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Article

Pregnancy Loss After Amniocentesis with Double-Needle Insertions in Twin Pregnancies

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Abstract

The aim of this study was to determine the pregnancy loss rate of amniocentesis with double-needle insertions in twin pregnancies. This was a retrospective study of twin pregnancies who underwent amniocentesis with double-needle insertion between 2010 and 2019 at a single center. The pregnancy loss rates were recorded as single or double fetal loss before 24 weeks' gestation and within 4 weeks after the procedure. Risk factors for pregnancy loss after amniocentesis were also assessed. A total of 678 twin pregnancies with amniocentesis were finally included. The pregnancy loss rates before 24 weeks' gestation and within 4 weeks after the procedure were 0.9% and 1.9%, respectively. Only one fetal loss was presumed to be a direct result of the procedure. All other cases were complicated by structural or chromosomal anomalies. Twin pregnancies with abnormal ultrasound findings had a significantly higher rate of pregnancy loss with a relative risk of 4.81 (95% CI [1.03, 22.2]). Our study showed a low pregnancy loss rate after amniocentesis in twin pregnancies with double-needle insertions technique of sampling, which can help decision making in prenatal screening and diagnosis for twin pregnancies.

Keywords: Amniocentesis; twin pregnancy; pregnancy loss

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Over the past decades, the incidence of twin pregnancies has increased mainly due to advanced maternal age and the broader use of assisted reproductive technologies (ART; Martin et al., 2012). As twins face higher risks of fetal chromosomal and structural anomalies than singletons (Chauhan et al., 2010), prenatal screening and invasive diagnostic testing are essential for twin pregnancies. However, it is hard for patients to choose from prenatal screening or diagnostic testing. Prenatal screening has the advantage of being a noninvasive approach, but the accuracy is limited in twin gestations. One meta-analysis concluded that the performance of cell-free DNA (cfDNA) testing for trisomy 21 in twin pregnancies was comparable to that in singleton pregnancies, whereas the number of cases of trisomies 18 and 13 was too small for estimation (Gil et al., 2019). The failure rate for cfDNA testing in twins was higher than that reported in singletons (Bevilacqua et al., 2015; Sarno et al., 2016). On the other hand, invasive diagnostic testing remains the standard for determining fetal genetic outcomes, but procedure-related complications, especially the risk of pregnancy loss, are of keen concern.

Amniocentesis is the most common diagnostic procedure that implements percutaneous transabdominal puncture of the uterus to obtain amniotic fluid. In the Cochrane database, the spontaneous miscarriage rate after amniocentesis in singleton gestations spans between 0.13% and 0.33%, while there are no data for twin gestations (Alfirevic et al., 2017). The risk of pregnancy loss in twin gestations reported in the last 20 years varies from 0% to 4.2% (Antsaklis et al., 2002; Cahill et al., 2009; Chen et al., 2020; Daskalakis et al., 2009; Dechnunthapiphat et al., 2020; Kalogiannidis et al., 2011; Kan et al., 2012; Kim et al., 2019; Krispin et al., 2019; Lenis-Cordoba et al., 2013; Millaire et al., 2006; Simonazzi et al., 2010; Sperling et al., 2019; Supadilokluck et al., 2009; Toth-Pal et al., 2004; Yukobowich et al., 2001). One recent systematic review demonstrated that the pooled pregnancy loss rate before 24 weeks and within 4 weeks after the amniocentesis procedure in twin pregnancies were 2.1% and 2.1%, respectively, which were as high as the background risk in twin pregnancy without undergoing the procedure (Di Mascio et al., 2020).

