

GYNECOLOGY

Does maternal age at retrieval influence the implantation potential of euploid blastocysts?



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BACKGROUND: Age-related decline in reproductive potential is mainly due to the increased incidence of aneuploidy. Furthermore, 2 recent studies have shown that euploid embryos of older women may have a lower implantation potential compared to those of younger women, suggesting that aging might compromise embryos beyond their ploidy status. However, the inherent limitations of these studies preclude solid conclusions.

OBJECTIVE: The aim of this study was to determine whether maternal age at retrieval affects the implantation potential of euploid blastocysts.

MATERIALS AND METHODS: This is a retrospective cohort study that was conducted at an academic medical center. Patients who underwent frozen–thawed euploid embryo transfers (FET) between 2013 and 2016 were included. Cycles were divided into the following 5 age groups: <35, 35–37, 38–40, 41–42, and >42 years of age. Blastocysts were assessed before biopsy and assigned the following morphological grades: excellent (3–6AA), good (3–6AB, 3–6BA), average (2–6BB), and poor (3–6BC, 3–6CB, 3–6CC). The main outcome measures were implantation (IR) and live birth (LBR) rates. Both χ^2 and Fisher exact tests were used to compare categorical variables. Odds ratios (ORs) with 95% confidence intervals (CIs) were calculated and controlled for confounders.

RESULTS: A total of 785 FET cycles (870 blastocysts) were included. Excellent-quality blastocysts were associated with a significantly higher

LBR compared with good-quality (78.8% vs 63.8%), average-quality (78.8% vs 54.2%), and poor-quality (78.8% vs 28.3%) counterparts. Poor-quality embryos yielded a higher spontaneous abortion (SAB) rate compared with average-, good-, and excellent-quality blastocysts (25.0%, 9.0%, 6.9%, and 2.4%, respectively). Embryos biopsied on day 5 had a significantly higher LBR compared with those biopsied on day 6 (60.0% vs 46.6%). The 5 age groups (<35, 35–37, 38–40, 41–42, and >42 years) had comparable IRs (56.5%, 52.9%, 55.4%, 59.1%, and 71.4%, respectively), LBRs (55.1%, 51.3%, 53.5%, 52.4%, and 61.9%, respectively), and SAB rates (8.8%, 7.9%, 8.3%, 14.3, and 13.3%, respectively). Older women had fewer euploid embryos, but they were of comparable morphology and developed at a similar rate to the blastocyst stage as compared to those of younger women.

CONCLUSION: Maternal age at retrieval influences the number of euploid embryos; however, contrary to previously published studies, it does not affect their implantation potential. The morphodynamic characteristics of embryos, as reflected by blastocyst morphology and speed of development, are critical for selecting among euploid embryos.

Key words: blastocyst morphological grading, day of trophectoderm biopsy, euploid embryo, maternal age, preimplantation genetic testing for aneuploidy (PGT-A)

Maternal age is the most critical factor determining the likelihood of conception either naturally or following in vitro fertilization (IVF).^{1,2} In fact, the live birth rate (LBR) after IVF decreases from >40% in women <35 years old to \leq 1% in those >44 years.² In addition, the miscarriage rate increases from 10% in young women to >50% in women >40 years old.² These challenges are becoming more relevant as the mean age of first-time mothers rises steadily worldwide.^{3,4}

Uterine receptivity is not a major contributor to the decrease in fecundity

associated with advanced maternal age, as older women who use donated oocytes have LBRs comparable to those of young women.^{2,5} The decline in fecundity is attributed to a lower oocyte yield and, most importantly, higher oocyte aneuploidy rates in older women.^{5–7} Chromosomal analysis of >15,000 trophectoderm biopsy specimens confirmed that aneuploidy rates increase steadily after age 30, reaching 88.2% by age 44 years.⁶ This explains the lower fecundability and higher miscarriage rates in women of this age group, as the majority of their aneuploid embryos either do not implant or lead to pregnancy loss.^{8–10}

