 or D5	Patients under 43 years, own oocytes (n = 300 / 235)	Selection: standard morphology and cc_2, t_5, s_2, s_3, tSB and cleavage abnormalities for TLM versus standard morphology in control group.	Clinical pregnancy rate (w6); implantation rate (IR); early pregnancy losses.
Kovacs et al., 2017. (continuation of Kovacs et al., 2013)	Multicentre RCT; randomization (1:1) before stimulation start; PV at 5% O2; eSET on D5	Patients under 36 years, first, second cycle (n = 161 / 139)	Selection: cc_1, cc_2, s_1, s_2, t_5 and BC morphology for TLM vs. D5 morphology in control group.	Biochemical pregnancy rate; ongoing pregnancy rate (w16); miscarriage rate; delivery outcome

BC, blastocyst; D3, day 3 of culture; D5, day 5 of culture; DET, double embryo transfer; ET, embryo transfer; ES, embryoscope; eSET, elective single embryo transfer; FHB, fetal heart beat; N, randomized patients/patients completed the protocol; OPR, ongoing pregnancy rate; PR, pregnancy rate; PV, Primo Vision (Vitrolife, Sweden); RCT, randomized controlled trial; TET, triple embryo transfer; TLM: time-lapse monitoring; w, the given week of gestation. Morphokinetic variables: cc_1, duration of first cell cycle, cc_2, duration of second cell cycle, t_5, timing to five discrete cells, s_1, duration of the first cytokinesis, s_2, the synchronicity of the two blastomere divisions within the second cell cycle, (calculated as t4-t3), s_3, the synchronicity of the four blastomere divisions within the third cell cycle, (calculated as t8-t5), tSB: timing of the initiation of blastulation. The first lines in each of the outcome column cells show the primary end-point of the given study, the forthcoming lines show secondary end-points.

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Table 2 – Risks of biases in individual studies.

Kahraman et al., 2013		
Bias	Judgment	Support for judgment
Random sequence generation (selection bias)	Low risk	Randomization according to a list generated on random.org.
Allocation concealment (selection bias)	Low risk	Sequentially numbered list with groups masked (personal communication).
Blinding of participants and personnel (performance bias)	Low risk	Patients were blinded (personal communication). Incomplete blinding, but investigators judged that the outcome was not likely to be influenced by lack of blinding.
Blinding of outcome assessment (detection bias)	Medium risk	No blinding of outcome assessment.
Incomplete outcome data addressed (attrition bias)	Low risk	Randomization applied for 76 patients; nine dropped-out because of insufficient numbers of good blastocysts on the day; 64 patients were analysed; three dropouts lacked information. The proportion of missing outcomes compared with observed event risk was insufficient to clinically affect the intervention effect estimate.
Selective reporting (reporting bias)	Low risk.	Embryo quality, blastocyst rate, pregnancy and miscarriage rates were reported.
Other bias	Low risk	–
Rubio et al., 2014		
Bias	Judgment	Support for judgment
Random sequence generation (selection bias)	High risk	Patients allocated to either TLM system (study group) or standard incubator (control group) using a computer-generated randomization table, which was handled by the embryologist at the laboratory in charge the day before the oocyte retrieval or oocyte donation. In a limited number of cases, patients could have a preference that would increase risk of bias, ending up with a patient distribution of 51.9 : 48.1.
Allocation concealment (selection bias)	High risk	Randomization was not carried out optimally, as the patient distribution to the two groups would have been expected to be closer to a 50:50 ratio than the reported 51.9:48.1. This deviation was explained by limited patient requests for time-lapse monitoring system culture.
Blinding of participants and personnel (performance bias)	Medium risk	The gynaecologist evaluating the primary effect was not aware to which group the patients had been assigned, and the statistician evaluating the results only knew the incubators by a binary code, not by type. Owing to patient request to allocation, patients were not blinded.
Blinding of outcome assessment (detection bias)	Low risk	The gynaecologist evaluating the primary effect was blinded.
Incomplete outcome data addressed (attrition bias)	High risk	Out of 856 patients, 13 dropped out (1.5%), all of which were detailed and explained. Some participants changed groups.
Selective reporting (reporting bias)	High risk	All data were reported; however, outcomes disclosed in the protocol were different than those reported in the study.
Other bias	Low risk	–
Siristatidis et al. 2015.		
Bias	Judgment	Support for judgment
Random sequence generation (selection bias)	High risk	Last digit of the patient file number was used for random sequence generation: time-lapse or conventional monitoring was offered according to the file number of each patient (0–2 versus 3–9 as a last digit).
Allocation concealment (selection bias)	High risk	Case record number was used to allocate the patient into TLM or control groups. Patients had to agree the allocation. If the patient did not accept the group assignment, they were excluded from the trial. Two cases in the TLM group and three in the control group were excluded.
Blinding of participants and personnel (performance bias)	High risk	Neither the patient, physician nor embryologist were blinded.
Blinding of outcome assessment (detection bias)	Low risk	Patients were checked by different gynaecologists who did not know the randomization.
Incomplete outcome data addressed (attrition bias)	Low risk	244 cycles were randomized, five dropped out: two from the TLM group, three from the control group. Causes were sufficiently explained.
Selective reporting (reporting bias)	Low risk.	All data were reported.
Other bias	Low risk	-

