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Original Article

Clinical characteristics and risk factors of twin pregnancies with single fetal malformation: a multi-center analysis in Southeastern China

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Abstract: Objective: Twin pregnancy, which has risen in the incidence across the world, causes an increased risk to both mother and babies. However, there is little knowledge on the clinical characteristics and risk factors of twin pregnancies with single fetal malformation. This multi-center analysis aimed to investigate the clinical characteristics and identify the risk factors of twin pregnancies with single fetal malformation in Fujian Province, Southeastern China. Methods: A total of 216 twin pregnancies with single fetal malformation diagnosed at 6 tertiary hospitals in Fujian Province during the period from October 1, 2013 through June 30, 2016 were sampled as the case group, while 216 normal twin pregnancies were selected as controls. The fetal malformations were identified from the subjects' medical records, and univariate and multivariate logistic regression analyses were employed to identify the risk factors of twin pregnancies with single fetal malformation. Results: The prevalence of twin pregnancies with single fetal malformation was 15.2% in Fujian Province, Southeastern China from 2013 to 2016. There were 225 types of fetal malformations diagnosed in the 216 twin pregnancies with single fetal malformation, and the most three common organs where the fetal malformation developed were the cardiovascular system, the skeletal system, and the urinary system. Univariate analysis revealed that the development of twin pregnancies with single fetal malformation was positively correlated with natural conception and monochorionic twins ($P = 0$), and multivariate logistic regression analysis identified monochorionic twins as an independent risk factor for twin pregnancies with single fetal malformation ($P = 0$). Conclusions: The results of the present study demonstrate a high rate of twin pregnancies with single fetal malformation in Fujian Province, southeastern China, and the fetal malformation mainly occurs in the cardiovascular system, the skeletal system and the urinary system. In addition, having monochorionic twins is an independent risk factor for twin pregnancies with single fetal malformation.

Keywords: Twin pregnancy, single fetal malformation, clinical characteristics, risk factor, multi-center study, southeastern China

Introduction

Twins are defined as two offspring produced by the same pregnancy, which account for 2%-3% of all births [1]. The incidence of twin pregnancy has recently risen across the world due to high maternal age, increasing use of assisted reproductive technology (ART) and other risk factors [2, 3]. Twin pregnancy causes an increased risk to both mothers and babies, including preterm delivery, intrauterine growth restriction, and pre-eclampsia [2]. In addition, monochorionic gestations confer an even higher rate of perinatal morbidity and mortality, which may lead to twin-to-twin transfusion syndrome or twin anaemia-polycythaemia sequence [4-7].

With the increasing incidence of twin pregnancy, the rate of twin pregnancies with single fetal malformation is also on a rise [8]. With the implementation of China's universal two-child policy [9], the use of ART has sharply increased [10], which may lead to an increase in the incidence of twin pregnancy in China. However, there is little knowledge on the clinical characteristics and risk factors of twin pregnancies with single fetal malformation. This multi-center analysis aimed to investigate the clinical char-

acteristics and identify the risk factors of twin pregnancies with single fetal malformation in Fujian Province, Southeastern China.

Subjects and methods

Subjects

Twin pregnancies with single fetal malformation diagnosed at 6 tertiary hospitals in Fuzhou City, Fujian Province during the period from October 1, 2013 through June 30, 2016 were sampled as the case group; including Fujian Provincial Hospital, Fujian Provincial Maternity and Children's Hospital, the First Affiliated Hospital of Fujian Medical University, Fujian Medical University Union Hospital, Fuzhou Municipal First Hospital and Fuzhou Municipal Second Hospital; while pair-matched normal twin pregnancies were selected as controls. All twin pregnancies with single fetal malformation were diagnosed by means of clinical diagnosis or auxiliary tools (ultrasonography, chromosomal examinations or X-ray scan) during the study period were included in this study, while singleton pregnancy, two or more gestations, normal twin pregnancies or twin pregnancies with both fetal malformations were excluded from the study.

Twin pregnancies were classified into monochorionic and dichorionic forms based on the chorionicity and grouped into natural conception and ART conception groups according to the type of conception [1]. All subjects' demographic and clinical characteristics were captured from the medical records.

