CHAPTER 10

GENERAL DISCUSSION AND FUTURE PERSPECTIVES

Marrit M. Hitzert
The aims of this thesis were threefold:

I. To investigate early motor development and the long-term neurodevelopmental outcomes of infants treated with postnatal corticosteroids.

II. To investigate early motor development in various groups at risk of abnormal neurological development.

III. To investigate the interrelationship between early motor development, early visual attention, and neurodevelopmental outcomes up to school age in preterm-born and healthy fullterm-born children.

With regard to our first aim, we determined the quality of general movements (GMs) until 3 months post term in infants treated with either hydrocortisone (*Chapter 2*), high-dose dexamethasone (*Chapter 2*) or low-dose dexamethasone (*Chapter 3*), and we established the motor, cognitive and behavioral outcomes at school age of children treated with high-dose dexamethasone (*Chapter 4*). Regarding our second aim, we explored early motor development of infants prenatally exposed to PCBs and OH-PCBs (*Chapter 5*), and we reported on the outcome of an infant with molybdenum cofactor deficiency (MoCD) type A whom we treated experimentally with cyclic pyranopterin monophosphate (cPMP) (*Chapter 6*). Concerning our third aim, we described associations between early motor development and early visual attention in preterm and healthy fullterm infants (*Chapter 7*), we determined the predictive value of early motor development for functional outcome in healthy fullterm-born children (*Chapter 8*), and finally we studied the relation between early visual attention and functional outcome in preterm and fullterm children (*Chapter 9*).

**Part I. Early motor development and outcome after corticosteroids**

Bronchopulmonary dysplasia (BPD) is still one of the major complications in mechanically ventilated premature infants resulting in significant mortality and morbidity. For decades, debate is ongoing about the most optimal (treatment) strategy for preterm infants at risk for BPD. Persistent inflammation of the lungs is the most likely cause of BPD and since corticosteroids have strong anti-inflammatory effects these are among the most commonly administered drugs in this specific patient population. Treatment with dexamethasone (DXM) has repeatedly been shown to be effective in weaning infants from the ventilator, lowering the rate of BPD, and in decreasing total duration of supplemental oxygen therapy, even when using lower doses. Adverse long-term neurological sequelae such as cerebral palsy, however, have been reported. Hydrocortisone, as an alternative to DXM, seems not to exert such adverse effects on neurodevelopment although the efficacy of hydrocortisone (HC) treatment remains questionable as infants treated with HC showed no improvements in important outcomes of mortality, or of rates of BPD or home oxygen dependence.

A difficulty in interpreting the results of these studies lies in the various methodologies with different types of corticosteroids used (dexamethasone or hydrocortisone), and widely differing...
timing and dosage schedules of corticosteroids. In addition, studies are often contaminated in that infants randomly assigned to placebo still receive open-label corticosteroids. Moreover, part of the infants treated with corticosteroids still go on to develop BPD, which in itself constitutes a risk factor for abnormal neurodevelopment. A crucial shortcoming of current studies is that they report on outcomes from the age of 12 months onwards while it is particularly this group of infants that might benefit from identification at the earliest possible age in order to start interventions when the brain is subject to a high degree of neuroplasticity.

**General movements**

One of the diagnostic tools that can be used already in early infancy to judge the integrity of the central nervous system is the qualitative assessment of general movements (GMs). In addition, it has proven to serve as the most reliable predictor for later neurodevelopment. In accordance with the adverse neurodevelopmental effects seen after treatment with high-dose DXM, we demonstrated a poorer motor optimality already in the first few days after birth (days 1 and 7) and at 3 months in infants treated with high-dose DXM (starting doses of 0.5 mg/kg/day) when compared to untreated infants (Chapter 2). After DXM had been started, the motor optimality declined within one day.