Given that the increased incidence of aneuploidy is the main culprit for age-related decline in reproductive potential, it would be logical to assume that euploid embryos of older women have implantation rates comparable to those of young women. However, a study evaluating the transfer of 133 embryos identified as euploid using single-

nucleotide polymorphism microarray-based analysis showed that women \geq 35 years old yielded significantly lower delivery rates compared to women <35 years old.¹⁰ The authors concluded that there are age-related factors other than aneuploidy affecting reproductive potential. Yet, their findings should be carefully interpreted because they included embryos that had undergone 3 biopsies (first polar body, second polar body, and blastomere or trophectoderm) and others that were biopsied once (blastomere or trophectoderm), and there was no adjustment made for the embryo stage at biopsy or the number of biopsies performed between the 2 age groups.¹⁰ This is pertinent because cleavage stage and repetitive biopsies may impair embryo viability.^{10,11}

A multicenter retrospective study including 343 euploid blastocyst transfer cycles, in which trophectoderm biopsies were analyzed with microarray comparative genomic hybridization, showed

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AJOG at a Glance

Why this study was conducted?

To determine whether maternal age at retrieval affects the implantation potential of euploid blastocysts.

Key findings

Although older women had fewer euploid embryos at retrieval, they were of comparable morphology and developed at a similar rate to the blastocyst stage as compared to those of younger women. Euploid embryos of older women had comparable implantation, live birth, and spontaneous abortion rates compared to those of younger women.

What does this add to what is known?

The findings of this study contradict previously published papers, which suggested that the implantation potential of euploid embryos may decline with advanced maternal age at retrieval. It also confirms that blastocyst morphology and speed of development are paramount for selecting among euploid embryos.

comparable ongoing pregnancy rates (OPRs) among 4 age groups (<35, 35–37, 38–40, and 41–42 years) ranging between 53.6% and 64.4%.¹² Although the authors of the study reported 25% OPRs in women >42 years old, they had only 16 cycles in this group. Therefore, the low OPR of euploid blastocysts in this group may not represent a true OPR of the general population due to limited sample size. In addition, this study lacked data on blastocyst morphology, which influences the implantation potential of euploid blastocysts.^{12,13} With this in mind, we aimed to determine the effect of maternal age at retrieval on the implantation potential of euploid blastocysts, while also including a larger sample size and taking blastocyst grading and other confounders into consideration.

Materials and Methods**Cycle selection**

The Institutional Review Board at Weill Cornell Medicine approved this study. Autologous frozen–thawed embryo transfer (FET) cycles of euploid blastocysts that occurred at the Ronald O. Perleman and Claudia Cohen Center for Reproductive Medicine between January 2013 and December 2016 were reviewed for inclusion. The only cycles that we excluded were those in which 2 blastocysts of different morphological grades or 2 blastocysts biopsied on different days (day 5 and day 6) were transferred.

Cycles were divided into the following 5 groups according to the patient's age at retrieval: <35, 35–37, 38–40, 41–42, and >42 years.

Clinical protocols

Controlled ovarian hyperstimulation protocol, monitoring of response to stimulation, final oocyte maturation trigger, oocyte retrieval, fertilization, embryo culture, and embryo transfer were performed per our standard protocols.¹⁴ The protocol and gonadotropin dosage were determined based on patient age and weight, assessment of ovarian reserve (antral follicle count and anti-Müllerian hormone levels), and prior response to stimulation. Exogenous gonadotropins (Gonal-F, EMD-Serono Inc.; Follistim, Merck, Kenilworth, NJ; and Menopur, Ferring Pharmaceuticals Inc., Parsippany, NJ) were administered daily. Serum estradiol (E2) levels and transvaginal ultrasounds were performed frequently to monitor the response to stimulation. Endogenous gonadotropins were suppressed with either GnRH-antagonist (Ganirelix acetate, Merck, Kenilworth, NJ; or Cetrootide, EMD-Serono Inc., Rockland, MA) or GnRH-agonist (leuprolide acetate, Abbott Laboratories, Chicago, IL). The trigger with human chorionic gonadotropin (Novarel, Ferring Pharmaceuticals Inc., Parsippany, NJ) and/or GnRH-agonist (leuprolide acetate, Abbott Laboratories, Chicago, IL) was

administered when ≥ 2 follicles reached 17 mm. Oocyte retrieval was performed under ultrasound guidance 35–37 hours after trigger.

