

The impact of a thin endometrial lining on fresh and frozen–thaw IVF outcomes: an analysis of over 40 000 embryo transfers

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STUDY QUESTION: Does each millimeter decrease in endometrial thickness lead to lower pregnancy and live birth rates in fresh and frozen IVF cycles?

SUMMARY ANSWER: Clinical pregnancy and live birth rates decline as the endometrial thickness decreases below 8 mm in fresh IVF-ET and below 7 mm in frozen–thaw embryo transfer (ET) cycles.

WHAT IS KNOWN ALREADY: Previous studies have been heterogenous and have shown conflicting results on the impact of endometrial thickness on IVF outcomes. Most studies do not include many patients with an endometrial thickness below 6 mm, and there are few studies of frozen–thaw ET cycles.

STUDY DESIGN, SIZE, DURATION: This study is a retrospective cohort analysis of all Canadian IVF fresh and frozen–thaw ET cycles from the CARTR-BORN database for autologous and donor fresh and frozen–thaw IVF-ET cycles from 1 January 2013 to 31 December 2015. A total of 24 363 fresh and 20 114 frozen–thaw IVF-ET cycles were reported during this timeframe.

PARTICIPANTS/MATERIALS, SETTING, METHODS: 33 Canadian clinics participated in voluntary reporting of IVF and pregnancy outcomes to the CARTR-BORN database. The impact of endometrial thickness on pregnancy, live birth and pregnancy loss rates were analyzed for fresh IVF-ET and frozen–thaw cycles.

MAIN RESULTS AND THE ROLE OF CHANCE: In fresh IVF-ET cycles, clinical pregnancy and live birth rates decreased ($P < 0.0001$) and pregnancy loss rates increased ($P = 0.01$) with each millimeter decline in endometrial thickness below 8 mm. Live birth rates were 33.7, 25.5, 24.6 and 18.1% for endometrial thickness ≥ 8 , 7–7.9, 6–6.9 and 5–5.9 mm, respectively. In frozen–thaw ET cycles, clinical pregnancy ($P = 0.007$) and live birth rates decreased ($P = 0.002$) with each millimeter decline in endometrial thickness below 7 mm, with no significant difference in pregnancy loss rates. Live birth rates were 28.4, 27.4, 23.7, 15 and 21.2% for endometrial thickness ≥ 8 , 7–7.9, 6–6.9, 5–5.9 and 4–4.9 mm, respectively. The likelihood of achieving an endometrial thickness ≥ 8 mm decreased with age (89.7, 87.8 and 83.9% in women < 35 , 35–39 and ≥ 40 , respectively) ($P < 0.0001$).

LIMITATIONS, REASONS FOR CAUTION: This study only included cycles which proceeded to ET, which may overestimate pregnancy outcomes. Approximately 8% of cycles could not be included in the analysis due to data irregularity related to data entry. Demographic data aside from age were unavailable but may be important as lower endometrial thickness may be associated with poor ovarian response.

WIDER IMPLICATIONS OF THE FINDINGS: Although pregnancy and live birth rates decrease with endometrial thickness, reasonable outcomes were obtained even with lower endometrial thickness measurements. These data provide valuable guidance for both physicians and patients when confronted with decisions related to a persistently thin endometrium.

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Introduction

The definition of a thin endometrium varies between studies, but is generally defined as <7 or <8 mm on the day of ovulation trigger in fresh IVF cycles and prior to the start of progesterone in frozen–thaw embryo transfer (ET) cycles (Al-Ghamdi et al., 2008, El-Toukhy et al., 2008, Shufaro et al., 2008, Aydin et al., 2013, Wu et al., 2014, Bu and Sun, 2015). The incidence of thin endometrium in ART has been reported to be between 1.5 and 9.1% (Al-Ghamdi et al., 2008, Shufaro et al., 2008, Aydin et al., 2013, Wu et al., 2014, Bu and Sun, 2015). Differences in incidence may be related to variation in measurement techniques, ultrasound equipment, ART protocols and the clinical threshold to proceed with ET.

Studies that have evaluated the impact of a thin endometrial lining on IVF outcomes are quite heterogeneous. In fresh cycles, some studies (Kovacs et al., 2003, Kumbak et al., 2009, Zhao et al., 2014, Yuan et al., 2016, Vaegter et al., 2017) have found decreased pregnancy and live birth rates with a thin endometrium, but other studies (Noyes et al., 1995, Gingold et al., 2015) have not found the same association. A recent systemic review (Kasius et al., 2014) of fresh cycles showed lower clinical pregnancy rates (OR = 0.42, 95% CI: 0.27–0.67) but uncertainty in live birth rates (OR = 0.38, 95% CI: 0.09–1.54) with an endometrial thickness <7 mm.