In Figure 1, we provide an overview of our findings and proposed mechanisms for the observed changes in GM quality. One has to keep in mind that these mechanisms are based on a combination of human and animal studies, and are speculative in nature. DXM is known to have forceful anti-inflammatory effects, which are caused by DXM’s strong binding to glucocorticoid receptors. In addition to these anti-inflammatory effects, activation of glucocorticoid receptors leads to inhibition of glucose uptake into muscle and nerve cells, an effect which is already evident within 24 hours. Activation of glucocorticoid receptors may not only result in acute metabolic effects but might even lead to temporary or permanent changes in the brain architecture. In rat studies it was investigated whether high doses of glucocorticoid altered the dendritic spinal turnover in cortical brain areas. In the first 24 hours after glucocorticoid treatment, the formation of dendritic spines in rats was not decreased. The poorer motor optimality that we observed at the first day after the start of treatment was mainly related to reduced speed of movements (unpublished data) and not by a poorer GM quality. This supports the view that the deterioration in motor optimality at the first day following treatment might be considered an acute metabolic effect rather than a result from a decrease in synaptic connectivity. After daily glucocorticoid injections for 5 to 21 days, however, a decrease in the formation of dendritic spines has been observed, indicating that glucocorticoid administration may alter the integrity of the brain after repeated exposure to glucocorticoids which is in accordance with the poorer quality of GMs that we observed at day 7 following treatment. Previous findings and the present results suggest that DXM treatment may disturb normal patterns of synaptic connectivity. DXM may also indirectly lead to a decrease in synaptic connectivity by causing a hypoglycemic condition in neuronal cells and/or by alterations in gene transcription.

Synaptic input is a prerequisite for neurons to survive. A decreased synaptic connectivity caused by prolonged exposure to DXM may eventually lead to neuronal cell death. This theory
has been confirmed in rats who received a tapering course of DXM and had a higher neuronal apoptosis rate than placebo-treated rats in different brain regions, including cortex, thalamus, hippocampus, cerebellum, and subventricular zone. This generalized and persisting effect of DXM on brain development might account for the observed poorer FM quality and lower MOSs at 3 months.

**Figure 1. Observational changes in quality of general movements (GMs), fidgety movements (FM) and motor optimality scores (MOS) in infants treated with dexamethasone (DXM) compared to untreated infants, and proposed mechanisms.**

**Functional outcome at school age**

To determine if and to what extent treatment with high-dose DXM may have an impact on outcome at school age, we performed a follow-up study at 9 years of age including preterm infants treated with high starting doses of DXM (Chapter 4). Where previous studies predominantly reported on motor outcome, we found poorer performance on multiple other functional domains at school age as well when compared to existing literature on outcome of preterm-born, untreated, children. We speculated that the combination of affected domains, i.e. fine motor skills, visuomotor integration, visual perception, and attention suggests impairment
of the dorsal visual stream, a neural network linking the occipital and posterior parietal cortices, and which is heavily connected to the prefrontal and premotor cortex, hippocampal regions, basal ganglia, and cerebellum. The dorsal visual stream has been suggested to be particularly vulnerable for perinatal medical complications in preterm-born children.

Furthermore, evidence is accumulating that the cerebellum, apart from its role in motor skills, might contribute to several higher cognitive functions including visual-spatial information processing. Rodent data indicate that DXM exposure produced apoptosis in neuronal progenitor cells and led to permanent cerebellar pathology. In accordance with these findings, neuroimaging studies have demonstrated reduced cerebellar growth at term-equivalent age after postnatal exposure to DXM. We, therefore, propose that a reduced cerebellar volume may further contribute to the deficits we found at school age.

To our surprise, DXM-treated children performed well within the range of a norm population on verbal long-term memory and recognition. Still, those infants with the longest treatment duration and highest cumulative doses had the lowest memory performance scores. Impairment in memory and learning has been linked to hippocampal damage. Recently, it was demonstrated that the neurons of the ventral part of the hippocampus were more affected by DXM exposure than the dorsal part. Interestingly, in fMRI studies the dorsal part of the hippocampus was more activated than the ventral part during recall of verbal memory. Although speculative, it might be that long-term verbal memory and recognition remained relatively spared due to the less neurotoxic effects on the dorsal part of the hippocampus.

Other risk factors, related to the use of DXM, might have contributed to the poorer school age performance. Our studies particularly hint at longer mechanical ventilation (Chapters 3, 4) and the presence of BPD (Chapter 4) to be related to poorer outcome, possibly by mechanisms involving disruption of the integrity of cerebral white matter and/or cerebellar impairment (Figure 1).
Different types and dosing schedules of corticosteroids
Since high-dose DXM does not seem to confer additional therapeutic benefit over lower doses, the American Academy of Pediatrics has recommended restricting routine use of high-dose DXM in 2002. Since then, researchers have attempted to find the lowest clinically efficacious dose of DXM, with a view to minimizing the adverse neurodevelopmental effects. Where some found that lower doses of DXM facilitated extubation without increasing short-term adverse neurodevelopment, others could not demonstrate a decreased risk of abnormal neurodevelopment.

