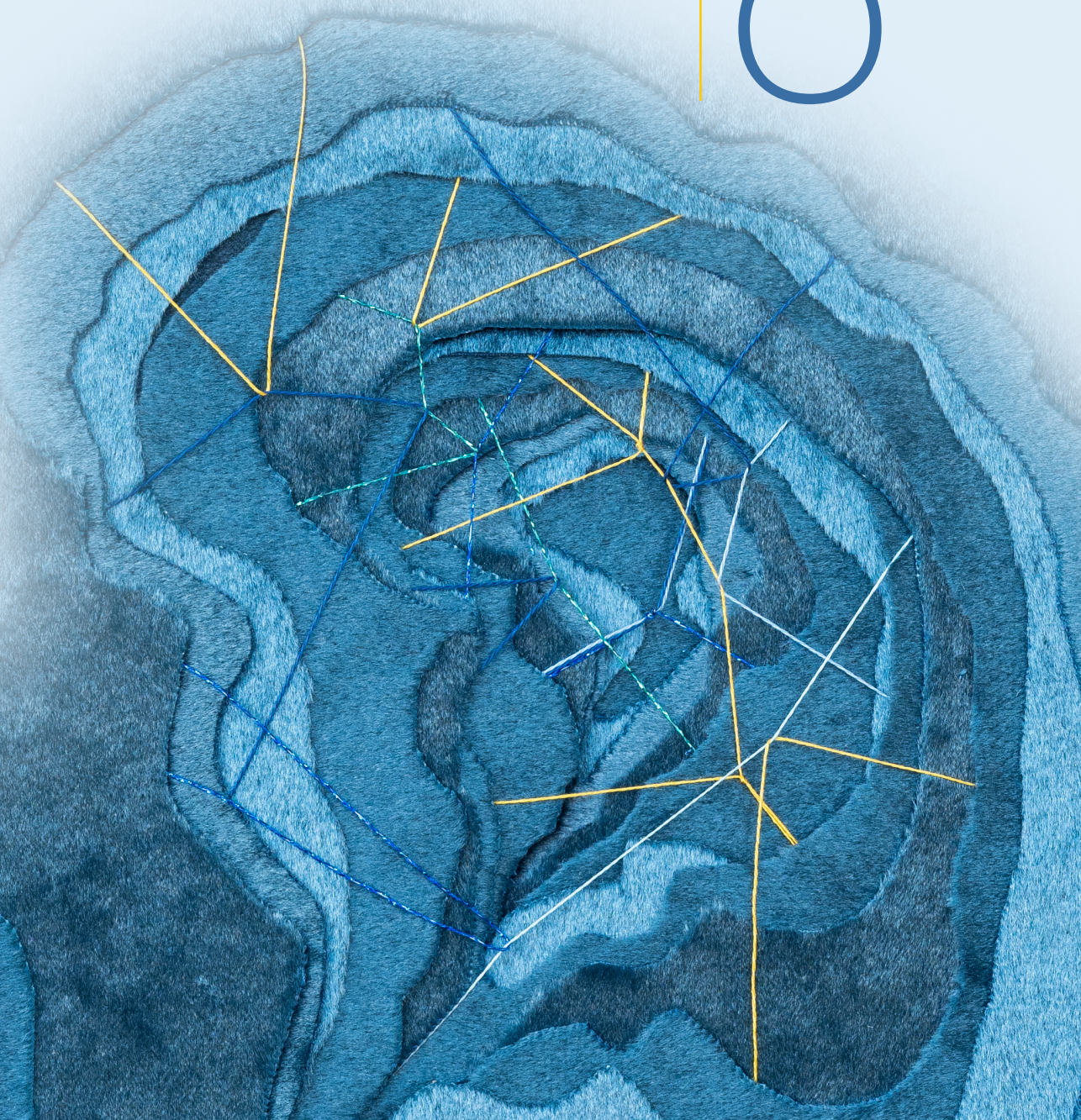
Imaging the prenatal brain in congenital heart defects

Everwijn, S.M.P.

General Discussion

CHAPTER

8



Congenital heart defects (CHD) are the most prevalent congenital defect with 6-8/1000 live births.