Classification of fetal malformation

All fetal malformation data were captured from the hospital-based birth defects registration cards and perinatal infant reports, and all fetal malformations were classified according to the International Classification of Diseases (ICD)-10 diagnostic categories [11]. Fetal malformations were classified into 11 categories according to the site where the malformation occurred, including the nervous system, cardiovascular system, digestive system, skeletal system, urinary system, acardiac twins sequence, face and neck, respiratory system, abdominal fissure, chromosome abnormality and conjoined twins. A malformation that occurred in two systems or more was defined as multiple malformations.

Ethical statement

This study was approved by the Ethical Review Committee of Fujian Provincial Maternity and Children's Hospital (date of approval: August 17, 2016; approval no.: 2016-0183). Written informed consent was obtained from all participants following a detailed description of the purpose of the study.

Data analysis

All data were entered into Microsoft Excel 2007 (Microsoft; Redmond, WA, USA), and all statistical analyses were performed using the statistical software SPSS version 13.0 (SPSS, Inc.; Chicago, IL, USA). All measurement data were expressed as mean ± standard deviation (SD). Differences of proportions were compared with chi-square test, and the risk factors of twin pregnancies with single fetal malformation were identified using univariate and multivariate logistic regression analyses.

Results

Subject characteristics

During the period between October 1, 2013 and June 30, 2016, a total of 1421 twin pregnancies were recruited at 6 tertiary hospitals in Fuzhou City, Fujian Province, and there were 216 twin pregnancies with single fetal malformations diagnosed, with a prevalence rate of 15.2%. Among all cases, there were 57.87% (125/216) diagnosed at Fujian Provincial Maternity and Children's Hospital, 13.42% (29/216) diagnosed at Fujian Provincial Hospital, 8.33% (18/216) diagnosed at the First Affiliated Hospital of Fujian Medical University, 5.56% (12/216) diagnosed at Fujian Medical University Union Hospital, 8.8% (19/216) diagnosed at Fuzhou Municipal First Hospital and 6.02% (13/216) diagnosed at Fuzhou Municipal Second Hospital, respectively.

There were 23.61% twin pregnancies with single fetal malformation (51/216) diagnosed during the antenatal period, and 76.39% (165/216) during the postnatal period, 86.27% (44/51) that received prenatal diagnosis at < 28 weeks of gestational age and 13.73% (7/51) at 28 weeks of gestational age and greater. The malformed fetuses included 115 males (53.24%), 95 females (43.98%) and 6 of unknown gender

Table 1. Types of conception and chorionicity of 225 malformations seen in 216 twin pregnancies with single fetal malformation

Site of fetal malformation	Type of conception		Chorionicity		Total
	Natural conception	Assisted reproductive technology	Monochorionic twin	Dichorionic twin	
Cardiovascular system	79	11	66	24	90
Skeletal system	30	4	23	11	34
Urinary system	27	5	24	8	32
Face and neck	19	2	14	7	21
Nervous system	15	0	14	1	15
Digestive system	9	0	7	2	9
Conjoined twins	8	0	8	0	8
Abdominal fissure	8	0	6	2	8
Acardiac twins sequence	3	0	3	0	3
Respiratory system	3	0	1	2	3
Chromosome abnormality	3	0	2	0	2
Total	203	22	168	57	225

(2.78%), and the prognoses included 180 live births (83.33%), 27 stillbirths (12.5%) and 9 deaths within 7 days upon diagnosis (4.17%). There were 125 twin pregnancies with single fetal malformation (57.87%) diagnosed using ultrasonography, 75 (34.72%) through clinical examinations, 11 (5.09%) by means of clinical inspections and ultrasonography, 1 (0.46%) using chromosomal test, 1 (0.46%) using ultrasonography combined with chromosomal test and 3 (1.39%) using other tools.

The case and control groups were balanced with the mean maternal age (29 ± 4.6 vs. 27 ± 5.7 years, $P > 0.05$).