In our study of infants treated with low doses of DXM (0.25 mg/kg/day) after the second postnatal week (Chapter 3) we demonstrated promising results in that the quality of GMs significantly improved with the majority of infants having a normal neurodevelopment at 12-36 months. As illustrated in Figure 2, low-dose DXM treated infants had a higher MOS at 3 months compared to high-dose DXM-treated infants from our previous cohort (Chapter 2). The latter results are in line with the previously shown differential impact of different DXM doses on brain structures. For instance, in rats hippocampal neuronal cell loss was demonstrated after treatment with high doses of DXM, but not after low-dose DXM treatment.

Despite these favorable outcomes, we found a high prevalence of BPD in our cohort (94%). As this is in contrast to previous studies of low-dose DXM, we proposed that this high prevalence of BPD might be related to our stringent treatment policy, which is we only started treatment in those infants who required rather high levels of respiratory support (ventilator dependency after the 10th day with high ventilatory pressures (mean airway pressure >12 cm H2O) and/or high fractional inspirational oxygen requirement (>0.50). It might therefore well be that the a priori risk of developing BPD was higher than in the other studies. Treatment indications in most previous studies were ventilatory dependence and/or the necessity to start treatment as judged by the clinician without reporting details on ventilator settings, which makes it difficult to compare the a priori risk of BPD between our cohort and those study groups.

In agreement with previous studies we demonstrated no adverse effects of hydrocortisone (HC) on neurodevelopment in the first three months after term. As elaborated in Chapter 2, the quality of GMs was comparable between HC-treated infants and untreated infants, and MOSs at 3 months were higher than in infants treated with high-dose DXM (Figure 2). HC differs from DXM in the lower potency for glucocorticoid activation and much higher potency for mineralocorticoid activation. The lower glucocorticoid activation of HC may account for its less forceful anti-inflammatory properties and consequently less beneficial effects on preventing BPD. In contrast, mineralocorticoid receptors protect against neuronal cell apoptosis by upregulation of anti-apoptotic proteins, which might explain the higher MOS at 3 months in HC-treated infants and the absence of adverse neurodevelopmental effects at school age.

The results provided in this part of the thesis suggest that low-dose DXM might be considered a potentially safer alternative treatment option than high-dose DXM in infants at risk for development of BPD. Nevertheless, randomized controlled trials are needed to investigate long-term respiratory outcome and to investigate whether the favorable neurological outcome persists up to school age and beyond.
Part II. Early motor development in various risk groups

The perinatal period is characterized by great plasticity of the brain, which renders the brain particularly vulnerable to injury. Prenatal exposure to environmental pollutants such as polychlorinated biphenyls (PCBs) and their hydroxylated compounds (OH-PCBs) may induce long-lasting neurological damage. In Chapter 5, we determined whether prenatal background exposure to (OH-)PCBs would affect the early motor development of three-month-old infants. We demonstrated that high background exposure to most PCBs and OH-PCBC-107 was associated with a reduced movement repertoire, whereas several other OH-PCBs tended to be associated with an age-adequate movement repertoire. Overall, our findings indicate that prenatal exposure to OH-PCBs may be less neurotoxic than does prenatal exposure to PCBs.

One of the potential mechanisms through which (OH-)PCBs may have an adverse effect on motor development is by disruption of thyroid hormone homeostasis. Thyroid hormones regulate migration, process outgrowth, synaptic development, and myelin formation in specific brain regions, as well as the timing of these processes. Previous studies on thyroid hormonal activity in vitro and in vivo suggested more neurotoxic effects of OH-PCBs than PCBs, which is in contrast to our results. We speculated that this might be related to the higher cord blood levels of PCBs than OH-PCBs in our cohort. Furthermore, exposure to PCBs continues after birth by maternal transfer of PCBs via breast feeding, while OH-PCBs are poorly transferred to human milk. Thus, the actual exposure to PCBs might have been higher than the reported concentrations based on cord blood.