A prenatal diagnosis (PD) of a CHD results in a decrease of mortality and morbidity, underlining the importance of screening for CHD. The vast majority of children with a CHD survive infancy nowadays due to optimized neonatal and peri-operative care, but these children still face an increased risk of morbidity. Deficits in neurocognitive development are increasingly recognized, even in purely isolated heart defects. These deficits are described in multiple domains such as cognition, behavior, planning, execution as well as academic achievements and are linked to white matter injury. White matter injury has been attributed as a complication of (open) heart and percutaneous surgeries in the first year of life, leading to periods of low cardiac output and hypoxia, which is associated with abnormal cerebral development and delayed neurological development. Most recently, smaller cerebral volumes and decreased head circumferences were also seen in imaging studies in infants with less severe cardiac defects which only required percutaneous intervention or no intervention at all. This shows that neurocognitive impairment in children with CHD might not only arise from peripartum or perioperative circumstances. Adjacent to this, studies that showed brain injury on MRI in CHD-children prior to surgery have led to the hypothesis that these alterations might originate earlier than previously thought. Furthermore, fetal data has even shown signs of altered cortical development in CHD fetuses, thus prior to birth. This poses the hypothesis: is it possible that neurodevelopmental delay in congenital heart defects originates in fetal life? Can fetal brain injury worsen by postnatal circumstances, for example cardiopulmonary bypass or low cardiac output (second-hit theory)? If the altered fetal circulation plays a role, does this mean that certain cerebral areas or certain cardiac defects are more prone to delay than others?
This thesis focusses on in-utero neurological development, as well as the prenatal detection of two common congenital heart defects, to prevent postnatal brain injury.

Prenatal detection of congenital heart defects

The prenatal detection of congenital heart defects has been a topic of great interest over the last decades. Ultrasound as an medical imaging modality was introduced in the sixties and became more widespread and was introduced in obstetrical care early after its development. At first, ultrasound was only applied to determine fetal position. Secondly, it became apparent that it was possible to diagnose severe malformations, with anencephaly as the first diagnosed abnormality. In the decades thereafter, it gradually became clear that the diagnosis of a congenital defect could lead to optimized care and thus better outcome in the majority of the defects. This

was the start of the introduction of routine screening for congenital abnormalities using ultrasound in pregnancy.

In the Netherlands, prenatal screening for fetal anomalies was introduced relatively late compared to the surrounding countries. This was mainly caused by the concerns of Christian-Democratic politicians, who feared an increase in pregnancy termination after the introduction of prenatal screening and considered abortion not as 'treatment' for a disease, which was the most important prerequisite for the introduction of a test in the population-health program according the Dutch law. Eventually, all parties agreed that expecting parents had the right of equal access to prenatal care and freedom in their reproductive choices [1]. Thus, the routine screening program was implemented in 2007 in the Netherlands and is monitored by the government. Health-care professionals performing the standard anomaly scan (SAS) are bound to an annual number of scans and regular quality monitoring. The officially registered uptake of the 20 weeks' anomaly scan reports around 95% in the Netherlands, while the majority of the remaining 5% is scanned in a department for PD because of increased risk. The majority of the ultrasonographers in the screening setting are midwives that have received a standardized training in ultrasound to detect fetal anomalies.

Evaluation of the fetal heart is considered one of the more difficult parts of screening for anomalies. Possibly, the movement of the organ, the small size and the necessity to evaluate the organ in different planes, might play a role in the historically low rates of prenatal detection of congenital heart defects. Furthermore, the large variation in morphologic variants that can present with a completely normal four-chamber view and the relative low exposure to abnormalities, makes it difficult to recognize CHD. Furthermore, an interview study among sonographers showed that they experience a high threshold in referring a patient to a tertiary center, because they want to be absolutely sure that there is an anomaly, to not produce unnecessary parental anxiety [2]. Anomalies visible in the four-chamber view are detected in > 93% in our geographic region (Amsterdam – Leiden) after introduction of the SAS[3]. This means that recognition of very abnormal planes is excellent when prenatal screening follows a uniform protocol. Cardiac abnormalities that present with a normal four-chamber view, which encompass mainly outflow tract anomalies, for example tetralogy of Fallot and transposition of the great arteries (TGA), are detected less often. Since these abnormalities can be more subtle on prenatal ultrasound, they are perceived as more difficult. In chapter 2, the detection rates of these two common outflow-tract anomalies is described, before and after the introduction of the three-vessel view in our screening program. The reason why we chose to study the detection rate of Fallot and TGA lies in the fact that in TGA, prenatal detection can