However, there is still no consensus on the definition of pregnancy loss in published studies. It varies in the mechanism of loss (fetal death or miscarriage) and the number of fetuses involved (one or both). The intervals of pregnancy loss are recorded as loss before 24–28 weeks' gestation, within 2–4 weeks after the procedure and overall loss rate at any gestational age. Some studies excluded fetuses with structural and chromosomal anomalies and defined outcomes as procedure-related pregnancy loss, while others included abnormal fetuses and estimated the pregnancy loss rate. In addition, some studies spanned a long time period and thus the amniocentesis technique was not conserved well. Sample sizes are also considered a contributing factor to this issue, ranging from 87 to 476, and are relatively small regarding the low chance of

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pregnancy loss. All those limitations make it hard for individual counseling.

This study aimed to assess the outcomes of twin pregnancies where amniocentesis was performed following the standard protocol in a tertiary referral center to give a more reliable pregnancy and procedure-related loss rate to help prenatal counseling.

Materials and Methods

This retrospective study was carried out at the Department of Fetal Medicine, Shanghai 1st Maternity and Infant Hospital of Tongji between December 2010 and December 2019. The study included patients with twin pregnancies who underwent amniocentesis between 15 and 24 weeks' gestation and had two live fetuses at the time of the procedure. Exclusions were monoamniotic twin pregnancies, intrauterine procedures before amniocentesis (e.g. selective fetal reduction or CVS, amniocentesis in other hospitals) and puncture made only in a single sac.

In the current study, the indications for amniocentesis were advanced maternal age (≥35 years), abnormal ultrasound findings (including thickening of nuchal translucency [NT] ≥3.0 mm, soft markers, structural anomaly or fetal growth restriction), suspected TORCH infection, chromosomal aberration in parents or abnormal pregnancy history with congenital anomalies, genetic disease or recurrent miscarriage. All pregnant women with indications were referred to the twins' clinic in the fetal medicine department and followed by a standard protocol. Serum analytes were assayed, including C reaction proteins, blood routine examinations and coagulation indexes. Patients also received detailed ultrasound assessments, including fetal biometry, sex, anatomy, position, placental location and cord insertion to help distinguish two fetuses. Cervical length was also measured before amniocentesis. Patients whose cervical lengths were below 25 mm were counseled with a higher risk of pregnancy loss (Conde-Agudelo et al., 2010; Conde-Agudelo & Romero, 2014). Genetic counseling was then offered regarding the process and possible risks of amniocentesis for twins, and the limitations and benefits of the diagnostic genetic testing. Patients who decided to perform amniocentesis were required to sign an informed consent sheet.

All amniocentesis procedures were performed between 15 and 24 weeks of gestation by three well-trained maternal–fetal medicine specialists at our center. The procedure was guided by continuous ultrasound and performed double insertion techniques using two disposable 22-gauge needles. Each amniotic sac was punctured separately regardless of chorionicity. The first 1–2 mL of amniotic fluid aspirated was discarded to avoid maternal cell contamination. The final amount of aspirated fluid was determined by the need for genetic tests. At our center, dyes were not used to confirm that separate sacs were sampled. After the procedure, fetal heart rates were assessed sonographically. Patients were asked to rest for 30 min to observe whether abdominal pain, severe uterine cramping, vaginal bleeding or fluid occurred.

The following baseline characteristics were obtained from the medical records: maternal age, body mass index (BMI), gravidity, parity, conception mode, chorionicity, cervical length and indications for amniocentesis. Pregnancy loss was defined as the loss of one or two fetuses prior to viability, and thus we only included amniocentesis cases performed until 24 weeks gestation. Pregnancy loss rates within 1, 2 or 4 weeks after amniocentesis were assessed. Cases that underwent selective feticide or termination of pregnancy within 1, 2 or 4 weeks after amniocentesis were excluded, respectively, when calculating. Pregnancy loss within 4 weeks after amniocentesis was stratified by advanced maternal age, conception mode, BMI, parity, chorionicity, placental position, cervical length and ultrasound results. We also estimated pregnancy loss rate before 24 weeks' gestation that underwent amniocentesis before 22 weeks' gestation as most published studies did (Antsaklis, 2002; Cahill et al., 2009; Daskalakis et al., 2009; Dechnunthapiphat et al., 2020; Kalogiannidis et al., 2011; Kan et al., 2012; Kim et al., 2019; Lenis-Cordoba et al., 2013; Millaire et al., 2006; Simonazzi et al., 2010; Toth-Pal et al., 2004) to help compare results. Follow-ups of pregnancy outcomes were informed by electronic medical records or by phone calls.