Fetal malformations

There were 225 types of fetal malformations diagnosed in the 216 twin pregnancies with single fetal malformation, and the most three common systems where the fetal malformation occurred included the cardiovascular system (40%), the skeletal system (15.1%) and the urinary system (14.2%). Ventricular septal defect was the predominant fetal malformation in the cardiovascular system (72.2%), talipese quinovarus was the predominant fetal malformation in the skeletal system (35.3%), and hypospadia was the predominant fetal malformation in the urinary system (34.4%). In addition, natural conception was the predominant conceptive type (90.2%) and monochorionic twins were the predominant type o twin pregnancies (74.7%) seen in the 216 twin pregnancies with single fetal malformation (**Table 1**). **Table 2** describes the types of fetal malformations diagnosed at the 6 tertiary hospitals included in this study.

Risk factors of twin pregnancies with single fetal malformation

Univariate analysis revealed that the development of twin pregnancies with single fetal malformation correlated with natural conception and monochorionic twins ($P = 0$), and had no associations with maternal age, number of pregnancies, residency or education levels ($P > 0.05$) (**Table 3**). Multivariate logistic regression analysis identified monochorionic twins as an independent risk factor for twin pregnancies with single fetal malformation ($P = 0$) (**Table 4**).

Discussion

Fetal malformation, including specific and non-specific malformation, is a major cause of perinatal mortality, which involves all systems across the body [12]. Previous studies have demonstrated a higher rate of fetal malformation in twin pregnancies than in single pregnancies, with single-twin malformation as the predominant form [13-18]. In this study, the prevalence of twin pregnancies with single fetal malformation was 15.2% in Fujian Province, southeastern China, which was far greater than the national prevalence in China (2.86%) [19], and in 14 European countries from 1984 to 2007 (10.7 per 10,000 births) [20]. The high rate of twin pregnancies with single fetal mal-

Table 2. Types of fetal malformations seen in 216 twin pregnancies with single fetal malformation in 6 centers included in this study

Center at diagnosis	Type of fetal malformation											
	Cardiovascular system	Skeletal system	Urinary system	Face and neck	Nervous system	Digestive system	Conjoined twins	Abdominal fissure	Acardiac twins sequence	Respiratory system	Chromosome abnormality	Total
Fujian Provincial Maternity and Children's Hospital	55	20	20	7	9	5	4	3	2	3	2	130
Fujian Provincial Hospital	10	5	1	5	2	2	2	2	1	0	0	30
The First Affiliated Hospital of Fujian Medical University	9	3	2	0	2	1	1	0	0	0	0	18
Fujian Medical University Union Hospital	3	3	2	1	1	1	0	1	0	0	0	12
Fuzhou Municipal First Hospital	8	3	5	3	1	0	1	1	0	0	0	22
Fuzhou Municipal Second Hospital	5	0	2	5	0	0	0	1	0	0	0	13
Total	90	34	32	21	15	9	8	8	3	3	2	225

Table 3. Univariate analysis of risk factors for 216 twin pregnancies with single fetal malformation

Demographic and clinical feature		Case group (n = 216)	Control group (n = 216)	χ^2 value	P value
Maternal age (years)	< 35	188	197	1.912	0.167
	≥ 35	28	19		
Number of pregnancy	1	88	95	0.464	0.496
	≥ 2	128	121		
Residency	Urban areas	116	95	0.337	0.562
	Rural areas	100	121		
Education level	Junior high school and lower	54	64	0.948	0.33
	Senior high school and higher	162	152		
Type of conception	Natural conception	194	161	16.213	0
	Assisted reproductive technology	22	55		
Chorionicity	Monochorionic twin	159	79	56.747	0
	Dichorionic twin	57	137		

Table 4. Multivariate analysis of risk factors for 216 twin pregnancies with single fetal malformation

Variable	*b*	*SE*	*Wald*	*P*	*OR*	95% *CI*
Natural conception	0.452	0.301	2.257	0.133	1.304	0.871, 2.837
Monochorionic twin	1.459	0.222	43.334	0	3.507	2.785, 6.639

formation may be attributed to the advanced maternal age [3]; however, the exact causes require further investigations.

