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Early spontaneous multiple fetal pregnancy reduction is associated with adverse perinatal outcomes in in vitro fertilization cycles

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Abstract

The primary objective of this study is to investigate whether early spontaneous multiple fetal pregnancy reduction, also known as vanishing twin syndrome, is associated with adverse perinatal outcomes in fresh in vitro fertilization cycles. This is a retrospective cohort study of women with live singleton births with and without an early vanishing twin after fresh in vitro fertilization. Characteristics compared included incidence of preterm birth, overall birth weight, overall low birth weight, overall very low birth weight, and term low birth weight. In all, 4049 patients with live singleton births were included—853 and 3196 with and without a vanishing twin, respectively. The vanishing twin group had a lower overall birth weight compared to those without (3279.5 ± 369.9 vs 3368.6 ± 567.5 g; $p < 0.01$). Early vanishing twin was also associated with an increased odds of overall low birth weight (odds ratio: 1.75; 95% confidence interval: 1.36–2.25; $p < 0.01$) and increased odds of term low birth weight (odds ratio: 3.44; 95% confidence interval: 2.14–5.53; $p < 0.01$). Our study suggests that early vanishing twin is associated with lower overall birth weight and higher odds of overall low birth weight and term low birth weight in live singleton births after fresh in vitro fertilization.

Keywords

in vitro fertilization, obstetric outcomes, perinatal outcomes, vanishing twin

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Introduction

Spontaneous multiple fetal pregnancy reduction (SMFPR), also known as vanishing twin (VT) syndrome, refers to the spontaneous in utero reduction in one or more twins.[1,2] Ever since its recognition in the 1970s, VT syndrome and its impact on the surviving twin has sparked the interest of investigators.[3] Previous studies have indicated that approximately 10%–12% of singleton pregnancies originate from a twin gestation.[4] Ultrasonographic studies have also shown that spontaneous reduction in one or more gestational sacs may occur in up to 19% of twin pregnancies before the 7th week of gestation.[5] The pathologic implications of the VT on the surviving twin have been debated, with some studies indicating a higher incidence of adverse obstetric and perinatal outcomes,[6–9] while others demonstrating similar outcomes to singleton pregnancies,[2,10,11] or even a protective effect.[12] Specifically, multiple studies have indicated an increased risk of low birth weight (LBW) in pregnancies with VT.[4,6–9] Furthermore, LBW has been associated with adverse long term and adult health outcomes.[13] These findings are particularly important in

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pregnancies arising from in vitro fertilization (IVF) where >1 embryo may be transferred to achieve a pregnancy.[4] Given that one out of every 10 singleton IVF pregnancies arises from a twin gestation,[4] we sought to investigate whether the presence of an early VT is associated with adverse perinatal outcomes in women undergoing IVF-embryo transfer (ET) cycles.

Materials and methods

Cycle inclusion criteria

This retrospective cohort study was approved by the institutional review board at Weill Cornell Medical College, New York (IRB#1503016064). Given that all data were obtained from a clinical database retrospectively, individual patient consent was not required. All fresh IVF-ET cycles occurring between January 2004 and June 2013 at our center were analyzed for potential inclusion. For the purpose of this study, only patients undergoing day-3 ET who subsequently had a live singleton birth were included. Patients who underwent ET with >1 embryo (grades 1, 1.5, or 2) were included in the analysis. All patients undergoing day-5 ET, frozen-thawed ET, or those utilizing donor oocytes were excluded.

Patients with a positive pregnancy test on cycle day (CD) 28, that is, 14 days after oocyte retrieval underwent transvaginal ultrasonography (TVUS) on CD 35 to record the number of gestational sacs, and on CD 49 to record the number of fetuses with cardiac activity within the respective gestational sacs. Patients with a singleton gestation with cardiac activity on CD 49 were then assigned to two groups: group 1 consisted of patients with spontaneous in utero reduction in one or more gestational sacs between CD 35 and CD 49, while group 2 consisted of patients without reduction in gestational sacs. The former group, that is, the early VT group was considered the study group, while the latter was considered the control group.