(*continued on next page*)

1.542; CI 1.211 to 1.965; $P < 0.001$). In parallel, early pregnancy loss ($n = 904$) was significantly reduced from 21.3% to 15.3% (OR: 0.662; CI 0.469 to 0.935; $P = 0.019$) (**Figure 1A and 1B**) (Goodman et al., 2016; Kahraman et al., 2013; Kovacs et al., 2017; Rubio et al., 2014; Siristatidis et al., 2015). Live birth rate ($n = 481$) increased significantly, from 31.3% to 44.2% (OR 1.668; CI 1.134 to 2.455; $P = 0.009$) if TLM was used, however, no difference was found between the groups in stillbirth rates (2.6% versus 4.7%; OR 2.483; CI 0.794 to 7.759) (**Figure 1C and 1D**) (Kahraman et al., 2013; Siristatidis et al., 2015; and Kovacs *et al.*, 2017). Chi-square analysis showed similarly a significant effect of TLM in case of ongoing pregnancy, early pregnancy loss and live birth, whereas stillbirth was not different between the groups (results are

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Table 2 – (*continued*)

Goodman et al. 2016

Bias	Judgment	Support for judgment
Random sequence generation (selection bias)	Low risk	Randomized 1:1 to conventional embryo selection versus embryoscope time-lapse morphokinetic selection with the use of a computer-generated random number sequence.
Allocation concealment (selection bias)	Low risk	The randomization list was held in the laboratory, where it was accessible only by research personnel not involved with the recruitment of patients.
Blinding of participants and personnel (performance bias)	Low risk	Patients, reproductive endocrinology physicians and staff, and sonographers were blinded to how embryos were selected.
Blinding of outcome assessment (detection bias)	Low risk	Sonographers were blinded to how embryos were selected.
Incomplete outcome data addressed (attrition bias)	Low risk	300 cycles were randomized: 31 dropped out from the TLM group and 34 from the control group. Two dropouts from the control group were not explained.
Selective reporting (reporting bias)	Low risk	All data were reported.
Other bias	Low risk	–

Kovacs et al. 2017 (continuation of Kovacs et al., 2013)

Bias	Judgment	Support for judgment
Random sequence generation (selection bias)	Low risk	Eligible patients who consented to participate were randomized in blocks of two, by selecting TLM or control assignments from closed, opaque envelopes.
Allocation concealment (selection bias)	High risk	Randomization was carried out by the principal investigator by selecting TLM or control assignments from closed, opaque envelopes. Using blocks of two raised the possibility for bias.
Blinding of participants and personnel (performance bias)	Low risk	Patients were blinded to their assignment. The embryologist was not blinded and it could not be ensured that the gynaecologist carrying out the transfer was blinded. The review authors judge that the outcome is not likely to be influenced by lack of blinding.
Blinding of outcome assessment (detection bias)	Medium risk	Most of the outcome assessment was made outside of the clinic.
Incomplete outcome data addressed (attrition bias)	Low risk	'A total of 161 patients were randomized. Twenty-two patients (12 in group 1 and 10 in group 2) dropped out for various reasons (group 1: double-embryo transfer requested: 2; no fertilization: 1; < 3 good embryos on day 3: 7; elective cryopreservation due to hyperstimulation risk: 2. For Group 2: < 3 good embryos on day 3: 8; no fertilization: 1; elective cryopreservation for hyperstimulation risk: 1) and 139 completed the trial.' The missing outcome data are balanced and unlikely to be related to true outcome.
Selective reporting (reporting bias)	Low risk	All data were reported.
Other bias	High risk	Interim monitoring took place, but it was compensated for in the analysis. There was a baseline imbalance (the average age of patients in the groups differed: 31.2 ± 2.7 versus 32.1 ± 2.5).