There are only a few studies that have evaluated the impact of a thin endometrium on frozen–thaw ETs (El-Toukhy et al., 2008, Gingold et al., 2015). One study (El-Toukhy et al., 2008) of 743 frozen–thaw ETs using autologous embryos showed a lower pregnancy rate in women between 7 and 8 mm compared to an endometrial thickness above 8 mm.

The objective of our study was to evaluate the impact of each 1-mm decrease in endometrial thickness below 8 mm on fresh and frozen–thaw IVF-ET cycles using the Canadian ART Registry (CARTR-BORN) data. We also sought to determine whether pregnancy outcomes, in the presence of a thin endometrium, were influenced by the day of ET (cleavage stage vs blastocyst) or the age of the patient.

Materials and Methods

The Canadian ART Registry (CARTR) has been collecting outcomes for all IVF cycles in Canada since 2001. Although participation is voluntary, virtually all Canadian clinics comply, which results in near complete data for all Canadian ART treatment cycles. In 2013, CARTR partnered with Better Outcomes Registry & Network (BORN) to allow for complete tracking of pregnancy and live birth data. In the province of Ontario, birth outcomes are automatically linked. Throughout the rest of Canada, birth outcomes are entered by individual clinics once available from the patients or

provincial birth records. Clinics submit anonymized data for all patients undergoing ART treatment including cycle information and outcomes.

This study is a retrospective cohort analysis of all Canadian IVF fresh and frozen–thaw ET (ET) cycles from the CARTR-BORN database for autologous and donor fresh and frozen–thaw IVF-ET cycles from 1 January 2013 to 31 December 2015 (the last year for which live birth data are currently available). During this period, data were available from 33 Canadian ART clinics. For fresh IVF-ET cycles, the endometrial thickness on the day of trigger was recorded. For frozen–thaw ET cycles, the last endometrial thickness prior to the initiation of progesterone or documentation of LH surge was used. Blastocysts transfers on Days 5 and 6 were grouped and analyzed together. Data in which total counts were <6 were not available for analysis in accordance with CARTR-BORN policies. Only cycles which proceeded to ET were included. Clinical pregnancy was defined as the presence of a gestational sac on first trimester ultrasound. Clinical pregnancy, pregnancy loss after documentation of a clinical pregnancy and live birth rates were calculated per ET.

When the initial dataset was reviewed, an anomaly in the data was noted. The incidence of ET occurring in each endometrial measurement decreased with each declining millimeter for all groups except the 1.0–1.9 and <1 mm groups. A significant number (2%) of ETs were found in these groups with most of these transfers coming from the same clinics. This seemed incongruent with clinical practice and clarification with the specific clinics confirmed that a data entry error had occurred in which these clinics reported their data using incorrect units of measurement (cm instead of mm). Because it was not clear if this error had impacted all or only some of the cycles in which data from these clinics was entered, a new dataset from CARTR-BORN was provided in which all of the data from the affected clinics was removed.

Statistical analysis was performed using SAS 9.4 (SAS Institute, North Carolina, USA). Chi-square tests were performed to assess the differences in rates, and Cochran–Armitage tests were applied to examine the trends in outcome rates over endometrial thickness strata. Ethics approval was obtained from the Mount Sinai Hospital Research Ethics Board (MSH REB 18-0095-C).

Results

Between 2013 and 2015, a total of 24 363 fresh and 20 114 frozen–thaw IVF-ET cycles were reported. In the fresh IVF-ET group, 2449 (10.1%) of the cycles were excluded due to incorrect data entry of the units of measurement for endometrial thickness as described above; therefore, 21 914 cycles were analyzed. For the same reason, 1172 (5.8%) of the frozen–thaw ETs were excluded and 18 942 cycles could be analyzed.

In the fresh IVF-ET cycles, 87.7% of patients had an endometrial thickness \geq 8 mm. For these patients, the clinical pregnancy rate was 43.2%, the live birth rate was 33.7% and the pregnancy loss rate after

clinical pregnancy was 22%. Both clinical pregnancy and live birth rates declined with each millimeter decrease in endometrial thickness under 8 mm (tests for trends, $P < 0.0001$ for CPR and LBR), while pregnancy loss rates after achieving a clinical pregnancy increased ($P = 0.01$; Table I). Less than 1% of patients who underwent an ET had an endometrial thickness between 4 and 5.9 mm; the clinical pregnancy rate was 25% in this group. Subgroup analyses of Days 5/6 and 3 transfers showed similar associations of endometrial thickness on clinical pregnancy and live birth rates (Tables II and III), although overall clinical pregnancy and live birth rates were lower in the Day 3 ET group.

