Functional MRI, structural MRI and school performance in extremely preterm/extremely low birth weight children

Silja Torvik Griffiths

Dissertation for the degree philosophiae doctor (PhD)

At the University of Bergen, Norway

22nd November 2013

SCIENTIFIC ENVIRONMENT

This thesis is part of the PhD program at the Department of Clinical Medicine. However, as the main research environment was located at the Department of Paediatrics, Haukeland University Hospital, there is also a strong link to the Department of Clinical Science, all in Bergen, Norway.

The thesis is based on a national cohort of preterm children called the Project Extreme Prematurity, and I was invited into the research group in January 2010. My main supervisor has been paediatrician and child psychiatrist Prof.Irene Elgen, co-supervised by paediatrician Prof Trond Markestad and paediatric neuro radiologist Stein Magnus Aukland (PhD).

A substantial part of the thesis has been performed in collaboration with Prof Kenneth Hugdahl, head of Bergen fMRI Group, assisted by Emanuel Neto, Hilde Gundersen, Alex Craven and Anne Marie Rød.

Evaluation of structural MRI data was performed by Kling Chong, Consultant Paediatric Neuroradiologist, Department of Radiology, Gt Ormond Street Hospital for Children, London, UK.

Parts of the work were carried out in collaboration with bio statistician Prof Geir Egil Eide, Centre for Clinical Research, Haukeland University Hospital, Bergen, Norway.

TABLE OF CONTENTS

1. ABBREVIATIONS

AAL – anatomical automatic labelling

ACC – anterior cingulate cortex

ADHD - attention deficit hyperactivity disorders

B - estimated regression coefficient

BA – Brodman areas

BOLD – blood oxygen level dependent

BPD – broncho pulmonal dysplasia

BW – birth weight

CC – corpus callosum

CI – confidence interval

CP – cerebral palsy

CT – computed tomography

DEHSI – diffuse excessive high signal intensities

DTI – diffusion tensor imaging

ELBW – extremely low birth weight (BW < 1000g)

EPI – echo planar imaging

EPT – extremely preterm (GA < 28 weeks)

FWE - family wise error

fMRI – functional MRI

GA – gestational age

GI – gastro intestinal

GM – grey matter

HUS – Haukeland University Hospital

IVH – intra ventricular haemorrhage

MBRN – Medical Birth Registry of Norway

MNI – Montreal Neurological Institute

MRI – magnetic resonance imaging

NDDs – neuro developmental disabilities

NEC – necrotizing enterocolitis

NICU – neonatal intensive care unit

OR – odds ratio

PEP – Project Extreme Prematurity

PVL - periventricular leukomalacia

RA – response accuracy

RDS - respiratory distress syndrome

ROP - retinopathy of prematurity

RT – reaction time

SD – standard deviation

SGA – small for gestational age

T – tesla

VPT – very preterm (GA 28 – 31 weeks)

WHO – World Health Organization

WM – white matter

2. SUMMARY

PURPOSE: Numbers of extremely preterm (EPT) children who survive is steadily increasing, and the majority of these children are in need of help at school.

The purpose of this regional, clinical controlled cohort study was to compare anatomy and function of the brain in EPT/extremely low birth weight (ELBW) and term born children at eleven years of age, and study the relation to school performance and cognitive skills. Brain anatomy findings were also compared to young adults born very preterm.

METHOD: A population based cohort of all EPT/ELBW (gestational age (GA) < 28 weeks or birth weight (BW) < 1000g) children born in Hordaland or Sogn- og Fjordane in 1999-2000, was the basis for this thesis. An additional cohort of very preterm (VPT, GA 28-31) young adults (19 years) was included. Both cohorts were compared to a randomly selected, age appropriate term born control group. In paper I, frequency and magnitude of cerebral brain pathology assessed by magnetic resonance imaging (MRI) in EPT, VPT and term born children/young adults were investigated. In paper II, a possible difference in blood oxygen level dependent (BOLD) activation, assessed by functional magnetic resonance imaging (fMRI) and performance during a working memory/selective attention task (the n-back/Stroop task), between EPT/ELBW and term born children was analysed. In paper III, a possible association between school performance, assessed by compulsory national school tests, scores on a working memory/selective attention task, BOLD activation during this task and prematurity was investigated.

RESULTS:

Paper I: An increased frequency of MRI pathology was found in both the EPT and the VPT cohorts compared to their respective term born control groups. The frequency was higher in the EPT than the VPT group, but also higher in the EPT controls than the VPT controls. Group differences were mainly limited to mild pathology.

Paper II: When performing a working memory/selective attention task, the eleven year old regional EPT/ELBW cohort showed the same pattern of brain activation as the term born control group, but the intensity was significantly reduced. The main areas of activation were the prefrontal and parietal areas and the anterior cingulate cortex (ACC). The EPT/ELBW children had fewer correct responses, particularly in the cognitively more demanding settings.

Paper III: There was no significant difference in school performance between the children born EPT/ELBW and at term. There was a significant positive association between correct responses on the n-back/Stroop task and school performance, independent of prematurity ($r=0.41$, $p=0.004$). BOLD activation was associated with response accuracy on the n-back/Stroop task, but not with school performance. The BOLD activation pattern in children scoring high versus low on the n-back/Stroop task was different compared to the pattern in preterm versus term born control children.

CONCLUSIONS: The same structural MRI pathology was found in subjects born EPT, VPT and at term, but less frequently in those born at term. MRI lesions were more common in the EPT than the VPT group, but also in the EPT control group compared to the VPT control group. Lack of objective criteria for differentiating mild pathology from normality at the different ages may be the cause of differences in the term born groups. The fMRI study demonstrated that working memory and selective attention capacity was reduced in EPT/ELBW children compared to term born controls, with a matching reduction in BOLD activation in areas responsible for cognitive control. High BOLD brain activation was associated with better cognitive performance. Although cognitive performance was positively associated with school performance, BOLD activation did not reflect school performance.

3. LIST OF PAPERS

3.1. Cerebral MRI findings in extremely preterm, very preterm young adults and term born controls. 2013, Pediatric Neurology

Silja T. Griffiths, Irene B. Elgen, W K Chong, Morten Duus Odberg, Trond Markestad, Emanuel Neto, Stein Magnus Aukland

3.2. fMRI – blood oxygen level dependent activation during a working memory/selective attention task in in extremely preterm/extremely low birth weight school children. 2013, Pediatric Research
Silja T. Griffiths, Hilde Gundersen, Emanuel Neto, Irene Elgen, Trond Markestad, Stein M. Aukland, Kenneth Hugdahl

3.3. Functional Magnetic Resonance Imaging, school performance and prematurity: a regional, clinical-controlled cohort study. Submitted,
Silja T.Griffiths, Stein Magnus Aukland, Trond Markestad, Geir Egil Eide, Irene Elgen, Alexander R Craven, Kenneth Hugdahl

4. BACKGROUND

4.1. Normal brain development

Anatomical development: The development of the central nervous system starts at the 16th day after fertilization with the appearance of the neural plate(1). The lateral edges of the neural plate become elevated to form the neural tube, which is the forerunner of the brain and spinal cord. Most neurons are produced between week four and week twenty, and several die before the child is even born. At the end of the fourth week, three major divisions of the brain appear (the forebrain/prosencephalon, the midbrain/ mesencephalon and the hindbrain/telencephalon), and during the 5th week secondary swellings appear and make a total of five major parts (the telencephalon (becomes the right and left cerebrum), diencephalon (becomes thalamus, epithalamus, hypothalamus and subthalamus), mesencephalon (becomes the midbrain), metencephalon (becomes pons and cerebellum) and myelencephalon (becomes medulla oblongata)). The lumen of the neural tube is the start of the ventricular system. The lateral ventricles develop on each side of the cerebral hemispheres, the third ventricle is centred in the diencephalon and the fourth ventricle boarders the medulla, pons and cerebellum. Narrow channels are formed connecting the ventricles to each other and to the lumen of the spinal channel (1).

Gyration: Folding of the brain, gyration, start at week 14, but the main process happens between week 26 and 36, which is mainly after time of birth in extremely preterm (EPT) children. What initiates and runs the process is uncertain. Several theories have been suggested, like gene control, cortical growth caused by different growth patterns of the inner and outer cortical layers, tension created by white matter (WM) axonal fibres or environmental factors (2). Being outside the uterus is thought to have a negative influence on the process, and is therefore an essential factor in brain development and function in EPT children.

Blood vessels: Around week seven to eight, the first blood vessels are developed in the human embryo. Blood vessels in the germinal matrix are fragile temporary vessels with a different microstructure to the more permanent vessels at term birth (3). The germinal matrix has the highest density of blood vessels and percentage of blood vessel area , followed by grey matter (GM) and then WM. Density and blood vessel area both increase with increasing GA. Haemorrhages in these areas are therefore more common in preterm than term born children (4). Lack of or immature auto regulation of cerebral blood flow has also been suggested as a possible mediating factors in rupture of blood vessels in the premature brain (5), though some authors suggest that the relationship between pCO_2 and blood pressure is more important than pressure and blood flow (6).

Brain size: At term birth, a normal brain weighs about 400g. Additional weight after this time is due to continued synaptic connections being made, development of more neuroglia, and development and thickening of the myelin sheath around the nerves. The brain contains about 100 billion neurons at birth, continuously making new synaptic connections and eliminating inefficient synapses (7). At three years of age the weight of the brain is almost that of the adult brain, but slow growth continues until 18 years. The total brain volume reaches adult size at six to eight years, and a decline in brain size starts after about 50 years (8).

Grey matter (GM), white matter (WM) and corpus callosum (CC): GM volume increases about 13% from early childhood (19-33 months) to late childhood (6-8yr), before the total GM volume decreases slowly throughout life. WM volume, however, increases with about 74% from early childhood to adolescence (12-15years) before it increases slowly to its maximum level around the fourth decade, and then very slowly decrease (about 13% total decrease at the age of 70-80yr)(8).
There is a trend towards lower brain volume, lower cortical surface area, lower WM volumes but equivalent sulcation in females compared to males (2).

4.2. Brain injury

A critical issue for preterm infants is how the immature tolerates the impact of birth and life outside the uterus, as the preterm brain is fragile and susceptible to injury. Particularly highly vascular areas and periventricular WM are at risk of haemorrhage and ischemia. As quoted: 'Infants born at between 22 to 25 weeks of gestation are fragile and vulnerable and at high risk for brain injury due to hypoxia and ischemia and malnutrition, as well as sepsis, which starts the cascade of events that increase the risk of brain injury with haemorrhage, white-matter injury (periventricular leukomalacia (PVL) and ventriculomegaly), and poor brain growth, and for subsequent neurodevelopmental impairment '(9).

Few theories on why intra-ventricular haemorrhages (IVH) appear mainly in the germinal matrix have been presented, and the pathogenesis of IVH in preterm children is not well understood. Ballabh et al found that vessel density and percentage of blood vessel area in the germinal matrix, WM and GM increase with increasing gestational age (GA) (3). The increased vascularity of the vessels found in the germinal matrix compared to WM and GM may explain why haemorrhages frequently appear in this area. The severity of the haemorrhage is normally classified according to Papile's classification of cerebral haemorrhages, although this is a computed tomography (CT) classification (10). PVL is the most common WM lesion in preterm infants/children, and is characterized by necrosis, inflammation and often cyst formation. The pathophysiology is poorly understood, but it is suggested to be caused by ischemia as the areas around the ventricles (the water-shed areas) has a relatively low vascularity (4, 5). PVL can be either cystic or non-cystic, and is normally classified according to De Vries ultrasound based classification (11). Multicystic PVL is a strong predictor of later cerebral palsy (CP) (12). Both haemorrhages and ischemia are associated with loss of brain volume, and subsequent increase in ventricular size (13).

Diffuse excessive high signal intensities (DEHSI) are common findings in MRI scans of EPT infants at term age, and will normally disappear within the first months of life. Possible causes are vasogenic oedema, reduced axonal diameter, oligodendrocyte

damage or delay in maturation (14). DEHSI are not thought to have any impact on later neurodevelopmental outcome.

4.3. Prematurity – low birth weight

In 1948, The World Health Organization (WHO) defined preterm birth as birth before week 37, and this definition is still valid (15). 15 million babies are born preterm every year, and it is the leading cause of new born deaths (within the first four weeks after birth) globally according to the WHO. North America and Africa have the highest preterm birth rate registered, with 10.6 and 11.9 per cent respectively, while Europe has the lowest rate with 6.2 per cent (16). Numbers of children actually registered as born premature (live and stillbirths) are increasing in most countries (both high and low income countries) due to better measurements of GA, increases in maternal age, underlying maternal health problems (like diabetes and high blood pressure), increased use of infertility treatment (initially a strong increase in rate of multiple births, but now declining again, Årstabeller for medisinsk fødselsregister 2011, Folkehelseinstituttet) and a higher use of caesarean section before term among others (15).

4.3.1. Definitions

Over the years there has been a difference in opinion concerning how to define prematurity. In 1948, WHO defined all living infants with birth weight (BW) below 2500g as immature, and GA below 37 weeks as premature. The two terms were more or less interpreted as equivalents. As ultrasound was not in common use to determine GA before the 1990s, BW became the commonly reported measure in studies of preterm children. Whether GA or BW is the best parameter for defining preterm children is debatable; some argue that neither one is sufficient on its own (17, 18). A

challenge using both GA and BW, however, is that the mechanisms behind abnormal brain anatomy or function can be based on very different degrees of maturation despite similar weight (19). Mature children born 'small for gestational age' (SGA, often defined as below the 10thcentile for weight), will be included in the same group as more immature children with the same BW. The most commonly used classification system at the present covers both GA and BW (Table 1).

Table 1 Definitions of prematurity according to gestational age or birth weight

	Gestational age, weeks	Birth weight, grams
Micro premature	< 26	< 750
Extremely low GA/extremely preterm	< 28	
Extremely low birth weight		< 1000
Very low GA/very preterm	28 - 31	
Very low birth weight		< 1500
Low GA/preterm	< 37	
Low birth weight		< 2500
Late preterm	34 - 36	
Term	37 - 42	≥ 2500
Post term	> 43	

In Norway, the number of EPT children born has been stable at 0.4% a year since 1970, but the number of EPT infants surviving the first year of life is increasing (Medical Birth Registry of Norway).

4.3.2. Aetiology

Preterm birth can start either with spontaneous labour (around 45%), with preterm and pre labour rupture of membranes (ca. 25%) or be started by medical personnel due to complications (ca. 30%) (20, 21). For most spontaneous preterm births the cause is unknown, but there are several known risk factors. Some of these are of maternal origin like socioeconomic demographics, systemic illness, nutritional status, stress and genetic factors. Other factors related to obstetrics and pregnancy like infections,

multiple pregnancies, cervical insufficiency, vaginal bleeding, previous preterm birth, uterine surgery, maternal smoking, maternal age and short interval between pregnancies can all influence length of pregnancies (20-22). Additionally, the fact that ultrasound scans are used more frequently as a measure for expected date of delivery have resulted in lower estimations of GA (23).

