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POPULATION STUDY ARTICLE

Early structural cardiovascular changes on neonatal echocardiography after adverse intrauterine circumstances in identical twins

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BACKGROUND: Studies on cardiovascular changes after fetal growth restriction (FGR) are limited by their design in which growth-restricted neonates are compared to appropriately-grown neonates. We aim to investigate early structural cardiovascular remodeling after FGR in identical twins, controlling for confounding of genetic and maternal factors.
METHODS: This study is part of a prospective cohort study including monochorionic twins from January 2019. Transthoracic echocardiography was performed within one week after birth. Z-scores for cardiac valve annuli diameters and left ventricle dimensions based on gestational age at birth were compared between smaller and larger twins. Z-score differences between birth weight and cardiac structure per twin were tested against the intercept.
RESULTS: Median gestational age at birth of the 100 included twin pairs was 33.8 (interquartile range (IQR) 30.8–36.1) weeks, with birth weights of 1729 (IQR 1200–2115) grams for smaller twins and 2058 (IQR 1643–2500) grams for larger twins. Smaller twins had a lower z-score for all structures. Z-score differences in birth weight and cardiac structure were higher than the intercept.
CONCLUSION: While cardiac structures are generally smaller for the twin with the lower birth weight, the deviation in birth weight tends to be more pronounced than the deviation in cardiac structure.

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IMPACT:

- Our study shows that in identical twins, the smaller twin at birth has a structurally smaller heart on neonatal echocardiography when compared to the larger twin. Yet, cardiac structures per individual twin are less affected than body size for their given gestational age at birth.
- We have used a unique natural experiment by studying a population of identical twins with varying degrees of birth weight discordance, eliminating any confounding of genetic, maternal and obstetrical factors.
- Our results are suggestive of early cardiovascular remodeling after adverse intrauterine circumstances. This provides insight into the fetal programming of cardiovascular disease.

INTRODUCTION

Adverse intrauterine circumstances are thought to negatively impact lifelong health. Previous research has demonstrated a strong association between fetal growth restriction (FGR), defined as an estimated fetal weight <10th centile, and an increased risk of cardiovascular disease (CVD) in adulthood.[1,2] The mechanism behind this association is thought to be fetal programming: structural and functional adaptations in fetal development in response to a suboptimal environment that are persistent throughout life.[3–5]

Cardiovascular remodeling can already be observed in fetuses with FGR.[3] It is thought that 'heart sparing' occurs, i.e. blood flow redistribution favoring major organs in a prolonged state of hypoxia.[6] Despite its positive connotation, the subtle changes invoked by heart sparing are considered disadvantageous and may persist after birth. Echocardiographic studies in children born after FGR have reported smaller cardiovascular dimensions and mass as well as greater arterial wall stiffness in comparison to appropriately-grown controls from early childhood until preadolescence.[7–9] Yet, neonatal cardiovascular changes after FGR are

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scarcely reported, though these form the basis for tracking the lifelong cardiovascular consequences of FGR.[10–13] The available studies are largely focused on cardiac function and limited by their study design in which small for gestational age (SGA) neonates are compared to appropriately-grown neonates, inherently subject to confounding of maternal (nutritional status, comorbidity, socio-economic status), obstetric (blood pressure, other complications) or genetic factors.[12,14] A population of identical twins offers a solution.

Monochorionic (MC) twins are genetically identical and share a single placenta during pregnancy. This shared placenta can give rise to a range of complications that may affect fetal hemodynamics. Twin-twin transfusion syndrome (TTTS) and twin anemia polycythemia sequence (TAPS) both result from unequal intertwin transfusion, the former in amniotic fluid and the latter in red blood cells. Both complications can also have a discordant antenatal growth pattern following from an unequally shared placenta.[15] When there is a discordant antenatal growth pattern resulting in a birth weight discordance (BWD) ≥ 20%, in the absence of TTTS or TAPS, it is described as selective fetal growth restriction (sFGR). Within-pair comparisons in a population of MC twins thereby control for any potential confounding of genetic and maternal factors. By including a range of complications, we can both enlarge the study population, as isolated selective fetal growth restriction sFGR is a rare complication occurring in only 10–15% of these twin pregnancies, and study within-pair differences in birth weight on a continuous scale.[16] Therefore, this study aims to investigate the effect of prenatal growth on the relative size of cardiac structures, by comparing structural cardiac measurements and aortic pulse-wave velocity (aPWV), a measure for aortic stiffness previously linked to an increased risk of CVD, between smaller and larger twins at birth and to assess whether heart sparing is present using neonatal echocardiography.