TLM, time-lapse monitoring.

Table 3 – Risk of biases.

	Random sequence generation	*Allocation concealment*	*Blinding, participants/ personnel*	*Blinding, outcome assessment*	*Incomplete outcome data*	*Selective reporting*	*Other bias*
Kahraman *et al.*, 2013	+	+	+	+/–	+	+	+
Rubio *et al.*, 2014	–	–	+/–	+	–	–	+
Siristatidis *et al.*, 2015	–	–	–	+	+	+	+
Goodman *et al.*, 2016	+	+	+	+	+	+	+
Kovacs *et al.*, 2017 (continuation of Kovacs *et al.*, 2013)	+	–	+	+/–	+	+	–

– High risk of bias + Low risk of bias +/– Medium risk of bias

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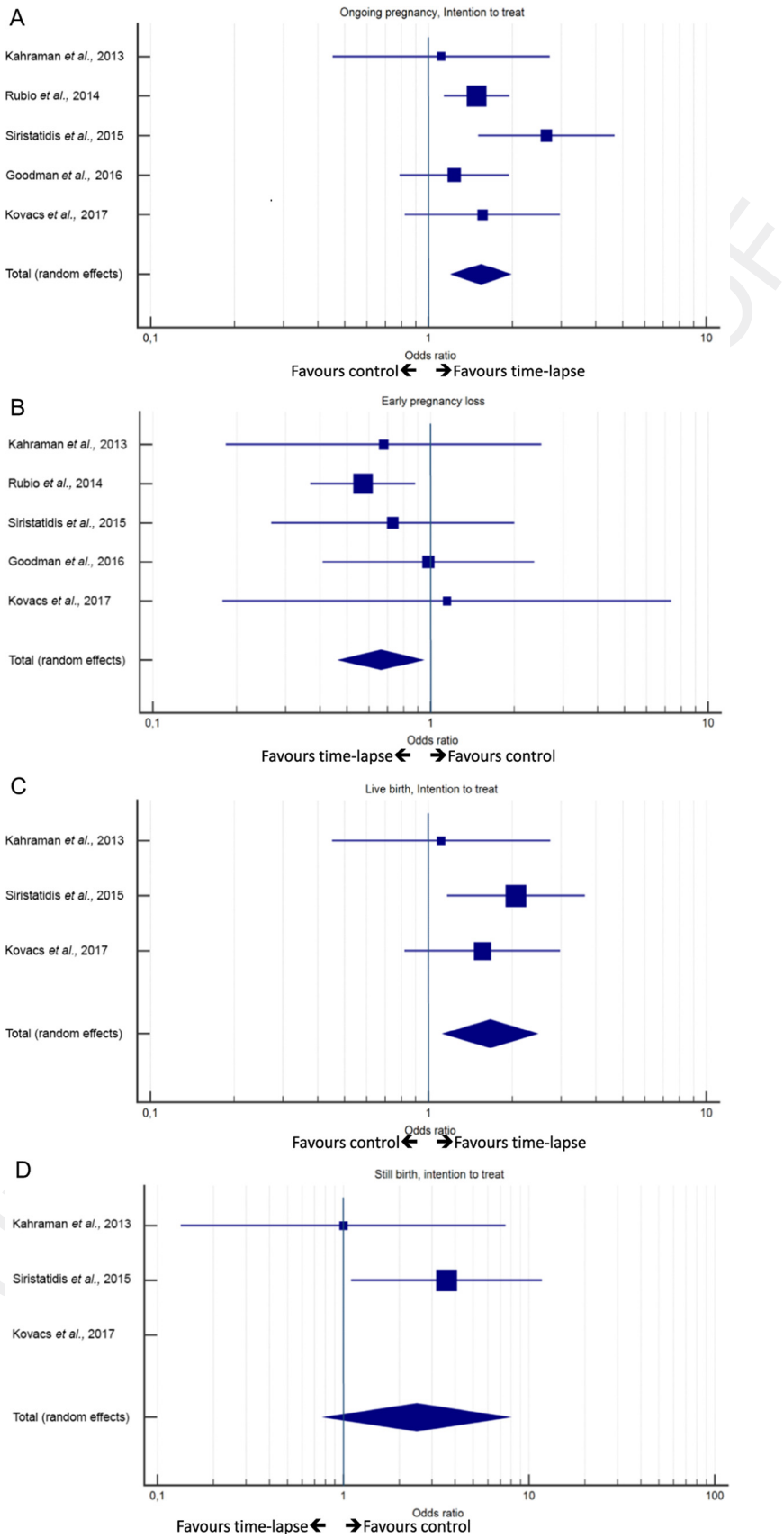