4.4. Prematurity and outcome

Challenges related to EPT/extremely low birth weight (ELBW) infants in the neonatal period are numerous, and often persist into childhood, adolescence and adulthood. However, children born EPT during the last two-three decades have better outcome than cohorts born earlier due to the major developments in neonatal medicine (24). As the rate of surviving preterm children is increasing, so does the economic costs for society due to neonatal morbidities and deviant long term outcome. Already in 1978-88, the cost of hospital admissions alone during the first ten years of life in children born earlier than 28 weeks GA ten times exceeded the price of a term born child, and will have increased along with improved advanced life support measures over the last two decades (25).

4.4.1. Neonatal morbidities

The chance of surviving the first year after preterm birth is highly dependent on place of birth. While 57% of EPT and 92% of very preterm (VPT) infants born between 2001 and 2010 in Norway survived the first week, according to the Medical Birth Registry of Norway (MBRN), children born before week 32 have limited chance of survival in many low income countries (16). Survival rates the first year of life if born EPT are lately reported to be 50 -75% (18, 26, 27). Year of birth is another important

factor, as preterm birth during the last two decades has improved compared to earlier years due to e.g. increased use of antenatal steroids, surfactant therapy, increased use of assisted ventilation and a change in attitude towards who to save (24, 28) .

The most common neonatal morbidities are infections (sepsis), respiratory problems, problems caused by immaturity of the gastro-intestinal (GI) tract, retinopathy of prematurity (ROP) and brain injury (chapter 4.2). Neonatal sepsis, or sepsis within the first week of life, is reported in two to seven per cent and is a major risk factor for death and neonatal morbidity in EPT/ELBW infants (29, 30). Late onset sepsis (> 6 days, before discharge from neonatal intensive care unit (NICU)) is reported in 20 % of preterm children in the Project Extreme Prematurity (PEP) and in 11 – 32 per cent in other studies, and the incidence increases with decreasing GA (30-32). Whether sepsis during an early stage can influence later neurodevelopmental outcome is still uncertain, but several studies have reported an association (32, 33).

Lung disorders are perhaps the most recognized neonatal morbidities. The initial stage is respiratory distress syndrome (RDS) where structural immaturity (as the formation of the alveoli is incomplete) and lack of alveolar surfactant lead to alveolar collapse. The incidence is increasing with decreasing GA (34). Bronco-pulmonary dysplasia (BPD) is defined as continued dependency on oxygen or assisted ventilation at four weeks post birth, or more commonly today as such dependency at 36 weeks' postmenstrual age. Risk factors are prematurity, oxygen treatment and mechanical ventilation (35). In the PEP cohort the incidence of BPD was 67% for children with GA < 26 weeks, and 37% for children with GA 26 - 30 (36). The use of postnatal steroids during treatment of infants with lung disease is common, but caution has been advised as an association with poorer neurodevelopmental outcome has been found in recent studies (37-39).

Immaturity of the GI tract can lead to necrotizing enterocolitis (NEC), causing necrosis of tissue in the GI tract. The reported incidence is eight to nine per cent, and it is thought to be related to later neurodevelopmental outcome when surgically treated (40, 41). Retinopathy of prematurity (ROP), the main cause of reduced vision and blindness in preterm children, is characterized by vascular abnormalities in the

developing retina, and it was found in 60 per cent of children with GA = 24 weeks, and 24 per cent in GA = 27 in the national PEP study (39). There is a strong association between oxygen treatment and ROP (42). Sensorineural hearing loss is found in about 6% of children with BW < 750g , and the recognized neonatal risk factors are asphyxia, ototoxic drug exposure, hyperbilirubinemia, neuro infections and IVH (43).

4.4.2. Neuro-developmental Disabilities (NDD)

Preterm children surviving the neonatal period have an increased risk of severe neurodevelopmental disabilities (NDDs) like CP, blindness, deafness and learning disabilities compared to the general population. IQ levels, cognitive capacity and learning disabilities versus difficulties are described under chapter 4.4.3. In a Swedish study, Johnson et al. found severe functional disability (CP, impaired neuro-motor function, vision- and hearing impairments) in 45% of children born < week 26 compared to 1% in classmates, and that only 50% of EPT are free of disability at 11 years of age (44). It has been suggested that the increased survival rate of EPT/ELBW children has led to an increase in the rates of impairments and disabilities, but recent studies have found the contrary (45). The incidence of CP increases with decreasing GA, and was found in 9 % EPT compared with 0,1% in term born children in a large Norwegian national epidemiological study of subjects born during 1967 - 82 (46). Other studies have found similar ratios, ranging from 7 to 17 per cent (44, 47). In the Norwegian PEP-study the incidence was 7% in children born EPT, but within this group the incidence was 4% for survivors born at GA 26-27 weeks vs 15% for those born at GA 23-25 weeks (48). Severe visual impairment in EPT children is normally a result of ROP, and is seen in 2 to 9 per cent depending on GA below 26 or 28, and at what time the cohorts in the studies were born (39, 44). Hearing impairment is reported in 1 to 6 per cent in recent studies, dependent on how the hearing impairment

is classified (39, 44, 47, 49). Most of the major NDDs are discovered before two years of age (39).

Mental problems from school age described as attentional, emotional and peer problems are also accepted long term risks in EPT children (50). Attention deficit hyperactivity disorders (ADHD)(51), autism spectrum disorders (46) and mental health problems in general (52) are all reported in increased ratios in preterm children compared to term born control children. Mental health problems and learning difficulties are mainly diagnosed at school age and later.

4.4.3. Cognitive skills – working memory/selective attention

Two important aspects of cognitive skills are working memory and selective attention, both essential to school performance. The term working memory can be interpreted as the ability to temporarily store and process incoming information in order to restore it within a short interval of time (53). Selective attention is the ability to ignore one incoming stimulus in order to focus on another incoming stimulus (54).

This thesis uses the term learning disabilities in its English interpretation, meaning mental retardation or IQ level below 70 IQ points ('Mental retardasjon' in Norwegian, http://www.nhs.uk/Livewell/Childrenwithalearningdisability/Pages/Whatislearningdisability.aspx), and must not be confused with learning difficulties, which refer to milder problems concerning the ability to learn (e.g. dyslexia, inattention) and are not necessarily linked to IQ level.

Impaired cognitive skills have been recognized as one of the main challenges for preterm children not suffering from any major NDDs. Cognitive performance at school age is directly proportional with the GA at birth (55). However, a review of cognitive skills in EPT children born from the 1960s and into this century concludes that being born after the year 2000 increases the chance of having better cognitive skills than being born earlier (56).

4.4.4. School performance in EPT/ELBW children

Thirty per cent of EPT/ELBW children born in 1985-1990 performed at grade level in school without educational support in a study from Australia (57), and two thirds of children with GA < 26 weeks needed extra help at school according to the Epicure study in Great Britain (58). A Norwegian national epidemiological cohort study of preterm children born in 1967 – 83 found that two thirds of EPT graduate from high school, compared to three fourths of term born children, and one fourth completed university or bachelor's degree compared to one third of term born controls (46). Several studies find low cognitive skills or low school performance in preterm children (39, 55, 59).

4.5. Cerebral magnetic resonance imaging (MRI)

MRI is primarily used for visualizing the organs and structure/tissues of the body. The unique benefit compared to CT is that there is no ionizing radiation and that it provides greater contrast between soft tissues and thereby improve image clarity. MRI is based on the magnetic moment and spin of the atomic nuclei of the hydrogen molecule (the brain consists to a large extent of water) when placed in a magnetic field. In a homogenous magnetic field, the spins will be parallel to the field axis, and applying additional energy in the form of high frequency radio pulses will dislocate the orientation of the spins. As the extra energy is gradually absorbed by the surroundings, the protons will realign with the magnetic field again, and the time it takes to realign can be measured and used to differentiate between tissue textures. Strong magnetic fields are used to create images of biological tissue, and the field strength is expressed as Tesla (T). Most scanners used for humans are 1,5T or 3T, but even 7T are now in use (in comparison, the magnetic field of the earth is 0,00005 T). To generate a static magnetic field, a series of large electromagnetic coils carrying large currents are placed around the core of the scanner (60).

The very start of MRI can be traced back to an Austrian physicist named Wolfgang Pauli in the 1920s, followed by the discoveries by the American Nobel Prize winners Isidor Rabi (1944), Felix Bloch and Edward Purcell (separately, in 1952) among others(60). Raymond Damadian first suggested using Nuclear Magnetic Resonance measurements for biological material and was the first to build a full body MRI scanner (23). Some say he was passed over for the Nobel Prize in Physiology and Medicine 2003 by Paul Lauterbur and Peter Mansfield who introduced magnetic field gradients, which made it possible to produce images (61) and developed the echo planar pulse sequence (62) necessary for the fast image acquisitions used in fMRI, respectively. As MRI scanners became commercially available to hospitals in the 1980s, the scanners became available for structural initially and then functional MRI research from 1992 (60).

4.5.1. Structural MRI and prematurity

For preterm infants, the most common pathological cerebral findings in MRI scans are haemorrhages, focal WM pathology including DEHSI, reduced size of CC and ventricular dilatation (14, 63-69). When the children grow older, the DHESI, haemorrhages and ischaemic areas vanish but often leave sequelae in the form of PVL, increased ventricular size (also from other causes) and focal WM pathology. Regarding long term outcome, PVL is one of the main predictors of later NDDs. Several other MRI outcome measures are also related to reduced neuro developmental outcome in preterm children, like increased ventricular size and diffuse WM abnormalities (14, 70). The connection between pathological MRI findings and mild cognitive impairment is not clear, but it is uncommon that preterm children with severe neuropsychological impairment or psychiatric disorders have a normal MRI scan (71). Reductions in the size of CC and differences in the ratios of WM and GM are other common cerebral MRI findings in childhood that are related to poorer cognitive outcome (72, 73).

As adolescents, signs of preterm birth may still be visible on MRI scans in the form of increased ventricular size, reduced size of CC, reduced bulk of WM and some focal WM pathology (71, 74-79).

So far, the MRI studies referred to have mainly been of VPT children or mixed groups of preterm children. Regarding the selected group of EPT children, information is limited as they enter school age. An overview of studies up to 2012 is given in Table 2.

Table 2 Cerebral MRI findings in extremely preterm infants and children found via a Pubmed search using the terms extremely preterm/premature, cerebral and MRI up to 2012

Author	Year	Cohort	Controls	Results
Maalouf et al.	1999	UK, 1997-98, <30weeks, infants, n=41	Yes	High incidence of persisting WM anomalies compared to term infants
Felderhoff-Mueser et al.	1999	UK, 23-28weeks, extremely sick infants, n=7	No	Conventional MRI was able to detect structural changes like haemorrhages, but not more subtle histological abnormalities
Ajayi-Obe et al.	2000	UK, Extremely preterm, infants, n=14	Term born	At 38-42 weeks, cerebral cortex had less cortical surface and was less complex than in term born infants
Messeschmidt et al.	2005	Austria, 1988-2004, <30weeks,<1500g, infants, n=28	No	Severe reduction in cerebellar volume with symmetric involvement of cerebellum
Bodensteiner et al.	2006	USA, 50 GA<28 and <1000g with CP, children, n=50	No	Decrease in cerebral WM without gliosis
Dyet et al.	2006	UK, 1997-2000, infants, GA<30weeks, n=119	No	Diffuse WM abnormalities and ventricular dilatation common at birth, correlate with reduced developmental quotients
Horsch et al	2007	Sweden, 2004-2005, <27weeks, infants, n=51	No	2%severe WM abnormalities, 16%moderate, 8% GM abnormalities
Fumagalli et al.	2009	Italy, <28weeks or <1000g, cerebellar bleeding, n=9	66 ELBW without cerebellar bleeding	Cerebellar haemorrhages seem to effect the development of pons in the youngest GA
Skiold et al.	2010	Sweden, 2004-07, <27weeks, n=129	16 term born, caesarean section	14% had moderate or severe WM abnormalities, subtle WM changes verified by DTI in the majority of the extremely preterms
Taylor et al.	2011	USA, 1982-86, <750g, infants, n=37	36 term born	Reduced volume subcortical GM, WM and cerebellum in preterm children

4.5.2. Functional fMRI and prematurity

In its traditional form, fMRI is a neuroimaging technique where the participant performs a task (cognitive, sensory, motor, emotional, etc.) while MRI scans are continuously acquired and synchronized to the presentation of a stimulus or instruction, and thereby to the initiation of a cognitive, sensory or other process. This allows for studying brain function over time, and for investigating neural correlates to mental processes. Choosing the right fMRI task in order to understand and be able to interpret the resulting brain activation maps is therefore of major importance.

fMRI is based on the so called Blood Oxygen Level Dependent (BOLD) contrast which reflects the differences in magnetic properties of oxygenated (diamagnetic) versus deoxygenated (paramagnetic) haemoglobin (Hb), or oxygen extraction. During baseline conditions, oxygenated Hb is converted to deoxygenated Hb at a constant rate in the capillary bed. Oxygen demand in the tissue is increased when neuronal activity is increased, thereby increasing the blood flow to the brain region where metabolic demands are rising (e.g. when the neurons in that region are engaged in processing a specific stimulus or task). The increase in blood flow to the critical region(s) exceeds the metabolic oxygen consumption about four-fold, causing a relative decrease in the amount of deoxygenated Hb, which can be recorded as a difference in magnetic susceptibility in MRI images (80). The BOLD contrast is therefore providing an indirect measure of neuronal activity, but with relatively high spatial resolution (60).

fMRI imaging has been used to study for example somatosensory (81), sensorimotor (82) and a resting state (83) network(s) in preterm infants, but there are natural limitations to which tasks one can apply in infants and small children. Peterson et al found that eight year old preterm children (GA < 33 weeks) processed a story with a semantic content the same way as term born children processed a nonsense story (84). Different neuronal pathways for language processing in preterm compared to term born children have been supported also by other authors (85-88). Although some studies report signs of alternative pathways, the overall message from fMRI studies of

preterm children is that they show reduced brain activation in task related areas of the brain compared to their term born peers, and they score below their term peers on cognitive tasks (84, 89). fMRI studies of preterm children up to 2012 is summarized in Table 3.

Table 3 Functional MRI studies in preterm children up to 2012.