We have to emphasize, however, that most associations were found for the age-adequacy of movement patterns. In our study on the predictive value of early motor development for school age outcome in healthy fullterm children (Chapter 8), we demonstrated that an age-adequate repertoire was related to poorer cognition. We therefore advocate these children to be followed up to clarify what impact prenatal exposure to (OH-PCBs) has on later functioning, and to determine how assessment of the early motor development may assist in predicting outcome. Furthermore, we recommend that future studies focus on the influence of prenatal exposure to mixtures of several compounds on neurodevelopment because humans are continuously exposed to a variety of endocrine-disrupting chemicals, which can act together, and lead to effects that are different from those of the individual pollutants.

In Chapter 6 we described the neurodevelopmental outcome of an infant with molybdenum cofactor deficiency (MoCD) type A who was experimentally treated with cyclic pyranopterin monophosphate (cPMP). In our patient, diagnosis was confirmed prenatally and birth was induced at 36+3 weeks PMA because of deterioration of MRI findings (signs of atrophy and edema; unpublished data). We were the first to demonstrate that starting cPMP treatment within 4 hours after birth leads to a favorable neurodevelopment as shown by improvement in aEEG patterns, improvement of GM quality and a normal neurodevelopmental outcome with only a mild delay in cognitive functioning at 21 months of age. To date, this patient is still responding well. Our findings suggest that if a diagnosis of MoCD has been confirmed by antenatal testing, early induction of birth should be taken in consideration to enable early treatment before the onset of
cerebral injury. An additional interesting finding was that the course of the MOS corresponded well to the course of SSC-levels, one of the most sensitive hallmarks for sulfite toxicity.\textsuperscript{61} Our data suggest that the assessment of GM quality may contribute in monitoring treatment efficiency in infants with MoCD type A. Extended follow-up is urgently needed to reveal whether the beneficial effects of cPMP on neurodevelopment last throughout childhood and continue into adulthood.

### Part III. Interrelationship between early motor development, early visual attention and functional outcome at school age

During prenatal development and early infancy, the brain is subject to considerable developmental changes. Neurons proliferate, migrate and an excessive amount of neurons and synaptic connections are formed. The fate of these excess neurons and synapses is determined by synaptic pruning, a process which involves selective elimination of neurons and synapses to form efficient neural networks.\textsuperscript{30} Until the stage of synaptogenesis, brain development is largely driven by genetic processes.\textsuperscript{62-64} Once the brain reaches the stage of synaptic pruning, however, the balance shifts towards activity-dependent processes: the activity of a neural pathway, driven by experience, affects whether a particular connection weakens or stabilizes as part of a permanent network.\textsuperscript{62,65} Although synaptic reorganization of the brain continues into early adult years, it is already during infancy that the foundations of the brain architecture are established that are critical to the many complex functions of the adult brain.\textsuperscript{62,65} In the earliest years of life, children’s experiences mainly depend on their ability to generate a large repertoire of varied movements and their visual exploration of surroundings. These early behaviors may therefore serve as important markers for brain integrity and, as a consequence, may also serve as potential predictors for later functioning.

**Early motor development and functional outcomes at school age**

The qualitative assessment of spontaneous movements, the so-called general movements (GMs), has proven to be a powerful predictor for later cerebral palsy (CP)\textsuperscript{66,67} and minor neurological dysfunction.\textsuperscript{68} Evidence is accumulating that the quality of GMs not only predicts later motor outcome but also has predictive value for later cognition such as intelligence and attention.\textsuperscript{69,70} Previous studies of the predictive value of early motor development for later functioning are drawn from specific populations such as high-risk fullterm infants or infants born preterm. These studies suggest that the predictive relations between early motor expression and later outcome are mediated by brain abnormalities, such as white matter abnormalities,\textsuperscript{71} basal ganglia and thalamic lesions,\textsuperscript{72} or more broadly, dysfunction or damage of the cortical subplate and/or its connections, which run through the periventricular white matter and basal ganglia.\textsuperscript{73,74}

To contribute to a better understanding about the role of spontaneous movement activity in the infant during normal brain development, we examined the early motor development and subsequent functional outcomes in a cohort of fullterm, healthy children at 6 years of age (Chapter 8).\textsuperscript{75} Since all but two infants had a normal quality of fidgety GMs we focused
on detailed aspects, and found that children with an age-adequate repertoire of movements had poorer cognitive outcomes at school age than children with a reduced motor repertoire. Detailed patterns that were associated with better outcomes included the presence of variable finger postures and the absence of a monotonous concurrent repertoire.