Figure 1 – (A) Ongoing pregnancies; (B) early pregnancy losses; (C) live births and (D) stillbirths in randomized controlled trials. (A) Analysis favours time-lapse with OR 1.542 (CI 1.211 to 1.965; $P < 0.001$) ($n = 1637$); (B) analysis favours time-lapse with OR: 0.662 (CI 0.469 to 0.935; $P = 0.019$) ($n = 904$); (C) analysis favours time-lapse with OR: 1.668 (CI 1.134 to 2.455; $P = 0,009$) ($n = 481$); (D) no difference between the groups (OR 2.483; CI 0.794 to 7.759; $P = 0.1180$) ($n = 481$).

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Table 4 – Chi-square analysis of the time-lapse monitoring effect on clinical outcome.

	Time-lapse monitoring, n (%)	Conventional culture and evaluation, n (%)	P-value
Ongoing pregnancy	400/784 (51.0)	340/853 (39.9)	<0.0001
Early pregnancy loss	72/472 (15.3)	92/432 (21.3)	NS
Live birth	84/190 (44.2)	91/291 (31.3)	0.0040
Stillbirth	9/190 (4.7)	7/291 (2.4)	NS

NS, not statistically significant.

presented in **Table 4**). Summary of findings, including the quality of evidence, is presented in **Table 5**.

Discussion

In-vitro culture of embryos and the in-process decision-making of whether to transfer, cryopreserve or discard is the most crucial and challenging task in human assisted reproduction. Although the first time-lapse analysis of the developing mammalian embryo was published in 1929 (Lewis and Gregory, 1929), a further 80 years passed until the first routine clinical use of time-lapse in the human IVF laboratory (Meseguer et al., 2011; Pribenszky et al., 2010).

Since then, many observational studies have evaluated time-lapse on a small scale, using different patient populations, different culture conditions, different approaches for the purpose of undisturbed culture, as a tool to deselect abnormal embryos, predict blastocyst formation, or embryo selection for transfer using morphokinetics. Different nomenclature was also used. This also applies to the currently available randomized studies, as these are y diverse in many aspects and some, by themselves, are not even adequate to draw a conclusion on utility of time-lapse monitoring. Summing up studies is needed at certain stages, however, as Cohen and Alikani (2013) wrote in their 2013 paper: systematic reviews can only be as strong as the work they include.

Notwithstanding the above caveats, the synthesis of this meta-analysis is different to the conclusions of previously published meta-analyses.

The first meta-analysis presented by Polanski et al. (2014) evaluated the possible benefits of time-lapse based on an interim report of a randomized controlled trial with 62 enrolled patients (Kovacs et al., 2013) and a non-inferiority study with 76 patients (Kahraman et al., 2013). These authors concluded that time-lapse embryo imaging does not alter the chance of achieving ongoing and clinical pregnancy and called for further evidence (Polanski et al., 2014). After the publication by Rubio et al. (2014), a Cochrane report was published that analysed the three studies with 994 patients altogether (Armstrong et al., 2015b). With some flaws in the analysis (positive beta-HCG of a study was added to the 'clinical pregnancy group'; two studies rather than one used an embryo selection algorithm based on morphokinetics), the conclusions were similar to the first meta-analysis: insufficient evidence for differences in live birth, miscarriage, stillbirth or clinical pregnancy to choose between time-lapse and conventional incubation, and called for further studies. After the study by Park et al. (2015), a further analysis by Racowsky et al. (2015) showed similar findings, based on 1358 patients, stating that 'While TLM has the potential to revolutionize clinical embryology, there are currently no high-quality data to support the clinical use of this technology for selection of human preimplantation embryos.'

