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Review

Placenta morphology and biomarkers in pregnancies with congenital heart disease – A systematic review

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ABSTRACT

Impaired placentation is an important contributing factor to intra-uterine growth restriction and pre-eclampsia in fetuses with congenital heart defects (CHD). These pregnancy complications occur more frequently in pregnancies with fetal CHD. One of the most important factors influencing the life of children with CHD is neurodevelopmental delay, which seems to start already in utero. Delayed neurodevelopment in utero may be correlated or even (partly) explained by impaired placentation in CHD cases. This systematic review provides an overview of published literature on placental development in pregnancies with fetal CHD. A systematic search was performed and the Newcastle-Ottawa scale was used to assess data quality. Primary outcomes were placenta size and weight, vascular and villous architecture, immunohistochemistry, angiogenic biomarkers and/or placental gene expression. A total of 1161 articles were reviewed and 21 studies were included. Studies including CHD with a genetic disorder or syndrome and/or multiple pregnancies were excluded. Lower placental weight and elevated rates of abnormal umbilical cord insertions were found in CHD. Cases with CHD more frequently showed microscopic placental abnormalities (i.e. abnormal villous maturation and increased maternal vascular malperfusion lesions), reduced levels of angiogenic biomarkers and increased levels of anti-angiogenic biomarkers in maternal serum and umbilical cord blood. Altered gene expression involved in placental development and fetal growth were found in maternal serum and CHD placentas. In conclusion, abnormal placentation is found in CHD. More extensive studies are needed to elucidate the contribution of impaired placentation to delayed neurodevelopment in CHD cases.

1. Introduction

With an incidence of five to eight per thousand newborns, congenital heart defects (CHD) are the most common congenital anomalies [1]. As approximately half of the CHDs are severe and treatment carries risks, CHDs are a large contributor to infant mortality worldwide [1,2]. Due to continuous improvement of ICU-care and cardiothoracic surgery, together with increased prenatal detection, survival rates of affected newborns have increased greatly. For that reason, the focus of innovation has shifted from increasing survival rates to improvement of long-term (neuro)developmental outcomes [1–3].

An important part of the morbidity in children and adolescents with CHD is neurodevelopmental delay, which is reported in a significant

amount of cases [4–9]. In earlier literature, this impairment was attributed to the adverse effects of complex cardiothoracic surgery in early life [4–6,8,9]. More recently, a decreased prenatal and postnatal head circumference and delayed cortical maturation on prenatal ultrasound and MRI were found in CHD fetuses and newborns before surgery [5–9]. This raises the question whether the hemodynamic effects of the CHD induce intrauterine exposure to decreased flow or low saturation in early neonatal life.

A second finding in pregnancies with fetal CHD is the increased incidence of intra-uterine growth restriction (IUGR), pre-eclampsia (PE) and pregnancy induced hypertension (PIH) [10–22]. These signs of impaired placental development are confirmed by an increased umbilical artery resistance and decreased global placental perfusion in fetuses

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with CHD [18–27]. The increased incidence of placental pathology in pregnancies with CHD, raises the question if this is a possible contributor to impaired neurodevelopment in these fetuses, as increased umbilical artery resistance in fetuses with CHD is associated with altered (neurodevelopmental) outcomes [24,25]. The relation between placenta characteristics, placental function and fetal neurodevelopment has not yet been studied in CHD cases and is still hypothetical.

In this systematic review, an overview of the literature on isolated CHD and placenta development is provided, aiming to explore the role of the placenta in the relation to CHD and fetal neurodevelopment.

2. Materials and methods

2.1. Search strategy

A systematic search was performed in Pubmed, Embase, Web of Science, Cochrane and Emcare on April 15th 2021. The search terms contained “congenital heart defect”, “placenta”, “genetics”, “fetus”, “neurodevelopment”, “outcome”, “long term outcome”, “angiogenic”, “biomarker”. Publications from all dates were included. The complete search string is available in appendix A.