For interpreting our findings regarding the age-adequacy, we must take into account that the variety of motor patterns is a complex phenomenon. Previous studies indicate that the number of different movement patterns increases between 6 and 24 weeks postterm in preterm and fullterm populations. We replicated this finding when we looked at the averaged developmental curves of movement patterns for a fullterm and a preterm group (Chapter 7). From our observations of individual longitudinal trajectories of movement patterns, however, it became clear that the majority of infants showed a drop in their number of different movement patterns at variable time points (for individual developmental trajectories, see Supplement 2 in Chapter 7). We might consider these drops in movement variety being related to either of two processes. The first is the period of neural reorganization that normally occurs around 3 months postterm age and has been shown to be preceded by a temporary decline in movement variation. The other is the selection phase in which an infant is engaged in one particular movement pattern for a prolonged period or shows repetition of a few particular patterns. This selection phase occurs at function-specific ages and is accompanied by a transient reduction in the overall motor repertoire.

We speculated that infants with fewer movement patterns represented those infants that started selection of specific movement patterns earlier due to advanced neurodevelopment resulting in a less variable repertoire than infants who had not reached this stage yet. Alternatively, infants with fewer movement patterns might have represented those infants that were in the period just before the neural transformation in contrast to infants who showed more movements and were thought to have passed this phase. If the latter would have been the case, though, we would have expected an association with poorer, rather than better, outcomes because of a delayed neurodevelopment. Thus, although we corrected for ages at recording, we cannot exclude the possibility that the observed differences in movement variation were attributed to the different timing of assessing motor development between infants rather than that we observed true differences in developmental progressing within the individual infant. In order to improve the interpretation of our findings and to be able to place our findings in a broader perspective, it is important that future studies focus on developmental trajectories of movement variety and its association with later neurological and functional outcome.

Despite these puzzling findings, it was striking that the detailed patterns that were associated with better outcomes, i.e. variable finger postures and the absence of a monotonous concurrent repertoire, were the ones that represented more qualitative aspects of motor development in contrast to the age-adequacy that comprised a more quantitative measure of motor development.

Together, our findings suggest that qualitative markers of early motor development, other than quality of GMs, may reflect those neural systems that underlie higher cognitive functioning and have already gained their permanent brain architecture around 3 months or remained more
or less stable over time.

Important to note is that in all studies on early predictive measures for later outcome other factors than the one investigated may have affected brain development and therefore future functioning. These factors probably include biological factors, such as genetic and epigenetic factors, and environmental factors, such as social environment, mother-infant interaction, motivation, and parental education.81

Early visual attention and functional outcomes at school age
An important exploratory behavior that may have an impact on later functioning is the looking behavior of infants. An infant’s ability to disengage and shift gaze from a fixated central stimulus to another stimulus in the periphery (‘visual attention’) is thought to reflect cognitive processing.82 It involves attention networks that have been shown to be closely associated with the widely distributed dorsal stream of visual-spatial processing, which underlies many cognitive and motor functions.83 In Chapter 9 of this thesis we investigated the longitudinal relation between the developmental rate of visual attention in the first 6 months and functional outcome at 10 to 11 years of age. Our data showed that infants who reached adult levels of visual attention at later postterm ages had a poorer performance on attention, motor skills, and handwriting at school age. These results should also be interpreted in the light of several other factors that may have contributed to neurodevelopment, as discussed in the previous paragraph.

The interrelationship between early motor development, early visual attention and functional outcome at school age
In the past years, a close interrelationship between motor development and cognitive development has been recognized with empirical evidence stemming from cross-sectional84,85 as well as longitudinal studies.86-88 Several theories have been posed regarding the link between motor and cognitive development. According to Gesell’s maturational theory,89 a child’s development including motor, intellectual, and behavioral development, proceeds according to a biological and genetically predetermined plan regardless of other potential environmental influences. Others, such as Piaget90 and Thelen,91 stress a more flexible, constructivist perspective in which brain development involves both preprogrammed processes and experience-based self-constructed shaping and fine-tuning of neural networks. Following the line of argumentation of a maturational factor underlying the link between motor and cognitive development, one might expect shared neural substrates for different motor and cognitive functions at varying stages of development. In the different Chapters of this thesis, we elaborated on the mechanisms underlying the relations between early motor development and early visual attention on the one hand and functional outcomes at school age on the other hand. Neurophysiological and neuroimaging evidence suggests the involvement of the prefrontal cortex, the cerebellum and their connecting structures (among others the basal ganglia and striatum) for the interrelation between motor and cognitive development. For an overview see Diamond et al. 2000.92