Clinical and laboratory protocols

Controlled ovarian stimulation (COS), human chorionic gonadotropin (hCG) trigger, and oocyte retrieval were performed per standard protocols.[14] Gonadotropin doses were based on patient age, weight, antral follicle count, serum anti-müllerian hormone (AMH) levels, and previous response to stimulation, if any. Patients underwent COS with gonadotropins (Follistim; Merck, Kenilworth, NJ, USA; Gonal-F, EMD-Serono, Rockland, MA, USA; and/or Menopur, Ferring Pharmaceuticals Inc, Parsippany, NJ, USA), with ovulation being suppressed with once daily 0.25 mg gonadotropin releasing hormone (GnRH) antagonist, that is, Ganirelix Acetate (Merck) injections.[14] hCG (Novarel; Ferring Pharmaceuticals Inc) or Pregnyl (Merck) was administered according to a sliding scale[14] and was given when the two lead follicles attained a mean diameter >17 mm. Oocyte retrieval was performed under conscious sedation using TVUS guidance approximately 34–35 h after hCG administration. Luteal support with 50 mg of intramuscular progesterone daily was begun the day after oocyte retrieval. Oocytes were fertilized with conventional insemination or intracytoplasmic sperm injection (ICSI) based on the couple's history and the male partner's semen analysis. Embryos were cultured using in-house culture media and assessed on day 2 (44–46 h after insemination or sperm injection) and day 3 (66–72 h insemination or sperm injection).[15,16] Day-3 embryos with grades 1, 1.5, or 2[17] were transferred with Wallace catheters (Smiths Medical, Dublin, OH, USA) at approximately 1 cm less than the uterine depth identified at prior trial transfer.

Outcome variables

Demographic characteristics recorded were age, parity, body mass index (BMI; kg/m^2), infertility diagnosis, and number of previous IVF attempts. COS parameters recorded were type of COS protocol (GnRH-agonist-based vs GnRH-antagonist-based), total COS days, total gonadotropins administered (intrauterine (IU)), peak endometrial stripe (mm), peak estradiol (E_2) level (pg/mL), number of oocytes retrieved, and number of mature oocytes. The number of day-3 embryos transferred was also recorded. Perinatal outcomes analyzed included mode of delivery, incidence of term birth, preterm birth (PTB), overall birth weight, LBW, very low birth weight (VLBW), and term LBW. Any live birth >37 weeks of gestational age was considered term birth, while live birth ⩽37 weeks of gestational age was defined as PTB. PTB <34 weeks of gestation was classified as early PTB, while PTB between >34 and ⩽37 weeks of gestation was defined as late PTB.[18] Birth weight <2500 g irrespective of gestational age was considered LBW.[19] VLBW was defined as birth weight <1500 g irrespective of gestational age.[19]

Statistical analyses

Categorical variables were expressed as number of cases (n) and percentage of occurrence (%). Non-parametric variables were expressed as median (interquartile range (IQR)). Continuous variables were checked for normality and expressed as mean ± standard deviation (SD). McNemar's chi-square (χ^2) test was used for categorical variables. Independent t-tests and Wilcoxon rank-sum tests were utilized for continuous variables and non-parametric variables, respectively. Odds ratios (OR) with 95% confidence intervals (CI) for the incidence of term birth, PTB, overall birth weight, LBW, VLBW, and term LBW were calculated, adjusted with multivariate logistic regression analysis when indicated (adjusted odds ratio (aOR)). Statistical analyses were performed using STATA version

Table 1. IVF cycle characteristics in patients with live singleton births with and without a vanishing twin ($n = 4049$).

Parameter	Group 1 ($n = 853$)	Group 2 ($n = 3196$)	p
Age (years)	36.1 (±4.08)	35.8 (±4.51)	0.63
Parity	0.61 (±0.26)	0.60 (±0.27)	0.33
BMI (kg/m^2)	22.7 (±5.70)	22.8 (±6.28)	0.67
Infertility diagnoses			0.60
Ovulatory	237 (27.8%)	727 (22.7%)	
Tubal	42 (4.92%)	196 (6.13%)	
Endometriosis	40 (4.69%)	210 (6.57%)	
Male factor	313 (36.7%)	1038 (32.5%)	
Idiopathic	52 (6.10%)	229 (7.17%)	
Other	169 (19.8%)	796 (24.9%)	
Previous IVF attempts	1.85 (±1.04)	1.92 (±1.08)	0.09
Protocol			0.61
GnRH agonist based	321 (37.6%)	1091 (34.1%)	
GnRH antagonist based	532 (62.4%)	2105 (65.9%)	
Total stimulation days	9.52 (±1.79)	9.62 (±1.90)	0.17
Total gonadotropins administered (IU)	3014.3 (±689.4)	2997.1 (±757.5)	0.55
Peak endometrial stripe (mm)	11.4 (±2.61)	11.3 (±2.38)	0.29
E_2 level on day of trigger (pg/mL)	1718.2 (±712.1)	1696.5 (±750.6)	0.45
Number of oocytes retrieved	11 (8–15)	11 (7–14)	0.99
Number of mature oocytes	9 (7–13)	9 (6–12)	0.99
ICSI rate (%)	68%	70.4%	0.71
Number of embryos transferred	3 (2–4)	3 (2–4)	0.99

BMI: body mass index; IVF: in vitro fertilization; GnRH-ant: gonadotropin releasing hormone-antagonist; GnRH-a: gonadotropin releasing hormone-agonist; E_2: estradiol; ICSI: intracytoplasmic sperm injection.
Group 1: with vanishing twin; group 2: without vanishing twin.
Data are presented as mean ± standard deviation, median (interquartile range), and *n* (%).