Author, year	Preterm cohort	Controls	Method - task	Results
INFANT STUDIES				
Heep et al. 2009	Germany, 24-30weeks, n=5, infants	No	passive sensorimotor stimulation	fMRI possible in infants
Ariachi et al. 2009	UK, 2008-2009, 25-36weeks, n=32, infants	Term born, n=8	passive somatosensory stimulus	Possible to reliably identify BOLD signals in infant the brain
Smyser et al. 2010	USA, <38weeks, n=90, infants	Term born, n=10	resting state	Identified resting state networks demonstrate a regionally age-specific pattern of development
Doria et al. 2010	UK, 25-35weeks, n=62, infants	Term born, n=8	resting state	Resting state networks develop with different trajectories, the repertoire of resting state dynamics during rapid neural growth in last trimester
Kalpakidou et al. 2012	UK, 1979-84, <33weeks, n=41, infants	Term born, n=17	verbal associate learning task	Right frontal and right parietal brain activation decrease as the severity of neonatal brain injury increase
Lee et al. 2012	Canada, mean 29weeks, n=36, infants	Term born, n=23	visual flash	No reliable BOLD signal from visual flash in infants, not appropriate for occipital lobe function testing.
Smith et al. 2011	USA, 2008-09, <30weeks, n=44, infants	No	stressors	Exposure to stressors associated with alterations in functional connectivity
LANGUAGE STUDIES				
Peterson et al. 2002	USA, 26-33weeks, n=24, 7-9yr	Term born, local community, n=13, + 4 adults	language	Preterm children used same pathway for semantic story as term children for meaningless story

Rushe et al. 2004	UK, 1979-80, preterm male with corpus callosum thinning, n=6, 18yr	Term born, n=6	phonological processing	Increased frontal and decreased occipital activation.
Ment et al. 2006	USA, 1987-89, 27-35weeks, n=14, 12yr	Term born, local community, n=24	passive language	Different neural systems for processing language
Ment et al. 2006	USA, 1987-89, <33weeks, n=47, 8yr	Term born, n=24	passive language test	Positive effect of indomethacin treatment on language function in preterm children
Schafer et al. 2008	USA, 1987-89, 600-1250g, n=22, 12yr	Term born, local community, n=26	semantic association task	No performance difference, similar activity, connectivity analysis revealed different neural pathways for lexical semantic pathways
Nosarti et al. 2009	UK, 1983-84, <33weeks, n=28, 20yr	Term born, n=26	letter fluency task	fMRI activation differences only partly explained by structural changes, may reflect functional plasticity.
Gozzo et al. 2009	USA, 1987-89, 27-35 weeks, n=54, 9yr	Term born, local community, n=24	auditory language task	Employ neural systems for auditory language function differently to term controls
Myers et al.2010	USA, 1987-89, 27-35weeks, n=31, 16yr	Term born, local community, n=36	language	Preterm adolescents engage a dorsal right hemisphere region for language.
Lawrence et al. 2010	UK, 1983-84, <33weeks, n=22,	Term born, advertisement, n=22	verbal associate learning task	Preterm birth leads to functional neuronal differences at adulthood
Barde et al. 2012	USA, 27-35weeks, n=18, 9-14yr	Term born, n=14	auditory sentence comprehension	Preterm birth modulates brain-behaviour relations in sentence comprehension as task demands increase
MEMORY STUDIES				
Maguire et al. 2001	UK, GA 26weeks, male with bilateral hippocampal damage, n=1, 22yr	Term born, n=6	memory retrieval	Despite 50%volume loss of hippocampus, the preterm male showed increased bilateral BOLD signal in hippocampus during task
Gimenez et al. 2004	Spain, 1982-84, <34weeks, n=14, 12-18yr	Term born, n=14	memory task	Contralateral compensatory activation mechanisms when volume decrease in left hippocampus in preterms

Curtis et al.	USA, 1987-89, 27-35weeks, n=9, 12-15yr	Term born, friends, n=9	memory task	Increased signal in right caudatus in preterms during encoding, no difference in in-magnet behavioural data
RESTING STATE STUDIES				
Damaraju et al.	USA, <1500g, n=16 at 18months, n=13 at 36months	Term born, n=9 at 18 and 36 months	resting state	Resting state networks well developed at 18months, no group differences at 18 or 36 months
Constable et al. 2012	USA, 27-35weeks, n=19, 20yr	Term born, local community, n=19	resting state	20years after preterm birth still alterations in functional organization of the brain
STUDIES OF EXECUTIVE FUNCTIONS				
Nosarti et al. 2003	USA, 1983-84, <33 weeks, male ,n=8, 14-15yr	Term born, right handed, advertisement , male, n=8	response inhibition	Different BOLD signals as task response in preterm sample, possibly alternative pathways
VISUAL AND AUDITORY STUDIES				
Santhouse et al. 2002	UK, <33weeks, corpus callosum damage, n=7, 18-20yr	<33weeks, no corpus callosum damage, n=9 and term born, n=7	auditory and visual	Early damage to corpus callosum in preterms gives a different neural strategy for forced callosal transfer
Narberhaus et al. 2009	UK, 1983-84, <33weeks, n=21, 20yr	Term born, n=22	visuo-perceptual learning	Activate different neural networks during mnemonic processing og visuo-perceptual material, indicating neural compensation

5. INTRODUCTION TO THE THESIS

5.1. Project Extreme Prematurity Norway (PEP Norway)

Project Extreme Prematurity Norway (PEP Norway), which this thesis is based on, is a national project initiated by Professor Trond Markestad (the Pediatric department of Haukeland University Hospital, 1998). In collaboration with the Norwegian Birth Registry, all children with GA < 28 weeks or with a BW < 1000g, born in Norway in 1999 or 2000, were registered. In this two year period, 638 EPT/ELBW children were born, 485 of them were born alive, and 373 were alive at two years of age. This national cohort has previously been invited to participate at two and five years of age (Figure 1).

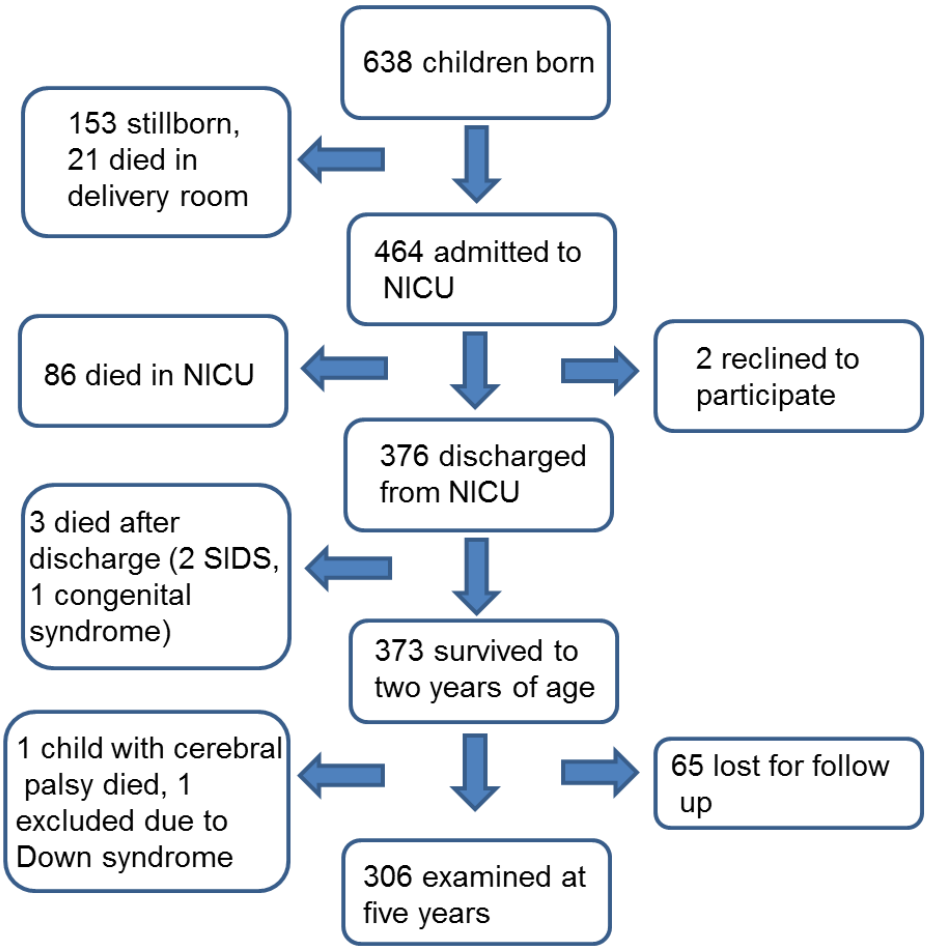


Figure 1 Included and excluded children from the national Project Extreme Prematurity: all children born in Norway in 1999 or 2000 with BW < 1000 or GA < 28 weeks.

Pre-, neo- and post natal data were recorded. The assessment at two and five years of age involved parental questionnaires regarding physical and mental health, and a general clinical and neurological examination by a local paediatrician. Pre-defined developmental milestones were used to assess developmental stage at two years of age. At five years of age the children were also subjected to cognitive tests (WPPSI-R) and parental assessment of the Strength and Difficulties Questionnaire (SDQ, www.sdqinfo.com).

At two and five years of age the rate of major NDDs (like CP, blindness and deafness) was 8 and 12 per cent, respectively, and the rate was increasing with decreasing GA (39, 48, 90). At five years of age, extreme prematurity was found to be associated with increased risk of mental health problems (38 per cent had a Total Difficulty Score above the 90th percentile on the SDQ compared to 11 per cent in term born control children)(52). The risk was higher in EPT children with NDD, but even preterm children without NDD had higher risk of mental health problems than term born control children.

5.2. Aims of the study

The overall purpose of this thesis was to investigate cerebral anatomy and function in extremely preterm (GA < 28 weeks) or ELBW (< 1000g) children at the age of eleven compared to term born children. The main focus was the following aspects:

I. To investigate possible differences in magnitude and frequency of MRI findings in a cohort of EPT and VPT compared to term born control subjects.

II. To describe group differences in fMRI BOLD activation patterns and task performance in EPT/ELBW 11 year old children during a working memory/selective attention task.

III. To investigate the association between school performance, working memory/selective attention task performance and BOLD activation during the task, and if prematurity *per se* is an influencing factor.

6. STUDY DESIGN

6.1 Population

All papers in this thesis are based on a regional cohort from the national PEP Norway study. At eleven years of age, 371 children from the original population were eligible for follow up, and 232 (63%) consented to participate. The regional EPT cohort included in this study comprised the 40 EPT/ELBW children born in the counties of Hordaland and Sogn- og Fjordane (Figure 2).

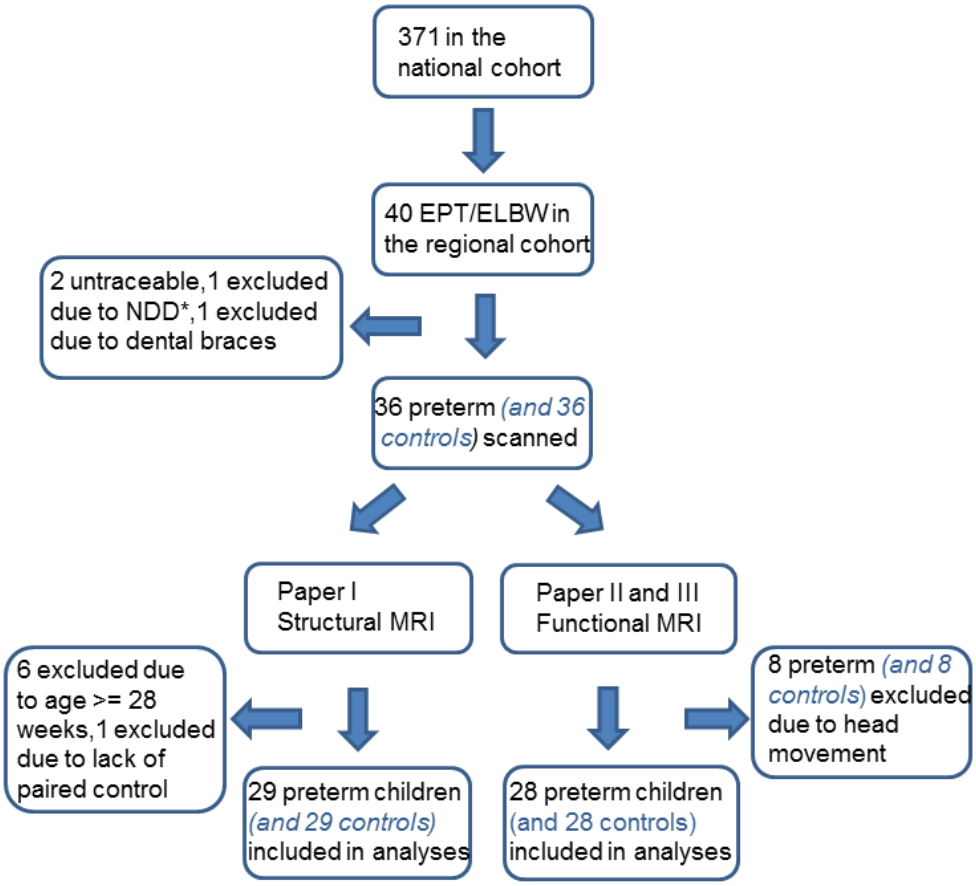


Figure 2 The regional cohort of EPT/ELBW children born in Hordaland or Sogn- og Fjordane, Norway 1999/2000, BW < 1000g or GA < 28 weeks from paper I-III, Project Extreme Prematurity.

*) Participant with NDD and a magnetic shunt.

From a total of 40 preterm children in the regional cohort, 38 agreed to have the MRI scan, and the last two we did not manage to trace. One child had to be excluded as she has a magnetic shunt, and therefore could not be scanned in a hospital without a neurosurgeon on call (additionally, her neurological disabilities would not enable her to follow the instructions of the fMRI task). Controls for the 37 preterm children were selected in cooperation with the Obstetric Department, Haukeland University Hospital

(HUS). The study nurse made a list of the three first children of the same sex born at the Obstetric Department, HUS, after the birth of a preterm child (all born at HUS) participating in the study (inclusion criteria: GA > 37, birth weight > 3000g). The family of the first term born child on the list was first invited to participate. If they declined the next child on the list was approached. All invited controls received an equivalent letter to the EPT/ELBW children, containing information, a questionnaire and a consent form inviting them to participate as a control. Out of 60 controls invited, 38 agreed to participate, but one child subsequently declined before scanning due to illness in the family, one child was excluded due to dental braces disturbing the images, and 36 were scanned.

Population Paper I

Structural anatomy of the brain was assessed in regional cohorts of EPT, VPT young adults and respective control groups.

The EPT group was the children with GA less than 28 weeks from the Norwegian PEP study born in Hordaland and Sogn og Fjordane. They were assessed at 11 years of age along with individually matched controls (date of birth, place of birth and sex). Out of 30 eligible preterm infants, one was excluded from analyses because of lack of a matched control Figure 2, Table 4).

The VPT cohort was the subjects born with GA 28-31 weeks (n =51) extracted from a cohort of 19 year old subjects with BW less than 2000g born in Hordaland county. They were examined by members of the research group at 19 years of age along with a control group born at term (78). The control group was the original control group of 100 subjects included in Odberg's study, i.e. they were of the same age and with the same distribution of first vs. later born children (78, 91). Descriptive data of the two preterm cohorts are given in Table 4.