To our knowledge, we are one of the first to show longitudinal associations for predictive measures obtained in the first 6 months of postnatal life, a period in which cortical or cerebellar
structures have not yet fully matured and brain development is predominated by the formation of cortico-subcortical circuits guided by the cortical subplate.\textsuperscript{93,94} This suggests that underlying mechanisms also involve connectivity between the cortico-subcortical circuits involved in motor and higher cognitive functioning.\textsuperscript{86,87}

In \textit{Chapter 7}, we elaborated on this question by examining whether early measures of motor development and cognition were associated. By exploring longitudinal trajectories we aimed at providing more insight into possible coupling between maturing pathways involved in early motor development (expressed in movement variety) and early cognitive processing (expressed in visual attention). Those pathways are considered to involve anatomically segregated basal ganglia-thalamo-cortical circuits.\textsuperscript{95,96} Despite their anatomical segregation within the neurological structures comprising the circuit, synaptogenesis in those neurological substrates follows synchronous patterns.\textsuperscript{97} In other words, it seems that these circuits develop in parallel, which led to our hypothesis that maturational changes in movement variety and visual attention might be coupled at an individual level.

However, we were unable to show a coupling between developmental trajectories of movement variety and visual attention, which suggests that movement variety and visual attention do not follow a synchronous development but develop independently during the first 6 months. The lack of finding a close association could be interpreted in several ways. First, one might conclude that motor and cognitive development are not correlated in such early stages of development and that overlapping or strong interconnectivity between cortico-subcortical circuits should not be considered the mediating factor in which motor and cognition are related at later ages. Referring to our previous findings that an age-adequate repertoire was associated with poorer cognitive outcomes, one might also speculate that motor development and cognitive development compete for similar resources during early development. In that case, we would expect to find opposing associations between developmental trajectories. Our data, however, provided no clues for such opposing associations. Finally, we should consider that the developmental events occurring in the first 6 months might have been too variable over time for associations between movement variety and disengagement to come to light within our small study sample.

\textit{The effect of preterm exposure to the extra-uterine environment on neurodevelopment}

Infants born preterm are exposed to the extra-uterine environment during a period when the brain is still developing rapidly. If and to what extent different brain regions and their behavioral correlates might be accelerated has been a topic of research for years. No clear answers have been provided so far.\textsuperscript{98-101}

We described in \textit{Chapter 8} that developmental trajectories of movement variety were similar for fullterm and low-risk preterm infants. Probably, a longer exposure to the extra-uterine environment has no beneficial, nor adverse, effect on cortico-subcortical circuits involved in the infant’s motor development during the first 6 months postterm. This strengthens previous findings in which it was suggested that the presence of various motor patterns, like the quality of the motor repertoire, are largely dependent upon intrinsic processes.\textsuperscript{102,103}
Regarding visual attention data, it was previously reported that our group of preterm infants had a temporarily faster development of visual attention at similar postterm ages than their fullterm counterparts, suggesting that maturation of cortical circuits involved in the preterm infants’ visual and attentional development was accelerated as a consequence of their early visual experience.\textsuperscript{104}

The question remains whether the faster development of visual attention affected the normal trajectory of brain development. It has been put forward that accelerated brain development may have deleterious effects on later neuronal differentiation,\textsuperscript{105} that it may compromise the development of related neural networks,\textsuperscript{106} and that it may interfere with normal cortical organization.\textsuperscript{105} Deleterious effects of accelerated neurodevelopment could only be confirmed for comprehensive reading skills. Within the group of preterm infants, the ones with the fastest development of visual attention, i.e. the ones that may have benefitted most from the additional exposure, had poorer comprehensive reading skills later on. This particular finding suggests that the accelerated maturation of visual attention networks had disruptive effects on neural networks involved in comprehensive reading. For most of the motor and cognitive outcome measures, however, the additional visual exposure seemed not to have interfered with the ongoing development of related neuronal networks.

The results presented in the last part of this thesis suggest that developmental changes in movement variety in the first postnatal months are mainly based on endogenous maturational processes, leaving a minor role for postnatal experience. In contrast, developmental changes in visual attention in the first postnatal months were influenced by the duration of exposure to the extra-uterine environment. The accelerated development of visual attention processes seen in preterm infants, however, did not last until 10 to 11 years of age and seemed not to interfere with the ongoing development of related neuronal networks.