2.2. Study selection

The screening of the title/abstract and subsequently the reading of the full-text articles was performed by two independent researchers (MS, MA). A third researcher (MH) was consulted when agreement was not reached. Studies were eligible for inclusion if 1) newborns with isolated congenital heart disease were included; and 2) placenta characteristics regarding macroscopic measurements, immunohistochemistry, vascular and villous architecture, immunoreactivity, angiogenic biomarkers and/or placenta genetics were provided and; 3) a control group or a comparison with reference values for the described placenta characteristics was present. As the presence of a genetic syndrome or multiple pregnancies have effect on intra-uterine growth and developmental outcomes [28,29], studies on congenital heart disease with a genetic disorder or syndrome and/or multiple pregnancies were excluded, in order to identify the sole effect of the congenital heart disease. Studies with less than ten cases were also excluded from the review. Cross-referencing was performed manually on the included studies.

2.3. Quality assessment and data extraction

Data quality was assessed using the Newcastle Ottawa Scale (NOS) for assessing the quality of non-randomized studies [30].

Patient characteristics and placenta characteristics regarding macroscopic measurements, immunohistochemistry, vascular and villous architecture, immunoreactivity, angiogenic biomarkers and/or placenta genetics were extracted and analyzed.

3. Results

3.1. Study selection

The conducted search generated 1288 articles. After screening on title/abstract, 136 articles were read full text and 21 articles were included (Fig. 1) [31–51]. No relevant studies were found by cross-referencing. Study characteristics are shown in Table 1.

Study groups were not always comparable for the different included studies and differed specifically on categorization of types of CHDs (Table 1). Most studies included all CHD, whilst others only included ‘major’ CHD (defined as CHD requiring cardiac intervention within the first year of life) or included only one specific congenital heart defect.

Of the included studies, 10 studies describe macroscopic placenta characteristics, 6 studies describe microscopic placenta characteristics, 4 studies describe angiogenic biomarkers in maternal and/or umbilical

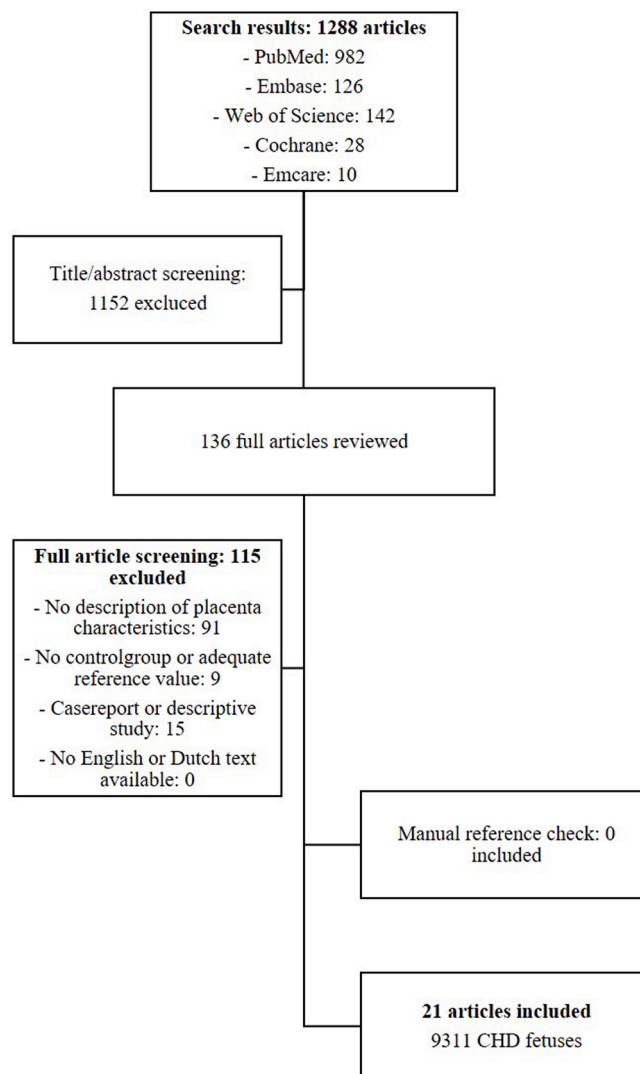


Fig. 1. a: Flowchart of included studies.

cord serum and 7 studies describe genetic expression in maternal serum and/or placental tissue.