Table 4 Clinical characteristics of the study cohorts

Gestational age (GA) group	GA < 28 weeks	GA 28 - 31 weeks
Number of subjects	**29**	**51**
GA, weeks (SD)	26 (±1.0)	30 (±1)
Birth weight, grams (SD)	900 (±168)	1436 (±308)
Males, n (%)	17 (59)	23 (45)
Scan age, years (SD)	11.1 (0.5)	19.1 (0.8)
Mothers education ≤ high school, n (%)	11/26 (42)	33/43 (77)

Population Paper II and III

From the original cohort of 40, 37 EPT/ELBW children were scanned. One child was excluded due to dental braces, and 8 preterm and 8 control children were excluded from the analysis during the pre-processing of the data due to excessive head movements disrupting the quality of the fMRI Images (translation and rotation was set to 5 mm each). Thus, 28 EPT/ELBW and 28 control children were included in the analyses (Table 5).

Table 5 Characteristics of the 28 EPT/ ELBW children and 28 controls (regional PEP cohort) included in the fMRI study.

	EPT/ELBW n=28		Controls n=28		p
Boys, *n (%)*	15	(54)	16	(57)	0.793
Gestational age, weeks[a]	27	(1.3)	>37	-	-
Birth weight, grams[a]	898	(154)	3737	(413)	<0.001
Scanning age, months[a]	132	(6)	133	(6)	-
Left handedness[a], *n (%)*	3	(11)	3	(11)	-
PVH[b], *n (%)*	4	(14)	-	-	-
Neurodevelopmental impairment[c], *n (%)*	1	(4)	-	-	-
High maternal education[d], *n (%)*	12	(43)	10	(36)	0.661
School performance[a,e],	52.5	(25.0)	57.1	(19.2)	0.487
Response accuracy[a]	310.5	(44.4)	341.3	(49.7)	0.018
Top 50 group[f], *n (%)*	9	(32)	19	(68)	0.015
BOLD[a,g]: cingulum	0.044	(0.014)	0.049	(0.013)	0.249
right inferior parietal	0.033	(0.011)	0.038	(0.012)	0.437
right inferior frontal	0.033	(0.009)	0.035	(0.010)	0.438
total 2-back minus 1-back	0.001	(0.016)	-0.003	(0.013)	0.264

a) Mean and one standard deviation, b) peri ventricular haemorrhage, all of mild degree, c) cerebral palsy, mental retardation or severe sensory deficits d) defined as at least four years of college or university training, e) National school results from 24EPT/ELBW and 23 control children, f) Children with response accuracy in the top 50% of scores on the n-back/Stroop task, g) Total BOLD activation in the three ROI's.

Previously obtained data on morbidities and outcome up to five years of age were available on all 37 scanned EPT/ELBW children (Table 6). There was no statistically significant difference between the 9 excluded subjects and the 28 participants regarding sex, handedness, GA, BW, degree of neurological disabilities or level of mother's education.

Table 6 Comparison of characteristics of the included versus the scanned cohort of EPT/ELBW children in paper II and III, recorded at five years of age. Numbers of children (per cent) with the different characteristics, and mean (SD) for IQ.

	37 EPT/ELBW children	28 included EPT/ELBW children
Boys	22 (59)	15 (54)
Cerebral palsy	2 (5)	1 (4)
Patent ductus arteriosus	21 (57)	16 (57)
Necrotizing enterocolitis	2 (5)	1 (4)
BPD	16 (43)	13 (46)
Impaired vision	9 (24)	7 (25)
Impaired hearing	6 (16)	3 (11)
SGA	10 (27)	5 (18)
IQ < 85	16 (43)	10 (36)

Though seven of the included children had impaired vision and three had reduced hearing, all children were able to see the computer screen clearly through the Lcd goggles, all could hear messages through the ear phones, and none of them revealed any sign of colour blindness during the pre-scanning computer trial.

6.2. MRI

6.2.1. MRI protocol

All the anatomical data from the EPT/ELBW group were collected at Haraldsplass Deaconess Hospital (Bergen, Norway) during the period of October 2010 to July 2011. Examinations were performed without sedation on a GE Signa HD 1.5 Tesla® (Milwaukee, USA) MRI scanner. The MRI protocol included a sagittal T1-3D fast spoiled gradient inversion recovery sequence and an axial T2 weighted propeller sequence using standard parameter settings. For the fMRI data, an EPI sequence was used for the BOLD responses. The parameters of the EPI sequence were: TR 3000 ms, matrix 64 x 64, FOV 240 mm, number of slices 28, with slice thickness 5 mm, no gap, voxel size 3.75 x 3.75 x 5 mm. There were fourteen EPI scans per block, and eight blocks, making a total of 112 scans. Five initial dummy scans were discarded before the data was analysed.

The anatomical MRI examinations of the VPT cohort (at a different time) were performed without sedation at the Department of Radiology, Haukeland University Hospital, Bergen, Norway during the period of January 2006 to May 2007, using a GE Signa Excite HD 3.0 T® (Milwaukee, USA) scanner. The MRI protocol included a sagittal T1 weighted volume sequence (Spoiled Gradient Echo (SPGR) and an axial T2 weighted sequence (Fast Spin Echo (FSE)).

Four cerebral parameters were evaluated in the MRI scans; 1) dilatation of the ventricles, 2) reduction in the bulk of WM, 3) the presence of focal WM pathology and 4) thinning of the CC. Reduction in WM bulk was judged on an overall assessment of all WM volumes including subcortical, periventricular, capsular, brainstem and cerebellar WM. The assessment was subjective, and performed by one person without any background knowledge of the subjects other than EPT and VPT cohort. Within each cohort patient vs. control status was not revealed. Each parameter was scored as being either normal or having a mild, moderate or severe degree of pathology, and as a summary each parameter was scored as either pathological or normal. A total pathology variable, called *Any of the above,* was defined as positive if the subject had any degree of pathology in either of the four parameters.

The scans from the two cohorts were obtained at two different MRI machines with different field strengths (1.5 T versus 3 T), but the same MRI sequences were assessed using the same Image Software Program (NordicICE®) and the same experienced paediatric neuroradiologist (blinded to GA status of each subject) evaluated the scans.

6.2.2. fMRI task: Selective attention – working memory

EPT/ELBW children are at increased risk of suffering from impaired working memory and selective attention (56, 92, 93). An fMRI paradigm combining working memory and selective attention was developed by professor Kenneth Hugdahl, and first used in Ole Gunnar Viken's Masters degree thesis (94). This paradigm is based on a combination of two well-known neuropsychological tests; the n-back test for working memory and the Stroop colour-word test for selective attention.

The n-back test: Working memory is thought of as a system with limited capacity, covering the tasks of storing, processing and retrieving information over a short time frame (95). In the n-back task, a subject is typically presented with a series of visual or auditory stimuli, and is asked to respond when a pre-instructed target stimulus is the

same as one presented at some point (1, 2 …or n presentations) previously in the sequence (96-99). Brain areas known to display BOLD activation as a response to the n-back task are found bilaterally in the prefrontal cortex, the inferior parietal lobule, the precuneus, the anterior cingulum/supplementary motor area, the lateral premotor cortex and the thalamus (98).

The Stroop colour-word test: The test was first introduced by Stroop in 1935 (54). In this task, the participant is presented with the names of colour words (e.g. red, blue) written in inks of conflicting colours, e.g. the word "RED" written in blue ink. The participant is instructed to either respond to the written words themselves (word task) or the colour of the ink in which the words are presented (colour task). Naming the ink colour of a stimulus is slowed if the printed word designates a conflicting colour because word reading is an automatic process that takes precedence over colour naming (for review see (100)). The increase in naming latency, known as the Stroop effect, is referred to as colour-word interference (101-103). This process is highly dependent on frontal and parietal lobe function, the anterior cingulum in particular (100), and is therefore a suitable measure for studying executive functions like working memory/selective attention (104).

The combination of the two tests used in this fMRI study consisted of visual presentations of different colour-words written in conflicting ink colour, and presented one by one, and is referred to as the n-back/Stroop task.

The fMRI session consisted of four runs of the words “RED”, “BLUE”, “GREEN” and “YELLOW”, all written in the three incongruent colours (e.g. red written in the ink colour blue, yellow or green, never red), making a total of 12 colour-words. The words were displayed through MRI compatible LCD goggles (NordicNeurolab,Inc) attached to the head coil while the participant was in the scanner (see Figure 3 for a schematic illustration of the stimulus set-up).

Figure 3 Example of the colour words written in incongruent ink colours in the n-back/Stroop task.

The words were written in the child's native language, Norwegian. The task was to either respond to the word, independent of the ink colour it was written in, or to the colour independent of the actual word displayed, and to press a button held in the dominant hand when a word or ink colour matched the one presented either 1- or 2-stimuli backwards in the sequence. The layout of the task comprised four different experimental conditions (word 1-back, colour 1-back, word 2-back, colour 2-back). As an example, in the Word 1-back and 2-back conditions, the participants were told to press the response button when the current word stimulus was the same as the word presented one or two words back in the stimulus sequence. In the Colour 1-back and 2-back conditions the subjects were asked to respond to the ink colour of the words, see Figure 4. The participants were introduced to the procedure through a short computer program test sampling all four research conditions 15 min in advance of the actual scanning. This also worked as a quick screening for possible colour blindness and reading skills that could otherwise interact with their capability of responding to the correct stimulus. The stimulus sequence is exemplified in Figure 4.

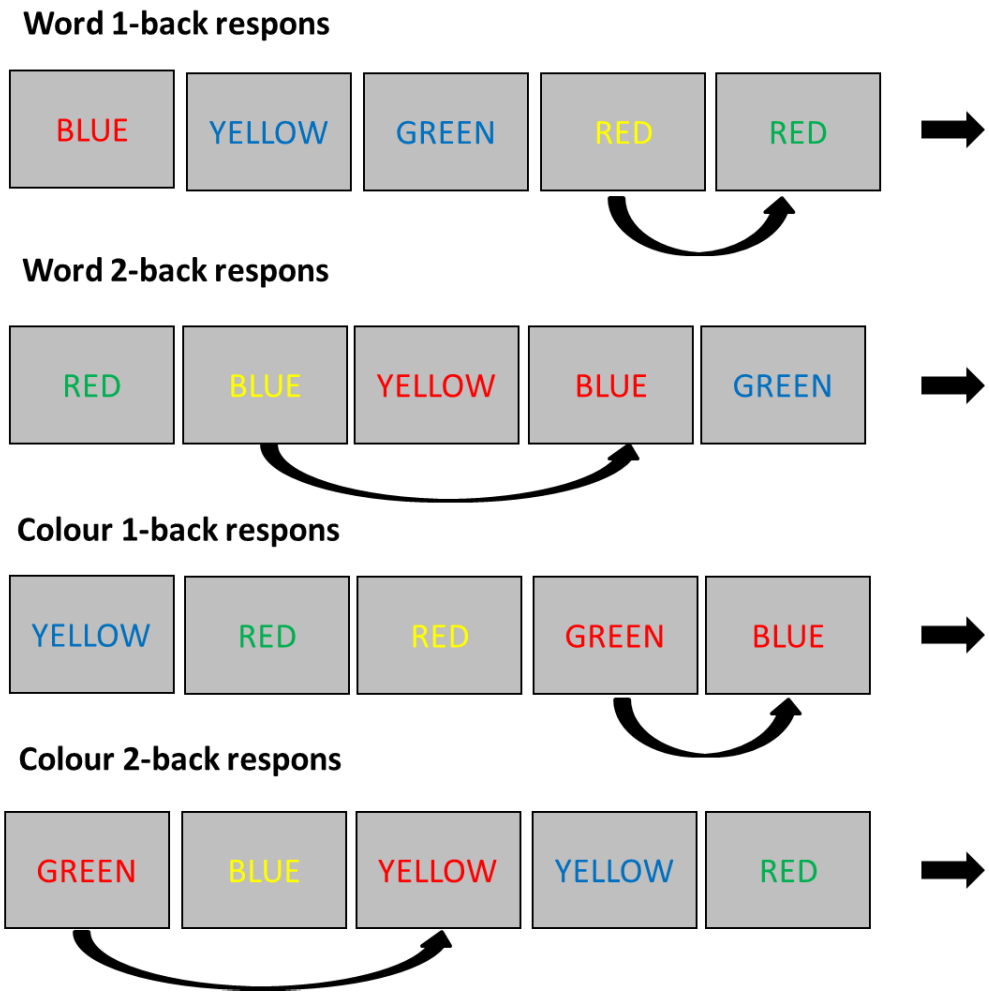


Figure 4 Example of task instructions: In the word 1-back condition the child was instructed to press the response button when the written word presented was the same as the stimulus presented one screen back in the presentation sequence. In the colour 2-back condition the response was based on the ink colour of the stimuli being the same as the one presented two screens back in the presentation sequence.

The stimulus sequences were presented via the E-prime software (Psychology Software Tools, Inc.). Responses were recorded by pressing a button held in the participant's dominant hand.

6.2.3. fMRI Protocol

Design: A three way (2 x 2 x 2) factorial design was chosen, based on the three independent variables Group (Preterm versus Control) x Load (1-back versus 2-back) x Instruction (word versus ink colour) organised in four separate runs. In Paper III, the Group level was also analysed for High versus Low rather than Preterm versus Control. Each run consisted of one experimental condition (Word 1-back condition = one run). For each run there were four ON blocks consisting of 16 stimulus presentations each and four OFF (stimulus absent) blocks. The OFF blocks were used as a baseline, in order to achieve proper contrast between task-related and task-unrelated processing, and were subtracted from the ON blocks in a standard subtraction procedure typically used in block design. In each ON block there were 3, 4 or 5 target stimuli randomly presented in the time series, where the subjects were instructed to press the response-key. Each stimulus was presented for 2.25 s followed by a blank interval of 0.3 s. The total time for each ON and OFF block was 40.8 s, (Figure 5).

Figure 5 The block design used in the study. Each block lasted 40.8s and alternated between OFF (stimulus absent) and ON (stimulus present). There were four blocks in each run.

The order of presentation of the four different experimental conditions was counter-balanced across participants, so that one quarter of the subjects was randomly assigned to one of the four different presentation iterations; (a) colour 1-back, word 1-back, colour 2-back, word 2-back, b) word 1-back, colour 1-back, word 2-back, colour 2-back, c) colour 2-back, word 2-back, colour 1-back, word 1-back and d) word 2-back,

colour 2-back, word 1-back, colour 1-back). This was done in order to cancel out any ordering-effects.

Analysis of fMRI data

The analyses of the fMRI data were performed using the SPM 8 software package (Statistical Parametric Mapping, Welcome Trust Centre for Neuroimaging, London, UK). SPM runs under MatLab (Mathworks, Natick, Mass.) and contains software particularly designed to analyse fMRI data.

Pre-processing: fMRI data are susceptible to movements (head movements, breathing, pulsation, non-homogenic magnetic fields, etc.), and the images must therefore be pre-processed in order to improve the quality. The first step is *Realignment* where the echo planar imaging (EPI) -images from each individual are realigned, typically such that all EPI images are realigned with the first scan, to control for variations caused by head movements. Translation and rotation movement were set to a maximum of 5 mm (any subject moving their head more than 5 mm sideways or up/down was excluded). The next step in the pre-processing is *Co-registration*, as EPI scans are of low resolution and show little anatomical contrast. In order to identify underlying brain structures in the activation data, the functional images are mapped onto high-resolution structural data. The *Normalization* process follows next to compensate for differences in anatomy between individuals. SPM 8 has an inbuilt function based on the Montreal Neurological Institute (MNI) stereotactic reference system, where each individual is "normalized" to fit an averaged brain volume.