METHODS

This study is part of the Twinlife study (Twin Longitudinal Investigation of FEtal discordance; International Clinical Trials Registry Platform ID NL7538), a prospective, longitudinal cohort study including all live-born MC twins born in the Leiden University Medical Center (LUMC) from January 2019 onwards, regardless of gestational age at birth.[17] All MC twin complications, both mono- and diamniotic were eligible for inclusion. This study was approved by the ethics committee of the LUMC (P18.184), and inclusion is ongoing. All parents are asked for informed consent. Exclusion criteria are triplet pregnancies, perinatal mortality, cases with twin reversed arterial perfusion or congenital abnormalities, including congenital heart defects. The sample size was previously determined based on the detection of a difference in within-pair DNA methylation derived from the previous Dutch Hunger Winter studies, as is one of the primary objectives of the Twinlife study.[17]

For each twin pair, the twin with the lowest birth weight was marked as the 'smaller twin' and the twin with the highest birth weight as the 'larger twin'. Baseline characteristics were collected: maternal age, gravidity, parity, type of complication (monoamnionicity, TTTS including Quintero stage,[18] TAPS[19] and sFGR including Gratacós type,[20] defined as a BWD ≥ 20% calculated as (birth weight larger twin – birth weight smaller twin)/birth weight larger twin ×100[21] in the absence of TTTS/TAPS), fetoscopic laser coagulation or other treatment modalities for TTTS/TAPS, gestational age at diagnosis of complication, time between diagnosis and birth, gestational age at birth, sex, delivery mode, birth weight with z-score (singleton curves based on gestational age and sex), proportion of neonates born SGA (birth weight <10th centile[22]), the presence of a patent ductus arteriosus (PDA) requiring either medical/surgical treatment and the presence of persistent pulmonary hypertension in the neonate (PPHN) requiring nitric oxide treatment. In our center it is standard practice to perform fetoscopic laser surgery for TTTS/TAPS up to 28 weeks of gestational age.[23–25] The postnatal definition for sFGR based on BWD was chosen rather than the antenatal diagnostic criteria based on estimated fetal weight, as birth weight is ultimately considered the true end-point of prenatal growth and estimated fetal weight has a margin of error.[26]

Transthoracic echocardiography was performed in neonates at a time of hemodynamic stability by a senior pediatric cardiologist (AAWR/ADJH, unblinded for size) within one week after birth according to the guidelines of the American Society for Echocardiography (ASE).[27,28] Respiratory support at time of ultrasound was documented, including mechanical ventilation, non-invasive support or no support. Mitral valve (MV) annulus diameter and tricuspid valve (TV) annulus diameter were measured in an apical four-chamber view. Aortic valve (AV) annulus diameter was measured in the parasternal long-axis view and pulmonary valve (PV) annulus diameter in the parasternal short-axis view. The following parameters were measured in M-mode: left atrium diameter (LA), aortic root diameter (Ao), LA/Ao ratio, left ventricular internal diameter in diastole (LIVDD) and systole (LVIDS), left ventricular posterior wall thickness in diastole (LVPWD) and systole (LVPWS), LV mass,[29] ejection fraction, fractional shortening calculated as (LVIDD – LVIDS)/LVIDD x 100, and relative wall thickness (RWT) calculated as (2 x LVPWD)/LVIDD).[28–30] All measurements were performed in accordance with ASE guidelines by a single investigator (SGG, unblinded), thereby excluding interobserver variability, under supervision of a senior pediatric cardiologist (AAWR, unblinded). Z-scores were calculated based on gestational age at birth, using the best available data set.[31,32] Z-scores per gestational age window or MC twin complication are currently unavailable.

To determine aPWV as a secondary outcome, a measure of aortic stiffness that was previously associated with adult cardiovascular disease, two pulsed Doppler recordings of one two-dimensional cross-section of the aorta were made at the level of the AV and the descending aorta as distal as possible (Fig. 1).[33–35] All recordings were obtained by a single senior pediatric cardiologist (AAWR). A clearly discernible identical time point in the electrocardiogram was chosen for both recordings and the time until onset of flow was measured three times in both recordings to calculate an average. The transit time was calculated as: average time of onset of flow at descending aorta – average time of onset of flow at AV. The length of the aorta from the location of the first Doppler recording to the other was determined using Osirix DICOM viewer.[36] aPWV was calculated as: (aortic length)/(100 × transit time).[35] Normative data for aPWV in neonates are currently unavailable.