\textit{Limitations of this thesis}

The studies described in this thesis encountered some potential limitations. The first is the explorative nature of some studies with a large number of comparisons resulting in a great potential for chance findings (\textit{Chapters 5, 8, 9}). Nevertheless, we believe that exploring several associations in the particular studies was justified as part of a careful evaluation of a rich data set in the context of hypothesis-driven research.\textsuperscript{107} A second limitation was the lack of detailed neuroimaging data. Although cranial ultrasound visualizes most large lesion that are associated with abnormal neurodevelopmental outcome,\textsuperscript{108,109} we may have missed more subtle lesions that may also have had an impact on neurodevelopment. A third limitation is that no control groups were available for the two studies in which we assessed neurodevelopment after treatment with either high (\textit{Chapter 4}) or low doses of DXM (\textit{Chapter 3}). We, however, intentionally refrained from including a preterm control group because of the difficulty of sorting out the effects of BPD and its associated neonatal morbidities from the effects of DXM treatment. Preterm control infants (thus having BPD without DXM treatment) would be less severely ill in terms of BPD and would have differed on many other neonatal characteristics what makes it difficult to disentangle the effects of DXM on neurodevelopmental outcome. A fourth limitation is that in our longitudinal studies
the sample sizes were small which may have caused an underestimation of true associations. A concluding remark for the studies described in this thesis is that our results are all obtained from behavioral studies and therefore do not allow for direct evidence on the involvement of neurological substrates. However, our data still provide valuable insight into motor, cognitive and behavioral performances at varying stages of development and its complex interrelationships.

Conclusions, implications and future perspectives
In the first two parts of this thesis we provide insight into the effect of several risk factors in the prenatal and early postnatal period on early motor development. The third part sheds light on the complex interrelations between early motor development, early visual attention and functional outcome.

Prenatal exposure to polychlorinated biphenyls (PCBs) and some hydroxylated PCBs (OH-PCBs) led to a reduced motor development around 3 months of age, probably by a mechanism involving the disruption of thyroid hormone homeostasis. Additional research is needed to fully elucidate the possible adverse effects of (OH-)PCBs on later functioning. Follow-up testing should not only include neurological, motor, cognitive and behavioral assessments but should also include current (OH-)PCB levels and thyroid hormone status to determine if and to what extent prenatal exposure to (OH-)PCBs has lasting effects. Moreover, it should include the assessment of pubertal development as OH-PCBs are considered to have disruptive effects on reproductive hormones as well.

In an infant with MoCD type A, we demonstrated that GM quality contributed in monitoring and predicting neurological functioning after experimental cPMP treatment. Even though we did not directly relate GM quality to outcome, the marked improvement in GM quality and normal FMs at 3 months of age well agreed with the favorable outcome at 21 months of age. These observations add to the growing body of evidence that the assessment of GMs quality might be a valuable tool not only to assess neurological functioning, but it may also facilitate in predicting outcome at much earlier ages than with traditional neurological examination. An important practical implication is that the assessment of GM quality may help clinicians to recognize early signs of neurodevelopmental impairment, and consequently, they may begin early treatment in those infants that suffer from diseases that, if left untreated, will lead to severe neurological abnormalities and ultimately death.

Postnatal exposure to glucocorticoids, in particular high doses of dexamethasone (DXM), led to a consistently poorer quality of GMs as compared to controls, possibly by mechanisms including metabolic changes, altered gene transcription, decreased synaptic connectivity, and ultimately neuronal apoptosis. These adverse effects on neurodevelopment lasted at least up to 9 years at which age children showed functional impairments on multiple domains. We proposed that permanent neuronal changes in dorsal visual stream, hippocampus and cerebellum underlie some of the deficits found at school age. Treatment with lower doses of DXM seemed not to exert adverse effects on neurodevelopment, at least not until the age of two years. Our findings contribute to the ongoing debate suggesting that the use of a tapering course of DXM in starting doses of 0.25 mg/kg/day might be a good candidate regimen for
infants at risk of BPD that deserves further evaluation. We advocate for randomized controlled trials without open-label use of DXM to investigate long-term respiratory and neurological outcome after low-dose DXM treatment.

