Dichorionic triamniotic triplets after two blastocysts transfer underwent multifetal pregnancy reduction: two case reports and literature review

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Abstract

Background The increase in the rate of multiple pregnancies in clinical practice is associated with assisted reproductive technology (ART). Given the high risk of dichorionic triamniotic (DCTA) triplet pregnancies, reducing DCTA triplet pregnancies to twin or singleton pregnancies is often beneficial.

Case presentation This article reports on two cases of DCTA triplet pregnancies resulting from two blastocyst transfers. Given the high risk of complications such as twin-to-twin transfusion syndrome (TTTS) in monochorionic diamniotic (MCDA) twin pregnancies, patients have a strong desire to preserve the dichorionic diamniotic (DCDA) twins. Multifetal pregnancy reduction (MFPR) was performed in both cases to continue the pregnancy with DCDA twins by reducing one of the MCDA twins. Both of the pregnant women in this report eventually gave birth to healthy twins at 37 weeks.

Conclusions For infertile couples with multiple pregnancies but with a strong desire to remain the DCDA twins, our report suggests that reducing DCTA triplets to DCDA twin pregnancies may be an option based on clinical operability and assessment of surgical difficulty.

Keywords Dichorionic triamniotic (DCTA) triplet, Monochorionic diamniotic (MCDA) twin, Dichorionic diamniotic (DCDA) twin, Multifetal pregnancy reduction (MFPR), In vitro fertilization and embryo transfer (IVF-ET)

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Introduction

The clinical utilization of assisted reproductive technology (ART) and ovulation induction drugs, coupled with the rise in maternal age, has resulted in a consistent increase in the occurrence of multiple pregnancies over the last four decades [1]. As mentioned earlier, the likelihood of a monozygotic twin (MZT) following ART treatment is significantly greater than that of natural conception [2, 3]. There are three types of MZT: monochorionic-monoamniotic, monochorionic-diamniotic and dichorionic-diamniotic. Monochorionic twin (MCT) pregnancies are at high risk of maternal and fetal complications, especially fetal complications. In addition to selective intrauterine growth restriction (SIGR) and TTTS, which are unique complications of MCT, common fetal complications include preterm birth, fetal malformations, and perinatal death [4, 5].

Multiple pregnancies are becoming more prevalent as a potential consequence of infertility treatment [6]. In particular, MZT that split into two embryos from one embryo may result in high-order multiple pregnancies (HOMPs), such as DCTA triplet pregnancies [7, 8]. HOMPs are associated with a higher risk of complications such as miscarriage and preterm birth compared to singleton and twin pregnancies [9, 10]. Multifetal pregnancy reduction (MFPR) was first reported 35 years ago as a novel approach to managing multiple pregnancies [11]. Fetal reduction techniques were developed to improve the clinical outcomes associated with high mortality and morbidity associated with multiple pregnancies due to infertility treatment [12]. With clinicians' increasing surgical experience, familiarity with the patient's clinical situation, and a better understanding of the risks and benefits of different fetal reduction methods, transabdominal reduction and transvaginal reduction have become the most commonly used fetal reduction methods to reduce the high risk of multiple pregnancies [13–16]. Transvaginal fetal reduction is less commonly used than transabdominal fetal reduction, however, it may be a better option when the patient wishes to undergo surgery during the first trimester of pregnancy, or when it is difficult or impossible to perform transabdominal fetal reduction due to extreme obesity, abdominal scarring, or if the fetus cannot be reached through the abdomen [17]. Transvaginal fetal reduction is usually performed by aspiration of the embryo under the guidance of vaginal ultrasound and is best used in the first trimester of pregnancy (usually 6 to 8 weeks) [18, 19]. Transabdominal fetal reduction is usually performed by ultrasound-guided injection of KCl with a needle inserted through the abdomen into the fetal chest cavity and is generally used in the early second trimester of pregnancy (usually refers to 11–14 weeks) [17]. Transabdominal intracranial injection of KCl has been reported as an alternative to achieve a reduction in multiple pregnancies, ultimately leading to good pregnancy outcomes [20, 21]. In addition, selective fetal reduction by radiofrequency ablation and umbilical cord coagulation in the second trimester of pregnancy has also been used in some institutions. Due to differences in equipment availability, technology preferences, and clinician experience, the above techniques are not common in our institution [22].