The final step of pre-processing is *Smoothing*, where high signal frequencies are reduced and low signal frequencies are increased using a Gaussian filter to reduce noise caused by hemodynamic changes. Smoothing reduces the amount of local cluster maxima showing significant activation and thereby reduces the chance of false significant positive clusters. The normalized and smoothed images are then used in the data analysis process in which the change in the BOLD response across time is estimated and tested for voxel-wise significance in a regression model with the time

course for the stimulus presentations, following the statistical principles of the General Linear Model.

1st level analysis: For each subject included in the study, the OFF blocks or baseline recordings in each run were subtracted from the following ON blocks. This was done to reduce BOLD activation signals from other origins than the cognitive process of answering the task at hand (reduce noise).

2nd level analysis: During the second level analysis, the processed 1st level images from each subject were grouped into the different conditions following the standard SPM 8 Full Factorial setup. The independent variable factors Group (Preterm versus Control), Load (1-back versus 2-back) and Instruction (Word versus Colour) were evaluated in a factorial design as shown in Figure 6.

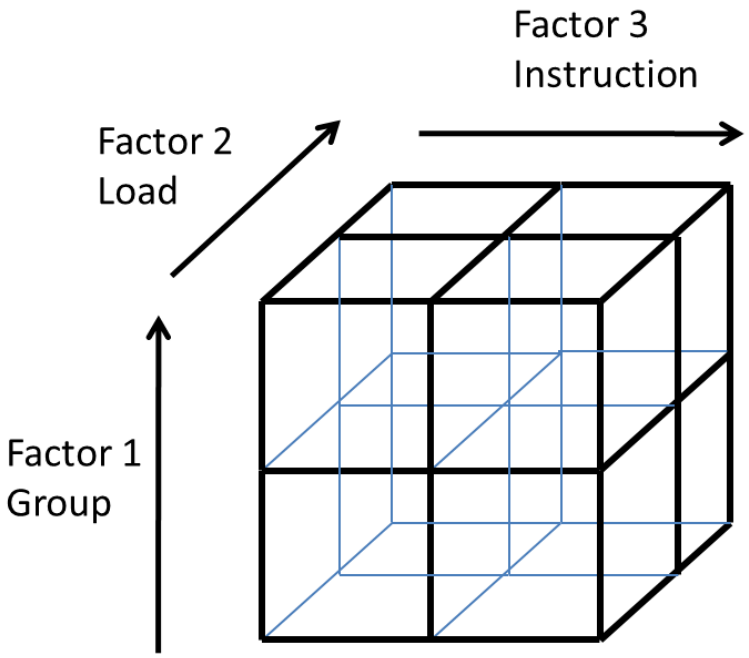


Figure 6 Diagram showing how the three different factors were arranged in a 2 x 2 x 2 manner.

In paper III, the second level analyses were repeated with Level 1 condition Group altered to consist of the groups “High” and “Low” scores on the RA task rather than “Preterm” and “Control”.

Group comparisons BOLD data

Voxel wise comparisons of fMRI data in SPM 8 consists of thousands of t- tests based on Gaussian field theory using multiple comparisons of average signals in the given condition (60). Different contrasts comparing the conditions are made by subtracting the average in one condition from the average of another condition. To correct for the multiple t-tests conducted by SPM 8, Family Wise Error (FWE) corrected significance threshold ($p < 0.05$) was used for the main affects activation within a group, and uncorrected threshold ($p < 0.001$) for contrasts between groups. FWE correction is also based on Gaussian field theory, where one corrects for the chance of finding false positive significances. Using uncorrected thresholds for the between group comparisons were chosen because FWE correction would be a too conservative test. Minimum cluster size was set to 10 voxels.

Region of interest (ROI) analysis BOLD data

Three regions of interest (ROIs) were chosen for a more detailed analysis of BOLD activation in each subject. The choice of ROIs was based on areas of activation with the highest overlap found when analysing the groups for whole brain volume activations. Two of the ROIs, located in the right inferior parietal and right inferior frontal gyrus, were defined by a 4 mm spherical area centred at coordinates ($x = 47$, $y = -42$, $z = 51$) and ($x = 40$, $y = 28$, $z = 30$) respectively. To make a third ROI in the ACC, a mask covering this region was made in order to define the area for signal extraction. Using in-house scripts based on the SPM 8 software package functionality, signal intensity within each ROI was averaged from spatially normalized frames for each individual subject, and then averaged along the time course, grouped by ON versus OFF blocks.

In- scanner behavioural data

Response accuracy (RA) and reaction time (RT), defined as time from stimulus presentation to response, were registered with the E-Prime software. In addition to the difference between the classic Stroop colour word test and the working memory part of the n-back/Stroop task used in this study was the presentation of single words and the time limit for responses as the subject only had time to respond until the new stimulus was presented.

6.3. School performance

In the 5th year of school (at eleven years of age), all children in Norway attend three mandatory national tests covering the topics reading, mathematics and English skills. The tests are adapted to children in the Norwegian society, and are performed in a standardised way throughout the country. These tests are meant as an evaluation of each pupil's skills in oral expression, writing, reading, mathematics, English skills and use of digital tools across several subjects (Utdanningsdirektoratet, www. Utdanningsdirektoratet.no). Each pupil is given a score in points and a general performance score, and all results are collected and stored by Statistics Norway. The results of these national tests for all consenting participants in this study were obtained from Statistics Norway. Although the tests are compulsory, not all children were able to attend all tests, and an average percentage score of points achieved (in relation to possible points) per attended tests was therefore computed and used as a measure of school performance.

6.4. Statistics

Statistical analyses of the behavioural data were performed using Statistical Product and Service Solutions version 20 (SPSS, Chicago, IL, USA), Cytel Studio 9 for the matched data, and SPM 8 for cluster analysis of fMRI data.

In paper I, descriptive statistics were reported using mean and standard deviations (SD), proportions and odds ratios (OR). Degree of pathology in each of the four parameters (dilatation of the lateral ventricles, reduction in the bulk of WM, the presence of focal WM pathology, thinning of the CC), sum of pathology and "Any pathology" were considered categorical variables. Group differences between EPT and their matched controls were calculated using the non-parametric McNemar's test. For comparison of VPT and controls, the Chi squared test was used, and risk estimates were expressed as OR and 95% confidence intervals (CI).

In paper II, the RA (correct responses given in per cent) and RT (time from stimulus presentation to response) data from the n-back/Stroop task were subjected to analysis of variance (ANOVA) significance testing according to the specified design. Simple main-effects were analysed with post hoc Tukey's HSD test. For the BOLD data, the MRICron software (http://www.cabiatl.com/mricro/mricron/index.html) was used to identify the anatomical location of significantly activated clusters, yoking the Anatomical Automatic Labelling (AAL) atlas 30 and Brodmann area (BA) templates. To test for normality, or equality of variances, in the two groups, we extracted the BOLD signal from three selected ROIs in all subjects. The three ROIs were chosen as representatives of overlapping activations between groups, and were spherical regions with a 5 mm radius around the peak voxel of the three MNI coordinates (x = 33, y = -85, z = -5) Occipital middle (left), (x = -21, Y = -91, z = 3) Occipital Inferior (right) and (x = 5, y = 19, z = 51) Supplementary motor area (right). The signal intensity area within each ROI was averaged throughout the volume on spatially normalized frames for each individual subject, and then averaged along the time course grouped by ON/OFF blocks, using in-house scripts built around functions in the SPM8. The relative change in signal intensity was calculated according to the formula ((ON/OFF) – 1). The difference in homogeneity of the variance/covariance matrices was tested using Levene's test. Non-significant ($p > 0.05$) differences between the groups was found for all tests in all three ROIs. The data was analysed for overlap between

expected normal values and the observed values for the preterm children and the controls, as normality is a statistical assumption in the General Linear Model implemented in SPM 8 (Figure in Paper II).

In paper III, SPM and MRICron were used the same way as in Paper II to localize clusters in the High versus Low group comparison. Mean values with SD (using t-tests), proportions and ORs (using the chi squared test) were computed in SPSS 20. To analyse school performance and/or RA according to BOLD activation and prematurity, a linear regression analysis using the General Linear Model procedure in SPSS was applied. Results were expressed as estimated regression coefficients (B) per unit change in each predictor variable with 95% CI.

6.5. Ethics

The project was approved by the Regional Ethics Committee for Medical Research in Western Norway (REK-Vest 2009/2271 - EPT cohort, REK 3.2005.1372 – VPT cohort). Informed written consent from the parents and oral consent from the children were collected for all EPT/ELBW participants. Written consent was obtained from all the 19 year old participants.

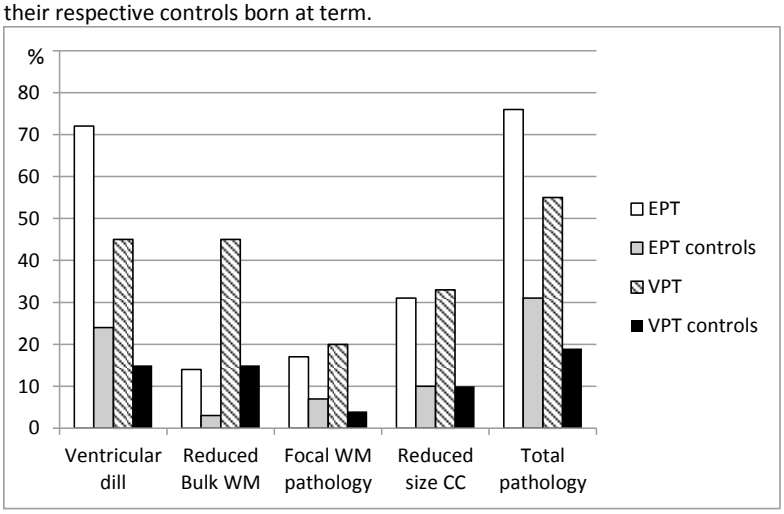
7. RESULTS

7.1. Summary of papers

Paper I: "**Cerebral Magnetic Resonance Imaging findings in children born extremely preterm, very preterm and at term"**

MRI pathology, (the summary score) was found in three out of four EPT children versus one out of three EPT controls, and in half of the VPT young adults versus one out of five VPT controls. A higher proportion of MRI pathology was found in the EPT group than the VPT group, as well as in the EPT control group compared to the VPT control group. The OR for the two preterm groups compared to their respective controls were therefore almost equal, and actually tended to be lower in the EPT group; (EPT – EPT controls: 4.3, 95% CI 1.5 to 137.5, VPT – VPT controls 5.2 CI 2.5 to 10.9). The types of MRI pathology were the same in both preterm groups, and to a lesser extent in the control groups, although less frequent. The most common pathological finding was mild pathology, and frequencies of moderate and severe pathology of any kind were too low to analyse. Ventricular dilatation was most common in the EPT cohort, reduction in bulk of WM most common in the VPT cohort and focal WM pathology and thinning of the CC was equally common in the EPT and VPT cohorts.

Figure 7 Structural MRI findings in children born EPT (GA < 28 weeks) or VPT (GA=28-31weeks) and their respective controls born at term.



Paper II: "fMRI – blood oxygen level dependent activation during a working memory-selective attention task in children born extremely preterm"

The pattern of BOLD activation during the task covered bilateral areas in the occipital inferior gyrus (BA 18/19), the right supplementary motor area (BA 6), bilaterally in an area extending from the ACC (BA 24/32) to the middle frontal and precentral gyrus (BA 6/44/45), bilaterally in the insula (BA 47/48) and bilaterally in the angular area extending into the parietal superior and inferior gyri (BA 7/40). The activation pattern found in the EPT group was the same as in the term born control children, but the intensity was significantly reduced. Increasing cognitive load (the highest cognitive load was in the colour 2-back condition) lead to increased BOLD activation, particularly in the ACC, prefrontal and parietal areas compared to controls, Figure 8.

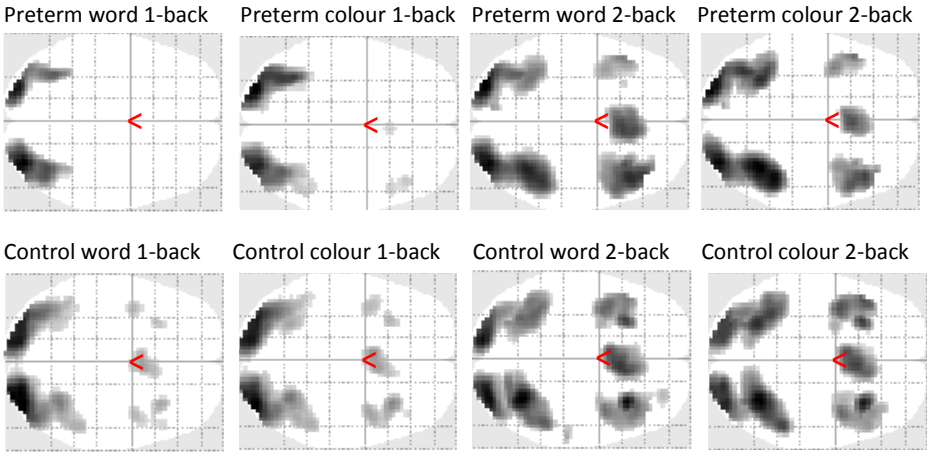


Figure 8 Main activation patterns during the fMRI task for all four conditions for preterm and control children, made in SPM 8, FWE corrected, $p < 0.05$.

RA results showed significantly fewer correct responses in the EPT group than in the control group. This difference was most pronounced with maximum cognitive load, i.e. the colour 2-back condition. RTs showed no significant group difference in any of the four experimental conditions of the test, although there was a trend towards longer RTs for the EPT/ELBW children (Table 8).

Table 8 Response accuracy scores (%) and reaction time (ms) for the n-back/Stroop task in preterm and control children.

	Word 1-back		Word 2-back		Colour 1-back		Colour 2-back	
	EPT	Controls	EPT	Controls	EPT	Controls	EPT	Controls
Response accuracy (%)	94	96	62	76	93	92	61	77
Reaction time (ms)	831	710	891	811	765	683	798	754

For EPT children, the mean group difference in BOLD activation patterns was increased in the colour 1- and 2-back conditions according to the SPM activation maps. This was consistent with the largest group difference in the RA measure being the colour 2-back condition.

In addition to the information reported in Paper II we performed an ROI analysis of the cingulate cortex, an area important for higher cognitive functioning. The results

showed significant mean difference in BOLD activation in EPT children compared to term born controls (Figure 9).