Euploid embryos were transferred in FET cycles. The uterine cavity was evaluated by saline infusion sonohysterography (SIS) within 1–3 months prior to embryo transfer, and a hysteroscopy was performed for patients who had abnormal SIS. Patients with regular menstrual cycles underwent natural FET cycles in which embryos were transferred 5 days after the LH surge. Based on physician preference, some women started vaginal progesterone supplementation (Endometrin, Ferring Pharmaceuticals Inc., Parsippany, NJ) 1 day after embryo transfer. Alternatively, women who were not ideal candidates for natural cycles underwent programmed cycles in which escalating doses of E2 were administered via transdermal patches for approximately 14 days. After an appropriate endometrial thickness (≥ 7 mm) was attained, the E2 was reduced, and intramuscular progesterone supplementation was initiated. Embryo transfers were performed after 5 days of progesterone.

Laboratory protocols

Embryos were cultured using the EmbryoScope (Vitrolife) time-lapse system and sequential culture media. Blastocysts were graded immediately before trophectoderm biopsies according to their degree of expansion, inner cell mass (ICM), and trophectoderm.^{15,16} The 6 grades of degree of expansion were as follows: 1, a blastocoel filling <50% of a nonexpanded embryo; 2, a blastocoel filling >50% of the blastocyst; 3, a blastocyst filling the whole embryo; 4, an expanded blastocyst surrounded by a thin zona pellucida; 5, a hatching blastocyst; and 6, a hatched blastocyst. The 3 ICM grades were as follows: A, firmly packed cells; B, loosely accumulated cells; and C, no detectable cells. The trophectoderm was graded as follows: A, several cells forming a cohesive epithelial layer; B, a few cells of different sizes creating a loose epithelium; and C, a few large cells pushed to the side.

TABLE 1

Demographic characteristics of women who underwent frozen – thawed euploid blastocyst transfer divided into 5 age groups

Parameters	Age (y)					Pvalue
	<35 (n = 227)	35–37 (n = 228)	38–40 (n = 185)	41–42 (n = 103)	>42 (n = 42)	
Age (y)	31.3 ± 2.0	35.9 ± 0.8	39.1 ± 0.8	41.4 ± 0.5	43.5 ± 1.0	<0.0001
BMI (kg/m ²)	23.4 ± 4.8	23.0 ± 3.7	23.9 ± 4.3	24.2 ± 4.4	23.8 ± 3.6	NS
Parity	0.5 ± 0.9	0.5 ± 0.7	0.5 ± 0.7	0.4 ± 0.6	0.5 ± 0.8	NS
Peak endometrial thickness (mm)	9.2 ± 1.8	9.2 ± 1.9	9.1 ± 1.9	9.2 ± 1.7	9.3 ± 1.8	NS
Number of transferred embryos	1.1 ± 0.3	1.1 ± 0.3	1.1 ± 0.3	1.1 ± 0.2	1.0 ± 0.0	NS
1	87.7%	86.4%	89.7%	93.2%	100%	0.03
2	12.3%	13.5%	10.3%	6.8%	0%	
Number of available euploid embryos	3.9 ± 2.4	3.6 ± 2.1	2.5 ± 1.5	1.8 ± 1.0	1.6 ± 1.1	<0.0001
% of Day 5 biopsies	51.1	53.9	57.3	45.6	42.9	NS
Morphological grading						
% of Excellent	8.8	6.1	5.4	3.9	9.5	NS
% of Good	15.9	11.8	11.3	14.6	14.3	NS
% of Average	64.3	68.4	69.7	62.1	64.3	NS
% of Poor	11.0	13.6	13.5	19.4	11.9	NS

Table also includes the differences in blastocyst morphology and day of trophectoderm biopsy among the 5 age groups.