prevent postnatal hypoxia and ultimately cardiac arrest, and in Fallot it creates the opportunity for genetic testing in pregnancy.
In TGA, the foramen ovale closes immediately after birth and the ductus arteriosus constricts within the first days of life, transitioning the heart to the circumstances outside the womb. If a fetus with TGA is undetected before birth, they have an increased risk of mortality and morbidity due to the severe hypoxia which is the result of the fact that the oxygenated blood cannot reach the systemic circulation.
It is known that Tetralogy of Fallot cases have an increased risk of genetic abnormalities. These cases benefit from increased detection prior to 24 weeks gestation (the legal limit for pregnancy termination in the Netherlands), providing the opportunity to offer additional genetic testing. When a genetic anomaly is present, the long term outcome for children with Fallot is worse than children with an isolated defect, thus a prenatal diagnosis allows parents to reach a decision on whether or not to continue with the pregnancy.

The current mandatory planes in the Dutch screening program consist of four cardiac planes; the four-chamber view, left- and right outflow tracts and the three-vessel view. This is quite sparse considering the fact that in the diagnostic setting, more than 60 minutes and over a hundred planes and sequences is no exception. Our study showed that the simple addition of the three-vessel view as a mandatory plane to the national screening program leads to the increased detection of TGA and Fallot. Following this example, the national screening guideline could be expanded with the 'V-sign/three vessel trachea view', the aortic arch in the sagittal plane and measurements of the semilunar valves to detect anomalies like aortic coarctation, pulmonary artery or aortic valve stenosis and total pulmonary venous return, which all present with very subtle changes in the planes. Preliminary research shows that the detection rate of aortic coarctation has been consistently low at a rate of 25% and has not risen in recent years, despite efforts in quality control and regular schooling of ultrasonographers. Children suffering from aortic coarctation benefit from an early detection, as a postnatal detection can lead to cyanosis and morbidity and even cardiac arrest, therefore we emphasize the need for a prenatal detection in these cases. Adding additional planes could increase the detection further. Moreover, to eliminate human error to some extent, we expect innovation and increased detection if automated image detection is implemented (with artificial intelligence). Studies regarding this topic are already being undertaken and show promising results.

Cortical development in fetal life

Prenatal brain development in CHD has gained increased interest as it became apparent that fetuses showed delayed Head Circumference (HC)-growth compared with normal controls. Secondly, pre-operative delays in cortical development in CHD children has furthered the interest in fetal cortical development for the etiology of altered neurodevelopment in CHD.

The human fetus follows a predictable pattern of emerging and specific temporal evolution of gyri and sulci. It is known from studies in brain malformations that cortical development is complex and is orchestrated by neuronal migration, which can be influenced by abnormal circumstances. Several studies showed that sustained or intermittent periods of cerebral hypoxia can lead to abnormal cerebral development[4]. This has been shown in lamb fetuses that were exposed to cerebral hypoxemia by transient bilateral carotid clamping, in which signs of decreased oligodendrocyte maturation were seen[5]. Oligodendrocytes synthesize myeline, which, if disturbed, results in white matter injury, since pre-oligodendrocytes are specifically prone to ischemic injury. The pre-oligodendrocytes in lamb fetuses with CHD are more easily damaged compared to those in healthy controls. Next to disturbances in oligodendrocyte maturation, studies with hypoxic animals also describe abnormal cortical development, such as reduced cortical volume and microstructural maturation. With the use of pre-operative MRI and neurological examinations in CHD neonates, more abnormalities were found compared to control neonates[6, 7], which supports the hypothesis that neurodevelopmental delays may be present before surgery and not all damage occurs in the peri-operative period.

In chapter 5 the ultrasonographic assessment of cortical development in fetal life showed delays in cortical maturation. This was, however, only statistically significant in the Sylvian, calcarine and the cingulate fissures. Alterations in the cingulate fissure area, are associated with attention deficit disorder, which is significantly more prevalent in CHD children. Therefore, it is possible the delay in maturation found in the Sylvian and cingulate fissures are associated with altered cognitive and behavioral problems in later life. The differences in cortical maturation found in the prenatal ultrasound studies described in this thesis (chapter 5,6 and 7) are, however, small. In chapter 6 we describe a brain maturation delay of four days using an brain-age estimation algorithm in all CHD's combined. After stratification, these delays were mainly seen in subgroups with TGA and cases with intra-cardiac mixing.