13 (StataCorp LP, College Station, TX, USA). Statistical significance was set at $p < 0.05$.

Results

In all, 4049 patients met inclusion criteria during the study period: 853 in group 1 and 3196 in group 2. Table 1 compares the demographics, baseline IVF characteristics, and COS parameters. Overall, there were no differences in the mean age, parity, BMI, number of previous IVF attempts, total COS days, total gonadotropins administered, peak endometrial stripe, or peak E_2 level. There was also no difference in distribution of infertility diagnoses, COS protocols within each group, total stimulation days, or total gonadotropins administered. E_2 on day of trigger as well as peak endometrial stripe was also comparable. The number of oocytes retrieved was comparable as was the median number of mature oocytes in group 1 (9 (7–13)) and group 2 (9 (6–12)). The number of embryos transferred in each group was also the same (3 (2–4)).

Table 2 summarizes the perinatal outcomes of the study cohort. The rates of vaginal and cesarean deliveries were comparable between the groups. There was no difference in the rate of term birth. In addition, no differences in the rates of late or early PTB were found between the two groups. The VT group had a lower overall birth weight compared to those without a VT (3279.5 ± 369.9 vs 3368.6 ± 567.5 g; $p < 0.01$). Furthermore, the findings of a VT was also associated with an increased odds of overall LBW (OR: 1.75; 95% CI: 1.36–2.25; $p < 0.01$) and increased odds of term LBW (OR: 3.44; 95% CI: 2.14–5.53; $p < 0.01$). There was no difference in rate of overall VLBW. The increased odds for LBW (aOR: 1.62; 95% CI: 1.19–2.08) and term LBW (aOR: 3.12; 95% CI: 2.01–5.13) remained unchanged even after controlling for age, BMI, total gonadotropins administered, and peak E_2 level.

Discussion

This retrospective cohort study aimed to investigate the perinatal outcomes of pregnancies associated with VT syndrome in fresh IVF-ET cycles. Previous studies have reported various associations of VT syndrome with adverse perinatal outcomes such as LBW, chronic hypertension, gestational diabetes, PTB, preterm labor, low APGAR scores, as well as fetal malformations.[20] However, many of the aforementioned studies have varied in their methodology (case–control vs retrospective cohort vs national registry), study population (natural conceptions vs IVF conceptions), and sample sizes. Table 3 summarizes some of the representative published studies regarding VT syndrome and associated perinatal outcomes. As evident from

Table 2. Comparison of perinatal outcomes in patients with live singleton births with and without a vanishing twin ($n = 4049$).

Parameter	Group 1 ($n = 853$)	Group 2 ($n = 3196$)	OR (95% CI)	p
Mode of delivery				0.81
Vaginal	444 (52.1%)	1717 (53.7%)	0.94 (0.54–1.63)	
Cesarean	409 (47.9%)	1479 (46.2%)		
Term birth	771 (90.4%)	2897 (90.6%)	0.98 (0.38–2.51)	0.82
Preterm birth	50 (5.86%)			0.95
Late preterm	32 (3.75%)	225 (7.04%)	0.52 (0.07–3.69)	
Early preterm		74 (2.32%)		
Overall birth weight (g)	3279.5 (±369.9)	3368.6 (±567.5)	–	<0.01
Overall LBW	99 (11.6%)	223 (6.98%)	1.75 (1.36–2.25)	<0.01
Overall VLBW	11 (1.29%)	28 (0.88%)	1.47 (0.10–22.4)	0.27
Term LBW	33 (4.28%)	37 (1.28%)	3.44 (2.14–5.53)	<0.01

LBW: low birth weight; VLBW: very low birth weight; OR: odds ratio; CI: confidence interval.
Group 1: with vanishing twin; Group 2: without vanishing twin.
Data are presented as mean ± standard deviation, median (interquartile range), and *n* (%).