In this multi-center study, 57.87% twin pregnancies with single fetal malformation were diagnosed at Fujian Provincial Maternity and Children's Hospital. As the only province-level maternity and children's healthcare center, more than 18 thousand pregnancies were delivered at the hospital, and most of the twin pregnancies received prenatal diagnosis and were delivered at this hospital. In addition, Fujian Provincial Center for Prenatal Diagnosis is built in this hospital, which undertakes the diagnosis of birth defects, notably the birth defects of twin pregnancies in Fujian Province. Therefore, more than a half of the twin pregnancies with single fetal malformation were diagnosed at this center. We found that the twin pregnancies with single fetal malformation were predominantly diagnosed by means of ultrasonography (57.87%). It has been shown that ultrasound detects over 90% of fetal malformations, including congenital heart disease, malformations of the nervous system, malformations of the skeletal system, abdominal fissure and acardiac twins [21]. Clinical inspection is a non-ignorable diagnostic approach, notably for the malformations on the fetal body surface, such as hypospadias, accessory ear, hemangiom and talipes equinovarus, and a careful physical examination may reduce the missing diagnosis of these malformations. In this study, the circulatory system was found to rank first as the site of fetal malformation development, which may be explained by the fact that most of the malformations in the circulatory system are diagnosed using ultrasound. These data indicate that improving the detection of malformations during pregnancy and the subsequent termination of pregnancy is of great importance to reduce fetal death and the birth of unstable and malformed infants. In addition, the 51 twin pregnancies with a single fetal malformation receiving prenatal diagnosis all had severe malformations, which mainly occurred at the nervous system (19.6%) and the circulatory system (19.6%). Therefore, selective fetal reduction may be done to reduce the delivery of the fetus with severe malformations in the context of the other normal fetus in twin pregnancies.

In the current study, the most three common organs where fetal malformation developed were the cardiovascular system, the skeletal system, and the urinary system, which was similar to previous reports [22, 23]. We found that congenital heart disease was predominant in cardiovascular disorders, and ventricular septal defect was the predominant fetal malformation in the cardiovascular system, which was in

agreement with previous findings that congenital heart defects are the most common type of birth defects in both single and twin pregnancies [24-26]. Malformations of the skeletal system include talipese quinovarus, polydactyly, limb shortening and deformity, spinal dysplasia, strephexopodia, ankylodactylia, rib deformity and muscular spasm, and most of these malformations may be identified by prenatal diagnosis during the pregnancy and have a satisfactory prognosis [20]. However, severe limb shortening and deformity and muscular spasm may cause neonatal disabilities, leading to psychological and physiological burdens. Malformations of the urinary system mainly included hypospadia, enorchia, hydrocele of tunica vaginalis and concealed penis, and most of these malformations are diagnosed after birth. Therefore, active postpartum follow-up and treatment should be given to these malformations and prevention is required at all three levels.

With great advances in maternal-fetal medicine [27], complex twin pregnancies are paid increasing attention [1], and remarkable improvements have been achieved in intrauterine therapy [28-31]. Most complex twin pregnancies are considered as specific complications of enzygotic twins [1]. In this study, we detected a low proportion of acardiac twins and conjoined twins in the study subjects; however, these highly fatal and unstable malformations require a lot of attention. Acardiac twins sequence mainly includes acephalocardia, acardius anencephalus, acardius amorphus and acardius acormus [32]. During the embryonic period, the twins may develop obvious placental vascular anastomosis, and the fetus with normal structure (blood pumper) pumps and transfers blood to the fetus with abnormal structure (blood receptor), which aggravates the cardiac burden in the blood pumper and leads to a high likelihood of developing heart failure, resulting in pericardial effusion and polyhydramnios [33, 34].

In the current study, we found that the development of twin pregnancies with single fetal malformation was associated with the chorionicity and type of conception ($P < 0.05$). Univariate analysis identified natural conception and monochorionic twins as risk factors for twin pregnancies with single fetal malformation ($P < 0.05$), and further multivariate logistic regression analysis revealed that monochorionic twins as an independent risk factor for twin pregnancies with single fetal malformation ($P < 0.05$).