The results of our studies show an important difference with other studies exploring this subject, describing a more severe delay of up to 3 weeks in ultrasound[8, 9] and MRI studies[10-12], however, in our studies, we only see a delay of 4 days.

This could have two reasons: Firstly, the generalizability of some MRI studies are questionable, as these studies included severe cases (for example HLHS) more often than

moderate or minor cases, which might have introduced selection bias[11]. Secondly, some of the studies concerned one or two measurements in a broad range of gestational ages, producing results with broad confidence intervals, this could have over-exaggerated their findings[10, 13, 14]. Furthermore, it is frequently mentioned that ultrasound has a lower accuracy than MRI to pick up brain abnormalities. We do not agree with this statement, as it has been proven that neurosonography performed by experienced sonographers in a tertiary setting is not inferior to MRI[15, 16].
On the other hand, we need to keep in mind that even small differences found prenatally could have a big impact on development in later life. Fetuses are known for an accelerated growth trajectory during pregnancy. If a disturbance takes place, it could mean this hampers physiological brain development. This could lead to a cumulative effect, since the foundation of the fetal brain has been flawed. In other words: the small deviations we have found prenatally with ultrasound studies might still have a clinical relevance in CHD-children.

Etiology of prenatal brain damage

The incidence and severity of neurologic deficits are still very difficult to predict, even if the cardiac defect is known prenatally, and birth is uncomplicated and timely, takes place in an center equipped for cardiac care. The multitude of published data on prenatal, neonatal and adolescent neurodevelopment is testament to the diversity of proposed etiologies. In this thesis we explore the theory on altered cerebral hemodynamics in CHD in several chapters. Also, altered placental function and different genetic make-up in CHD are possible etiologies that might play a role.

Children born with CHD are prone to brain damage in the first weeks of life and during the pre-operative period. Prolonged ventilation, cardiopulmonary-bypass and ICU admission are all, on itself, associated with white matter injury. *Pre*-operative brain abnormalities, such as white matter injury and arterial ischemic stroke, are seen in 19-52% of cases with different imaging modalities. Furthermore, in *fetuses* with CHD, head growth and intracerebral blood flow are found to be abnormal, even in-utero[17]. This poses the theory that altered neurodevelopment in CHD children might have its origin in prenatal life. Theoretically, fetuses with left obstructive lesions (Aortic arch anomalies, HLHS) have diminished flow, and fetuses with intra-cardiac mixing lesions (TGA, DORV, Fallot) have diminished oxygenation of the brain. In these lesions it was shown that impaired development of the fetal cortex in a small group of severe CHD cases (mainly TGA and left-sided obstructive lesions) with MRI[18]. The largest finding in this thesis (chapter 7) is a delay in cortical maturation of almost 4 days is found in fetuses with TGA. Yet, these findings in fetuses with TGA are in large contradiction to the neurologic(al) follow-up studies in children with TGA;

they are known to have normal cognitive development with a normal IQ. Yet they show a somewhat higher prevalence of behavioral disorders (for example attention deficit disorder)[19]. Furthermore, in our review of the literature on prenatal neurodevelopment in CHD (chapter 3), we have included studies in which MCA measurements (middle cerebral artery) were available. A total of 1412 fetuses with different CHD raging from moderate to severe were included. In one of the largest prospective studies, 72 fetuses with single ventricle morphology, increased cerebral blood flow was correlated with better ND-outcome in later life [20]. Theoretically, decreased resistance in the middle cerebral artery due to vasodilatation should be present in the case of a CHD, to enhance cerebral oxygen and nutrient delivery. In growth restricted fetuses, this is called the brainsparing effect, but in growth restriction this correlated with a worse long-term outcome. Since brainsparing in CHD-cases was correlated with a more favorable neurological outcome, a different etiology than growth restricted fetuses might be playing a role. The contradictory effect of the same intracerebral flow pattern (i.e. brain perfusion) in different pathologies, makes it questionable if the proposed theory that altered neurodevelopment is the effect of altered brain perfusion is correct.
Finally, as mentioned previously, the methodology of several imaging studies in CHD-fetuses, show selection bias. Fetuses with severe CHD (univentricular heart defects, HLHS) are included in these studies more often than moderate/minor CHD. These children are prone to more complex interventions and lengthy ICU stays as compared to children in the minor end of the CHD spectrum, with subsequent worse neurological outcome. Furthermore, genetic abnormalities are more often present in severe CHD-cases. This makes it difficult to draw conclusions on brain development of the full CHD -spectrum. Also, severe delays in-utero are not always correlated to postnatal findings, meaning that the clinical relevance of these findings still remains uncertain.