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Blastocysts were grouped into the following 4 categories based on their morphology before biopsy: excellent (3AA, 4AA, 5AA, and 6AA), good (3AB, 4AB, 5AB, 6AB, 3BA, 4BA, 5BA, and 6BA), average (2BB, 3BB, 4BB, 5BB, and 6BB), and poor (2BC, 3BC, 4BC, 5BC, 6BC, 2CB, 3CB, 4CB, 5CB, 6CB, 2CC, 3CC, 4CC, 5CC, and 6CC). Embryos that reached the blastocyst stage on day 5 were biopsied that day, whereas delayed embryos were observed for 1 more day and biopsied on day 6 if they developed into blastocysts. After immobilizing the embryo with a holding pipette, laser pulses (ZI-LOS-tk Laser) were used to perforate the zona pellucida. Three to 7 cells were aspirated using a biopsy pipette 20 μ m in internal diameter. After exposing the cells to wash buffer, they were loaded into 0.2-mL polymerase chain reaction tubes with 2 μ L lysis buffer. The specimens were analyzed by the preimplantation genetic screening team at Cornell using the Illumina (BlueGnome, Cambridge, UK) 24SureV3 chip (array comparative genomic hybridization). Embryos were

vitrified after the biopsy using the Kitazato-based method.¹⁷

Study variables

The study's primary outcomes were LBR and implantation rate (IR). The secondary outcome was spontaneous abortion (SAB) rate. LBR was defined as the proportion of transfers resulting in a live birth beyond 24 weeks of gestation. IR was defined as the proportion of transferred embryos that resulted in intrauterine gestational sacs seen on transvaginal ultrasound. The SAB rate was defined as the proportion of clinical pregnancies that resulted in first-trimester pregnancy loss. Demographic parameters and outcomes were compared among the 5 age groups.

Statistical analysis

Categorical variables were compared with the χ^2 and Fisher exact tests. Continuous variables were tested for normality. They were expressed as mean \pm standard deviation, and parametric data were compared with the Student *t* test. We calculated the odds ratios (ORs)

with 95% confidence intervals (CIs) and controlled for body mass index (BMI), number of transferred embryos, peak endometrial thickness, type of FET (natural or programmed), blastocyst grading, and day of trophectoderm biopsy (day 5 or 6). The analysis was accounted for repeated measures using generalized estimating equations. *P* < .05 was considered statistically significant. Data analysis was performed with STATA statistical software version 14 (StataCorp LP, College Station, TX).

Results

A total of 785 FET cycles in which 870 blastocysts were transferred were included. Cycles were divided into the following 5 age groups: <35 (n = 227), 35–37 (n = 228), 38–40 (n = 185), 41–42 (n = 103), and >42 (n = 42) years old. The demographic parameters of the women in these groups are summarized in Table 1. There were no significant differences in BMI, parity, or peak endometrial thickness among the 5 groups. There were comparable proportions of cycles in which 1 or 2

TABLE 2

Implantation rates (IRs) and live birth rates (LBRs) of euploid blastocysts categorized by maternal age at retrieval, embryo morphology, and day of trophectoderm biopsy

Parameter	Category	Number of cycles	LBR			IR		
			LBR (%)	LBR aOR (95% CI)	Pvalue ^a	IR (%)	IR aOR (95% CI)	Pvalue ^a
Blastocyst	Excellent	52	78.8	vs Excellent	vs Excellent	78.6	vs Excellent	vs Excellent
Grade	Good	105	63.8	0.3 (0.1–0.9)	0.03	68.8	0.5 (0.2–1.3)	NS
	Average	522	54.2	0.1 (0.1–0.5)	0.001	55.7	0.3 (0.1–0.7)	0.01
	Poor	106	28.3	0.1 (0.0–0.2)	<0.001	35.4	0.1 (0.1–0.3)	<0.001
Day of TE	Day 5	410	60.0	vs Day 5	vs Day 5	63.9	vs Day 5	vs Day 5
Biopsy	Day 6	375	46.6	0.7 (0.5–0.9)	0.03	47.8	0.6 (0.4–0.8)	0.003
Maternal	<35	227	55.1	vs <35	vs <35	56.5	vs <35	vs <35
Age (y)	35–37	228	51.3	0.8 (0.5–1.2)	NS	52.9	0.8 (0.5–1.2)	NS
	38–40	185	53.5	0.9 (0.6–1.5)	NS	55.4	1.0 (0.6–1.6)	NS
	41–42	103	52.4	1.1 (0.6–1.9)	NS	59.1	1.3 (0.7–2.3)	NS
	>42	42	61.9	1.5 (0.6–3.5)	NS	71.4	2.2 (0.9–5.5)	NS