Statistical analysis was performed using SPSS Statistics version 23.0 (IBM SPSS Statistics for Windows, IBM Corp., Armonk, NY, USA). Continuous variables were compared using the t test, and categorized variables were compared using chi-square or Fisher's exact test, as appropriate. Two-sided p values $\leq .05$ were considered statistically significant. This retrospective database study was considered exempt from ethical approval.

Results

Out of 838 twin pregnancies that underwent amniocentesis through double needle technique, 678 were performed between 15 and 24 weeks' gestation. Complete follow-ups were achieved in 575/678 (84.8%) of patients, as presented in Figure 1. Baseline characteristics of our population are shown in Table 1. The average age of patients was 33.65 ± 4.50 years, and the proportion of advanced maternal age (≥35 years) was 40.8%; 495 (73.0%) patients were nulliparous and 388 (60.7%) were conceived after ART. Regarding chorionicity, 527 (77.7%) were dichorionic diamniotic (DCDA) and 151 (22.3%) were monochorionic diamnioti (MCDA). All patients underwent cervical length measurement just before the procedure, while 22 (1.9%) cases were not recorded. The mean cervical length was 38.19 ± 6.15 mm, ranging from 23 to 64 mm. The gestational age at procedure was 19.99 ± 1.69 weeks of gestation. Indications for performing the procedure are also illustrated in Table 1. Abnormal ultrasound findings were the most common indication.

After amniocentesis, 10 cases experienced intrauterine fetal death (IUFD) of one fetus within 4 weeks after the procedure, of which 7 cases were DCDA, including 3 cases complicated with severe FGR, 2 cases with structural anomalies, and 1 case with fetal genetic result of 21 trisomy. One case had preterm premature rupture of the membrane (PPROM) and subsequent IUFD of one fetus 3 days after amniocentesis at 19 weeks' gestation, and the remaining twin was delivered at 37 weeks' gestation. The patient's cervical length before the procedure was 30 mm. Among the other three MCDA cases, two were complicated with selective intrauterine growth restriction and one was complicated with structural anomalies. As illustrated in Table 2, the pregnancy loss rate was 0.5%, 0.7% and 1.9% within 1, 2 and 4 weeks after the procedure, respectively. The pregnancy loss before 24 weeks' gestation was 0.9%. Only one fetal loss case was regarded as merely related to the procedure due to PPROM.

When risk factors of pregnancy loss rate within 4 weeks after the procedure were investigated, twin pregnancies with abnormal ultrasound findings had a significantly higher rate of pregnancy loss with a relative risk of 4.81 (95% CI [1.03, 22.2], $p = .049$), whereas other risk factors, including maternal age, conception mode, chorionicity, placental position and cervical length, were not associated with the rate of pregnancy loss (see Table 3).