3.2. Quality assessment

Quality assessment is shown in Supplement 1. Most included studies in this systematic review were rated with an adequate quality. According to the NOS quality assessment, 16 articles obtained five or more out of six stars. One study obtained only one star, due to inadequate representativeness of the cohort and inadequate selection of the controls. Three studies obtained three stars, as they had no control group and compared the measurements with reference values.

3.3. Study results

3.3.1. Macroscopic placenta characteristics

3.3.1.1. Placental weight. In Tetralogy of Fallot (ToF), double-outlet right ventricle (double-outlet RV), major ventricular septal defects (VSDs) [39] and hypoplastic left heart syndrome (HLHS), a reduced absolute placental weight and placenta weight percentiles were found at birth (Table 1) [36]. Placenta volumes measured by MRI between 18 and 39 weeks of gestation, tended to be smaller in a study with mixed CHD cases, but the found differences did not reach statistical significance

Table 1
Overview of included studies.

	Study design	Single- or multi-center	Number of patients	Number of controls	Type of CHD	Time of measurement	Outcome measure(s)
Arcelli (2010), Italy	Retrospective case-control	S	40	48	All CHD	Second trimester; Postpartum	mRNA species in maternal serum; gene expression in placental tissue
Llurba (2013), Spain	Prospective case-control	S	68	340	Major congenital cardiac defects	First trimester	PIGF and PAPP-A in maternal serum
Andescavage (2014), USA	Prospective case-control	S	41	94	All CHD	Second and third trimester	Placental volume (fetal MRI)
Llurba (2014), Spain	Prospective case-control	S	65	204	Major congenital cardiac defects	Second trimester; Postpartum	PIGF, sFlt-1, sEng in maternal blood; PIGF, sFlt-1, sEng in umbilical cord blood
Curti (2015), Italy	Prospective case-control	S	10	50	All CHD	Third trimester	PIGF in maternal blood
Jones (2015), USA	Retrospective case-control	M	16	18	HLHS	Postpartum	Placenta weight, umbilical cord insertion, parenchymal morphology of the placenta
Albalawi (2016), USA	Retrospective case-control	S	200	200	CHD requiring cardiac surgery within 6 months	Postpartum	Placental cord insertion, placenta-to-birth weight ratio
Curti (2016), Italy	Prospective case-control	S	39	31	All CHD	Second trimester	mRNA species in maternal serum
Matthiesen (2016), Denmark	Retrospective cohort	M	7569	916,853	All CHD	Postpartum	Placenta weight
Contro (2017), Italy	Retrospective case-control	S	36	42	CNTRA, LVOT	Second trimester	Gene expression (MAPK1, IQCAP1, Visfatin) in maternal serum
Fantasia (2018), UK	Prospective cohort	S	196	49,898	Major congenital cardiac defects	First trimester	PIGF and PAPP-A in maternal serum
Morano (2018), Italy	Retrospective case-control	M	10	30	VSD	Second trimester	mRNA Tenascin-X gene in maternal serum
Rychik (2018), USA	Prospective cohort	S	120	No controls ^a	Complex CHD	Postpartum	Placenta weight, placenta-to-birth weight ratio, placenta pathology
Takemoto (2018), Japan	Prospective case-control	S	37	626	All CHD	Postpartum	Placenta-to-birth weight ratio
Miremberg (2019), Israel	Retrospective case-control	S	32	34	Severe CHD with termination of pregnancy	Postpartum	Placenta weight, placenta-to-birth weight ratio, maternal and fetal vascular malperfusion lesions of the placenta, inflammatory lesions of the placenta, umbilical cord abnormalities
Radhakrishna (2019), USA	Prospective case-control	S	8	10	VSD	Postpartum	Gene expression in placental tissue
Russell (2019), USA	Prospective cohort	S	133	No controls ^a	CHD requiring surgery in infancy	Postpartum	PRA genes damaging variants in placental tissue
Schlatterer (2019), USA	Prospective cohort	S	51	No controls ^a	All CHD	Postpartum	Placenta weight, placenta-to-birth weight ratio, placenta pathology
Courtney (2020), USA	Retrospective case-control	S	24	18	HLHS, TGA	Postpartum	Gene expression in placenta tissue
Giorgione (2020), Italy	Retrospective case-control	S	480	456	Major congenital cardiac defects	Postpartum	Placenta-to-birthweight ratio, parenchymal morphology of the placenta
Ozcan (2021), USA	Retrospective cohort study	S	47	92	Major congenital cardiac defects	Postpartum	Placenta disorders (placenta praevia, placental abruption, placenta accrete/increta/percreta, cord insertion abnormalities)
							Placenta weight, placenta-to-birthweight, placenta perfusion defects

^a Compared to population reference values.