The difference between the z-score of birth weight and the z-score of each cardiac structure was calculated per twin to assess heart sparing. A value > 0 indicates that the deviation of the cardiac structure is less than the deviation of birth weight in relation to gestational age at birth, i.e. the cardiac structure is less affected than birth weight (heart sparing). A value < 0 indicates that the deviation of the cardiac structure is more than the deviation of birth weight in relation to gestational age at birth, i.e. the cardiac structure is more affected than birth weight. The relationship between the z-scores of birth weight and the cardiac structures was plotted with a line depicting x = y (representing a one-to-one relationship). Values above this line indicate that the cardiac structure was less affected than birth weight relative to gestational age at birth, and values below the line indicate that the cardiac structure was more affected than birth weight relative to gestational age at birth.

To test for association between 1) twin size (smaller/larger) and the structural cardiovascular measurements and 2) the difference in z-scores of birth weight and cardiac structures and the intercept (representing the absence of a difference in these z-scores), a Generalized Estimating Equation (GEE) was used. This method accounts for the fact that we study paired data (twin pairs) while also allowing us to control for covariates. It is well-known that in MC twins TTTS can influence fetal cardiac development, differentially for donors and recipients, due to hemodynamic imbalances.[37] The same is presumed for TAPS. As prenatal growth is our primary focus, we have chosen to examine the effect of twin size and regard the group of included twins as a whole. To take into account any potential influences of the different types of complications on the cardiac parameters, we first tested this association using a GEE. When there was a statistically significant association, the analysis of twin size for that parameter was corrected for the type of complication. A subgroup analysis per complication (TTTS/TAPS/sFGR) was performed for the differences in structural cardiovascular measurements between donors and recipients in TTTS/TAPS and smaller and larger twins in sFGR. A subgroup analysis was performed for within-pair differences in cardiac z-scores in twin pairs in which the smaller twin was born SGA. Statistical analyses were performed using IBM Statistics Version 25.0 (SPSS, Inc. an IBM company, Chicago, IL) and R Version 4.1.2 (R Foundation for Statistical Computing, Vienna, Austria). A p-value of <0.05 was considered statistically significant.