In the frozen–thaw ET cycles, 85.9% of patients had an endometrial thickness greater than or equal to 8 mm. Both clinical pregnancy and live birth rates declined with each millimeter decrease in endometrial thickness (Table IV; $P = 0.02$ for CPR and $P = 0.01$ for LB). However, there was no difference between patients with an endometrial thickness >8 mm and those with an endometrial thickness between 7.0 and 7.9 mm ($P = 0.83$ for CPR and $P = .27$ for LBR). A small number of patients (0.6%) who underwent an ET had an endometrial thickness between 4 and 5.9 mm, their clinical pregnancy and live birth rates were 28.3 and 16.8%, respectively. For frozen–thaw cycles with Day

Table I Clinical and live birth rates in autologous and donor fresh IVF-ET.

Endometrial thickness at trigger	# Embryo transfers	Incidence %	Clinical pregnancy rate (n)	Pregnancy loss rate (n)	Live birth rate (n)
≥ 8 mm	19 220	87.7	43.2 (8309)	22.0 (1831)	33.7 (6478)
7.0–7.9 mm	1837	8.4	34.6 (636)	26.4 (168)	25.5 (468)
6.0–6.9 mm	647	3.0	33.7 (218)	27.1 (59)	24.6 (159)
5.0–5.9 mm	155	0.7	25.8 (40)	30.0 (12)	18.1 (28)
4.0–4.9 mm	29	0.1	20.7 (6)	N/A ^a	N/A ^a
<4 mm	26	0.1	N/A ^a	N/A ^a	N/A ^a
Total	21 914	100%			
<i>P</i> for difference ^b			$P < 0.0001$	$P = 0.01$	$P < 0.0001$
<i>P</i> for trend ^c			<0.0001	0.002	<0.0001

^aData for cell counts <6 was suppressed and not available (N/A).

^b*P* values in Chi-square tests for differences in all groups with available data.

^c*P* values in Cochran–Armitage tests for trends in rates across endometrial thickness strata in all groups with available data.

Table II Clinical and live birth rates in autologous and donor fresh Day 5/6 IVF-ET.

Endometrial thickness at trigger	# Embryo transfers	Proportion %	Clinical pregnancy rate (n)	Pregnancy loss rate (n)	Live birth rate (n)
≥ 8 mm	9860	89.0	50.8 (5006)	20.0 (1002)	40.6 (4004)
7.0–7.9 mm	848	7.7	44.7 (379)	25.6 (97)	33.3 (282)
6.0–6.9 mm	290	2.6	41.7 (121)	24.0 (29)	31.7 (92)
5.0–5.9 mm	71	0.6	25.4 (18)	33.3 (6)	16.9 (12)
4.0–4.9 mm	9	0.1	N/A ^a	N/A ^a	0% (0)
Total	12 433	100			
<i>P</i> for difference ^b			<0.0001	0.02	0.0001
<i>P</i> for trend ^c			<0.0001	0.006	<0.0001

^aData for cell counts <6 was suppressed and not available (N/A).

^b*P* values in Chi-square tests for differences in all groups with available data.

^c*P* values in Cochran–Armitage tests for trends in rates across endometrial thickness strata in all groups with available data.

Table III Clinical and live birth rates in autologous and donor fresh Day 3 IVF-ET.

Endometrial thickness at trigger	# Embryo transfers	Proportion %	Clinical pregnancy rate (n)	Pregnancy loss rate (n)	Live birth rate (n)
≥8 mm	8001	87.6	37.0 (2960)	24.8 (734)	27.8 (2226)
7.0–7.9 mm	777	8.5	28.6 (222)	24.8 (55)	21.5 (167)
6.0–6.9 mm	272	3.0	29.8 (81)	32.1 (26)	20.2 (55)
5.0–5.9 mm	68	0.7	29.4 (20)	30.0 (6)	20.6 (14)
4.0–4.9 mm	11	0.1	N/A ^a	N/A ^a	N/A ^a
Total	9129	100%			
<i>P</i> for difference ^b			<0.0001	0.47	0.0001
<i>P</i> for trend ^c			<0.0001	0.21	<0.0001

^aData for cell counts <6 was suppressed and not available (N/A).

^b*P* values in Chi-square tests for differences in all groups with available data.

^c*P* values in Cochran–Armitage tests for trends in rates across endometrial thickness strata in all groups with available data.