The incidence of DCTA triplet pregnancies has been reported to be 0.004% [23]. MCT pregnancies are known to have a high risk of maternal and fetal complications including miscarriage, preterm birth, low birth weight, TTTS and perinatal death [4]. Reducing DCTA triplet pregnancies to twin or singleton pregnancies can significantly reduce the risk of complications for both mother and baby. However, there are few reports on fetal reduction protocols and pregnancy outcomes for DCTA triplet pregnancies [24]. There are even fewer studies on the reduction of DCTA triplet pregnancies to DCDA twin pregnancies. This article reports two cases of final pregnancy outcomes following the reduction of DCTA triplet pregnancies to DCDA twin pregnancies.

Case elaboration

Case 1

A 31-year-old woman complaining of secondary infertility underwent IVF-ET treatment after trying to conceive without contraception for 3 years. The woman has a history of two pregnancies that ended in miscarriage, one as a result of an induced abortion and the other due to embryonic development cessation. The patient was treated with a 10-day antagonist protocol with 150 IU of recombinant follicle-stimulating hormone (FSH) injected daily. 36 h before oocyte extraction, a total of 4000 IU of recombinant human chorionic gonadotropin (HCG) was injected to promote oocyte maturation. A total of 28 oocytes were retrieved vaginally under ultrasound guidance, of which 26 oocytes matured and successfully fertilized and cleavaged. There were 13 high-quality embryos on the third day. Twelve and six embryos reached the blastocyst stage on days 5 and 6, respectively, resulting in a total of 18 blastocysts, all of which were subsequently cryopreserved. In the subsequent frozen embryo transfer cycle, two day 5 blastocysts (Gardner classification: 4BA, 4BB) were transferred to the patient's uterus in response to the patient's request to increase the success rate of embryo implantation. The luteal phase was supported with progesterone (60 mg/day).

The patient's urine HCG test result was positive on the 14th day of the embryo transfer. We chose to measure fetal heartbeat status by transvaginal ultrasonography at 6 to 8 weeks of gestation to determine chorionicity and fetal number. The first ultrasound was performed on day 30 after embryo transfer and showed two chorionic sacs,

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suggesting a dichorionic pregnancy. One of the sacs contained germs that were 6.7 mm and 7.0 mm long, both of which showed a heartbeat. One germ in another sac was 7.3 mm long and exhibited pulsation (Fig. 1A). Follow-up ultrasound at 8 weeks gestation (42 days after oocyte transplantation) showed a dichorionic pregnancy with three distinct amniotic sacs, confirming a DCTA triplet pregnancy with MCDA twins. Ultrasonography revealed three embryos with heartbeats in the uterine cavity of the pregnant woman with lengths of 19.7 mm, 18 mm, and 20.1 mm (see Fig. 1B). After informing the patient and her family about the risks associated with DCTA triplet pregnancy and fetal reduction, the final selective reduction of the number of fetuses and the specific protocol is decided by the patient after a detailed consultation, and then the patient signed the informed consent form.