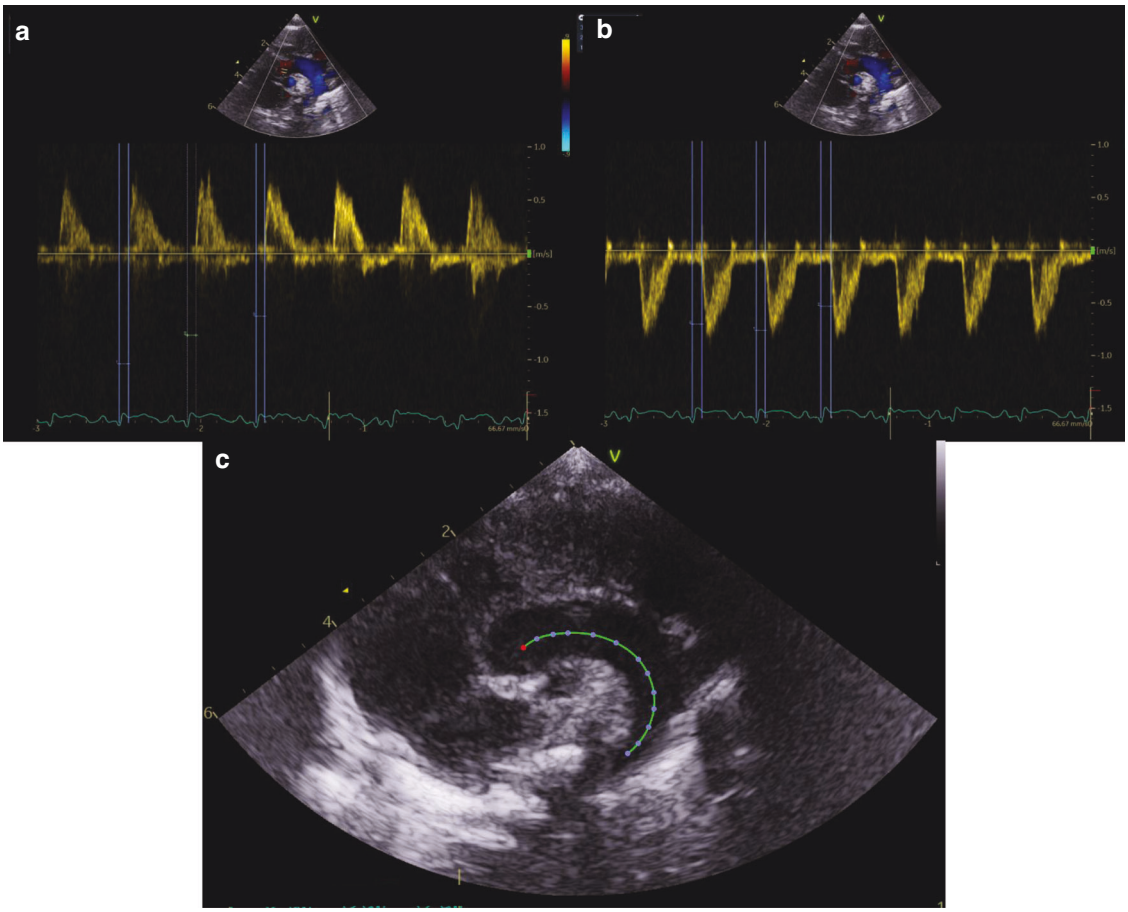

Fig. 1 Measurement of the aPWV based on two separate Doppler recordings of one single two-dimensional cross-section of the aorta. **a** Pulsed Doppler recording at the level of the AV with three measurements of time until flow, **b** pulsed Doppler recording at the descending aorta at the level of the diaphragm (or as distal as possible if unattainable) with three measurements of time until flow, **c** length of the aorta from the location of pulsed Doppler recording A to pulsed Doppler recording B. As time intervals were studied rather than the height of the signal, central placement was not relevant.

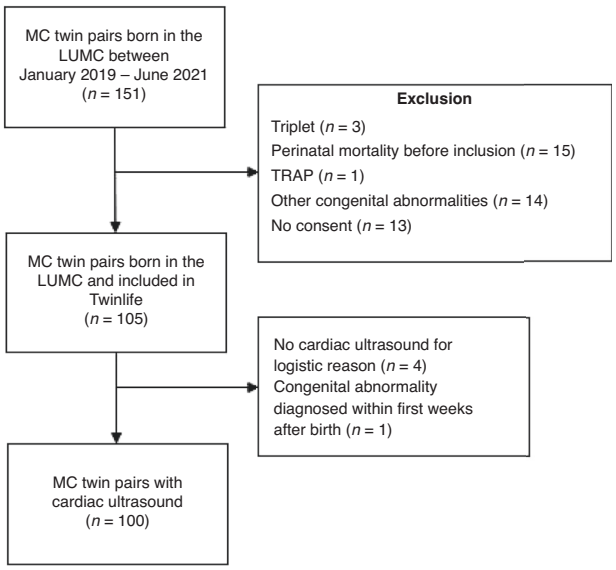

Fig. 2 Flowchart of study inclusion.

RESULTS

Between January 2019-June 2021, 100 parents participated in the study (Fig. 2). aPWV was missing in 24 cases due to unavailability of staff trained to perform this measurement.

Median gestational age at birth was 33.8 (30.8–36.1) weeks (Table 1). PDA requiring treatment was present in four cases (three smaller twins, one larger twin ($p = 0.340$)). PPHN requiring nitric oxide was present in eleven cases (five smaller twins, six larger twins, $p = 0.739$)). The presence of a PDA or PPHN did not influence the effect size of the structural measurements. Of all pregnancies, 41% was complicated by TTTS, of which 90% (37/41) were treated with fetoscopic laser coagulation which was successful in 36/37 cases; 17% was complicated by TAPS of which 13% (2/17) were successfully treated with fetoscopic laser coagulation in the TAPS trial (ClinicalTrails.gov NCT04432168); 19% was complicated by sFGR; and 23% was uncomplicated.

Median postnatal age at ultrasound was 3 (2–4) days. All cardiac valve annuli diameters showed the same association in which smaller twins had a lower z-score (Table 2). Similarly, both the LA diameter and Ao diameter were smaller in smaller twins. A similar association was observed for the z-scores of the LV dimensions with lower z-scores for smaller twins. The analysis of LVPWD was corrected for type of complication, following the significant association primarily caused by the untreated TTTS cases (Supplementary Table S1). LV mass was lower for smaller twins as opposed to larger twins, as was RWT. No significant difference was found for aPWV. A subgroup analysis per complication (TTTS/TAPS/sFGR) for the differences in structural cardiovascular measurements is presented in Supplementary Table S2. The subgroup analysis of twin pairs with a smaller twin born SGA ($n = 63$) demonstrated more pronounced within-pair differences in structural measurements (Supplementary Table S3).

Table 1. Baseline maternal, obstetrical and neonatal characteristics for included twins.