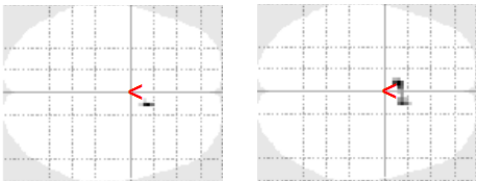


Figure 9 fMRI BOLD activation in the cingulate cortex in Control minus Preterm contrasts for the colour 1-back condition (left) and colour 2-back conditions (right). The opposite contrasts, Preterm minus Control, and the word conditions gave no significant activation.

Paper III: "Functional Magnetic Resonance Imaging, school performance and prematurity: a regional, clinical-controlled cohort study"

There was a linear relationship between school performance, based on national school test data, and RA on the n-back/Stroop task. RA was associated with BOLD activation, and being born preterm enhanced this association. There was, however, no association between school performance and BOLD activation.

8. DISCUSSION

The present thesis was an evaluation of structural and functional MRI findings during a working memory/selective attention task in a regional EPT cohort of eleven year old children compared to control children.

8.1. Summary

EPT children displayed more pathology on structural MRI scans than term born children, mainly due to mildly enlarged ventricles, and only limited numbers had moderate or severe pathology. The ratio of pathology in EPT children compared to term controls was not higher than in VPT young adults to their controls born 12 -14 years earlier, which may imply a very positive development for EPT children with time. The pattern of pathology was not specific for preterm children, but pathology was mainly found in the form of increased ventricular size, reduced bulk of WM, focal WM pathology or reduced size of CC. The cingulum, an area in close proximity to the ventricles and CC, as well as prefrontal and parietal areas, showed significantly reduced BOLD activation in EPT/ELBW compared to term born children. No signs of alternative pathways unique to EPT/ELBW children were found. Reduced BOLD activation is often associated with reduced cognitive outcome in fMRI studies, as in this study. Despite the link between cognitive function (RA scores on the fMRI task) and school performance, only cognitive function was associated with BOLD activation. BOLD activation maps revealed that the difference between EPT and term born children was not the same as the difference between children scoring high or low on the cognitive test.

8.2. Population

8.2.1. Selection criteria

The selection criteria for the national PEP cohort were based on both GA and BW (105). Many previous studies have classified preterm children according to BW, but lately GA has been considered a more precise measure of a preterm child's maturity (17). Children born SGA, e.g. with a low BW for GA, is more mature and therefore has a better outcome in several aspects than children with the same BW but lower GA. Using both GA and BW as criteria when selecting the national cohort was chosen to enable comparison to preterm cohorts in other studies. Paper I included children selected only by GA. They were extracted from two different cohorts of different ages; 11 year old children with GA less than 28 weeks from the PEP study and 19 year old young adults with GA 28-31 weeks from a cohort with BW less than 2000 g. In Paper II and III, both the EPT and ELBW children from the PEP study were included because a further reduction from the 28 included preterm children to obtain a pure EPT group would mean a simultaneous reduction in statistical power which we felt to be a greater scientific loss than a possible benefit gained by a pure GA group. To our knowledge, all other fMRI studies on preterm children included groups of mixed maturity and BW.

8.2.2. Number of participants

The study population in all three papers included a relatively small number of subjects, which increased the likelihood of the population not being representative for the true EPT/ELBW population. As an example, mothers of the EPT/ELBW children in this thesis had the same level of education as the mothers in the control group, Table 5. Most previous studies report that mothers of preterm children have a slightly lower level of education than mothers of term born children (106). Also, the lack of significantly lower school performance in the EPT/ELBW group compared to the

control group was an unexpected finding, as it is generally accepted that preterm children score below term born children on cognitive tasks and at school tests (55, 56). Whether the higher than expected school performance in this study is caused by the EPT mothers' higher than normal educational level is not known, but possible as the mother's educational level is a known strong predictor of the child's cognitive outcome (107). On the other hand, the similarities in maternal education abolished a potential significant confounder and the similarities in school performance and relatively small differences in all outcomes may confirm earlier suggestions that prematurity per se, is a weaker predictor of long term outcome than parental factors which may include both genetic and nurturing elements (108).

8.2.3. PEP and other preterm cohorts

The number of subjects included in the study could be a limiting factor when it comes to comparing this cohort to other EPT/ELBW cohorts. While 9% of the national PEP cohort had NDDs (CP, deafness, blindness) at five years of age (39), a relatively low rate, the number was only four per cent in the regional cohort. Gender distribution in the regional cohort was coherent with the national cohort (48) and a Swedish national cohort of extremely immature children (GA < 26 weeks) (109).

Regarding brain injury, the rate of PVH > grade 2 in the national PEP cohort was 6% (105), a slightly higher number than in the British EPICure study (110), possibly due to different grading systems.

All in all, the PEP cohort, both the regional and the national, is probably representative of other EPT/ELBW cohorts in other European and North American countries with similar economy (28, 110).

8.3. Methodological considerations

8.3.1. The working memory/selective attention task

The fMRI task chosen in this thesis reflects both working memory (the n-back part) and selective attention (the Stroop part) (54, 111). Both components are important for cognitive function, and are found to be an overall challenge to preterm children (56). In clinical terms, preterm children are often reported to have difficulties when given several instructions at the same time, and are known to have higher rates of attention problems than term born children (112). As both selective attention and working memory are of importance in daily function (for children and adults in general), the aim of the n-back/Stroop task was to look at selective attention in a working memory setting.

In adult studies, the n-back task normally includes1-back, 2-back and 3-back. When planning and piloting the study, the 3-back situation was assessed too difficult for eleven year old children in general, and therefore only the 1- and 2-back conditions were chosen.

8.3.2. School performance

School performance is an easy and reliable measure to evaluate how well a child is managing daily activities and life in general. In this thesis, only the academic aspect was included in the evaluation, and the mandatory national tests at eleven years of age were used as a measure. According to the Department of Education in Norway, the tests are meant as an evaluation of how well Norwegian schools succeed in developing the core educational skills of reading, mathematics and parts of the English language in their pupils (www.udir.no/nasjonaleprover). As an alternative, standard IQ tests instead of school performance could have been used. However, IQ is a very specific measure, and is not adapted to the requirements of function in daily life as well as school tests. The tests are standardised, adapted to the Norwegian society at eleven years of age, and compulsory. Therefore, no extra burden of costs, time or other

disadvantages would be put on the children in order to attend them, and the national school tests were deemed the most appropriate outcome measure in this thesis.

School performance is thought to be closely linked to cognitive function, which was confirmed in the present study in the form of a working memory/selective attention task. It is surprising, though, that the difference in school performance scores between EPT and control children was smaller than the difference in RA scores on the cognitive task. The brain activation unique to high compared to low achievers on the n-back/Stroop task covered different areas of the brain than control compared to EPT children. This supports the notion that underachievement at school in EPT children is caused by more than purely cognitive skills.

8.3.3. Data collection

All the EPT/ELBW children and families contacted in this study agreed to participate, while 22 of the invited controls reclined. A possible explanation is that EPT/ELBW families know the benefits from research on this group of children, while families of term born children do not have the same experience. Additionally, EPT/ELBW children are more likely to have been through structural MRI scanning previously and therefore less likely to be afraid of the scanning procedure. During the pre-scanning task introduction, the typical EPT/ELBW child would agree to understanding the task without asking questions, but still fail to press the pretend response button during the pre-scanning test. The typical term born control child, however, understood or asked questions, and then managed the pre-scanning practice procedure. This information is based on the observer's clinical impression, and is only mentioned as a curious observation, but it may reflect differences in cognitive abilities.
The main challenge during data collection was to make sure the eleven year old children did not move during the scanning procedure (45 minutes). All children were instructed to not move their head or body during the scanning session. Despite this, one child aborted the scanning procedure after ten minutes unable to lie still, and seven

other EPT/ELBW children and eight term born controls were lost to further analysis due to excessive head movements.

8.3.4. Statistics

Paper I: It was challenging to compare two cohorts of different ages and GA, and that had different criteria for control selection. The EPT cohort was individually matched for sex and time of birth at time of scanning, while the VPT control group was selected at a different time for a larger low BW cohort, and therefore not individually matched. This resulted in different statistical methods when comparing the cohorts to their respective controls. As far as we know, however, no other papers have reported cerebral MRI findings in EPT children of this age, or compared MRI findings of subjects born EPT and VPT. Although selected differently, we suggest that both control groups were representative of subjects born at term and that the results were valid and of importance

.

Paper II and III: We chose to evaluate the fMRI data in a parametric approach using the standard General Linear Approach implemented in SPM 8. This model was used to generate statistics in every single voxel, and then the resulting statistical map for significant brain areas was tested using distributional estimates for continuous random fields with the same distribution and smoothness. Behind this model lies an assumption that the data are obtained from stationary homogeneous discrete Gaussian random fields. If the intra individual variability in the BOLD data is large, it is difficult to know if the preterm data at each voxel is normally distributed. Another way to analyse the data, however, would have been to use a permutation based approach, which is a method that is not routinely available or used (113) but possibly performs better in small groups where the assumptions of classical parametric analysis are hard to test. We tested for differences in homogeneity of the variance/covariance matrices using Levene's test(114). The Levene's test statistically tests the null hypothesis that variances in two samples are equal, and thus assesses the assumption of homoscedasticity in tests of significance using a GLM approach. The results of the

Levene's test showed non-significant ($p > 0.05$) differences between the groups for all three ROIs (Figure 7). We therefore conclude that comparing the premature and control groups using a GLM approach, as implemented in SPM8 does not violate the assumption of homogeneity of variances between the groups.

8.4. Main results

8.4.1. Structural MRI

To our knowledge this is the first report comparing structural MRI findings in eleven year old EPT children with term born controls, and with a group of VPT participants in unselected cohorts. We find only very young EPT children referred to in the literature, making comparisons of the findings a challenge.

The neuroradiologist performing the assessments in this study had long experience with MRI data and was blinded to the preterm-control status, but the subjective nature of the grading process may have introduced a systematic difference in drawing a line between a normal or mildly abnormal scan. Also, the time lapse between the evaluations of the EPT and VPT groups could potentially have inflicted some change in evaluations. This may explain some of the difference in frequencies between the eleven and the nineteen year old subjects. All anatomical MRI scans were evaluated by one neuroradiologist, rather than two or more. Although we have no evaluation of inter-rater agreement in this study, one of the co-authors has shown in a previous study that the inter-rater agreement for ventricular dilatation is acceptable (115). We do however not know for certain whether this also applies to WM changes.

It is difficult to conclude whether the ratio of mild pathology in the MRI scans was higher in the EPT than the VPT group. The overall per-cent of pathology was higher in the EPT group, but the ratio of pathology in the EPT control group was equally higher than in the VPT control group. As a result, the OR of having a pathological structural MRI scan compared to term born controls was about equal in the EPT and the VPT group. This could be caused by a higher ratio of e.g. increased ventricular size in term

born eleven year old children than in term born nineteen year old young adults, or due to the subjective scoring. Term children have more prominent ventricles than teenagers (ventricles get smaller with age until the end of the second decade), and subjective assessment to separate mild ventriculomegaly from normal findings in children is therefore challenging.

All in all the frequency of moderate and severe pathology in EPT children was lower than expected, which is a positive development in this group. The main cause of the high pathology frequency in the EPT group was the high number of mild ventricular dilatation, which is not known to have any implications for neuro development.

8.4.2. Functional MRI

The main outcome of the first fMRI study (paper II), that EPT children displayed reduced BOLD activation compared to term born control children during the working memory/selective attention task, was in line with previous research of preterm children applying different cognitive tasks (89, 116). The n-back/Stroop task has not been used in an fMRI study of preterm children previously. To our knowledge, no previous fMRI paper has focused on differences in brain activation in the selected group of EPT/ELBW children versus term born children, only in mixed preterm groups. A large range of GA or BW values indicate a morphologically different group of children when it comes to brain development (SGA etc. (17)), and the results of this thesis will therefore describe changes more directly connected to prematurity, and have less confounding factors.

Accuracy of BOLD data in children is a critical issue for this thesis. Several previous fMRI studies of children have found that neuronal networks during cognitive tasks are more or less similar to what is seen in adults when the children are older than five years. The development rate of the brain is different in individual children (myelination, synaptic pruning, volumes), however, and whether these factors could influence the BOLD signal in minor ways is uncertain (117). Additionally, due to the on-going development, the perfusion and metabolism rate is higher in children than in

adults. This could have reduced the difference in BOLD signal between task and baseline, leading to mean group differences below significance level (118).

The areas of increased BOLD activation included areas expected from results of n-back and Stroop tasks in previous studies (94, 98, 102). The reduced activation in EPT/ELBW children in these areas is interpreted as indications of reduced or different activity compared to term born children. Other studies of preterm children have also found reduced BOLD activation in selected brain areas according to which fMRI task was chosen. Also, studies of children with dyslexia, ADHD or autism report reduced activation in given areas compared to controls, and some of these areas overlap with the findings in this study, i.e. when related to working memory and attention (119-121). Activation unique to preterm compared to term born children, indicating the existence of alternative pathways, has also been reported (86, 89). No evidence of alternative pathways was found in this study. It could be that there are no alternative neural pathways in preterm children for working memory/selective attention tasks. An alternative explanation is that the high baseline metabolism in children could have masked the existence of increased BOLD signal in alternative areas.

No association between school results and BOLD activation during the working memory/selective attention task was found. As school results are known to be closely linked to cognitive skills, and there was an association between the cognitive task (working memory and selective attention) and BOLD activation, this lack of significant results was surprising. Possible explanations may be that the low number of participants resulted in insufficient statistical power to reveal an existing association, that the baseline metabolism in children is so high that it masks a possible BOLD signal difference, or that there truly was no association, maybe because the national school tests and working memory/selective attention are not covering the same brain areas/functions. It seems reasonable that the low number of participants and high metabolic rate in children is the most likely cause of the lack of significant association.

The pattern of BOLD activation in the high achieving children compared to the low achieving children did not involve any part of the prefrontal cortex. This was surprising as the prefrontal cortex is the main area for processes involving working memory (94). It was interesting, however, that the precuneus was a main area of difference between high and low achievers as the precuneus is involved in a large range of highly integrated tasks (122). That the BOLD signal difference between EPT/ELBW and term born children being located in prefrontal and parietal areas and the ACC was less surprising as these are areas known to be involved in working memory and selective attention.

When analysing BOLD activation in all the EPT/ELBW children as a group, a significantly overall reduced BOLD activation was found compared to control children, but not when analysing the three ROIs with most overlapping areas of activation (in both groups) separately. Yet again it is possible that analysing the data individually, and only looking at a smaller area of the brain, reduced the strength of the data and masked an existing association. As this connection was found when comparing groups (EPT versus controls) and including the whole brain, it may perhaps be possible to find it in a larger scale study.