Table IV Clinical and live birth rates in autologous and donor frozen–thaw ET.

Peak endometrial thickness	# Embryo transfers	Proportion %	Clinical pregnancy rate (n)	Pregnancy loss rate (n)	Live birth rate (n)
≥8 mm	16263	85.9	38.4 (6245)	26.0 (1621)	28.4 (4624)
7.0–7.9 mm	2130	11.2	38.3 (816)	28.4 (232)	27.4 (584)
6.0–6.9 mm	413	2.2	31.7 (131)	25.2 (33)	23.7 (98)
5.0–5.9 mm	80	0.4	28.8 (23)	47.8 (11)	15.0 (12)
4.0–4.9 mm	33	0.2	27.3 (9)	22.2 (2)	21.2 (7)
<4 mm	23	0.3	N/A ^a	N/A ^a	N/A ^a
Total	18942	100			
<i>P</i> for difference ^b			0.02	0.09	0.01
<i>P</i> for trend ^c			0.007	0.1	0.002

^aData for cell counts <6 was suppressed and not available (N/A).

^b*P* values in Chi-square tests for differences in all groups with available data.

^c*P* values in Cochran–Armitage tests for trends in rates across endometrial thickness strata in all groups with available data.

5/6 ET, clinical pregnancy and live birth rates were not significantly different when all groups of endometrial thickness were compared but were significantly lower when endometrial thickness ≥7 mm was compared with the remaining groups (Table V). The lack of significance when all groups were analyzed is likely because there was no difference in clinical pregnancy and live birth rates between the ≥8 and 7–7.9 mm groups comprising 97% of all the cycles.

When patients were analyzed by age group in fresh IVF-ET, patients were more likely to have an endometrial thickness ≥8 mm at age <35 (89.7%) and 35–39 years (87.8%) than patients at age >40 years

(83.9%; *P* < 0.0001). Lower clinical pregnancy and live birth rates were noted in all age groups as the endometrium became increasingly thinner (Table SI). The same trend was noted in frozen–thaw ET cycles, but this failed to reach statistical significance, likely due to the small numbers of transfers with a lining <7 mm in each age group (Table SII).

Discussion

To the best of our knowledge, this is the largest study to date evaluating the effects of decreased endometrial thickness on IVF outcomes in

Table V Clinical and live birth rates in autologous and donor frozen–thaw Day 5/6 ET.

Peak endometrial thickness	# Embryo transfers	Proportion %	Clinical pregnancy rate (n)	Pregnancy loss rate (n)	Live birth rate (n)
≥8 mm	11 951	85.3	40.6 (4858)	25.7 (1249)	30.2% (3609)
7.0–7.9 mm	1644	11.7	40.4 (664)	28.5 (189)	28.9% (475)
6.0–6.9 mm	325	2.3	35.4 (115)	26.1 (30)	26.2% (85)
5.0–5.9 mm	58	0.4	29.3 (17)	41.2 (7)	17.2% (10)
4.0–4.9 mm	27	0.2	29.6 (8)	12.5 (1)	25.9% (7)
Total	14 005				
<i>P</i> for difference ^a			<i>P</i> = 0.009	<i>P</i> = 0.43	<i>P</i> = 0.07
<i>P</i> for trend ^b			0.02	0.78	0.009

^a*P* values in Chi-square tests for differences comparing groups >7 mm with remaining groups.

^b*P* values in Cochran–Armitage tests for trends in rates across endometrial thickness strata comparing groups >7 mm with remaining groups.

both fresh and frozen–thaw ET cycles. In our study, both clinical pregnancy and live birth rates decreased significantly for each millimeter increment below 8 mm in the fresh IVF-ET group and below 7 mm in the frozen-ET group. It is interesting to note that the threshold in which a decrease in pregnancy was noted was different between the fresh IVF-ET and frozen–thaw ET groups. This may be due to differences in receptivity or endometrial advancement between these protocols. We also found that women ≥ age 40 are less likely to achieve an endometrial thickness ≥8 mm in fresh IVF-ET cycles.

We noted that although clinical pregnancy and live birth rates were lower when the endometrial thickness was below 8 mm, the outcomes in these groups were still reasonably acceptable. For fresh IVF-ET, clinical pregnancy and live birth rates were 26 and 18% with an endometrial thickness of 5–5.9 mm. Clinical pregnancy rates were 21% at 4–4.9 mm (live birth data not available due to low numbers). Similarly, for frozen–thaw ET, clinical pregnancy and live birth rates were 29 and 15% for endometrial thickness of 5–5.9 mm, and 27 and 21% at 4–4.9 mm, respectively. Currently these represent only a small proportion of ETs in our study. These outcomes may provide reassurance for physicians and patients when faced with a persistently thin endometrium.