After completing the necessary preoperative examinations, an ultrasound-guided transvaginal fetal reduction was performed on the 43rd day after transplantation. To satisfy the desire of infertile couples who want to remain the DCDA twin, one of the MCDA twins was selectively reduced after assessing the difficulty of the surgery and informing them of the risks of the surgery. Intravenous anesthetics and ceftriaxone sodium (as a prophylactic antibiotic) were administered before surgery, followed by a comprehensive vaginal wash. The puncture needle was attached to a syringe, inserted into the vaginal wall using a needle guide, and then inserted into the gestational sac (GS). The puncture needle was positioned near the germ and used to aspirate and halt the activity of the embryonic heart. The surgery went smoothly. The patient bled less and experienced minor pain during the operation. The postoperative follow-up ultrasound revealed that one of the three fetuses had been reduced, and the embryonic cardiac tube pulse of the reduced fetus was no longer detectable (see Fig. 1C). The patient underwent an emergency cesarean section after fetal distress caused by premature rupture of membranes (PROM) at 36 weeks
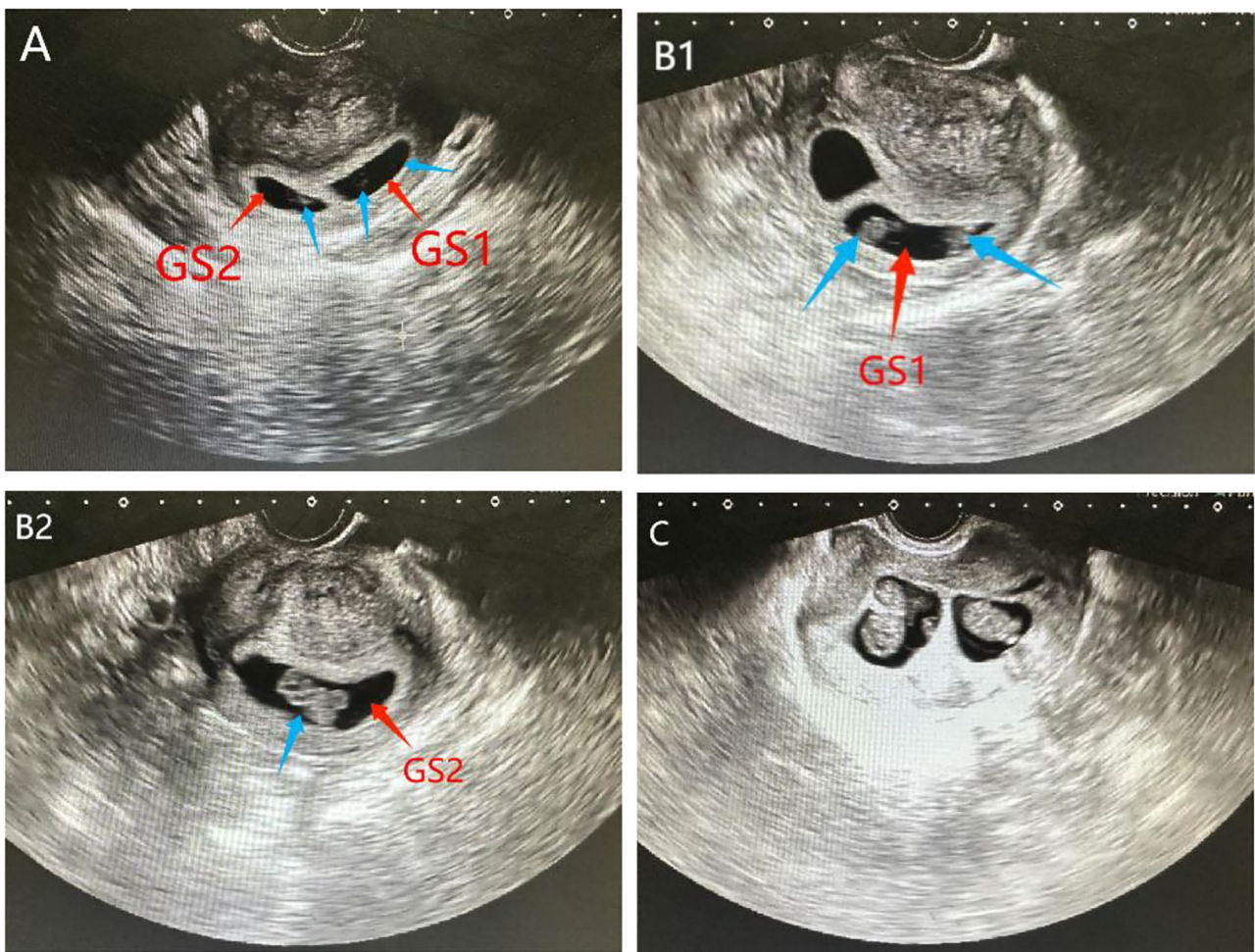

Fig. 1 Vaginal ultrasound reveals dichorionic triamniotic triplets with two gestational sacs (GS, red arrow) and three germs (blue arrow) in the uterus. **A**) Ultrasound images were taken for the first time 30 days after transplantation. **B1-2**) Ultrasound image of the second pregnancy on day 40 after embryo transfer. **C**) Ultrasound image after selective fetal reduction

and 6 days of gestation and successfully delivered a boy and a girl. The boy was born weighing 2800 g and was 50 cm in length, while the girl weighed 2500 g and measured 49 cm in length. Both infants scored 10 points on their 1-minute and 5-minute Apgar assessments. Subsequent follow-up visits with the children also confirmed that the twins remained healthy.