Lower birth weight and delayed HC-growth have been reported in CHD-fetuses, and is in itself associated with poor neurodevelopmental outcome as well. This has led to the thought that placental function in CHD cases might be abnormal and might also be influenced by the cardiac lesion. An association between non-genetic severe CHD cases (for example HLHS) and decreased HC-growth was not found, however, placental related complications, such as: growth restriction and pre-eclampsia, are seen more often in CHD-cases. Therefore, placental pathology might play a role in the etiology of altered neurodevelopment in CHD. Underlining this hypothesis, it has been shown that growth-factors that promote growth and angiogenesis, like VEGF (Vascular Endothelial Growth Factor), PIGF (Placental Endothelial Growth Factor) and soluble fms-like tyrosine kinase-1 (sFlt-1) are abnormal in mothers carrying

CHD fetuses. Also, the development of the placenta and the heart share the same regulatory pathways, such as Wnt (Wingless and Int-1). Wnt signaling pathways are involved in embryonic development and are necessary for normal cardiogenesis, they also play an important role in implantation, decidualization and placental differentation[21]. The strongest indication that placenta formation and heart development are intertwined, was seen in studies with knock-out mice. They were given specific genetic defects that are associated with congenital heart defects and showed abnormal placentation. After post-mortem examination, researchers found that these mice died because of severe growth restriction due to abnormal placentation[22].

Furthermore, a combination of aforementioned etiologies are involved in the presence of altered neurodevelopment in CHD, and therefore, we must consider it to be a multifactorial problem.

Parental counseling on neurodevelopment in CHD

The above described knowledge on the appearance and variation of neurodevelopmental delay in CHD cases, makes the prediction of outcome in later life extremely imprecise and difficult.

If parents are faced with the fact that the heart defect of their fetus may also influence brain development, but the extent of this delay cannot be predicted, this uncertainty may be very hard to cope with.

The American heart association (AHA) published a statement in 2012 stating that 'children with CHD are at increased risk of developmental disorder or disabilities' after evaluation of literature published on the matter between 1966 – 2011, advising physicians to strive for early detection of problems, in order to support them accordingly. For fetal medicine, the ISUOG released a statement in 2016 that is much more conservative than the statement of the AHA. They state that deficiencies in neurodevelopment are more prevalent in CHD children. The prenatal findings must, however, be correlated to postnatal neurologic testing, in order to validate the hypothesis that these findings really cause delay in neurodevelopment in later life. Since the release of this statement, a quick search reveals more than 200 peer-reviewed published articles on the subject. The association has been thoroughly confirmed, and in our opinion a new recommendation should amend the previous ISUOG statement. Numerous imaging studies have shown alterations in cortical development, decreased brain volume and white matter injury. Although the extent and timing of occurrence has not been sufficiently clarified, the link between neurodevelopment and congenital heart defects has been well-established. We would suggest to inform the parents on the multifactorial theory on neurodevelopmental delay in CHD children. Moreover, decreased cerebral oxygenation in-utero might be the

most obvious originator, but there must be more factors, seeing as the expression of altered cerebral development in CHD presents in a broad spectrum of defects. Although the timing might not be fully understood, exacerbating factors like preterm birth, perioperative complications, CPB and prolonged ventilation (e.a.) must be avoided if possible. Physicians should be aware of the increased vulnerability, and provide expecting parents with tailored information on altered neurodevelopmental outcome in CHD. This message should of course be delivered with some caution, because prenatal alterations have not sufficiently been correlated to postnatal outcome yet. Fortunately, the most frequent findings in neurodevelopmental testing concern learning difficulties, behavioral problems and executive functions, and not severe neurodevelopmental impairment (NDI) leading to complete dependency of medical care.