Since the last published review, two more relevant studies have been published, one of them including 300 patients, adding a considerable number of cases to the pool (Goodman et al., 2016). Moreover, Kovacs *et al.* (2017) completed their study after an initial

Table 5 – Findings.

Outcomes	Assumed risk (conventional incubation, median risk population)	Corresponding risk (time-lapse, median risk population)	Relative effect (95% CI)	Number of participants (studies)	Quality of evidence (GRADE)
Ongoing pregnancy	410/1000	517/1000 (457 to 577)	OR 1.542 (1.211 to 1.965)	1637 (5 RCTs)	Moderate[a,e]
Early pregnancy loss	196/1000	139/1000 (103 to 186)	OR 0.662 (0.469 to 0.935)	904 (5 RCTs)	Moderate[b,e]
Live birth	321/1000	441/1000 (349 to 537)	OR 1.668 (1.134 to 2.455)	481 (3 RCTs)	Moderate[c,e]
Stillbirth	29/1000	69/1000 (23 to 188)	OR 2.483 (0.794 to 7.759)	481 (3 RCTs)	Low[d,e]

[a] Direction of the effect is consistent in all five studies. Confidence interval for the pooled estimate is consistent with benefit. Test of heterogeneity is not significant. In 2% of cases in one study (52.3% weight), patients could switch groups; in one study (9.8% weight), allocation was in blocks of two, and the average age of the female differed slightly between the groups and was terminated before reaching full enrollment; one study (14.9% weight) had improper blinding, random sequence generation and allocation.

[b] Direction of the effect is consistent except for one study (7.2% weight). Confidence interval for the pooled estimate is consistent with benefit. Test of heterogeneity for all outcomes is not significant. In 2% of cases in one study (55.2% weight) patient could switch group; in one study (7.2% weight) allocation was in blocks of two, average age of the female differed slightly between the groups, and was terminated before reaching full enrollment; one study (12.4% weight) had improper blinding, random sequence generation and allocation.

[c] Direction of the effect is consistent in case of all studies. Confidence interval for the pooled estimate is consistent with benefit. Test of heterogeneity is not significant. In one study (33.5% weight) allocation was in blocks of two, the average age of the female differed slightly between the groups and was terminated before reaching full enrollment; one study (50.7% weight) had improper blinding, random sequence generation and allocation.

[d] Direction of the effect is not consistent. Test of heterogeneity is not significant.

[e] Confidence intervals overlap. Ages of women varied between the studies. More than 80% of participants were enrolled in trials included in the analysis. Data were reported consistently for the outcome of interest.

GRADE Working Group grades of evidence: high quality: further research is very unlikely to change our confidence in the estimate of effect; moderate quality: further research is likely to have an important impact on our confidence in the estimate of effect and may change the estimate; low quality: further research is very likely to have an important impact on our confidence in the estimate of effect and is likely to change the estimate; vvery low quality: we are very uncertain about the estimate.

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report of their interim analysis (Kovacs et al., 2013), including altogether 161 patients. Combining all five studies, the number of patients is 1637. On the other hand, we have defined time-lapse as the essential combination of undisturbed culture and the morphokinetic information provided by continuous monitoring. For this reason, we excluded those studies from the analysis, where, although embryos had been cultured in a time-lapse device, embryo evaluation was made solely based on single time-point morphology, and the visual information provided by time-lapse was not considered for selection (Park et al., 2015; Wu et al., 2016).

The studies included in this present analysis are heterogeneous in patient population, days of transfer, the way the visual information from the time-lapse devices were used to support embryo evaluation and end-points. Also, different biases are tangible across the studies.

The study by Kahraman et al. (2013) reached low risks in the bias analysis; however, it was designed as a non-inferiority trial. Time-lapse information was a secondary tool to augment the primary classic morphological assessment. For the kinetic assessment of the embryos in the TLM group, the hierarchical model proposed by Meseguer et al. (2011) was used.