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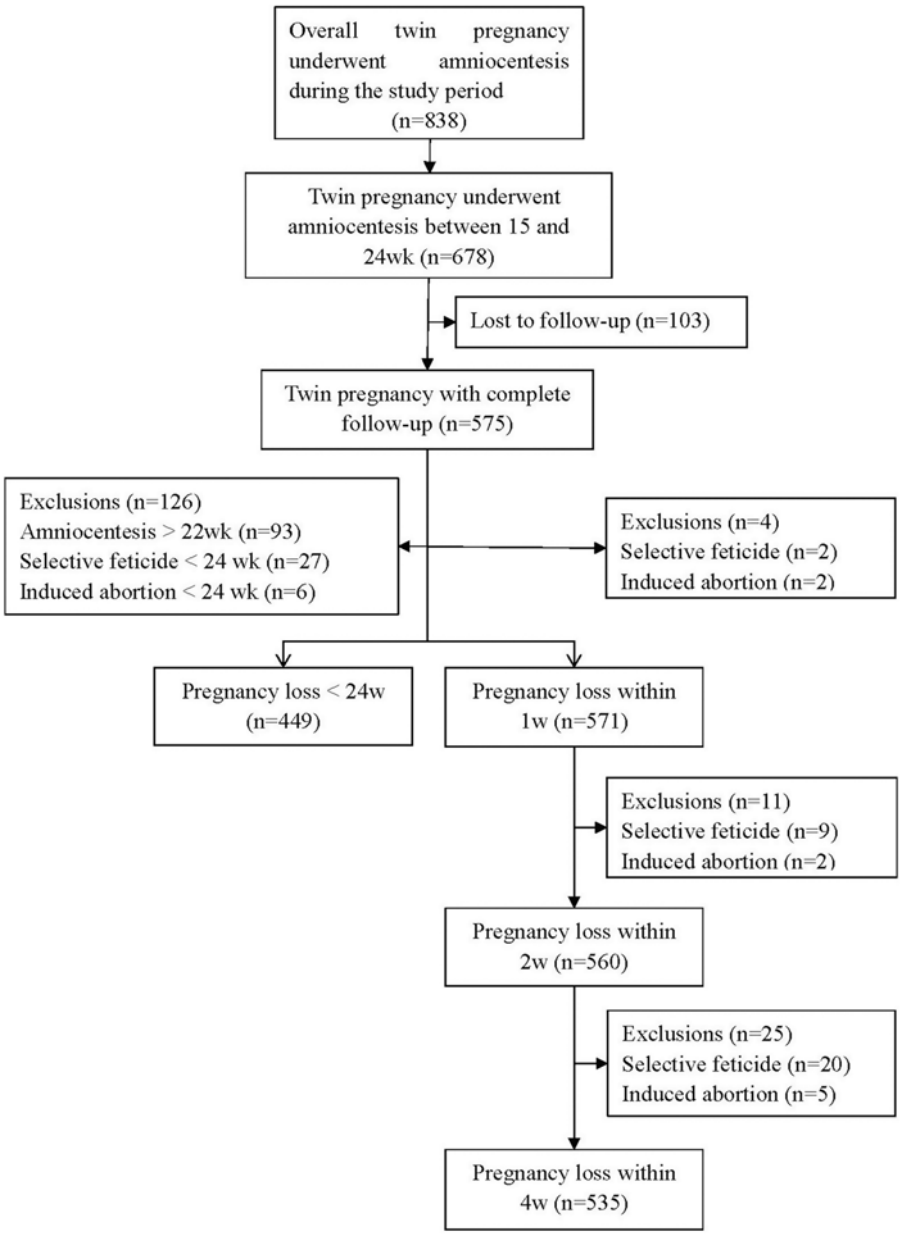

Fig. 1. Flow chart of subject eligibility.

During the internal study period from 2014 to 2017, there were 1065 twin pregnancies without invasive testing and intrauterine interventions at our center. Twenty-three patients had miscarriage of both twins, and two patients had one fetus intrauterine death between 15 and 24 weeks of gestation. The background pregnancy loss rate (one or both fetuses) was 2.3% (25/1065).

Discussion

In the era of noninvasive prenatal testing, invasive diagnosis techniques have the advantage of obtaining precise genetic information of each fetus through karyotyping, chromosomal microarray analysis, or even whole exome sequencing. Since the fetal risk for genetic abnormalities must be carefully weighed against the risk of pregnancy loss associated with amniocentesis, it is essential to counsel pregnant women using procedure-related risks based on data from local centers. Our study reviewed data in the past 10 years, and the results suggested that the pregnancy loss rates within 4 weeks after amniocentesis and before 24 weeks' gestation in twin pregnancies were 1.9% and 0.9%, respectively. The pregnancy loss rate only related to the procedure was 0.2%.