[33].

As placenta weight does not differentiate between cases with and without IUGR, it is important to identify placenta weight in relation to the birthweight. Five studies reported on placenta-to-birthweight ratio's, a measure to assess placental weight by correcting for birthweight, and described conflicting results. Four studies, of which three with a retrospective design, found no significant difference in placenta-to-birthweight ratio in CHD cases [37,44,45,51], suggesting that smaller placentas in CHD are only present in cases with a low birthweight. Three prospective studies did find a significant lower placenta-to-birthweight ratio in CHD cases [43,48,49], which means that the placentas were even smaller than expected, based on the birthweight of these cases. If CHD placentas are smaller in comparison to birthweight, fetus were able to reach their growth potential, indicating preservation of the placental function.

3.3.1.2. Umbilical cord insertion. Abnormal umbilical cord insertion is

associated with adverse pregnancy outcomes, such as IUGR and intra-uterine demise [52]. Significant elevated rates of abnormal umbilical cord insertion (marginal and velamentous insertions) were found in one study including 200 CHD cases [37]. No elevated rates of abnormal umbilical cord insertions were found in two smaller studies in 16 HLHS cases [31] and 32 cases with different types of CHD (HLHS, transposition of the great arteries, atrioventricular canal defect, ToF, double-outlet RV and coarctation of the aorta) [45], presumably because of the small number of inclusions. In major CHD cases, 4.5% had 'placental disorders' other than macroscopic morphology (defined as placenta praevia, placental abruption, abnormally invasive placenta), compared to 3.3% in healthy controls ($p = 0.046$) [50].

3.3.2. Microscopic placenta characteristics

Histological findings indicating impaired fetal-maternal exchange are more frequently found in pregnancies with CHD. It was first reported in HLHS cases, where fibrin deposition, distal villous hypoplasia, lower

vascular area and lower vasculo-syncytial membrane counts were described [36,49]. Later this was confirmed in placentas of a group of mixed CHD cases, as fetal placental thrombosis, placental infarction, choriangiomas, infarction and delayed villous maturation were frequently reported, whilst these are phenomena that are not found in placentas of healthy newborns [43,45,48,51]. There was no significant difference in incidence of microscopic placenta abnormalities in cases with and without PE, PIH and IUGR [48].

3.3.3. Angiogenic biomarkers

Angiogenic biomarkers in pregnancy, such as Placental Growth Factor (PlGF) and pregnancy-associated plasma protein A (PAPP-A) are mainly produced in the placental trophoblasts [53,54]. Altered expression of angiogenic biomarkers in CHD cases may modify vascular pathways in the placenta, as concentrations of the angiogenic biomarkers Placental Growth Factor (PlGF) and pregnancy-associated plasma protein A (PAPP-A) were significantly lower in maternal serum of CHD cases [32,34,38,41]. Soluble fms-like tyrosine kinase (sFlt-1) and soluble endoglin (sEng) are anti-angiogenic biomarkers and were significantly higher in maternal serum of CHD cases [34]. sFlt-1 and sEng levels were higher in umbilical cord blood of CHD cases [34]. None of the studies describing angiogenic biomarkers corrected for placenta weight. Alterations in expression of these (anti-)angiogenic biomarkers suggest placental dysfunction and impaired placental perfusion. These findings may explain the decreased birthweight and placenta weight in isolated CHD cases [41].