The chorionicity of twin pregnancies includes dichorionic and monochorionic twins; dichorionic twins mainly develop from dizygotic twins, while monochorionic twins, which only develop in monozygotic twins, arise from the division of a single fertilized egg [35]. Most of dichorionic twins occur in natural conception; however, they may also develop from unnatural conception. Following single fetal growth arrest in twin pregnancies, the surviving fertilized eggs develop cleavage, and then develop into monozygotic twins [36]. It is theoretically speculated that monochorionic twins share almost consistent genetic materials, and have an equal possibility of malformations; therefore, monochorionic twins are more likely to develop malformations, or exhibit a lower possibility to have a single fetal malformation than dichorionic twins [37]. Currently available data show a stronger correlation between the chorionicity and fetal malformation in monochorionic twins than in dichorionic twins [24, 33], which is in agreement with the findings from the present study. It is hypothesized that because of uneven cell distribution during the division of monozygotic twins, there is arteriovenous and placental sharing discordance in the placenta, which leads to unequal fetal nutritional distribution or impact of teratogens on a single fetus, thereby resulting in inconsistent growth trends of twins [38, 39]. Nevertheless, dichorionic twins have their respective independent placentas, and there is no vascular communication between the twins, thereby resulting in a low possibility of fetal malformations [40].

Currently, whether the conception by ART increases the incidence of fetal malformations remain in dispute, and the underlying mechanisms have not been fully demonstrated [41-44]. Twins conceived by natural ways had a greater possibility of anencephalus than those conceived by ART, which was considered to be attributed to ovarian hyperstimulation [23]. However, ART was also reported to have no associations with the development of fetal malformations [45-47]. In this study, our data showed that natural conception was the predominant type of conception in twin pregnancies with single fetal malformation, and natural conception was identified as a risk factor for

twin pregnancies with single fetal malformation. This may be explained by the fact that twins conceived by natural ways are mostly monochorionic, while the pregnancies conceived by ART are mostly dichorionic [48]. In addition, some diseases may be screened and diagnosed prior to implantation with the advances in ART, which reduces the development of malformations [49, 50].

Fetal malformation is a major cause of intrauterine fetal death in dichorionic twins [51]. In monochorionic twins, however, the single fetal malformation may cause poor pregnancy outcomes in the second healthy fetus due to vascular communication, including abortion, premature birth, stillbirth and cerebral injuries [1]. Therefore, the decision on the malformed fetus should be made based on a systematic assessment of the maternal and fetal conditions. Reduction of a malformed fetus is recommended for fetuses with deadly or severe disabling malformations following definitive diagnosis, notably exclusion of chromosome abnormality in the other fetus. Currently, intrauterine therapy has shown effective and safe for the treatment of severe fetal malformations, including acardiac twins sequence, and improvements in the outcomes of the normal fetus [25-27].

In summary, the results of the present study demonstrate a high rate of twin pregnancies with single fetal malformation in Fujian Province, southeastern China, and the fetal malformation mainly occurs in the cardiovascular system, the skeletal system and the urinary system. In addition, monochorionic twin is an independent risk factor for twin pregnancies with single fetal malformation. More health education and primary prevention is strongly recommended to reduce the development of malformations in twin pregnancies. Furthermore, precision diagnosis and management of twin pregnancies with single fetal malformation is of great significance to improve the outcomes [52].

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Disclosure of conflict of interest

None.

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References

[1] Ferriman E, Stratton S and Stern V. Twin pregnancy. Obstet Gynaecol Reprod Med 2018; 28: 221-228.

[2] Vogel JP, Torloni MR, Seuc A, Betrán AP, Widmer M, Souza JP and Merialdi M. Maternal and perinatal outcomes of twin pregnancy in 23 low- and middle-income countries. PLoS One 2013; 8: e70549.

[3] McLennan AS, Gyamfi-Bannerman C, Ananth CV, Wright JD, Siddiq Z, D'Alton ME and Friedman AM. The role of maternal age in twin pregnancy outcomes. Am J Obstet Gynecol 2017; 217: 80.e1-80.e8.