8.4.3. In-scanner behavioural data

The RA scores revealed that increasing the working memory load (going from 1-back to 2-back) made a larger impact on cognitive load than changing the task from word to colour (Figure 9). There was a significant difference in response accuracy and reaction times overall between the 1-back and the 2-back conditions. The colour 2-back was expected to be the most difficult, or cognitively most demanding task, and was also the only condition where we found a significant difference between the preterm and control group. The reaction time was longer during the 2-back conditions in both groups as expected, see Figure 10, but there was no significant group difference between term and preterm children, only a trend towards longer reaction times in the preterm group. Yet again, it is possible that the number of participants was too low to

show an existing group difference in reaction times. Some authors suggest that increased RT found in children with attention problems like ADHD etc. are due to a lack of attention in a few responses, while most recorded RTs are average length (123, 124). This could have been a likely explanation for the trend of longer RTs in this study, but the recorded RTs showed longer duration for most responds. More studies are necessary to investigate whether RTs are longer in EPT children and, if so, - why.

8.8. Implications

The number of surviving preterm children increases, particularly children born EPT. They are born at a critical time of the development of the brain, and numerous factors in the neonatal period can cause deviant development. It is important to disclose such developmental patterns in order to understand and possibly prevent deviant development, and also in order to support these children through school and into independent and happy lives as adults. Sadly, it is still a common belief in society that preterm children should have 'caught up' with their term born peers by school age, and their difficulties at school can easily be misinterpreted as inattentiveness or lack of dedication by teachers as well as parents. Confirming and describing differences in brain anatomy and function as in this thesis can make it easier for this group of children to get sufficient help and understanding, particularly at school.

9. CONCLUSIONS

EPT children without severe NDD have more cerebral pathology as seen in structural MRI scans, have lower response accuracy on a working memory/selective attention task and display reduced fMRI BOLD activation in selected brain areas compared to term born control children, although not associated with school performance in this study.

- The increased MRI pathology in EPT children is mainly due to mild ventricular dilatation
- The risk of finding pathology in cerebral MRI scans in EPT children compared to term born controls is not higher than for VPT young adults compared to young adult controls
- EPT/ELBW children display reduced BOLD signal response to a working memory/selective attention task compared to term born control children, but the same brain areas were involved, i.e. no alternative pathways were detected
- RA on the working memory/selective attention task is reduced in the EPT/ELBW group compared to the control group when cognitive load is increased
- School performance is associated with RA on a working memory/selective attention task, but not with the fMRI BOLD signal invoked by the task
- The area of reduced fMRI BOLD signal intensity in EPT/ELBW compared to control children is not the same as in low compared to high achievers on the working memory/selective attention test, indicating that cognitive function is not the whole explanation for underachievement in EPT/ELBW children

10. FUTURE CHALLENGES

- Convey to physicians, and particularly radiologists, the wide distribution of ventricular size in MRI scans in order to avoid overemphasis of its clinical relevance.
- A larger number of participants is needed to answer whether there is an association between BOLD activation and school results.
- Further studies on neuronal correlates for other cognitive functions e.g. executive functions, in EPT children to learn more about the cause of reduced cognitive functions in this group are need. A Diffusion-tensor-imaging (DTI) study describing the neural connections between implicated brain areas when solving the working memory/selective attention task could provide valuable knowledge on how the preterm brain differs in anatomy to the brain of the term born child.

11. ACKNOWLEDGEMENTS

Though frustrating at times, working on this thesis has been more fun than I expected, I have realized research is even more useful than I assumed, and I have met and worked with many very talented and inspiring people. It has been a steep learning ladder, and I am very grateful to all of you who have helped me along the way.

My main supervisor, Irene Elgen, you have supported and advised me throughout the project, listened to my frustrations and helped me get an overview when I got stuck in details. You are a great confidence booster, I have always felt highly prioritized, and I am very grateful for all the work you have done. Trond Markestad, founder of the preterm research environment at Barneklinikken and my co-supervisor, your extensive knowledge and understanding span over many areas, which I have benefited from in all my work. Thank you for inviting me to be a part of your group, and always being helpful along the way. Stein Magnus Aukland, my other highly valued co-supervisor and talented MRI expert, you are always ready to help – which has been much appreciated as I was all new to MRI and research in general. I am very grateful.
Kristian Sommerfelt, my supervisor while working as a doctor at Barneklinikken, you made me realize that research was a possible option for me, you always 'lend a helping thought', and often understand my questions before I do. Thank you.
Head of the Department of Pediatrics, Britt Skadberg, my sincere gratitude for your positive attitude to the project, for helping me and Maria find a practical work solution at the start of the project, and for always making me feel that I am welcome back/belong at BKB.

The most exciting part of the project has been the cooperation with Bergen fMRI Group. Kenneth Hugdahl, thank you for letting us into the world of fMRI, for listening to me despite my lack of fMRI expertise and explaining the parts I found difficult (and there are still many…), and for always replying very quickly to my mails. Emanuel Neto, you sacrificed many weekends and evenings to be by my side while scanning the children (I am afraid the ca.100 hours looking at the scanner would have been very

boring without you), helped me understand what to do with the data, and on many occasions later – thank you! I would also like to thank Anne Marie Rød for help with fMRI statistics, and Alex Craven for all the work you did on preparing the data for paper III. The very talented, helpful and friendly radiographers at Haraldsplass Hospital, Grethe Austgulen, Sverre Østerhus and Irene Vik also deserve mentioning and gratitude, together with Morten Odberg who collected the data for the VPT cohort, and Kling Chong (Great Ormond Street, London) who analysed the structural MRI scans.

Our study nurse, Lise Lønning, you did a great job organizing questionnaires, participants and controls etc. Thank you for all your hard work. Thanks also to everyone else in the PEP group for help and advice along the way, and particularly to Maria Vollsæter and lately also Hege Clemm for good companionship, advice and highly valued coffee breaks. Thanks to all my fellow researchers and colleagues, without you my days would have been rather lonely. My friend and former lecturer from Wales, Professor Alan Beaton, you helped me with statistics, English language and have been my personal 'reviewer' on one of the papers out of pure kindness. I am very grateful. Geir Egil Eide, your statistical guidance and advice have been highly appreciated and very necessary (as you know) – thank you.
Then, of course, the very basis of all my data: I would like to express my gratitude to all the preterm children, control children and their families for taking the time to participate and make this project possible. Thanks also to collaborators at the Norwegian Birth Registry and Statistics Norway.
I also want to thank the financial supporters of the study: the Norwegian Extra Foundation for Health and Rehabilitation through The Premature Society (with particular help from Marit K.Hagemoen), Helse Vest Hospital Trust and the Bergen fMRI Group via the Norwegian Western Health Authorities (Helse-Vest).

At the end I would like to thank my family and friends in Norway and Britain. Having a happy and fulfilling social life made it so much easier to focus on my research work when necessary, so thank you for all your support and care. My supporting husband

and best friend Dylan, and my children Emil, Maja and Ella – you are my life, nothing can get me down for long with you around!

12. REFERENCES

1. Kiernan JA. The Human Nervous System, An Anatomical Viewpiont. 8th ed: Lippincott Williams & Wilkins; 2005 2005. 470 p.

2. Dubois J, Benders M, Cachia A, Lazeyras F, Ha-Vinh Leuchter R, Sizonenko SV, et al. Mapping the early cortical folding process in the preterm newborn brain. Cereb Cortex. 2008 Jun;18(6):1444-54. PubMed PMID: 17934189. Epub 2007/10/16. eng.

3. Ballabh P. Intraventricular hemorrhage in premature infants: mechanism of disease. Pediatr Res. 2010 Jan;67(1):1-8. PubMed PMID: 19816235. Pubmed Central PMCID: 2799187. Epub 2009/10/10. eng.

4. Ballabh P, Braun A, Nedergaard M. Anatomic analysis of blood vessels in germinal matrix, cerebral cortex, and white matter in developing infants. Pediatr Res. 2004 Jul;56(1):117-24. PubMed PMID: 15128918. Epub 2004/05/07. eng.

5. Rezaie P, Dean A. Periventricular leukomalacia, inflammation and white matter lesions within the developing nervous system. Neuropathology : official journal of the Japanese Society of Neuropathology. 2002 Sep;22(3):106-32. PubMed PMID: 12416551. Epub 2002/11/06. eng.

6. Greisen G. Autoregulation of cerebral blood flow in newborn babies. Early Hum Dev. 2005 May;81(5):423-8. PubMed PMID: 15935919. Epub 2005/06/07. eng.

7. Toga AW, Thompson PM, Sowell ER. Mapping brain maturation. Trends Neurosci. 2006 Mar;29(3):148-59. PubMed PMID: 16472876. Pubmed Central PMCID: 3113697. Epub 2006/02/14. eng.

8. Courchesne E, Chisum HJ, Townsend J, Cowles A, Covington J, Egaas B, et al. Normal brain development and aging: quantitative analysis at in vivo MR imaging in healthy volunteers. Radiology. 2000 Sep;216(3):672-82. PubMed PMID: 10966694. Epub 2000/08/31. eng.

9. Vohr BR, Allen M. Extreme prematurity--the continuing dilemma. N Engl J Med. 2005 Jan 6;352(1):71-2. PubMed PMID: 15635115. Epub 2005/01/07. eng.

10. Papile LA, Burstein J, Burstein R, Koffler H. Incidence and evolution of subependymal and intraventricular hemorrhage: a study of infants with birth weights less than 1,500 gm. J Pediatr. 1978 Apr;92(4):529-34. PubMed PMID: 305471. Epub 1978/04/01. eng.

11. de Vries LS, Eken P, Dubowitz LM. The spectrum of leukomalacia using cranial ultrasound. Behavioural brain research. 1992 Jul 31;49(1):1-6. PubMed PMID: 1388792. Epub 1992/07/31. eng.

12. Bax M, Tydeman C, Flodmark O. Clinical and MRI correlates of cerebral palsy: the European Cerebral Palsy Study. JAMA. 2006 Oct 4;296(13):1602-8. PubMed PMID: 17018805. Epub 2006/10/05. eng.

13. Pierrat V, Duquennoy C, van Haastert IC, Ernst M, Guilley N, de Vries LS. Ultrasound diagnosis and neurodevelopmental outcome of localised and extensive cystic periventricular leucomalacia. Arch Dis Child Fetal Neonatal Ed. 2001 May;84(3):F151-6. PubMed PMID: 11320039. Pubmed Central PMCID: 1721251. Epub 2001/04/26. eng.

14. Dyet LE, Kennea N, Counsell SJ, Maalouf EF, Ajayi-Obe M, Duggan PJ, et al. Natural history of brain lesions in extremely preterm infants studied with serial magnetic resonance imaging from birth and neurodevelopmental assessment. Pediatrics. 2006 Aug;118(2):536-48. PubMed PMID: 16882805. Epub 2006/08/03. eng.

15. World Health Organization. Preterm birth 2012. Available from: http://www.who.int/mediacentre/factsheet/fs363/en.

16. Beck S, Wojdyla D, Say L, Betran AP, Merialdi M, Requejo JH, et al. The worldwide incidence of preterm birth: a systematic review of maternal mortality and morbidity. Bulletin of the World Health Organization. 2010 Jan;88(1):31-8. PubMed PMID: 20428351. Pubmed Central PMCID: 2802437. Epub 2010/04/30. eng.

17. Saigal S, Doyle LW. An overview of mortality and sequelae of preterm birth from infancy to adulthood. Lancet. 2008 Jan 19;371(9608):261-9. PubMed PMID: 18207020. Epub 2008/01/22. eng.

18. Tyson JE, Parikh NA, Langer J, Green C, Higgins RD, National Institute of Child H, et al. Intensive care for extreme prematurity--moving beyond gestational age. N Engl J Med. 2008 Apr 17;358(16):1672-81. PubMed PMID: 18420500. Pubmed Central PMCID: 2597069. Epub 2008/04/19. eng.

19. Arnold CC, Kramer MS, Hobbs CA, Mclean FH, Usher RH. Very-Low-Birth-Weight - a Problematic Cohort for Epidemiologic Studies of Very Small or Immature Neonates. Am J Epidemiol. 1991 Sep 15;134(6):604-13. PubMed PMID: ISI:A1991GL64600006. English.

20. Voltolini C, Torricelli M, Conti N, Vellucci FL, Severi FM, Petraglia F. Understanding Spontaneous Preterm Birth: From Underlying Mechanisms to Predictive and Preventive Interventions. Reproductive sciences. 2013 Mar 14. PubMed PMID: 23493416. Epub 2013/03/16. Eng.

21. Goldenberg RL, Culhane JF, Iams JD, Romero R. Epidemiology and causes of preterm birth. Lancet. 2008 Jan 5;371(9606):75-84. PubMed PMID: 18177778. Epub 2008/01/08. eng.

22. Ruiz RJ, Fullerton J, Dudley DJ. The interrelationship of maternal stress, endocrine factors and inflammation on gestational length. Obstetrical & gynecological survey. 2003 Jun;58(6):415-28. PubMed PMID: 12775946. Epub 2003/05/31. eng.

23. Damadian R. Tumor detection by nuclear magnetic resonance. Science. 1971 Mar 19;171(3976):1151-3. PubMed PMID: 5544870.

24. Lorenz JM. Survival and long-term neurodevelopmental outcome of the extremely preterm infant. A systematic review. Saudi medical journal. 2011 Sep;32(9):885-94. PubMed PMID: 21894349. Epub 2011/09/07. eng.

25. Petrou S. The economic consequences of preterm birth during the first 10 years of life. BJOG. 2005 Mar;112 Suppl 1:10-5. PubMed PMID: 15715587. Epub 2005/02/18. eng.

26. Doyle LW, Victorian Infant Collaborative Study G. Evaluation of neonatal intensive care for extremely low birth weight infants in Victoria over two decades: I. Effectiveness. Pediatrics. 2004 Mar;113(3 Pt 1):505-9. PubMed PMID: 14993541. Epub 2004/03/03. eng.

27. Group E, Fellman V, Hellstrom-Westas L, Norman M, Westgren M, Kallen K, et al. One-year survival of extremely preterm infants after active perinatal care in Sweden. JAMA. 2009 Jun 3;301(21):2225-33. PubMed PMID: 19491184. Epub 2009/06/06. eng.

28. Hack M, Fanaroff AA. Outcomes of children of extremely low birthweight and gestational age in the 1990's. Early Hum Dev. 1999 Jan;53(3):193-218. PubMed PMID: 10088988. Epub 1999/03/24. eng.

29. Ronnestad A, Abrahamsen TG, Medbo S, Reigstad H, Lossius K, Kaaresen PI, et al. Septicemia in the first week of life in a Norwegian national cohort of extremely premature infants. Pediatrics. 2005 Mar;115(3):e262-8. PubMed PMID: 15687417. Epub 2005/02/03. eng.

30. Stoll BJ, Hansen NI, Bell EF, Shankaran S, Laptook AR, Walsh MC, et al. Neonatal outcomes of extremely preterm infants from the NICHD Neonatal Research Network. Pediatrics. 2010 Sep;126(3):443-56. PubMed PMID: 20732945. Pubmed Central PMCID: 2982806. Epub 2010/08/25. eng.

31. Ronnestad A, Abrahamsen TG, Medbo S, Reigstad H, Lossius K, Kaaresen PI, et al. Late-onset septicemia in a Norwegian national cohort of extremely premature infants receiving very early full human milk feeding. Pediatrics. 2005 Mar;115(3):e269-76. PubMed PMID: 15687416. Epub 2005/02/03. eng.