3.3.4. Genetic expression in the placenta

Altered expression of (m)RNA and alterations in genes associated with cardiac function and development were found in both placental tissue and maternal serum of CHD cases. A significant part of the described genes with altered expression are expressed in both placental tissue and other (fetal) organs. Most of the described proteins have intracellular or direct extracellular effects, though altered expression is found in both placenta tissue and maternal serum. Altered expression of

genes associated with trophoblast development, placental development and fetal growth was also described (Table 2), demonstrating a presumable relation between CHD, placenta genetic expression and placental function [31,35,40,42,46,49]. Damaging variants in genes with positive regulation of angiogenesis (PRA) were found, suggesting an altered angiogenic pathway in placentas of CHD cases [47].

4. Discussion

This systematic review found that CHD is associated with lower placental weight, placental abnormalities that suggest impaired placentation and altered genic expression in placental tissue. Similar characteristics are observed in placentas of IUGR, PE and PIH cases, all finding their origin in abnormal trophoblast invasion and placental insufficiency [55–66]. This suggests a relation between CHD and impaired placentation and possibly a decreased fetal-maternal exchange of oxygens and nutrients as a result of this in these cases. Remarkably, even in absence of PE and IUGR, characteristics of altered placentation and placental insufficiency are found in CHD cases.

A possible origin of these findings could be directed to altered protein and RNA expression of genes that are likewise involved in the origin of the CHD. These alterations may be responsible for the placenta pathological changes that partly overlap the phenotype of classic placenta dysfunction based on insufficient trophoblast invasion.

Altered molecular expression in vascular developmental pathways impacts vascular development in both the placenta and fetal organs. Changes in protein expression of genes with a local effect on cardiac or placental tissue, as well as changed expression of proteins with a more distant effect were described in placental tissue and maternal serum. These findings advocate an effect on vascular developmental pathways in other fetal organs, in addition to the effect on placental and cardiac tissue. The origin of CHD is multifactorial and both environmental factors and genetic predisposition play an important role in this development [67]. One can hypothesize that angiogenic imbalance and altered vascular formation in the placenta of CHD cases may be due to

Table 2
Overview of described aberrant genes.

Study	Location	Type of CHD	Results	Aberrant genes	Clinical associations
Arcelli (2010)	Maternal serum, placental tissue	All CHD	mRNA expression	ACTN4; COLIA 2, FBLN1, HPSE, JARIDIB, P4HA2, PAPP-A; ET3, MYL7; FSCN1; MAP4; RAB36; SAVI, TNXB, TXN.	Contractile activity Extracellular matrix biosynthesis Congestive heart failure Contractile activity Contractile dysfunction Rhabdomyoma/aetiopathogenesis Morphogenesis
Curti (2016)	Maternal serum	All CHD	mRNA expression	FALZ; PAPP-A; PRKACB; SAVI; STK4; TNXB2.	Brain development Placental function Pattern formation and morphogenesis Tumor suppressor Trophoblast differentiation Heart rhythm disorders and connective tissue disease
Contro (2017)	Maternal serum	CNTRA, LVOT	mRNA expression	MAPK1; IQGAP1, Visfatin.	Trophoblast development Cardiomyocyte hypertrophy activation
Morano (2018)	Maternal serum	VSD	mRNA expression	Tenascin-X (TNXB)	Cardiac development (coronary vasculogenesis, valve development), collagen production
Radhakrishna (2019)	Placental tissue	VSD	DNA methylation	HEY2; ISL1; ACTC1; BMP4, TGFB1, PDGFRA, TBX2, EGFR1, CXCR4, TLX3, DPP6, FGFR1.	Cardiac development Cardiac septum formation Cardiac muscle development Heart development, cardiac anomalies
Russell (2019)	Placental tissue	CHD requiring surgery in infancy	Frequency of damaging PRAs	117 damaging PRA variants	Positive regulation of angiogenesis
Courtney (2020)	Placental tissue	HLHS, TGA	RNA expression	393 genes, a.o. IGF2; CITED2; GLUT1, GLUT3, SNAT2.	Placental development, cardiac function Placental capillary patterning, body axis IUGR

epigenetic influences from vascular and circulatory alterations or due to hypoxic stress in early embryogenesis of CHD cases [43,49,68]. The hemodynamic effects of the CHD may cause epigenetic changes, resulting in altered vascular developmental pathways. This may result in angiogenic alterations, placental abnormalities and its associated diseases, nonetheless this hypothesis can also be the other way around. Factors contributing to the pathogenesis of the CHD appear to also effect the development of the placenta and influence epigenetics of placental tissue, as common genetic contributors are found [47,49,69]. Similarly, the described altered levels of angiogenic biomarkers could be related to placenta size, as lower placental weight is common in CHD cases. Future studies on angiogenic biomarkers in placental tissue should index for placental weight, to get a better understanding of the effect of these biomarkers in the developmental pathway of CHD, angiogenic factors and placentation.