Future perspectives and conclusion

This thesis presents a number of prospective, well-executed studies in a cohort of consecutive cases with isolated CHD ranging from minor to severe defects of disease. These studies were specifically conducted to thoroughly explore brain development with multiple measurements in pregnancy, because previous studies had various methodological pitfalls (e.g. single measurements, selection bias, non-isolated caseload). Our studies showed significant but slight variations in development of the brain compared to normal fetuses. Repeated examinations allowed for the analysis of development during pregnancy and these analyses did not see a worsening during the third trimester as suggested by other publications.

We propose the following implementation in all tertiary centers in the Netherlands to gather a large set of CHD-types for research purposes.

The questions that still need answering are the following: Do children that display worse neurodevelopmental outcome later in life, have abnormal brain development in their fetal life? Can the severity of impaired neurodevelopment be predicted in pregnancy?

Is there an association between placental pathology and CHD, and also: does placenta pathology lead to worse neurodevelopmental outcome in CHD?

The methodology for research projects that will provide the answers to these knowledge gaps is a multicenter prospective cohort study according to the following guidelines:

Thorough neurosonography needs to be performed in the second and third trimester including: fetal growth, hemodynamics and cortical development. Specific attention should be aimed towards deviations from the normal sulcation patterns and structural anomalies. Furthermore, an MRI needs to be performed in moder-

ate/severe CHD cases using a uniform protocol (including the assessment of cortical development, volumetry and white matter injury) at least prenatally (around 34 weeks) and pre-and postoperatively. These MRI's need to be assessed by an experienced team, and use a standardized method for analysis of cortical development [23]. Whole exome sequencing (WES), has to be added as a standard genetic testing offer (now consisting of QF-PCR and SNP-array). Furthermore, placental tissue needs to be analyzed by a specialized pathologist to investigate signs of hypoxic-ischemic damage, and tissue needs to be stored in biobanks for further research. The infrastructure for such a collaboration has already been established in CAHAL (Center for congenital heart defects Amsterdam – Leiden), and can easily be rolled out in the rest of the Netherlands.

The focus of further study possibilities in cerebral development in CHD cases should address etiology and if prevention is deemed impossible, therapies should be developed to prevent further damage to the brain.
To further the understanding of the etiology of cerebral dysgenesis in CHD fetuses, we propose to look into placental pathology in combination with CHD. Fetal growth restriction and placental diseases like pre-eclampsia are more common in pregnancies complicated by CHD. A correlation between delayed neurodevelopment in fetuses with CHD with abnormal placentation might be explanatory in this issue. Therefore, biobanking of placental tissue is being conducted, which might clarify the contribution of the placenta further. In such a prospective cohort, all prenatal and perioperative data should be correlated to neuro-cognitive examinations and intellectual performance in school-aged children, to interpret the minor delays we have found with the presented studies. This requires the collaboration of maternal-fetal medicine specialists with pediatric cardiologists, and although this is not common practice everywhere, within CAHAL the collaboration is well-established. Ongoing research is being performed in our group, to gather an even more robust cohort of CHD, which allows for stronger subgroup analyses.

Although the etiology of prenatal brain damage in fetuses with congenital heart disease might not be completely understood, researchers are looking in to protective measures in peri-operative and neonatal ICU-care. Preventive measures such as the prenatal administration of Allopurinol[24, 25] or intranasal bone marrow cell therapy[26], might possibly be available in the future, to diminish the aforementioned 'second hit' caused by peripartum or perioperative circumstances.

Neurocognitive deficits in children with congenital heart defects are common and the mechanism behind it remains unclarified. A combination of genetics, hemo-

dynamics, placental factors and peri-operative complications are considered risk factors for worse neurodevelopmental outcome. One of the largest protecting factors is prenatal detection of a congenital heart defects. Uniform screening protocols with standard planes are known to increase detection of CHD, and we have demonstrated that adding new planes to this uniform program adds to the recognition of CHD. From a global perspective, other countries could model after our well organized national screenings program, since it has led to one of the highest detection rates of CHD. Although we are currently still dependent on human identification of imaging, automated image analysis has shown promising results and will definitely play a role in the future. This would benefit both prenatal detection of CHD and identify children who are prone to worse neurodevelopmental outcome.
Attention for the brain development is crucial in children with CHD as the increased vulnerability for altered neurodevelopment has been well established. With the right support, families, teachers and other caretakers of children with CHD could help them to thrive by meeting their specific needs.

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