Case 2

A couple who complained of primary infertility for 6 years came to our hospital for IVF-ET treatment due to the sequelae of pelvic inflammatory disease and oligospermia, asthenospermia and teratozoospermia. The patient is a 34-year-old woman with a hysterosalpingogram (HSG) showing obstruction of the right fallopian tube and irregularity of the left fallopian tube. The patient was also treated with an antagonist protocol. A total of 21 oocytes were retrieved vaginally under ultrasound guidance, of which 19 oocytes matured and 17 oocytes were successfully fertilized and cleavaged. In vitro, embryo culture showed seven high-quality embryos on day 3. All embryos were cultured in vitro to day 5 or 6, resulting in a total of 13 blastocysts. All blastocysts were cryopreserved, including seven day 5 embryos and six day 6 embryos. In the subsequent frozen embryo transfer cycle, two day 5 blastocysts (Gardner classification: 4BB*2) were transferred to the patient's uterus in response to the patient's request to increase the success rate of embryo implantation. The luteal phase was supported with progesterone (60 mg/day).

The patient's urine HCG test was positive on the 14th day after the embryo transfer. The first ultrasound, performed on day 29 after embryo transfer, revealed two chorionic sacs in the uterine cavity of the pregnant woman, one of which contained germs measuring 7.2 mm and 7.0 mm in length, both exhibiting a heartbeat. One germ in another sac was 6 mm long and had a heartbeat (see Fig. 2A). A repeat ultrasound on day 40 after oocyte transplantation confirmed a DCTA triplet pregnancy, with two of the fetuses being an MCDA twin. Ultrasound revealed three distinct amniotic sacs and three embryos with lengths of 19.8 mm, 19 mm, and 19.9 mm in length within the uterine cavity (see Fig. 2B). All three embryos were found to have a heartbeat. We informed the couple about the risks associated with multiple pregnancies and advised them to consider selectively reducing embryos. Eventually, the couple chose to selectively reduce the DCTA triple pregnancy to a DCDA twin pregnancy. After completing the necessary preoperative examinations, an ultrasound-guided transvaginal fetal reduction was performed on the 45th day after transplantation. Under ultrasound guidance, a puncture needle was used to puncture and rapidly aspirate one of the embryos in the MCDA twin, causing the embryonic echo to disappear. The operation went smoothly, and the patient experienced minimal bleeding and no significant pain during the procedure. The postoperative follow-up ultrasound revealed that one of the three fetuses had been reduced, as the fetal embryonic cardiac tube pulse of the reduced fetus was no longer detectable. The remaining embryos were growing normally. The patient was diagnosed with diabetes during subsequent pregnancies. Considering the great preciousness of the fetus to the couple, the pregnant woman and her family requested a cesarean section. Two girls were delivered by cesarean section at 37 weeks and 3 days of gestation. At birth, one of the girls weighed 2800 g and measured 48 cm in length, while the other weighed 2400 g and measured 46 cm in length. Both infants received Apgar scores of 10–10 at 1 min and 5 min after birth. Follow-up in the neonatal period also indicated that the twins were in good health.

Discussion and conclusions

As previously reported [25], an increasing number of women are choosing to delay marriage and childbirth, with the majority not starting a family until after the age of 30. Late marriage and delayed childbearing have become interrelated factors that affect women's reproductive health. Consequently, there has been an increase in the demand for assisted reproductive technologies.