[4] D'Antonio F, Odibo AO, Prefumo F, Khalil A, Buca D, Flacco ME, Liberati M, Manzoli L and Acharya G. Weight discordance and perinatal mortality in twin pregnancy: systematic review and meta-analysis. Ultrasound Obstet Gynecol 2018; 52: 11-23.

[5] Rizwan N, Abbasi RM and Mughal R. Maternal morbidity and perinatal outcome with twin pregnancy. J Ayub Med Coll Abbottabad 2010; 22: 105-107.

[6] Santana DS, Cecatti JG, Surita FG, Silveira C, Costa ML, Souza JP, Mazhar SB, Jayaratne K, Qureshi Z, Sousa MH and Vogel JP; WHO Multicountry Survey on Maternal and Newborn Health Research Network. Twin pregnancy and severe maternal outcomes: the world health organization multicountry survey on maternal and newborn health. Obstet Gynecol 2016; 127: 631-641.

[7] Santana DS, Silveira C, Costa ML, Souza RT, Surita FG, Souza JP, Mazhar SB, Jayaratne K, Qureshi Z, Sousa MH, Vogel JP and Cecatti JG; WHO Multi-Country Survey on Maternal and Newborn Health Research Network. Perinatal outcomes in twin pregnancies complicated by maternal morbidity: evidence from the WHO multicountry survey on maternal and newborn