Characteristics	MC twins ($n = 200$; 100 pregnancies)
Maternal age – *years*	32 (29–34)
Gravidity	2 (1–3)
Parity	1 (0–1)
MC twin pathology	
Uncomplicated	23 (23)
MA	4 (4)
TTTS	41 (41)
GA at diagnosis – *weeks*	21.6 (17.9–24.4)
Quintero stage	
I	13 (32)
II	14 (34)
III	13 (32)
IV	1 (2)
V	0 (0)
Fetoscopic laser coagulation	37 (90)
Other treatment	4 (10)
Time between diagnosis and birth – *weeks*	12.0 (7.9–15.1)
TAPS	17 (17)
GA at diagnosis – *weeks*	27.9 (22.9–31.3)
Fetoscopic laser coagulation	2 (13)
Other treatment	15 (87)
Time between diagnosis and birth – *weeks*	6.1 (2.5–9.5)
sFGR	19 (19)
GA at diagnosis – *weeks*	17.7 (16.8–19.8)
Gratacós type	
Type I	9 (47)
Type II	4 (21)
Type III	6 (32)
Time between diagnosis and birth – *weeks*	13.9 (10.8–16.5)
GA at birth – *weeks*	33.8 (30.8–36.1)
Female	96 (48)
Caesarean	123 (62)
Birth weight discordance – %	13 (7–27)
TTTS	11 (5–28)
TAPS	14 (8–26)
sFGR	28 (24–34)
Uncomplicated	8 (4–12)
Birth weight discordance ≥ 20%	38 (38)
Birth weight – *grams*	
Smaller twin	1729 (1200–2115)
z-score	−1.1 (−1.7–−0.7)
Larger twin	2058 (1643–2500)
z-score	−0.3 (−0.7–0.1)
Small for gestational age	
Smaller twin	63 (63)
Larger twin	17 (17)

Outcomes are presented as median (interquartile range (IQR)).
MC monochorionic, *MA* monoamniotic, *TTTS* twin-twin transfusion syndrome, *GA* gestational age, *TAPS* twin anemia-polycythemia sequence, *sFGR* selective fetal growth restriction.

Table 2. Structural and functional cardiac parameters for the smaller and larger twin.

Outcomes	Smaller twin ($n = 100$)	Larger twin ($n = 100$)	*p*-value
Postnatal age at ultrasound - *days*	3 (2–4)		
Respiratory support at ultrasound			
Mechanical ventilation	4 (4)	6 (6)	
Non-invasive support	27 (27)	28 (28)	
No support	69 (69)	66 (66)	
Cardiac valve annuli diameter			
MV – mm	9.9 (9.1–11.1)	10.8 (9.8–11.9)	
z-score	−0.8 (−1.6–−0.2)	−0.2 (−0.9–0.5)	**<0.0001**
TV – mm	10.4 (8.9–11.7)	11.0 (9.8–11.9)	
z-score	−1.3 (−2.4–−0.3)	−0.8 (−1.6–−0.1)	**<0.0001**
AV – mm	5.9 (5.3–6.5)	6.1 (5.8–6.7)	
z-score	−0.5 (−1.2–0.2)	0.0 (−0.7–0.7)	**<0.0001**
PV – mm	6.6 (5.9–7.4)	6.9 (6.2–7.5)	
z-score	0.8 (−0.2–1.7)	1.1 (0.2–2.0)	**0.019**
LA diameter – mm	10.7 (9.4–12.1)	11.5 (10.2–12.7)	**0.002**
Ao diameter – mm	8.2 (7.2–9.0)	8.6 (7.7–9.7)	**0.002**
LA/Ao ratio	1.3 (1.2–1.5)	1.3 (1.1–1.5)	0.347
Left ventricular dimensions			
LVIDD – mm	14.8 (13.3–16.2)	15.5 (13.7–16.8)	
z-score	0.5 (−0.1–1.3)	0.8 (0.2–1.7)	**0.001**
LVPWD – mm	2.5 (2.2–2.9)	2.7 (2.3–3.3)	
z-score	−0.2 (−0.9–−0.1)	−0.1 (−0.8–0.2)	**0.020**[a]
LVIDS – mm	9.6 (8.8–11.4)	10.7 (9.4–11.9)	
z-score	0.4 (−0.4–1.1)	0.7 (0.1–1.4)	**0.002**
LVPWS – mm	3.6 (3.1–4.0)	3.7 (3.3–4.6)	
z-score	−0.5 (−1.1–0.0)	−0.4 (−1.2–0.9)	**0.010**
LV mass – grams	5.1 (4.0–6.6)	6.0 (4.5–7.7)	**<0.0001**
LV ejection fraction – %	64.0 (57.7–70.5)	61.2 (56.0–69.2)	0.108
Fractional shortening – %	31.6 (27.9–36.6)	30.0 (26.6–35.7)	0.220
RWT	0.34 (0.29–0.40)	0.36 (0.29–0.47)	**0.020**
aPWV – m/s	3.1 (2.4–5.3)	3.3 (2.5–4.4)	0.259