aOR, adjusted odds ratio; CI, confidence interval; NS, not significant; TE, trophectoderm.

^a Adjusted for age, body mass index, parity, peak endometrial thickness, number of transferred embryos, number of available euploid embryos, day of biopsy, type of frozen–thawed euploid embryo transfer (FET) cycle, and blastocyst morphology.

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embryos were transferred among the following 4 age groups (<35, 35–37, 38–40, and 41–42 years) (Table 1). Although there was no significant difference in the mean number of transferred embryos among the 5 groups, none of the women >42 years old had a transfer of 2 embryos, which was a significantly lower proportion than women in the 3 youngest groups (Table 1). Therefore, the ORs were adjusted for the number of transferred embryos.

Blastocyst grading and day of trophectoderm biopsy correlated significantly with LBRs and IRs (Table 2).

Excellent-quality blastocysts were associated with a significantly higher LBR compared with good-quality (78.8% vs 63.8%; $P = .03$), average-quality (78.8% vs 54.2%; $P = .001$), and poor-quality (78.8% vs 28.3%; $P < .001$) blastocysts. Poor-quality embryos also yielded a lower LBR compared with good-quality (28.3% vs 63.8%; $P < .001$) and average-quality (28.3% vs 54.2%; $P < .001$) embryos. These differences remained significant after controlling for maternal age, BMI, number of transferred embryos, type of frozen cycles, number of available euploid embryos, and day of trophectoderm biopsy

(Table 2). Excellent-quality blastocysts were also associated with a significantly higher IR compared with average- and poor-quality blastocysts (Table 2). However, the difference in IRs between excellent- and good-quality blastocysts did not reach statistical significance, possibly because of the relatively small number of cycles in which excellent-quality blastocysts were transferred. Furthermore, poor-quality embryos yielded a higher SAB rate compared to their average-quality (25.0% vs 9.0%; $P = .003$), good-quality (25.0% vs 6.9%; $P = .01$), and excellent-quality (25.0% vs 2.4%; $P = .01$) counterparts.

Embryos biopsied on day 5, or faster-developing embryos, were associated with a higher LBR compared to those biopsied on day 6 (60.0% vs 46.6%; $P < .001$). The odds remained significant after adjusting for all confounders, including blastocyst morphology (adjusted OR, 1.4; 95% CI, 1.1–1.9; $P = .02$) (Table 2). The same trend was observed in the IRs (63.9% vs 47.8%; $P < .001$). There were no significant differences in the SAB rates between the 2 groups (9.6% vs 9.3%; $P = .9$). Of note,

TABLE 3

Distribution of morphological grading of embryos biopsied on different days

	Day 5 biopsy	Day 6 biopsy	Pvalue
Excellent-quality blastocysts	9.5	3.1	<.001
Good-quality blastocysts	16.3	8.4	<.001
Average-quality blastocysts	69.0	67.1	NS
Poor-quality blastocysts	5.3	21.4	<.001

NS, not significant.

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the majority of euploid embryos biopsied on day 5 or 6 were of average quality (69% vs 67.1%; $P = .5$). Embryos biopsied on day 5 had significantly higher percentages of excellent- and good-quality blastocysts and lower percentages of poor-quality blastocysts compared to those biopsied on day 6 ($P < .001$) (Table 3).