Previous studies include a heterogeneous mix of prospective and retrospective studies, which have used different cut-offs to define thin endometrium (Noyes *et al.*, 1995, Kumbak *et al.*, 2009, Dain *et al.*, 2013, Kasius *et al.*, 2014, Gingold *et al.*, 2015, Vaegter *et al.*, 2017). These studies have yielded inconsistent results regarding the impact of endometrial thickness below 8 mm for ART outcomes. Many studies used different thresholds of 8 or 7 mm to define thin endometrium (Zhao *et al.*, 2014, Yuan *et al.*, 2016, Vaegter *et al.*, 2017). A recent systematic review found an impact of thin endometrium on clinical pregnancy and live birth rates; however, the studies had different definitions for thin endometrium (Kasius *et al.*, 2014). In addition, most studies included very few patients with an endometrial thickness <7 mm (Noyes *et al.*, 1995, Kovacs *et al.*, 2003).

The strengths of our study include the large cohort size and completeness of the data to reflect all Canadian ART cycles from all clinics over a 3-year period, allowing robust analysis of outcomes grouped by endometrial thickness per millimeter. Many previous studies (Noyes *et al.*, 1995, Kumbak *et al.*, 2009, Dain *et al.*, 2013, Gingold *et al.*, 2015, Yuan *et al.*, 2016, Vaegter *et al.*, 2017) only analyzed outcomes above or below a specific threshold; however, a gradient response with decreasing endometrial thickness would be expected and is clearly demonstrated in our study. Previous studies also had very few ETs with endometrial thickness <6 mm (Kovacs *et al.*, 2003, Gingold *et al.*, 2015) or have canceled patients with an endometrial thickness considered thin (El-Toukhy *et al.*, 2008). Although these accounted for <1% of cycles, there were 253 patients who underwent ETs with endometrial thickness below 6 mm in our cohort.

There are several limitations in our study. The vast majority of ETs (96%) did occur with an endometrial lining ≥7 mm. Although the study did include >1200 ETs with an endometrial thickness <7 mm, the findings for outcomes in <7 mm sub-strata could benefit from confirmation in even larger prospective cohort studies. Further studies using other datasets such as the SART with, potentially, a greater number of ETs at endometrial thickness <7 mm may provide further insight on the outcomes for cycles with linings <7 mm.

We did not have detailed information on demographic and cycle characteristics. It is possible that a lower endometrial thickness may reflect a higher proportion of poor responders or patients with a poorer prognosis for pregnancy due to other factors. Our dataset also did not include the number of embryos to transfer. Physicians may adjust the number of embryos to transfer if a patient has a thin endometrium.

It is also important to note that our study only included cycles which proceeded to ET. This may reflect some bias favouring patients with certain prognostic factors to proceed with ET compared to other patients with a thin endometrium who were canceled. If so, the true adverse impact of thin endometrium in IVF may be underestimated. A

prospective study with ETs proceeding regardless of endometrial thickness would provide a more complete picture. However, such a study would most likely be difficult to implement since both patients and clinicians would have to consider the risks of, and alternatives to, proceeding forward with an ET. In addition, there was no standardization of ultrasound technology, equipment or measurement techniques across clinics. This, however, accurately reflects what is encountered in clinical practice. An additional limitation is that 8% of cycles were excluded due to incorrect data entry. However, as this was a systematic error unrelated to the outcomes, we suspect that these cycles would have had a similar distribution across groups and outcomes and should not have biased the results.

Conclusion

Clinical pregnancy and live birth rates decrease for each millimeter of endometrial thickness below 8 mm in fresh IVF cycles and below 7 mm for frozen–thaw IVF cycles. Nevertheless, viable pregnancy rates remain reasonably acceptable in patients with an endometrial thickness between 4 and 6 mm.

Supplementary data

Supplementary data are available at *Human Reproduction* online.

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Authors' roles

K.L. was responsible for study design and conception, data analysis, first draft of article, review and approval of revisions and the final article. M. H. was responsible for study design and conception, data analysis, review and approval of first draft, revisions and the final draft. A.H. was responsible for study design, review and approval of first draft, revisions and the final article. Z.-C.L. was responsible for study design, data and statistical analysis, review and approval of first draft, revisions and the final article. N.M. was responsible for study design and conception, data analysis, review and approval of first draft, revisions and the final article.

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Conflict of interest

The authors had no conflicts of interest to declare.

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