RWT calculated as (2× LVPWD)/LVIDD.
Outcomes are presented as median (IQR) or *n* (%).
MV mitral valve, *TV* tricuspid valve, *AV* aortic valve, *PV* pulmonary valve, *LA* left atrium, *Ao* aortic root, *LV* left ventricular, *LVIDD* end-diastolic left ventricular internal diameter, *LVPWD* end-diastolic left ventricular posterior wall thickness, *LVIDS* end-systolic left ventricular internal diameter, *LVPWS* end-systolic left ventricular posterior wall thickness, *RWT* relative wall thickness, *aPWV* aortic pulse-wave velocity.
[a]Corrected for type of complication.

Table 3. The difference in z-score of birth weight and z-score of the cardiac structure tested against the intercept.

Differences in z-scores	MC twins ($n = 200$)	*p*-value	Smaller twin ($n = 100$)	*p*-value	Larger twin ($n = 100$)	*p*-value
Cardiac valve annuli diameter						
Birth weight - MV	0.2 (−0.6–1.2)	**0.011**	0.3 (−0.6–1.3)	**<0.0001**	0.2 (−0.7–1.1)	0.329
Birth weight - TV	−0.5 (−1.2–0.5)	**0.002**	−0.2 (−1.0–0.8)	0.166	−0.7 (−1.4–0.4)	**<0.0001**
Birth weight - AV	0.5 (−0.4–1.3)	**<0.0001**	0.7 (−0.1–1.5)	**<0.0001**	0.2 (−0.6–1.1)	**0.020**
Birth weight - PV	1.6 (0.6–2.7)	**<0.0001**	1.9 (0.8–3.0)	**<0.0001**	1.2 (0.4–2.4)	**<0.0001**
Left ventricular dimensions						
Birth weight - LVIDD	0.4 (−0.2–1.1)	**<0.0001**	1.7 (1.1–2.4)	**<0.0001**	1.1 (0.3–1.9)	**<0.0001**
Birth weight - LVPWD	0.8 (0.1–1.4)	**<0.0001**	0.8 (0.1–1.4)	**<0.0001**	0.0 (−0.5–0.6)	0.529
Birth weight - LVIDS	1.3 (0.4–2.1)	**<0.0001**	1.5 (0.8–2.3)	**<0.0001**	1.1 (0.1–1.9)	**<0.0001**
Birth weight - LVPWS	0.3 (−0.4–1.4)	**<0.0001**	0.7 (−0.1–1.4)	**<0.0001**	−0.1 (−0.6–1.1)	0.057

Outcomes are presented as median (IQR). Significant *p* values, i.e., $p < 0.05$, are in bold.
MV mitral valve, *TV* tricuspid valve, *AV* aortic valve, *PV* pulmonary valve, *LVIDD* end-diastolic left ventricular internal diameter, *LVPWD* end-diastolic left ventricular posterior wall thickness, *LVIDS* end-systolic left ventricular internal diameter, *LVPWS* end-systolic left ventricular posterior wall thickness.

The cardiac structures were less affected than birth weight in relation to gestational age at birth, indicated by a significantly higher z-score difference compared to the intercept in all but one parameter (Table 3). This finding was more pronounced in smaller twins. These associations are also depicted in Fig. 3, illustrating that the majority of twins is above the x = y line.

DISCUSSION

In MC twins, smaller twins present with a structurally smaller heart (including cardiac valve annuli, LA, Ao and LV dimensions) at birth when compared to larger twins. Despite having smaller cardiac structures, the heart (except for the TV) appears to be less affected by insufficient prenatal growth than body size for a given gestational age. This finding may be indicative of heart sparing.

A fetus experiencing FGR adapts by redistributing blood flow towards major organs. This 'sparing' has been elaborately described in the context of the brain.[38–40] We have now observed a similar phenomenon for the heart in which the cardiac structures are less affected by FGR than body size. Redistribution of cardiac output is facilitated by vasodilation and increased blood flow towards essential vascular beds and vasoconstriction and decreased blood flow towards non-essential vascular beds.[39] When these hemodynamic changes continue for an extended period of time, structural cardiovascular remodeling occurs that can in turn result in functional deficits.[3,39,41] Another explanation for the observed structural changes may be prolonged fetal cardiac dysfunction as a result of FGR. Both stroke volume and cardiac output increase, accounting for the relatively larger size of the cardiac structures in smaller twins rather than heart sparing.[41] Both of these hypotheses align with our findings.