Table 3, conflicting results regarding the pathologic implications of the VT on the surviving twin exist. For example, a case–control study including 46 pregnancies with VT syndrome showed a lower overall birth weight and a greater frequency of small-for-gestational age (SGA) and LBW in singleton survivors of VT syndrome[8] In contrast, another case-control study including 84 singleton births arising from VT syndrome showed no difference in gestational age at delivery, birth weight, PTB, very PTB, LBW, and VLBW as compared to 602 singleton controls.[2]

In our study, we were able to include 853 cases of live singleton births affected by early VT syndrome fresh IVF-ET cycles with a control group of 3196 live singleton births not affected by VT syndrome. Our findings suggest that singleton pregnancies affected by early VT syndrome were associated with lower overall birth weight. We also found higher odds of overall LBW and term LBW in the VT group. While an association to lower birth weight as an adverse outcome in pregnancies affected by VT has been shown in the past, our study was able to achieve larger numbers for further validation of this finding, in a homogeneous IVF population.

Several theories exist regarding the etiology of the association of VT syndrome with LBW and other adverse perinatal outcomes. La Sala et al.[2] designate VT syndrome as a first-trimester missed abortion of one twin, and therefore, propose that this phenomenon should be considered as a subtype of single fetal demise in twins. This leads to blood shunting from vascular anastomoses in the placenta of the surviving twin, thereby leading to a deleterious effect of a VT on the ongoing pregnancy.[2] Furthermore, data have shown increasing adverse outcomes with increasing gestational age at vanishing. Specifically, VT syndrome occurring at greater than 8 weeks of gestation was associated with increasing odds of LBW, VLBW, and higher incidence of neurologic sequelae.[4] Mansour et al.[12] describe a significantly lower miscarriage rate in pregnancies with VT syndrome. For singleton pregnancies complicated by VT syndrome compared to control singleton pregnancies, miscarriage rate was found to be 5% and 20%, respectively. For twin pregnancies which started as triplets compared to control twin pregnancies, this trend was also found to be significant at 2% and 11%, respectively. This trend indicates increased capacity of the uterus for implantation and early embryonic development.[12] While seemingly in contrast to the idea of a VT asserting a negative effect on the survivor, this finding actually points to a potential stronger link between the VT itself and adverse outcomes rather than a uterine or placentation etiology. Another proposition regarding the impact of the VT on the surviving twin is due to chronic inflammation resulting from the reduction in the VT.[21] While adverse outcomes have now been substantiated by multiple studies, the etiology behind them remains to be elucidated.

Limitations of this study include those inherent due to its retrospective nature. In addition, we did not analyze the pregnancies arising from blastocyst transfers. Finally, the pregnancies included in our study were also not evaluated for chorionicity. Thus, it is important to note that adverse outcomes associated with VT syndrome could possibly be related to monochorionic gestations, that is, fetal rather than embryonic demise are responsible for adverse outcomes.[2]

In conclusion, our study provides confirmation of the increased odds of overall LBW, LBW, and term LBW in live births after fresh IVF-ET cycles affected by VT syndrome. It is important to understand the implications of adverse outcomes in regard to prenatal care and counseling of patients. Given these results, it is reasonable to consider closer ultrasonographic surveillance of twin pregnancies both in the first trimester to potentially diagnose a VT and once diagnosed, in the late trimester to confirm adequate growth. In addition, single ETs should be performed (when possible) to mitigate some of the aforementioned adverse outcomes arising from >1 ET.

Table 3. Summary of published literature regarding vanishing twin syndrome and associated perinatal outcomes.

Reference	Study type/sample size	Diagnosis of vanishing twin	Gestational age	Birth weight	Other
Dickey et al.[1]	Prospective observational/709	Spontaneous reduction in one or more gestational sacs or embryos before 12 weeks	After spontaneous reduction, the average length of gestation was shortened by 4–250 days	Birth weight was inversely related to the initial number of gestational irrespective of the final birth number	None
La Sala et al.[2]	Retrospective/686	Pregnancies that began as twins but in which one embryo with positive heartbeat was lost during the first trimester or found to be lost in a subsequent ultrasonogram	Rates of preterm birth were similar among survivors of vanishing twin (16.7%) and control singletons (15.9%)	Rates of LBW were similar among survivors of vanishing twin (10.7%) and control singletons (12.9%)	Similar results were found when conventional IVF and IVF plus ICSI pregnancies were separately evaluated
Pinborg et al.[4]	National registry/642 survivors of a vanishing co-twin, 5237 singletons from single gestations, and 3678 twins from twin gestations	One fetus with positive fetal heart rate plus a gestational sac/fetus without fetal heart beat at 8 weeks	Shorter length of gestation in survivors (38.9 weeks) compared to control singletons (39.5 weeks)	1.7 times and 2.1 times higher odds of LBW and VLBW in singleton survivors of a vanishing twin after 8 weeks of gestation, respectively	Correlation between spontaneous reduction later in pregnancy and risk of neurological sequelae
Pinborg et al.[7]	National registry/642 survivors of a vanishing co-twin, 5237 singletons from single gestations, and 3678 twins from twin gestations	One fetus with positive fetal heart rate plus a gestational sac/fetus without fetal heart beat at 8 weeks	Not reported	The odds of SGA infants were 1.5 times higher in survivors than in singletons. There was a significant inverse correlation between SGA and the gestational age at the time of vanishing	None
Shebl et al.[8]	Case–control/794	One fetus with positive fetal heart rate plus a gestational sac/fetus without fetal heartbeat during a first trimester ultrasonogram	No differences were observed in terms of duration of gestation	The frequency of LBW (26.1% vs 12.0%) and SGA (32.6% vs 16.3%) was significantly higher in vanishing twin survivors	None