Our results are in accordance with or even lower than those reported in recent studies. In the recent meta-analysis, the pooled rates of fetal loss in twin pregnancies after amniocentesis within 4 weeks and before 24 weeks' gestation were 45/1932 (2.1%, 95% CI [1.5, 2.9]) and 59/2439 (2.1%, 95% CI [1.4, 2.9]), respectively (Di Mascio et al., 2020). A more recent study of twin pregnancies not in the meta-analysis included 332 twin pregnancies undergoing amniocentesis, and the pregnancy loss rate before 24 weeks was 3.0% (Dechnunthapiphat et al., 2020). Chen et al. (2020) reported that the pregnancy loss rate before 28 weeks in 418 twin pregnancies was 1.91%. The pregnancy loss rate only related to the procedure in our study was the lowest among all the studies,

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Table 1. Baseline characteristics of women with twin pregnancies who underwent amniocentesis

Variables	$N = 678$
Age, years (advanced age, %)	33.65 ± 4.50(40.8%)
Body mass index, kg/m^2 (>24, %)	21.88 ± 3.00 (21.4%)
Gravidity	2.17 ± 1.34
Parity (Nulliparity%)	0.29 ± 0.50 (73.0%)
Conception mode,	
Spontaneous *n* (%)	290 (42.8%)
IVF *n* (%)	241 (35.5%)
ICSI *n* (%)	139 (20.5%)
PGT *n* (%)	8 (1.2%)
Chorionicity,	
Dichorionic *n* (%)	527 (77.7%)
Monochorionic *n* (%)	151 (22.3%)
Placental position,	
Anterior *n* (%)	645 (47.6%)
Posterior *n* (%)	671 (49.5%)
Lateral *n* (%)	32 (2.3%)
Fundal *n* (%)	8 (0.6%)
Cervical length, mm	38.19 ± 6.15 (23–64)[‡]
Gestational age at procedure, weeks	19.99 ± 1.69
Indications	
Abnormal noninvasive prenatal test	44 (6.5%)
Abnormal ultrasound scan	330 (48.7%)
Chromosomal aberration in the family	31(4.6%)
Previous history[†]	42 (6.2%)
Advanced maternal age without other indications	164 (24.2%)
ICSI or PGT without other indications	28 (4.1%)
Maternal request	39 (5.7%)

Note: [†]Previous child or fetal with chromosomal or congenital anomaly or RSA; [‡]22 missing data. ICSI, intracytoplasmic sperm injection; PGT, preimplantation genetic testing.

Table 2. Procedure-Related pregnancy loss of amniocentesis

Time of pregnancy loss	Amniocentesis pregnancy loss[†] *n/N* (%)
Within 1 week after amniocentesis	3/571 (0.5%)
Within 2 weeks after amniocentesis	4/560 (0.7%)
Within 4 weeks after amniocentesis	10/535 (1.9%)
< 24 weeks[‡] ($n = 482$)	4/449 (0.9%)

Note: [†]Pregnancy loss, defined as the loss of one or two fetuses; [‡]<24 weeks, only included women who underwent amniocentesis before 22 weeks' gestation.

Table 3. Relative risk of pregnancy loss rates (within 4 weeks after amniocentesis) based on univariate analysis

Variables	Pregnancy loss rate	RR (95% CI)	*p* value
Overall	10/535 (1.9%)	–	–
AMA[†]	4/222	0.94 (0.27, 3.29)	1.000
Non-AMA	6/313		
ART	7/323	1.53 (0.40, 5.85)	.747
Spontaneous conception	3/212		
BMI > 24	2/118	0.88 (0.19, 4.10)	1.000
BMI ≤ 24	8/417		
$p > 0$	3/139	1.22 (0.32, 4.65)	.725
$p = 0$	7/396		
Monochorionic	3/105	1.76 (0.46, 6.67)	.404
Dichorionic	7/430		
Anterior placenta	4/515	0.72 (0.20, 2.531)	.745
Placenta in other position	6/555		
Cervical length ≤ 30 mm[‡]	2/49	2.42 (0.53, 11.11)	.238
Cervical length > 30 mm	8/475		
Abnormal ultrasound	8/243	4.81 (1.03, 22.2)	.049
Normal ultrasound	2/292		

Note: [†]AMA, advanced maternal age; [‡]11 missing data; RR, relative risk.

mainly due to sticking to the standard protocol that helped exclude potential infection and cervical incompetence.