health. BMC Pregnancy Childbirth 2018; 18: 449.
[8] Saito K, Ohtsu Y, Amano K and Nishijima M. Perinatal outcome and management of single fetal death in twin pregnancy: a case series and review. J Perinat Med 1999; 27: 473-477.
[9] Zeng Y and Hesketh T. The effects of China's universal two-child policy. Lancet 2016; 388: 1930-1938.
[10] Qiao J and Feng HL. Assisted reproductive technology in China: compliance and non-compliance. Transl Pediatr 2014; 3: 91-97.
[11] Jordanova V, Wickramesinghe C, Gerada C and Prince M. Validation of two survey diagnostic interviews among primary care attendees: a comparison of CIS-R and CIDI with SCAN ICD-10 diagnostic categories. Psychol Med 2004; 34: 1013-1024.
[12] Yu VY. Global, regional and national perinatal and neonatal mortality. J Perinat Med 2003; 31: 376-379.
[13] Melcer Y, Svirsky R, Vaknin Z, Levinsohn-Tavor O, Feldman N and Maymon R. Fetal abnormalities leading to termination of twin pregnancies: the 17-year experience of a single medical center. J Matern Fetal Neonatal Med 2017; 30: 347-351.
[14] Meyer R, Orvieto R, Israel A, Mohr-Sasson A, Timerman Y, Gorodesky T, Toussia-Cohen S, Hendler I, Simchen MJ and Machtinger R. Outcomes of singleton versus twin pregnancies in the fifth and sixth decades. Eur J Obstet Gynecol Reprod Biol 2018; 231: 255-261.
[15] Francisco C, Wright D, Benkő Z, Syngelaki A and Nicolaides KH. Hidden high rate of pre-eclampsia in twin compared with singleton pregnancy. Ultrasound Obstet Gynecol 2017; 50: 88-92.
[16] AboEllail MAM, Kanenishi K, Mori N, Noguchi J, Marumo G and Hata T. Ultrasound study of fetal movements in singleton and twin pregnancies at 12-19 weeks. J Perinat Med 2018; 46: 832-838.
[17] Goswami D, Rani R, Saxena A, Arora MS, Batra S and Sreenivas V. Maternal and neonatal vitamin-D status in twin versus singleton pregnancies. J Obstet Gynaecol Res 2016; 42: 1250-1257.
[18] Gupta S, Fox NS, Feinberg J, Klauser CK and Rebarber A. Outcomes in twin pregnancies reduced to singleton pregnancies compared with ongoing twin pregnancies. Am J Obstet Gynecol 2015; 213: 580, e1-5.
[19] Li H, Zhao YX, Li SR, Xu YH, Huang BY, Cui MH and Zheng GY. Epidemiology of twin and twin with birth defects in China. Natl Med J Chin 2002; 82: 163-166.
[20] Boyle B, McConkey R, Garne E, Loane M, Addor MC, Bakker MK, Boyd PA, Gatt M, Greenlees R, Haeusler M, Klungsøyr K, Latos-Bielenska A, Lelong N, McDonnell R, Métneki J, Mullaney C, Nelen V, O'Mahony M, Pierini A, Rankin J, Rissmann A, Tucker D, Wellesley D and Dolk H. Trends in the prevalence, risk and pregnancy outcome of multiple births with congenital anomaly: a registry-based study in 14 European countries 1984-2007. BJOG 2013; 120: 707-716.
[21] Campaña H, Ermini M, Aiello HA, Krupitzki H, Castilla EE and López-Camelo JS; Latin American Collaborative Study of Congenital Malformations Study Group. Prenatal sonographic detection of birth defects in 18 hospitals from South America. J Ultrasound Med 2010; 29: 203-212.
[22] Lai CQ, Wang CH, Yang Y, Li YZ, Ju YL and Cheng L. Analysis of 70 cases with malformations in the fetuses of twin pregnancy. J Int Obstet Gynecol 2015; 42: 229-232.
[23] Zhong SL, Fang Q, Han ZY, Luo YM, Huang X, Chen JH and Huang AL. Distribution and risk factors of fetal malformation in twin pregnancies. Chin J Prenat Diagn 2012; 4: 6-10.
[24] Best KE and Rankin J. Increased risk of congenital heart disease in twins in the North of England between 1998 and 2010. Heart 2015; 101: 1807-1812.
[25] Panagiotopoulou O, Fouzas S, Sinopidis X, Mantagos SP, Dimitriou G and Karatza AA. Congenital heart disease in twins: the contribution of type of conception and chorionicity. Int J Cardiol 2016; 218: 144-149.
[26] Vaknin Z, Ben-Ami I, Reish O, Herman A and Maymon R. Fetal abnormalities leading to termination of singleton pregnancy: the 7-year experience of a single medical center. Prenat Diagn 2006; 26: 938-943.
[27] Fanos V, Atzori L, Makarenko K, Melis GB and Ferrazzi E. Metabolomics application in maternal-fetal medicine. Biomed Res Int 2013; 2013: 720514.
[28] Jaeggi E, Renaud C, Ryan G and Chaturvedi R. Intrauterine therapy for structural congenital heart disease: contemporary results and Canadian experience. Trends Cardiovasc Med 2016; 26: 639-646.
[29] Isnard M, Kohler A, Kohler M, Vayssière C and Favre R. Successful intrauterine therapy for congenital cystic adenomatoid malformation of the lung. A case report. Fetal Diagn Ther 2007; 22: 325-329.
[30] Wagner N, Kagan KO, Haen S, Schmidt S, Yerlikaya G, Maden Z, Jahn G and Hamprecht K. Effective management and intrauterine treatment of congenital cytomegalovirus infection: review article and case series. J Matern Fetal Neonatal Med 2014; 27: 209-214.
[31] Kuwabara Y, Sawa R, Otsubo Y, Yoneyama Y, Asakura H, Araki T and Takeshita T. Intrauterine therapy for the acutely enlarging fetal cys-