Fig. 2 Vaginal ultrasound reveals dichorionic triamniotic triplets with two gestational sacs (GS, red arrow) and three germs (blue arrow) in the uterus. **A**) Ultrasound images were taken for the first time 29 days after transplantation. **B1-2**) Ultrasound image of the second pregnancy taken on day 40 after transfer

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While utilizing ART, there are potential risks associated with IVF. The study data suggests that the likelihood of monochorionic twins after natural conception was 0.4%, whereas the probability after ART ranged from 1.57 to 13.2% [2, 3]. The use of ART significantly increases the likelihood of multiple pregnancies. However, it remains unclear how embryos undergo division after ART. Studies on this issue suggest that factors such as maternal age, age of oocytes, zona pellucida manipulation techniques, prolonged embryo culture to the blastocyst stage, and embryonic morphology may strongly correlate with an increased incidence of monochorionic twins [8, 26–28]. However, there is still no unified explanation of how these factors influence the occurrence of monochorionic twins. Given the high risk of multiple pregnancies and their influencing factors, we consider the transfer of no more than two day 3 cleavage embryos or one blastocyst to be an advantageous option.

Globally, the proportion of triplets resulting from IVF-ET treatment is 0.7% or greater [29], and the incidence of DCTA triplets is only 0.004% [23]. Compared to singleton pregnancies, triplet pregnancies carry a high risk of extremely preterm birth, low birth weight, fetal malformations, miscarriage, and perinatal death [4, 5]. It is worth noting that DCTA triplets also carry a high risk of SIGR, TTTS, and neurological disorders associated with MCDA twins. However, there is little follow-up of women with these conditions, resulting in limited data on pregnancy outcomes and inadequate follow-up of their children.

MFPR is often considered a favorable option to reduce the risk of multiple pregnancies. Selectively reducing triplet pregnancies or HOMPs to twin or singleton pregnancies is an effective approach to reduce the risks and enhance clinical outcomes for multiple pregnancies. It must be mentioned that MFPR, while used as a means to control the number of developing fetuses and thereby reduce the high risk of multiple pregnancies, also involves numerous ethical and clinical issues. Considering fetal reduction strategies alone, there are three options for reducing DCTA triplets: reducing the singleton to preserve the MCDA twins, reducing the MCDA twin to preserve the singleton, or reducing one of the MCDA twins to continue the pregnancy with the DCDA twins. Regarding the different strategies for MFPR, is conservative management a visible option, or must a DCTA triplet pregnancy be reduced to twin pregnancy or singleton pregnancy? There are still relatively few studies on this topic. Previous studies have shown that the complete miscarriage rate and preterm birth rate of DCTA triplets are 29.41% and 66.67% [30]. Reducing DCTA triplet pregnancies to MCDA twin pregnancies has been reported to result in lower miscarriage rates and higher live birth rates compared to expectant management [31]. However,

our previous study showed that reducing DCTA triplets to singleton pregnancies resulted in lower miscarriage and preterm birth rates, as well as higher live birth rates, compared to reducing DCTA triplets to twin pregnancies [30]. In addition, reducing DCTA triplets to singleton pregnancies is associated with a significant reduction in perinatal mortality and neonatal morbidity [32, 33]. These findings are consistent with previous reports that reducing DCTA triplets to MCDA twins was associated with higher rates of preterm birth, low birth weight, perinatal mortality, and higher prevalence of cesarean section compared with reduction of DCTA triplets to singletons [34, 35]. Therefore, we believe that reducing DCTA triplets to singleton pregnancies results in better pregnancy outcomes. This is also consistent with the results of most studies that reducing DCTA triplets to singleton pregnancies is a safe and reliable strategy [31, 36]. However, some infertile couples have expressed their desire to remain the twins. It is well known that the risk of complications such as TTTS in MCDA twin pregnancies is extremely high, so patients have a strong desire to preserve DCDA twins based on the risk of MCDA twins. Given this, better fetal reduction strategies need to be considered to avoid the risks associated with MCDA and to meet patient wishes. Selectively reducing one fetus in MCDA twins can prevent potential complications associated with MCDA twin pregnancy. However, this fetal reduction strategy leads back to another increased risk that when selectively reducing one of the MCDA twins may result in the death of the other surviving fetus due to the shared placenta [37]. Therefore, for infertile couples who strongly want to remain DCDA twins, we should fully inform them of this risk to choose the best strategy for fetal reduction.