Only a few studies introduced comparison groups of twin pregnancies without undergoing amniocentesis, and usually the maternal baseline of the control group did not match that of the amniocentesis group. Sperling et al. (2019) reported a multicenter cohort including 861 women with twin pregnancies screened positive. They were all offered amniocentesis, while 274 (31.8%) accepted and the rest declined. The rates in the accepted and declined group were 8.8% versus 6.8% with adjusted *OR* 1.32 (0.66−1.91), suggesting that amniocentesis did not appear to increase the risk of pregnancy loss further. Moreover, the recent meta-analysis reported that there was no significant difference between twin pregnancies who underwent amniocentesis and those who did not when focusing on either fetal loss before 24 weeks of gestation (*OR* 1.59, $p = .06$) or fetal loss within 4 weeks from the procedure (OR 1.38, $p = .3$), indicating that the risk of fetal loss following amniocentesis is lower than that previously reported (Di Mascio et al., 2020). Our study results were lower than the background pregnancy loss rate of 2.3% in the same center and that were published in other literature (range 0.6%−2.8%), suggesting that amniocentesis did not add to increased risk of pregnancy loss.

We also explored some potential risk factors, including maternal (advanced maternal age, ART conception mode, hither BMI, nulliparity, shorter cervical length) and fetal characteristics (monochorionicity, placental position and abnormal ultrasound findings) that might account for an increased risk of fetal loss in twin pregnancies. Only abnormal ultrasound findings were associated with increased risk, similar to that reported by Cahill et al. (2009). Likewise, some other studies also reported that advanced maternal age, parity, conception mode and chorionicity did not influence pregnancy loss after amniocentesis (Cahill et al., 2009; Chen et al., 2020; Daskalakis et al., 2009; Dechnunthapiphat et al., 2020; Lenis-Cordoba et al., 2013). To the authors' knowledge, there are no earlier reports on cervical length as a risk factor in twin pregnancies after amniocentesis. However, in our cohort, only two

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patients' cervical lengths were shorter than 25 mm, and these two patients did not have pregnancy loss after amniocentesis. Thus, more data are needed to answer whether amniocentesis will increase the risk of pregnancy loss in patients with shorter cervical lengths.

One strength of our study is a reliable estimation of the pregnancy loss rate after amniocentesis based on the largest sample size to date. Additionally, all procedures were mainly performed by two well-trained experts of double-needle insertions who followed the standard protocol. The comprehensive maternal baseline and the same performers' skills through the study period enhanced the reliability of our estimation of pregnancy loss rate and the speculation of potential risk factors. Finally, our study carefully assessed the reasons for pregnancy loss and stratified the risks according to potential risk factors, which helped individual counseling.

There are some limitations in our study. The pregnancy loss rate might be underestimated by the incomplete follow-up (Halliday et al., 1992). Second, our study lacked a well-designed and baseline-matched control group. Despite our large sample size, the relatively small number of final events limited the precision of stratified risk estimates.

In conclusion, our study showed a low pregnancy loss rate after amniocentesis in twin pregnancies, comparable with results in other international counterparts. The double-needle insertion technique of sampling in twins is a relatively safe procedure for prenatal diagnosis.

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Conflict of Interest. None.

References

Alfirevic, Z., Navaratnam, K., & Mujezinovic, F. (2017). Amniocentesis and chorionic villus sampling for prenatal diagnosis. *Cochrane Database of Systematic Reviews*, *9*, CD003252.