With regard to previous literature on neonatal cardiac remodeling, our results are largely similar. Both Altin et al. and Fouzas et al. found smaller LV, LA and Ao dimensions in thirty SGA infants compared to appropriately-grown controls.[10,11] Seghal et al. found that when corrected for body surface area, the LV dimensions were significantly larger in preterm SGA infants compared to appropriately-grown controls.[12,13] It is difficult to properly compare our findings as the other studies have primarily focused on cardiac function, e.g., using tissue Doppler imaging and reporting on outcomes such as myocardial performance index. In light of our hypothesis on the long-lasting effects of FGR, we chose to report on structural rather than functional parameters as our primary outcome measures, as the former are less affected by short-term loading conditions/volume imbalances.

It should be considered that neonatal structural cardiac remodeling is multifactorial in origin and dependent on the type of intrauterine insult, the duration of the exposure to the insult and the gestational age at birth. By design, our study considers the effect of prenatal growth across the entire pregnancy. Birth weight can be considered the outcome of prenatal growth. To accommodate the impact of gestational age, we have adopted using z-scores accounting for gestational age at birth. While this is a common approach, we acknowledge that using z-scores of uncomplicated twin pregnancies per gestational age window is preferable, yet this data is unavailable at present.

Tracking cardiovascular structure and functioning over a longer period of time is crucial to understand the pathophysiological processes that underlie CVD risk.[27,42] We have now focused on neonatal cardiovascular remodeling. It is plausible that changes in some parameters, such as the aPWV, are yet to occur if this adaptive process is more gradual.[27,43] Furthermore, normative data for a premature neonatal population are currently unavailable, and measurements were missing at random in 25% of cases.[35] We do not yet know whether the observed structural cardiovascular changes at birth will also persist into childhood or if these are (partially) reversible. This information is essential in devising targeted interventions to induce this reversal. It also remains to be seen if the relatively modest within-pair differences in the sizes of cardiac structures in our study indeed lead to clinically relevant cardiovascular deficits at a later age. To discover this, we will have to track these twins well into adulthood.

Our study has limitations. We have included a range of complications that can have a different impact on cardiovascular structure. TTTS and TAPS twins experience hemodynamic imbalances in utero, which can affect postnatal cardiac dimensions despite reversibility after fetal therapy.[37,44–48] Similarly, in TAPS, donors experience anemia and recipients polycythemia, resulting in distinct circulatory adaptations.[19] In sFGR, hemodynamic changes are different in origin and expressed by abnormal umbilical artery Doppler flow patterns.[49] Additionally, prenatal growth is affected differently by each complication. Where isolated sFGR is primarily caused by unequal placental sharing, growth discrepancies in TTTS and TAPS can also occur due to oliguria/anemia in donors. This influences extrapolation of our results to FGR in singletons, as this is caused by placental insufficiency. Yet, it is currently unknown whether a different etiology in FGR does in fact influence outcomes. To control for heterogeneity in our population, we assessed whether there was an association between the complication and each cardiac measurement. This was only the case for LVPWD that stayed