We generally advise patients to reduce DCTA triplets to singleton pregnancies for better pregnancy outcomes, however, some infertile couples have a strong desire to preserve the twins. For infertile couples with a strong desire to preserve the twins, there are two fetal reduction strategies: reducing monozygotic singletons to preserve the MCDA twins and reducing one of the MC twins to preserve the DCDA twins. However, there is little research on whether MCDA twins or DCDA twins are preserved. Existing studies have shown that the perinatal mortality, stillbirth, and neonatal mortality rates associated with MCDA twin pregnancies are higher than those of DCDA twin pregnancies [38, 39]. In addition, the study also found that there was a 28.6% miscarriage rate after reducing the DCTA triplets to MCDA twin pregnancies, which was higher than the miscarriage rate after the DCTA triplets were reduced to DCDA twin pregnancies, and most of them showed no signs of poor fetal condition [39, 40]. Studies on neonatal outcomes have shown that DCDA twins result in higher birth weight and lower

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neonatal morbidity (e.g., digestive morbidity) compared with MCDA twins [39]. To provide a more complete picture of pregnancy outcomes after elective reduction of triplets with DCTA, we performed a retrospective analysis of patients who were reduced to MCDA and those who were reduced to DCTA. Our recent study found that DCTA triplets to DCDA twin pregnancies resulted in lower rates of complete miscarriage, preterm birth rates, and very low birth weight compared to DCTA triplets to MCDA twin pregnancies [41]. In addition, the full-term delivery rate and neonatal survival rate after reducing the DCTA triplets to the DCDA twin pregnancy were also higher than the DCTA triplets to the MCDA twin pregnancy [41]. In neonates, the mean birth weight and twin weight after reducing the DCTA triplets to DCDA twins were also significantly higher than those after the DCTA triplets were reduced to MCDA twins [41]. Studies showed that the pregnancy outcomes and neonatal outcomes of DCTA triplets to DCDA twin pregnancies were better than those of DCTA triplets to MCDA twins [41]. However, it is important to note that our small sample size may lead to biased results. Due to the unique characteristics of monochorionic twins, selective reduction of one fetus in monozygotic twins is not considered feasible. Because the risk of early TTTS occurs due to 96% of the blood vessel's anastomosis on the placental bed, reducing one of the monochorionic twins is likely to result in a miscarriage of the other. Patients should be fully informed about the risks associated with this fetal reduction strategy, and the final number of fetal reductions and strategies should be decided by the patients after the patients are fully aware of the risks of different fetal reduction strategies. In summary, we believe that reducing DCTA triplets to DCDA twin pregnancies may be an acceptable option, also based on clinical feasibility, for infertile couples who have a strong desire to remain twins and want to minimize or reduce the risk of complications. Given the scarcity of reports of elective fetal reduction in DCTA triplet pregnancies, a definitive fetal reduction regimen remains unclear. Therefore, the optimal fetal reduction strategy is based on the patient's preference, assessment of the difficulty of the procedure, and the clinician's technique.

Several methods of fetal reduction have been reported, including ultrasound-guided transabdominal puncture injection of lethal substances such as KCl or hypertonic saline, radiofrequency ablation, cord occlusion, and transcervical puncture [42, 43]. However, there are some technical difficulties associated with these methods. For example, transcervical puncture requires cervical dilation and transabdominal puncture for injection of cardiotoxic substances may necessitate multiple punctures or multiple surgeries. When one of the MCDA twins is chosen to be reduced, the placenta of MC is rich in blood vessels,