Antsaklis, A., Souka, A.P., Daskalakis, G., Kavalakis, Y., & Michalas, S. (2002). Second-trimester amniocentesis vs. chorionic villus sampling for prenatal diagnosis in multiple gestations. *Ultrasound in Obstetrics & Gynecology*, *20*, 476–481.

Bevilacqua, E., Gil, M. M., Nicolaides, K. H., Ordonez, E., Cirigliano, V., Dierickx, H., Willems, P. J., & Jani, J. C. (2015). Performance of screening for aneuploidies by cell-free DNA analysis of maternal blood in twin pregnancies. *Ultrasound in Obstetrics & Gynecology*, *45*, 61–66.

Cahill, A. G., Macones, G. A., Stamilio, D. M., Dicke, J. M., Crane, J. P., & Odibo, A. O. (2009). Pregnancy loss rate after mid-trimester amniocentesis in twin pregnancies. *American Journal of Obstetrics and Gynecology*, *200*, 257, e251–256.

Chauhan, S. P., Scardo, J. A., Hayes, E., Abuhamad, A. Z., & Berghella, V. (2010). Twins: prevalence, problems, and preterm births. *American Journal of Obstetrics and Gynecology*, *203*, 305–315.

Chen, J., Liu, L., Xia, D., He, F., Wang, Q., Li, T., Lai, Y., Liu, S., & Zhang, Z. (2020). Comparison of spontaneous fetal loss rates between women with singleton and twin pregnancies after mid-trimester amniocentesis ¾ a historical cohort study. *Prenatal Diagnosis*, *40*, 1315–1320.

Conde-Agudelo, A., Romero, R., Hassan, S. S., & Yeo, L. (2010). Transvaginal sonographic cervical length for the prediction of spontaneous preterm birth in twin pregnancies: A systematic review and meta-analysis. *American Journal of Obstetrics and Gynecology*, *203*, 128.e1–128.e12.

Conde-Agudelo, A., & Romero, R. (2014). Prediction of preterm birth in twin gestations using biophysical and biochemical tests. *American Journal of Obstetrics and Gynecology*, *211*, 583–595.

Daskalakis, G., Anastasakis, E., Papantoniou, N., Mesogitis, S., & Antsaklis, A. (2009). Second trimester amniocentesis in assisted conception versus spontaneously conceived twins. *Fertility and Sterility*, *91*, 2572–2577.

Dechnunthapiphat, R., Sekararithi, R., Tongsong, T., Wanapirak, C., Piyamongkol, W., Sirichotiyakul, S., Tongprasert, F., Srisupundit, K., Luewan, S., Jatavan, P., & Traisrisilp, K. (2020). Comparisons of pregnancy outcomes between twin pregnancies with and without second-trimester amniocentesis. *Prenatal Diagnosis*, *40*, 1330–1337.

Di Mascio, D., Khalil, A., Rizzo, G., Buca, D., Liberati, M., Martellucci, C. A., Flacco, M. E., Manzoli, L., & D'Antonio, F. (2020). Risk of fetal loss following amniocentesis or chorionic villus sampling in twin pregnancies: A systematic review and meta-analysis. *Ultrasound in Obstetrics & Gynecology*, *56*, 647–655.

Gil, M. M., Galeva, S., Jani, J., Konstantinidou, L., Akolekar, R., Plana, M. N., & Nicolaides, K. H. (2019). Screening for trisomies by cfDNA testing of maternal blood in twin pregnancy: Update of the Fetal Medicine Foundation results and meta-analysis. *Ultrasound in Obstetrics & Gynecology*, *53*, 734–742.

Halliday, J. L., Sheffield, L. J., Lumley, J., Robinson, H. P., Renou, P., & Carlin, J. B. (1992). Importance of complete follow-up of spontaneous fetal loss after amniocentesis and chorion villus sampling. *The Lancet*, *340*, 886–890.