tic hygroma. Fetal Diagn Ther 2004; 19: 191-194.
[32] Sullivan AE, Varner MW, Ball RH, Jackson M and Silver RM. The management of acardiac twins: a conservative approach. Am J Obstet Gynecol 2003; 189: 1310-1313.
[33] Assunção RA, Liao AW, BrizotMde L, Krebs VL and Zugaib M. Perinatal outcome of twin pregnancies delivered in a teaching hospital. Rev Assoc Med Bras (1992) 2010; 56: 447-451.
[34] Baron M, Verspyck E, Diguet A, Eurin D and Marpeau L. Sonographic findings of acardiac twin in the first trimester of pregnancy. J Gynecol Obstet Biol Reprod (Paris) 2000; 29: 684-686.
[35] Machado M, Lima Teixeira E, Ferreira LM, Rodrigues F, Henriques R and Afonso E. Perinatal outcome in relation to chorionicity in twin pregnancy. Acta Med Port 2017; 30: 12-16.
[36] Kaku S, Kimura F and Murakami T. Management of fetal growth arrest in one of dichorionic twins: three cases and a literature review. Obstet Gynecol Int 2015; 2015: 289875.
[37] Moldenhauer JS and Johnson MP. Diagnosis and management of complicated monochorionic twins. Clin Obstet Gynecol 2015; 58: 632-642.
[38] Arınkan SA, Arısoy R and Api M. Assessment of pregnancy outcomes among twin pregnancies with single fetal demise regarding chorionicity and fetal death time. J Turk Ger Gynecol Assoc 2019; 20: 147-153.
[39] D'Antonio F, Thilaganathan B, Dias T and Khalil A; Southwest Thames Obstetric Research Collaborative (STORK). Influence of chorionicity and gestational age at single fetal loss on risk of preterm birth in twin pregnancy: analysis of STORK multiple pregnancy cohort. Ultrasound Obstet Gynecol 2017; 50: 723-727.
[40] Shi X, Li L, Huang X, Chen B, Zhou Y and Fang Q. Fetal aneuploidy: a comparison of dichorionic twins and monochorionic twins. Fetal Diagn Ther 2018; 44: 124-128.
[41] Qin J, Sheng X, Wang H, Liang D, Tan H and Xia J. Assisted reproductive technology and risk of congenital malformations: a meta-analysis based on cohort studies. Arch Gynecol Obstet 2015; 292: 777-798.
[42] Hoorsan H, Mirmiran P, Chaichian S, Moradi Y, Hoorsan R and Jesmi F. Congenital malformations in infants of mothers undergoing assisted reproductive technologies: a systematic review and meta-analysis study. J Prev Med Public Health 2017; 50: 347-360.
[43] Davies MJ, Rumbold AR and Moore VM. Assisted reproductive technologies: a hierarchy of risks for conception, pregnancy outcomes and treatment decisions. J Dev Orig Health Dis 2017; 8: 443-447.
[44] Chen M and Heilbronn LK. The health outcomes of human offspring conceived by assisted reproductive technologies (ART). J Dev Orig Health Dis 2017; 8: 388-402.
[45] Zhao J, Yan Y, Huang X and Li Y. Do the children born after assisted reproductive technology have an increased risk of birth defects? A systematic review and meta-analysis. J Matern Fetal Neonatal Med 2020; 33: 322-333.
[46] Hansen M, Bower C, Milne E, de Klerk N and Kurinczuk JJ. Assisted reproductive technologies and the risk of birth defects--a systematic review. Hum Reprod 2005; 20: 328-338.
[47] Hansen M, Kurinczuk JJ, Milne E, de Klerk N and Bower C. Assisted reproductive technology and birth defects: a systematic review and meta-analysis. Hum Reprod Update 2013; 19: 330-353.
[48] Ludwig M. Is there an increased risk of malformations after assisted reproductive technologies? Reprod Biomed Online 2005; 10: 83-89.
[49] Lu L, Lv B, Huang K, Xue Z, Zhu X and Fan G. Recent advances in preimplantation genetic diagnosis and screening. J Assist Reprod Genet 2016; 33: 1129-1134.
[50] Chen HF, Chen SU, Ma GC, Hsieh ST, Tsai HD, Yang YS and Chen M. Preimplantation genetic diagnosis and screening: current status and future challenges. J Formos Med Assoc 2018; 117: 94-100.
[51] Russo FM, Pozzi E, Pelizzoni F, Todyrenchuk L, Bernasconi DP, Cozzolino S and Vergani P. Stillbirths in singletons, dichorionic and monochorionic twins: a comparison of risks and causes. Eur J Obstet Gynecol Reprod Biol 2013; 170: 131-136.
[52] Van Mieghem T, De Heus R, Lewi L, Klaritsch P, Kollmann M, Baud D, Vial Y, Shah PS, Ranzini AC, Mason L, Raio L, Lachat R, Barrett J, Khorsand V, Windrim R and Ryan G. Prenatal management of monoamniotic twin pregnancies. Obstet Gynecol 2014; 124: 498-506.