The next step in this hypothesis is the idea that delayed fetal brain maturation and delayed neurodevelopment at child age are caused by the same vascular alterations, as the placental changes are also found in CHD cases without PE and IUGR [7,8]. The process of impaired placenta perfusion, altered vascular genetic pathways and altered angiogenic expression is presumably comparable in CHD cases and cases of PE and IUGR, as these findings were also found in the absence of these diseases in CHD cases. Impaired placenta function and altered vascular expression in the placenta can cause impaired fetal organ perfusion and hypoxia.

Hypoxia and reduced organ perfusion may specifically contribute to delayed development of the brain, as this organ is extremely sensitive to hypoxia [70,71]. This effect is demonstrated in fetuses with CHD, as reduced cerebral oxygenation and impaired brain growth on MRI was found in these fetuses [72] and dysregulation of the angiogenesis was found in cerebral tissue of fetal CHD cases [73]. In addition, more postnatal brain injuries (white matter injury, intraparenchymal hemorrhage, intraventricular hemorrhage, subdural hemorrhage) within the first five days of life were found in CHD cases with abnormal placenta pathology [48]. Moreover, this hypothesis is supported by the findings in a fetal sheep model, in which placental hypoxia levels similar to hypoxia in CHD cases were imitated. In brain tissue of these sheep,

microglial morphology was altered, indicating an effect of hypoxia on brain pathology [74]. An altered angiogenic environment could furthermore contribute to hypoxic stress and impaired microvascular development of the brain. This may therefore contribute to the impaired neurodevelopment and delayed brain maturation in CHD fetus.

In this review, placental function is assessed by examining placental weight, placental morphology and genic expression in placental tissue. These measures provide an adequate representation of fetal-maternal exchange and placenta perfusion, yet true placental function is only measurable by assessing volume and oxygen content of the umbilical venous blood volume. In previous studies by Sun et al. (2015) and Ho et al. (2020), impaired umbilical venous volume and impaired fetal cardiac output were demonstrated in CHD cases [72,75]. The morphological placental abnormalities described in this review, combined with the found decreased placental blood flow in CHD cases by Sun et al. (2015) and Ho et al. (2020), suggest abnormal placental function in CHD cases.

As previously delineated, we hypothesize that fetal CHD, placental development, angiogenic alterations and neurodevelopment are all closely related. The directed acyclic graph for these hypothetical associations is shown in Fig. 2. The causal pathway of CHD and neurodevelopmental outcomes is complex and consists of numerous causal and biasing paths. In order to further clarify the causative pathway, more research is needed. In future research, we suggest a focus on the contribution of impaired placentation on neurodevelopmental delay in found CHD cases.

Another important consequence of abnormal placentation in fetal life is the susceptibility to cardiovascular disease and insulin resistance in later childhood and adult life. Abnormal placentation is related to poor fetal growth and low birthweight. Following Barker's hypothesis, these circumstances in fetal life are linked to diabetes, obesity, hypertension and coronary artery disease in adulthood [76,77]. As CHD is related to abnormal placentation, these fetuses might be more prone to developing these conditions in later life, next to their congenital heart defect. In order to identify patients at risk for cardiovascular disease and prioritize preventive measures in these cases, is important to investigate cardiovascular outcomes of CHD cases in later life.