Kalogiannidis, I., Petousis, S., Prapa, S., Dagklis, T., Karkanaki, A., Prapas, Y., & Prapas, N. (2011). Amniocentesis-related adverse outcomes in diamniotic twins: Is there a difference compared to singleton pregnancies? *European Journal of Obstetrics & Gynecology and Reproductive Biology*, *155*, 23–26.

Kan, A. S., Lee, C. P., Leung, K. Y., Chan, B. C., Tang, M. H., & Chan, V. H. (2012). Outcome of twin pregnancies after amniocentesis. *Journal of Obstetrics and Gynaecology Research*, *38*, 376–382.

Kim, M. S., Moon, M. J., Kang, S., Jung, S. H., Chang, S. W., Ki, H. J., Kim B., & Ahn, E. (2019). Obstetrical outcomes of amniocentesis or chorionic villus sampling in dichorionic twin pregnancies. *Journal of Korean Medical Science*, *34*, e142.

Krispin, E., Wertheimer, A., Trigerman, S., Ben-Haroush, A., Meizner, I., Wiznitzer, A., & Bardin, R. (2019). Single or double needle insertion in twin's amniocentesis: Does the technique influence the risk of complications? *European Journal of Obstetrics & Gynecology and Reproductive Biology: X*, *3*, 100051

Lenis-Cordoba, N., Sanchez, M. A., Bello-Munoz, J. C., Sagala-Martinez, J., Campos, N., Carreras-Moratonas, E., & Cabero-Roura, L. (2013). Amniocentesis and the risk of second trimester fetal loss in twin pregnancies: results from a prospective observational study. *Journal of Maternal-Fetal and Neonatal Medicine*, *26*, 1537–1541.

Martin, J. A., Hamilton, B. E., & Osterman, M. J. (2012). Three decades of twin births in the United States, 1980–2009. *NCHS Data Brief*, *80*, 1–8.

Millaire, M., Bujold, E., Morency, A. M., & Gauthier, R. J. (2006). Mid-trimester genetic amniocentesis in twin pregnancy and the risk of fetal loss. *Journal of Obstetrics and Gynaecology Canada*, *28*, 512–518.

Sarno, L., Revello, R., Hanson, E., Akolekar, R., & Nicolaides, K. H. (2016). Prospective first-trimester screening for trisomies by cell-free DNA testing of maternal blood in twin pregnancy. *Ultrasound in Obstetrics & Gynecology*, *47*, 705–711.

Simonazzi, G., Curti, A., Farina, A., Pilu, G., Bovicelli, L., & Rizzo, N. (2010). Amniocentesis and chorionic villus sampling in twin gestations: Which is the best sampling technique? *American Journal of Obstetrics and Gynecology*, *202*, 365.e361–365.

Sperling, J. D., Zlatnik, M. G., Norton, M. E., & Currier, R. J. (2019). Pregnancy loss after amniocentesis in monochorionic and dichorionic twin

https://doi.org/10.1017/thg.2022.1 Published online by Cambridge University Press

pregnancies: results from a large population-based dataset. *Prenatal Diagnosis*, *39*, 896–900.

Supadilokluck, S., Tongprasert, F., Tongsong, T., Wanapirak, C., Piyamongkol, W., Sirichotiyakul, S., & Srisupundit, K. (2009). Amniocentesis in twin pregnancies. *Archives of Gynecology and Obstetrics*, *280*, 207–209.

Toth-Pal, E., Papp, C., Beke, A., Ban, Z., & Papp, Z. (2004). Genetic amniocentesis in multiple pregnancy. *Fetal Diagnosis and Therapy*, *19*, 138–144.

Yukobowich, E., Anteby, E. Y., Cohen, S. M., Lavy, Y., Granat, M., & Yagel, S. (2001). Risk of fetal loss in twin pregnancies undergoing second trimester amniocentesis. *Obstetrics & Gynecology*, *98*, 876–877.

https://doi.org/10.1017/thg.2022.1 Published online by Cambridge University Press