More recently, other treatment options for preterm infants at risk of BPD have become a topic of forthcoming research. These include inhaled nitric oxide therapy (iNO) combined with vitamin A supplementation,\textsuperscript{112} and stem cell-based therapies.\textsuperscript{113} Combined treatment with iNO and vitamin A supplementation in a sample of low birth weight preterm infants showed beneficial effects in reducing the risk of BPD and improving neurodevelopmental outcomes at 1 year of age when compared to infants treated with iNO alone.\textsuperscript{112} Very recently, the first clinical trial was published in which 9 extremely preterm infants at very high risk of developing BPD received an intratracheal allograft transplantation of mesenchymal stem cells derived from human umbilical cord blood.\textsuperscript{114} They had significantly lower BPD severity when compared with a historically matched comparison group. Although both new treatment strategies seem encouraging, future studies should be undertaken not only to assess long-term safety but also to compare those new treatment strategies with conventional corticosteroid treatment, in a randomized-controlled setting and sufficiently powered to control for potential confounders such as timing and dosages of corticosteroids and neonatal comorbidities.

In the third part of this thesis we demonstrated that early measures of both motor development and visual attention predicted several, and some overlapping, domains of functional outcome at school age. The mechanism by which these longitudinal associations can be explained might involve the establishment of relatively stable neural networks in the first postnatal months that are engaged in several motor, neurocognitive and behavioral functions in later life. However, in the first 6 months postnataally, we found no correlations between longitudinal trajectories of early motor development and cognitive processing, expressed in visual attention measures. This suggests that the strong association between motor and cognition development as described in literature has not been established yet in the first 6 months of life by overlapping of or a strong interconnectivity between cortico-subcortical circuits.

Both prenatal and early postnatal experiences are fundamental for the development of the central nervous system. In preterm infants, the development of their central nervous system might be accelerated by their earlier, and therefore additional, sensory-motor experiences. Previously reported findings lend support for a temporary accelerated development of visual attention processes.\textsuperscript{104} We now added that this accelerated development did not have a clear impact on later motor, behavioral or cognitive functioning, arguing against disruptive effects on related neural networks. Developmental trajectories of movement variety were not accelerated nor delayed, indicating that endogenous maturational processes predominate early motor development, leaving a minor role for postnatal experience.

Although the data presented in the final part of our thesis have enhanced our understanding about the complex interrelationships between early motor development, early visual attention and functional outcome, it has raised many questions in need of further investigation. One of
those questions is what range of movement variation should be considered normal during early
development. Future longitudinal studies on individual trajectories of movement variety and its
association with later neurological and functional outcome would be particularly helpful, as this
might not only help in interpreting the present findings but might also shed light on the issues
of intra- and interindividual variability. These are aspects of development that are considered to
be part of normal development, but if, and if so to what extent, they contribute to neurological
changes throughout infancy has yet to be determined.

Another important question that remains is whether the assessment of early motor
development and early visual attention measures might have practical implications. Our findings
on early motor development suggest that in particular the absence of variable finger postures
could help to identify those infants that might experience visual-spatial information processing
difficulties later on. Those infants might benefit from strategies aimed at improving visual spatial
skills, such as practicing manipulating and constructing objects and exploring new environments.
These should be employed at the earliest ages possible when infants might benefit most from
interventions. To improve school performance, teachers should not only encourage practicing
visual-spatial skills but they should also place more emphasis on verbal teaching instructions
rather than relying on visual displays, such as graphs and diagrams.

Our findings on early visual attention measures implicate that developmental changes in
visual attention measures may guide the identification of infants who will develop difficulties
in attentional, handwriting and motor skills. Advancements in technology have led to the
development of less complex, less expensive, and sufficiently accurate technological devices
capable of measuring eye movements, which may hold promise as a predictive tool to use in a
practical setting.

Finally, a key area of modern research should include longitudinal studies that combine early
behavioral measures with neuroimaging data to increase our knowledge about how behavioral
functions, such as early motor development and visual attention, relate to developmental
changes in the brain throughout infancy.
References


47. Yates HL, Newell SJ. Minidex: very low dose dexamethasone (0.05 mg/kg/day) in chronic lung disease. Arch Dis Child Fetal Neonatal Ed 2011;96(3):F190-4.
55. Porterfield SP, Hendry LB. Impact of PCBs on thyroid hormone directed brain development. Toxicol Ind Health 1998;14(1-2):103-120.


82. Lewis M, Brooks-Gunn J. Visual attention at three months as a predictor of cognitive functioning at two years of age. Intelligence 1981;5:131-140.
235.