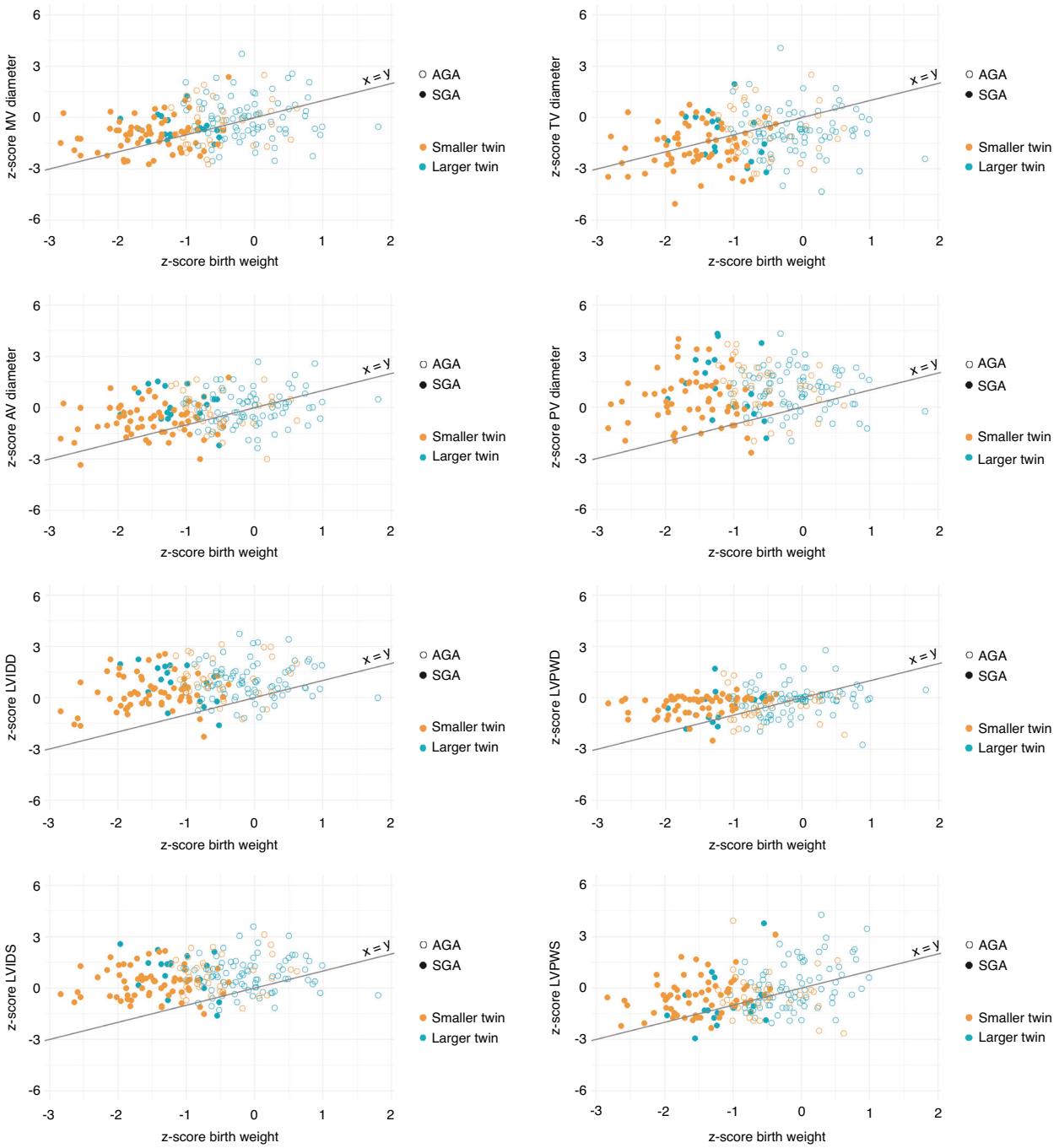

Fig. 3 Scatterplots of the relationship between the z-score of the birth weight (x-axis) and the z-score of the cardiac structures (y-axis), with the line depicting x = y. Values above this line indicate that the cardiac structure was less affected than birth weight relative to gestational age at birth and values below the line indicate that the cardiac structure was more affected than birth weight relative to gestational age at birth.

significant even after correction for the complications. Similarly, our subgroup analyses revealed little differences between donors and recipients in TTTS and TAPS with similar effect sizes for the majority of measurements in the sFGR group, illustrative of a limited influence. Only 38% of twin pairs presented with a BWD ≥ 20%. We may have a slight underrepresentation of discordant twin pairs. Yet, including concordant twin pairs allowed us to investigate the effect of BWD on a wide continuous scale. Moreover, 63% of the smaller twins were SGA, which is considered a postnatal expression of FGR. So, there was an adequate representation of FGR cases among smaller twins.

CONCLUSIONS

To conclude, we report a structurally smaller heart for smaller twins, of which approximately 60% were born SGA, in our population. While cardiac structures are smaller, the deviation in birth weight tends to be more pronounced than the deviation in cardiac structure. We used an identical twin cohort controlling for genetic and maternal factors as well as gestational age at birth, in which epigenetic profiling and cardiovascular follow-up will be performed in the near future. Tracking cardiovascular structure and functioning over a longer period of time is crucial to understand the pathophysiological processes that underlie CVD risk.[27,42]

DATA AVAILABILITY

Individual participant data (including data dictionaries) that underlie the results reported in this article will, after de-identification, be available beginning 3 months and ending 10 years following article publication. The study protocol will also be made available. The data will be shared with researchers who provide a methodologically sound proposal and whose proposed use of the data has been approved by an independent review committee identified for this purpose and the medical ethical committee of the Leiden University Medical Center. Proposals should be directed to s.g.groene@lumc.nl. To gain access, data requestors will need to sign a data access agreement.

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