The 5 age groups (<35, 35–37, 38–40, 41–42, and >42 years) had comparable IRs (56.5%, 52.9%, 55.4%, 59.1%, and 71.4%, respectively; $P = .2$) and LBRs (55.1%, 51.3%, 53.5%, 52.4%, and 61.9%, respectively; $P = .7$) (Table 2, Figure 1). The SAB rates were also comparable (8.8%, 7.9%, 8.3%, 14.3%, and 13.3%, respectively; $P = .5$) among the 5 groups (Figure 1). As we expected, older women had a significantly lower number of euploid embryos compared with younger women ($P < .001$) (Table 1). However, the euploid blastocysts from older women had similar morphology and embryo growth rates, as reflected by the day of biopsy, compared with those from younger women (Table 1).

Comment

This study investigated the effect of maternal age at the time of retrieval on euploid blastocysts. Although the number of euploid embryos diminished with advanced maternal age, they developed at a pace similar to those from younger women and yielded a comparable implantation potential. The SAB rates, which correlate with blastocyst grading, were not affected by maternal age or the day of trophoctoderm biopsy. Our data also confirm that the day of trophoctoderm biopsy, which reflects the speed of embryo development, and blastocyst morphology influenced the implantation potential of euploid embryos.

Preimplantation genetic testing for aneuploidy (PGT-A) has been suggested as a method to overcome the detrimental effects of advanced maternal age on SAB rates and failed IVF attempts.^{18–22} The original test was performed either as a polar body biopsy or on a blastomere of a cleavage-stage embryo using fluorescence in situ hybridization analysis. Several retrospective studies showed promising

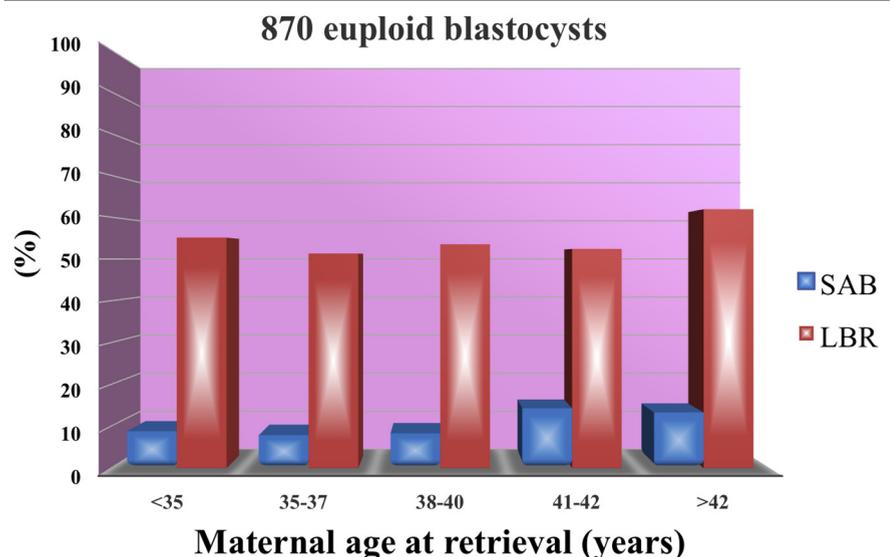
results stemming from this practice.^{23–25} However, subsequent randomized trials revealed that the technique may in fact be harmful.^{26–28} Thus, the procedure was enhanced by performing comprehensive chromosome screening on trophoctoderm cells, which has been shown to be beneficial for certain couples.^{18–20,22} The current generation of PGT-A allows for better selection of euploid embryos, thereby increasing the implantation rate per transfer and decreasing the SAB rate. It is worth noting that PGT-A is not perfect, as the mosaic nature of the embryos and the inherent technical errors associated with these platforms can lead to false results.^{29–31}