(Continued)

Table 3. (Continued)

Reference	Study type/sample size	Diagnosis of vanishing twin	Gestational age	Birth weight	Other
Almog et al.[9]	Retrospective case control study/1373 singleton deliveries and 57 cases of vanishing syndrome	Vanishing twin syndrome was defined in cases where two fetal heart beats were demonstrated between 6 and 7 weeks and one fetal heart beat was demonstrated thereafter, but before 12 weeks of gestation	Mean gestational age was lower (35.1 weeks) in compared to singleton controls (38.2 weeks)	Incidence of LBW (33.3%) and VLBW (3.5%) was higher in vanishing twin survivors compared to singleton controls, respectively	None
Rodríguez-González et al.[11]	Retrospective chart review of donor oocyte pregnancies/399	Spontaneous loss of one or more embryos after identifying heart activity during the first trimester	Gestational age at delivery was similar in both groups	Birth weight at delivery was similar in both groups	Preterm and term spontaneous rupture of membranes was increased in vanishing twin survivors
Mansour et al.[12]	Retrospective cohort study/2829	Presence of one or more additional empty gestational sacs or fetuses with no heartbeat at 6–7 weeks and 11–12 weeks of gestation	Not reported	There was no statistically significant difference in the mean gestational age between the groups	Live birth rate was higher significantly higher in singleton pregnancies after vanishing fetuses
Evron et al.[20]	Retrospective/278 vanishing twins were compared with 1801 dichorionic twins and 252,994 singletons	Pregnancies that started with double fetal sacs and spontaneously reduced into one during the first trimester	Not reported	Increased odds of IUGR (2.7 times) and LBW (6.9 times) in vanishing twin survivors compared to controls	Increased odds of gestational diabetes, low APGARS and perinatal mortality in vanishing twin survivors
Luke et al.[21]	Society for Assisted Reproductive Technology national database/21,535 singleton deliveries	First trimester loss of fetal cardiac activity	Increased odds of late PTB (1.73 times) and early PTB (2.56 times) in vanishing twin survivors compared to controls	Increased odds of LBW (2.09 times) and VLBW (1.94 times) in vanishing twin survivors compared to controls	None

IVF: in vitro fertilization; LBW: low birth weight; VLBW: very low birth weight; SGA: small-for-gestational age; PTB: preterm birth; ICSI: intracytoplasmic sperm injection; IUGR: intrauterine growth restriction.

Executive summary

Background

- SMFPR, also known as VT syndrome, refers to the spontaneous in utero reduction in one or more twins.
- Previous studies have reported various associations of VT syndrome with adverse perinatal outcomes such as LBW, chronic hypertension, gestational diabetes, preterm birth, preterm labor, as well as fetal malformations.
- The pathologic implications of the VT on the surviving twin has been debated, with some studies indicating a higher incidence of adverse obstetric and perinatal outcomes, while others demonstrating similar outcomes to singleton pregnancies.

Results

- Our findings suggest that singleton pregnancies affected by VT syndrome were associated with higher odds of overall LBW and term LBW in the VT group.
- VT was not associated with a higher incidence of preterm birth or overall very LBW.

Conclusion

- Our study provides confirmation of the increased odds of overall LBW, LBW, and term LBW in live births after fresh IVF-ET cycles affected by VT syndrome.
- Given these results, it is reasonable to consider closer ultrasonographic surveillance of twin pregnancies both in the first trimester to potentially diagnose a VT and once diagnosed, in the late trimester to confirm adequate growth.
- In addition, single ETs should be performed (when possible) to mitigate some of the aforementioned adverse outcomes arising from >1 ET.

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