and the flow through the placental vessels after KCl injection may lead to acute hemodynamic changes in the surviving fetus, and ultimately lead to the death of the other surviving fetus [44]. Other studies have shown that 84.7% of MC twins will die after KCl is injected into one of the MC twins [45]. The study found that the majority of MC twin deaths occurred within 1 day of injecting KCl into one of the MC twins [46]. KCl injection into one of the MC twins resulted in extremely high MC twin mortality. To what extent is this result related to the dose of KCl injections and the number of fetal punctures? There are very few studies on this issue, which may need to be confirmed in studies with larger sample sizes. This method may also be difficult to perform due to the position of the fetus. Intrafetal laser ablation has also been reported to result in up to 50% miscarriage after fetal reduction [47]. In addition, the clinical application of umbilical cord occlusion mainly depends on the availability of equipment and the experience or expertise of clinicians, which is not common in our country. Studies have shown that there is a possibility of spontaneous fetal reduction or single intrauterine fetal demise during the pregnancy of DCTA triplets, which refers to the natural cessation of one or both fetuses in DCTA [30, 48]. Studies have shown that a single intrauterine fetal death in multiple pregnancies results in a 30–50% risk of death for surviving twins [49, 50]. In this case, the patient should be adequately informed of the risks and communicated to the patient about the pros and cons of different fetal reductions. The decision of whether to perform MFPR and the number and specific strategy of fetal reduction is ultimately up to the patient.

Fetal reduction by transvaginal aspiration of embryos has been reported as an effective method for fetal reduction in DCTA triplets [36], both patients we reported opted for transvaginal ultrasound-guided embryo aspiration to reduce the number of embryos. We chose to diagnose chorionic and amniotic pregnancy in twin pregnancies by vaginal ultrasound in the first trimester of pregnancy (typically between 8 and 11 weeks of gestation, i.e., between 6 and 8 weeks after embryo transfer) to assess the appropriate timing of fetal reduction. This decision is based on the fact that at 7–9 weeks and 11–14 weeks of gestation, transvaginal ultrasound is reliable and very accurate in the identification and diagnosis of chorionic and amniotic membranes [51, 52]. Transvaginal ultrasound-guided embryo puncture overcomes the technical difficulties of transabdominal or transcervical puncture to a certain extent and also has obvious advantages. First, the high-frequency probe can greatly increase the image resolution of the gestational sac and its contents to make the imaging clear. Secondly, the tip of the sensor probe can be close to the uterine wall and the gestational sac to be aspirated with the help of the elasticity of

the vagina, and compared with the transabdominal route, the puncture process is more definite and the puncture route is shorter, which reduces the risk of inadvertent damage to the gestational sac. Furthermore, it is allowed to be carried out in the first trimester of pregnancy, such as 6 weeks of gestation, when the size of the gestational sac and its contents to be absorbed is smaller, and the embryo is easier to be sucked out. Early intervention and management of patients is psychologically beneficial. Additionally, repeated puncture of the embryo or injection of lethal substances such as KCl is avoided, making the whole process faster, more accurate, and easier, and also reducing the risk of vaginal bleeding and infection due to repeated vaginal insertion.

Currently, there are few studies on reducing DCTA triplets to twin pregnancies, especially DCDA twin pregnancies. Depending on the patient's preference and specific clinical situation, reducing DCTA triplets to DCDA twins may be an option. This case report describes the successful outcome of two DCTA triplets that were reduced to DCDA twins. Both twins had good pregnancy outcomes. This will serve as a clinical reference for triplet pregnancies in pregnant women who need selective fetal reduction. Our report will be beneficial for infertile couples with DCTA triplets who have a strong desire to retain the DCDA twins.

Abbreviations
ART	Assisted reproductive technology
DCTA	Dichorionic triamniotic
MCDA	Monochorionic diamniotic
DCDA	Dichorionic diamniotic
TTTS	Twin-to-twin transfusion syndrome
IVF-ET	In vitro fertilization and embryo transfer
MFPR	Multifetal pregnancy reduction
MZT	Monozygotic twin
MCT	Monochorionic twin
SIGR	Selective intrauterine growth restriction
HOMPs	High-order multiple pregnancies
FSH	Follicle-stimulating hormone
HCG	Human chorionic gonadotropin
GS	Gestational sacs
HSG	Hysterosalpingogram
PROM	Premature rupture of membranes
HSG	Hysterosalpingogram

Declarations

Consent for publication
The authors affirm that human research participants provided informed consent for the publication of the images in Figs. 1 and 2.

Competing interests
The authors declare no competing interests.

Ethics approval
This study was approved by the Ethics Committee of Anhui Medical University (PJ20180707). All methods were carried out in strict accordance with the relevant guidelines and regulations.

Consent to participate
Informed consent was obtained from all the parents.

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