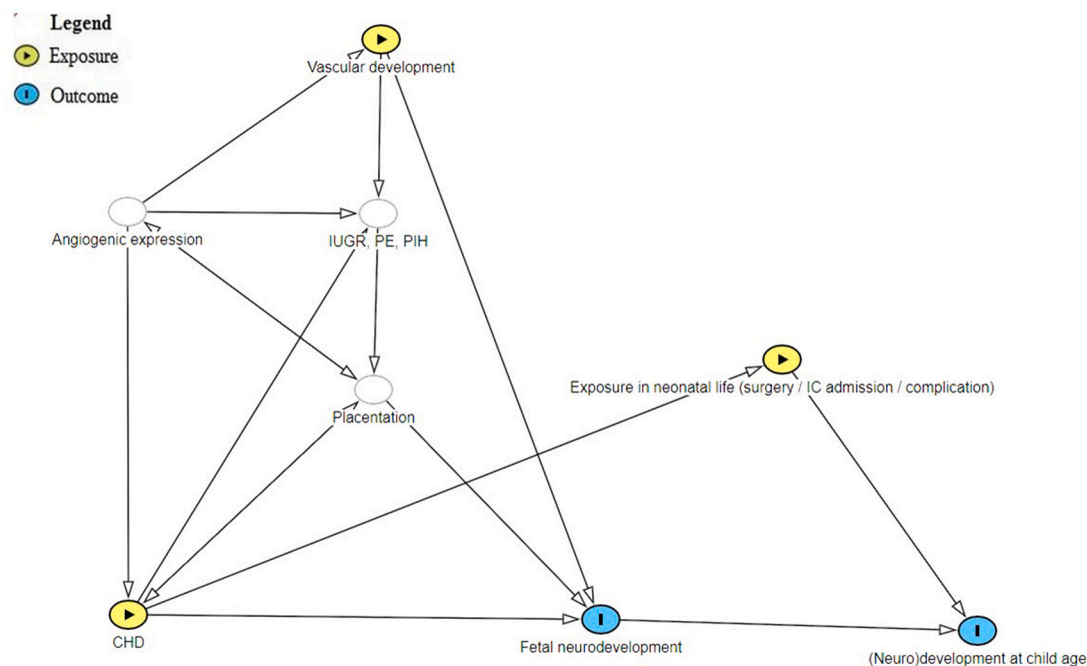


Fig. 2. The directed acyclic graph of hypothetical associating factors, available on <http://www.dagitty.net>. The directed acyclic graph of hypothetical associating factors demonstrates the complex causal pathway of CHD, angiogenic factors, placentation and neurodevelopment and the relation between the different associating factors.

A limitation of this review is the heterogeneity in the patient characteristics of the included studies. Most studies included all CHD cases, whereas other studies only included ‘severe’ or ‘critical’ cases. Most studies excluded cases with known genetic abnormalities, but not all studies described the methodology in this detail. Heterogeneity in the study populations may have caused differences in outcomes and error in the results. By excluding studies on fetuses with known syndromes or genetic abnormalities, a significant group of CHD cases is left out and the effect might be major in these cases. As this review seeks for the pure effect of the consequences of a CHD on placental development, patients with known genetic abnormalities were excluded, in order to efface the additional effect of the genetic abnormality. Most included studies, however, did not provide information on Whole Exome Sequencing, a technique that is developing fast and identifies single-gene abnormalities in CHD cases [78,79]. It is therefore possible that a part of the cases included in these studies which were considered as isolated CHD, may actually have a single gene disorder. Secondly, some of the studies had relatively small study populations, impeding the results to reach significant differences.

5. Conclusion

This systematic review shows that CHD is associated with lower placental weight, macroscopic and microscopic placental abnormalities, altered angiogenic biomarkers and altered genetic expression in placental tissue and maternal serum. These findings suggest impaired vascular development of both the placenta and fetal organs.

As impaired neurodevelopment is an important part of morbidity in CHD cases, future research should focus on the contribution of abnormal placentation to neurodevelopmental outcomes of these cases. Hemodynamic consequences of the different types of CHDs should be related to placental morphology and function as well as to neurodevelopmental outcomes in utero, neonatal life, childhood and adult life. This knowledge could contribute to the development of preventive measures for CHD cases both in utero and in the entire lifespan.

Ethics statement

Not applicable (systematic review of literature).

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Data available statement

Data used for the current review are available from the corresponding author on reasonable request.

Declaration of competing interest

The authors have no conflicts of interest to disclose.

Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.placenta.2021.07.297>.

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