Despite the advances made with PGT-A, a considerable proportion of euploid embryos still fails to implant due to as yet unknown etiologies. Poor-quality euploid embryos have been correlated with lower IRs and higher SAB rates compared to better-quality blastocysts, which partially explains some of the failed euploid cycles.¹³ Consistent with the literature, our data confirm that the morphology of euploid blastocysts

matters and can aid in embryo selection. Ooplasmic dysmaturity or abnormal gene expression of some euploid embryos may explain their poor morphological grading and relatively low implantation potential. Furthermore, the present study shows that faster-developing euploid blastocysts biopsied on day 5 have a higher chance of implanting compared to their slower counterparts biopsied on day 6, even after controlling for morphology. The differences between day-5 and day-6 untested blastocysts have been explored over the last 2 decades.^{32–38} Initial studies showed that fresh transfer of day-5 blastocysts is associated with superior pregnancy outcomes compared with day-6 embryos.^{32–34} A subsequent retrospective study including 377 fresh and 106 FET cycles showed that although day-6 blastocysts were associated with lower IRs compared to their day-5 counterparts in fresh cycles, comparable outcomes were seen in FET cycles.³⁵ This suggests that the poor outcomes of day-6 blastocysts in fresh cycles were attributed mainly to the

FIGURE 1

Spontaneous abortion (SAB) rate and live birth rate (LBR) following the transfer of euploid blastocysts categorized by maternal age at retrieval. There was no significant difference in SAB rate or LBR among the 5 age groups



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suboptimal embryo–endometrial synchrony observed in fresh cycles.³⁵ However, a more recent study including 699 FET cycles reported higher IRs and LBRs for day-5 compared with day-6 blastocysts.³⁶ Other studies have also reported better FET outcomes of day-5 compared with day-6 blastocysts.^{37,38} This has been attributed partially to the higher aneuploidy rates of embryos that require a longer time to start blastulation.^{39,40} Although that suggestion might be correct, our data, which comprise only euploid embryos, affirm that day-5 blastocysts have a higher implantation potential than their day-6 counterparts not only because of better embryo–endometrial synchrony and higher euploidy rates, but maybe also because of another embryo-related factor. This factor might be the embryo's overall metabolic or epigenetic health.

The fact that older oocyte recipients have pregnancy rates similar to those of young women proves that uterine receptivity is not altered by maternal age.² Indeed, the detrimental influence of advanced maternal age on human fertility has been linked to the increased aneuploidy rates due to meiotic nondisjunction.⁵ However, a question still remains as to whether aneuploidy is the only age-related factor determining reproductive potential. A few studies with limited sample sizes, which did not take blastocyst morphology or the day of trophectoderm biopsy into consideration, suggested that advanced maternal age might be associated with lower implantation rates of euploid blastocysts.^{10,12} The authors posited that aging might compromise embryos beyond their ploidy status, which was a reasonable conclusion before it was understood that morphology indeed affects euploid embryo viability.^{10,13} In fact, patients in the older groups of these studies might have had transfers of poorer-quality euploid blastocysts that led to the inferior pregnancy outcomes. Our findings confirm that aneuploidy is the major factor linked to the poor outcomes seen in older women; indeed, when euploid embryos are selected and transferred in older patients, the success rates are equal to those seen in young women. Although

older women had smaller cohorts of euploid embryos, the quality of these embryos, assessed by blastocyst morphology and day of trophectoderm biopsy, was not affected by maternal age. In addition, the SAB rates, which correlate with ploidy and blastocyst morphology, were comparable between embryos of the different age groups. Therefore, the effect of maternal age on embryos seems to be almost exclusively through the impairment of oocyte meiosis.

This study's large sample size and the inclusion of all major identifiable confounding factors demonstrate the impact of maternal age, blastocyst morphology, and day of trophectoderm biopsy on the competence of euploid embryos. However, it is important to acknowledge the inherent subjectivity of blastocyst grading as well as the variations in embryo assessment among different laboratories. In addition, 95% of women in the oldest age group were 42 or 43 years old, which may limit our conclusions to women who undergo oocyte retrieval at age <45 years.

In conclusion, maternal age at retrieval influences the number of euploid blastocysts, but it does not impair the implantation potential of euploid embryos. Blastocyst morphology and the pace of embryo development appear to correlate with the metabolic health of the conceptus and are of paramount importance in the selection of all embryos. ■

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