The study with the largest number of patients recruited (Rubio et al., 2014) concluded that time-lapse culture and selection provides superior ongoing pregnancy rate and significantly fewer losses compared with control group. Nevertheless, the study combined day 3 and 5 culture, autologous and donor oocyte sources. The culture conditions were different between the time-lapse group and the control group not just based on the different, undefined standard incubators in the control group, but also because of the different volume of culture media used between the groups. One might also criticize that the control group was removed from the incubator for morphology checks at least twice, whereas the time-lapse group was not, but this feature accounts for the beneficial nature of culturing embryos under time-lapse monitoring, which minimizes embryo handling. The Meseguer model was applied for the morphologically normal embryos for embryo selection, which, in principle, is similar to the approach applied in the study by Kahraman et al. (2013).

Siristatidis et al. (2015) was classified having high level of bias in random sequence generation, allocation and blinding. The study population was classified into time-lapse or control groups according to the last digit of the patient file, in 3:7 ratio, which in itself has limitations. There was no blinding, as the classification was actually offered to the patients so both the patient and the attending physician was aware of the allocation. On the other hand, the patient was excluded from the study if she did not accept the allocation, which happened in case of two patients (3%) in the TLM group and in case of three patients (2%) in the control group. Unlike in the Rubio study, patients did not have the chance to switch group. The embryologists used morphokinetic information from the time-lapse device for classic morphology assessment through the scrolling of the sequence of the digital images, and, in addition, kinetic events were used to define optimal times for cleavages and interphases.

The Goodman study (2016) showed low risk of bias in all categories. For embryo evaluation, the authors first determined the top-quality embryos based on morphologic grade and then a morphokinetic score was used to rank the best embryo for transfer.

Although Kovacs et al. (2017) stopped recruitment before the number of patients had reached the target number, they also showed a clear trend towards the benefit of using a time-lapse algorithm for embryo evaluation and selection versus the routine morphological investigation. The interim analysis of the study was published in 2013 (Kovacs et al., 2013), and later in 2015 (Matyas et al., 2015). In 2017, the final results were made public in clinicaltrials.gov. The study was biased at the allocation concealment as allocation was in blocks of two. On the other hand, an imbalance was observed between the ages of women in the groups (31.2 for TLM versus 32.1 for control), bringing another potential bias. For embryo evaluation, the authors scored classic morphology through viewing of time-lapse video footage and also added kinetic scores for normality of cleavages, thus constructing a composed score for selection.

It is also important to note that, because of the various definitions used in the studies, it was not possible to differentiate whether a pregnancy loss occurred after evaluation of the gestational sac in the study by Kahraman et al. (2013). The studies selected for this meta-analysis, however, randomized patients to time-lapse or conventional culture and selection, and assessed the benefit of this new intervention on laboratory performance and clinical outcome. Although they differ methodologically and in power, they can be used to combine results and derive an estimate to find the value of time-lapse.

The combined outcome of these studies shows significant difference between the time-lapse and control groups for ongoing pregnancy and early pregnancy loss and live birth. Although the studies did not weight equally in the combined dataset (4.6%; 52.3%; 14.9%; 18.3%; 9.8%), all showed individually a benefit.

In accordance with the results of this analysis, the significant benefit of time-lapse monitoring and its aid in embryo selection has also been shown in a recent concurrent cohort-controlled prospective study (Adamson et al., 2016). With 319 patients included, they present a 46% clinical pregnancy rate in the time-lapse group compared with 32% in the control group and conclude that TLM adds valuable information to traditional morphologic grading.

The present analysis was carried out using different relevant statistical methods (relative risk and also per protocol analysis; data not presented). The results had similar outcomes, which underlines the robustness of the analysis and confirms the findings.

In conclusion, this meta-analysis supports the growing evidence for the clinical benefit of using imaging systems in human IVF. The combined effect of reduced early pregnancy loss, higher ongoing pregnancy and higher live birth rate after embryo assessment by time-lapse results in a clear clinical benefit, with the potential to also shorten the time to pregnancy. On the basis of the pooled data from the available randomized controlled trials, change of routine practice from standard observation at fixed time-points to continuous observation, together with morphokinetic judgment, is supported by the findings. Nevertheless, general conclusions cannot be made at the moment, as the studies included in this analysis were carried out in selected populations and the quality of some studies included can be questioned.

Uncited references

Desai et al, 2014

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