32. Schlapbach LJ, Aebischer M, Adams M, Natalucci G, Bonhoeffer J, Latzin P, et al. Impact of sepsis on neurodevelopmental outcome in a Swiss National Cohort of extremely premature infants. Pediatrics. 2011 Aug;128(2):e348-57. PubMed PMID: 21768312. Epub 2011/07/20. eng.

33. Stoll BJ, Hansen NI, Adams-Chapman I, Fanaroff AA, Hintz SR, Vohr B, et al. Neurodevelopmental and growth impairment among extremely low-birth-weight infants with neonatal infection. JAMA. 2004 Nov 17;292(19):2357-65. PubMed PMID: 15547163. Epub 2004/11/18. eng.

34. Baraldi E, Filippone M. Chronic lung disease after premature birth. N Engl J Med. 2007 Nov 8;357(19):1946-55. PubMed PMID: 17989387.

35. Jobe AH, Bancalari E. Bronchopulmonary dysplasia. American journal of respiratory and critical care medicine. 2001 Jun;163(7):1723-9. PubMed PMID: 11401896. Epub 2001/06/13. eng.
36. Farstad T, Bratlid D, Medbo S, Markestad T, Norwegian Extreme Prematurity Study G. Bronchopulmonary dysplasia - prevalence, severity and predictive factors in a national cohort of extremely premature infants. Acta Paediatr. 2011 Jan;100(1):53-8. PubMed PMID: 20653607. Epub 2010/07/27. eng.
37. Ambalavanan N, Carlo WA, Tyson JE, Langer JC, Walsh MC, Parikh NA, et al. Outcome trajectories in extremely preterm infants. Pediatrics. 2012 Jul;130(1):e115-25. PubMed PMID: 22689874. Pubmed Central PMCID: 3382921. Epub 2012/06/13. eng.
38. Committee on F, Newborn. Postnatal corticosteroids to treat or prevent chronic lung disease in preterm infants. Pediatrics. 2002 Feb;109(2):330-8. PubMed PMID: 11826218. Epub 2002/02/05. eng.
39. Leversen KT, Sommerfelt K, Ronnestad A, Kaaresen PI, Farstad T, Skranes J, et al. Prediction of neurodevelopmental and sensory outcome at 5 years in Norwegian children born extremely preterm. Pediatrics. 2011 Mar;127(3):e630-8. PubMed PMID: 21321031. Epub 2011/02/16. eng.
40. Martin CR, Dammann O, Allred EN, Patel S, O'Shea TM, Kuban KC, et al. Neurodevelopment of extremely preterm infants who had necrotizing enterocolitis with or without late bacteremia. J Pediatr. 2010 Nov;157(5):751-6 e1. PubMed PMID: 20598317. Pubmed Central PMCID: 2952050. Epub 2010/07/06. eng.
41. Salhab WA, Perlman JM, Silver L, Sue Broyles R. Necrotizing enterocolitis and neurodevelopmental outcome in extremely low birth weight infants <1000 g. Journal of perinatology : official journal of the California Perinatal Association. 2004 Sep;24(9):534-40. PubMed PMID: 15254558. Epub 2004/07/16. eng.
42. Saugstad OD. Oxygen and retinopathy of prematurity. Journal of perinatology : official journal of the California Perinatal Association. 2006 May;26 Suppl 1:S46-50; discussion S63-4. PubMed PMID: 16482198. Epub 2006/02/17. eng.
43. Martinez-Cruz CF, Garcia Alonso-Themann P, Poblano A, Ochoa-Lopez JM. Hearing loss, auditory neuropathy, and neurological co-morbidity in children with birthweight <750 g. Archives of medical research. 2012 Aug;43(6):457-63. PubMed PMID: 22960856.
44. Johnson S, Fawke J, Hennessy E, Rowell V, Thomas S, Wolke D, et al. Neurodevelopmental disability through 11 years of age in children born before 26 weeks of gestation. Pediatrics. 2009 Aug;124(2):e249-57. PubMed PMID: 19651566. Epub 2009/08/05. eng.
45. Schlapbach LJ, Adams M, Proietti E, Aebischer M, Grunt S, Borradori-Tolsa C, et al. Outcome at two years of age in a Swiss national cohort of extremely preterm infants born between 2000 and 2008. BMC pediatrics. 2012;12:198. PubMed PMID: 23272671. Pubmed Central PMCID: 3546845. Epub 2013/01/01. eng.
46. Moster D, Lie RT, Markestad T. Long-term medical and social consequences of preterm birth. N Engl J Med. 2008 Jul 17;359(3):262-73. PubMed PMID: 18635431. Epub 2008/07/19. eng.
47. Farooqi A, Hagglof B, Sedin G, Serenius F. Impact at age 11 years of major neonatal morbidities in children born extremely preterm. Pediatrics. 2011 May;127(5):e1247-57. PubMed PMID: 21482612. Epub 2011/04/13. eng.
48. Leversen KT, Sommerfelt K, Ronnestad A, Kaaresen PI, Farstad T, Skranes J, et al. Predicting neurosensory disabilities at two years of age in a national cohort of extremely premature infants. Early Hum Dev. 2010 Sep;86(9):581-6. PubMed PMID: 20800392. Epub 2010/08/31. eng.
49. Marlow N, Wolke D, Bracewell MA, Samara M. Neurologic and developmental disability at six years of age after extremely preterm birth. N Engl J Med. 2005 Jan 6;352(1):9-19. PubMed PMID: 15635108. Epub 2005/01/07. eng.
50. Gardner F, Johnson A, Yudkin P, Bowler U, Hockley C, Mutch L, et al. Behavioral and emotional adjustment of teenagers in mainstream school who were born before 29 weeks' gestation. Pediatrics. 2004 Sep;114(3):676-82. PubMed PMID: 15342838. Epub 2004/09/03. eng.

51. Hille ET, den Ouden AL, Saigal S, Wolke D, Lambert M, Whitaker A, et al. Behavioural problems in children who weigh 1000 g or less at birth in four countries. Lancet. 2001 May 26;357(9269):1641-3. PubMed PMID: 11425366. Epub 2001/06/27. eng.
52. Elgen SK, Leversen KT, Grundt JH, Hurum J, Sundby AB, Elgen IB, et al. Mental health at 5 years among children born extremely preterm: a national population-based study. European child & adolescent psychiatry. 2012 Jun 30. PubMed PMID: 22752364. Epub 2012/07/04. Eng.
53. Baddeley A, Logie R, Bressi S, Della Sala S, Spinnler H. Dementia and working memory. The Quarterly journal of experimental psychology A, Human experimental psychology. 1986 Nov;38(4):603-18. PubMed PMID: 3809575. Epub 1986/11/01. eng.
54. Stroop J. Studies of interference in serial verbal reactions. J Exp Psychology. 1935;18:643-62.
55. Bhutta AT, Cleves MA, Casey PH, Cradock MM, Anand KJ. Cognitive and behavioral outcomes of school-aged children who were born preterm: a meta-analysis. JAMA. 2002 Aug 14;288(6):728-37. PubMed PMID: 12169077. Epub 2002/08/10. eng.
56. Baron IS, Rey-Casserly C. Extremely preterm birth outcome: a review of four decades of cognitive research. Neuropsychology review. 2010 Dec;20(4):430-52. PubMed PMID: 20512418. Epub 2010/06/01. eng.
57. Bowen JR, Gibson FL, Hand PJ. Educational outcome at 8 years for children who were born extremely prematurely: a controlled study. Journal of paediatrics and child health. 2002 Oct;38(5):438-44. PubMed PMID: 12354257.
58. Costeloe K. EPICure: facts and figures: why preterm labour should be treated. BJOG. 2006 Dec;113 Suppl 3:10-2. PubMed PMID: 17206960. Epub 2007/01/09. eng.
59. Doyle LW, Saigal S, Streiner DL. Prematurity and later cognitive outcomes. JAMA. 2002 Nov 27;288(20):2542-3; author reply 3. PubMed PMID: 12444858. Epub 2002/11/28. eng.
60. Huettel SA, Song, Allen W., McCarthy, Gregory. Functional Magnetic Resonance Imaging. 2nd ed: Sinauer Associates, Inc; 2009. 542 p.
61. Lauterbur P. Image formation by induced local interactions: Examples emplying nuclear magnetic resonance. Nature. 1973;242:190-1.
62. Mansfield P. Multi-planar imaging formation using NMR spin echoes. J Physics C Solid State Phys. 1977;10.
63. Battin MR, Maalouf EF, Counsell SJ, Herlihy AH, Rutherford MA, Azzopardi D, et al. Magnetic resonance imaging of the brain in very preterm infants: visualization of the germinal matrix, early myelination, and cortical folding. Pediatrics. 1998 Jun;101(6):957-62. PubMed PMID: 9606219. Epub 1998/06/02. eng.
64. Brown NC, Inder TE, Bear MJ, Hunt RW, Anderson PJ, Doyle LW. Neurobehavior at term and white and gray matter abnormalities in very preterm infants. J Pediatr. 2009 Jul;155(1):32-8, 8 e1. PubMed PMID: 19394041. Epub 2009/04/28. eng.
65. de Bruine FT, van den Berg-Huysmans AA, Leijser LM, Rijken M, Steggerda SJ, van der Grond J, et al. Clinical implications of MR imaging findings in the white matter in very preterm infants: a 2-year follow-up study. Radiology. 2011 Dec;261(3):899-906. PubMed PMID: 22031710. Epub 2011/10/28. eng.
66. Horsch S, Hallberg B, Leifsdottir K, Skiold B, Nagy Z, Mosskin M, et al. Brain abnormalities in extremely low gestational age infants: a Swedish population based MRI study. Acta Paediatr. 2007 Jul;96(7):979-84. PubMed PMID: 17524026. Epub 2007/05/26. eng.
67. Inder TE, Wells SJ, Mogridge NB, Spencer C, Volpe JJ. Defining the nature of the cerebral abnormalities in the premature infant: a qualitative magnetic resonance imaging study. J Pediatr. 2003 Aug;143(2):171-9. PubMed PMID: 12970628. Epub 2003/09/13. eng.
68. Maalouf EF, Duggan PJ, Rutherford MA, Counsell SJ, Fletcher AM, Battin M, et al. Magnetic resonance imaging of the brain in a cohort of extremely preterm infants. J Pediatr. 1999 Sep;135(3):351-7. PubMed PMID: 10484802. Epub 1999/09/15. eng.
69. Thompson DK, Inder TE, Faggian N, Johnston L, Warfield SK, Anderson PJ, et al. Characterization of the corpus callosum in very preterm and full-term infants utilizing MRI.

Neuroimage. 2011 Mar 15;55(2):479-90. PubMed PMID: 21168519. Pubmed Central PMCID: 3035727. Epub 2010/12/21. eng.
70. Skiold B, Vollmer B, Bohm B, Hallberg B, Horsch S, Mosskin M, et al. Neonatal Magnetic Resonance Imaging and Outcome at Age 30 Months in Extremely Preterm Infants. J Pediatr. 2011 Nov 3. PubMed PMID: 22056283. Epub 2011/11/08. Eng.
71. Skranes J, Evensen KI, Lohaugen GC, Martinussen M, Kulseng S, Myhr G, et al. Abnormal cerebral MRI findings and neuroimpairments in very low birth weight (VLBW) adolescents. Eur J Paediatr Neurol. 2008 Jul;12(4):273-83. PubMed PMID: 17933566. Epub 2007/10/16. eng.
72. Bodensteiner JB, Johnsen SD. Magnetic resonance imaging (MRI) findings in children surviving extremely premature delivery and extremely low birthweight with cerebral palsy. J Child Neurol. 2006 Sep;21(9):743-7. PubMed PMID: 16970878. Epub 2006/09/15. eng.
73. Peterson BS, Vohr B, Staib LH, Cannistraci CJ, Dolberg A, Schneider KC, et al. Regional brain volume abnormalities and long-term cognitive outcome in preterm infants. JAMA. 2000 Oct 18;284(15):1939-47. PubMed PMID: 11035890. Epub 2000/10/18. eng.
74. Cooke RW, Abernethy LJ. Cranial magnetic resonance imaging and school performance in very low birth weight infants in adolescence. Arch Dis Child Fetal Neonatal Ed. 1999 Sep;81(2):F116-21. PubMed PMID: 10448179. Pubmed Central PMCID: 1720984. Epub 1999/08/17. eng.
75. Eikenes L, Lohaugen GC, Brubakk AM, Skranes J, Haberg AK. Young adults born preterm with very low birth weight demonstrate widespread white matter alterations on brain DTI. Neuroimage. 2011 Feb 1;54(3):1774-85. PubMed PMID: 20965255. Epub 2010/10/23. eng.
76. Ment LR, Kesler S, Vohr B, Katz KH, Baumgartner H, Schneider KC, et al. Longitudinal brain volume changes in preterm and term control subjects during late childhood and adolescence. Pediatrics. 2009 Feb;123(2):503-11. PubMed PMID: 19171615. Pubmed Central PMCID: 2679898. Epub 2009/01/28. eng.
77. Nosarti C, Al-Asady MH, Frangou S, Stewart AL, Rifkin L, Murray RM. Adolescents who were born very preterm have decreased brain volumes. Brain. 2002 Jul;125(Pt 7):1616-23. PubMed PMID: 12077010. Epub 2002/06/22. eng.
78. Odberg MD, Aukland SM, Rosendahl K, Elgen IB. Cerebral MRI and cognition in nonhandicapped, low birth weight adults. Pediatr Neurol. 2010 Oct;43(4):258-62. PubMed PMID: 20837304. Epub 2010/09/15. eng.
79. Stewart AL, Rifkin L, Amess PN, Kirkbride V, Townsend JP, Miller DH, et al. Brain structure and neurocognitive and behavioural function in adolescents who were born very preterm. Lancet. 1999 May 15;353(9165):1653-7. PubMed PMID: 10335784. Epub 1999/05/21. eng.
80. Fox PT, Raichle ME. Focal physiological uncoupling of cerebral blood flow and oxidative metabolism during somatosensory stimulation in human subjects. Proc Natl Acad Sci U S A. 1986 Feb;83(4):1140-4. PubMed PMID: 3485282. Pubmed Central PMCID: 323027. Epub 1986/02/01. eng.
81. Arichi T, Moraux A, Melendez A, Doria V, Groppo M, Merchant N, et al. Somatosensory cortical activation identified by functional MRI in preterm and term infants. Neuroimage. 2010 Feb 1;49(3):2063-71. PubMed PMID: 19854281. Epub 2009/10/27. eng.
82. Heep A, Scheef L, Jankowski J, Born M, Zimmermann N, Sival D, et al. Functional magnetic resonance imaging of the sensorimotor system in preterm infants. Pediatrics. 2009 Jan;123(1):294-300. PubMed PMID: 19117895. Epub 2009/01/02. eng.
83. Doria V, Beckmann CF, Arichi T, Merchant N, Groppo M, Turkheimer FE, et al. Emergence of resting state networks in the preterm human brain. Proc Natl Acad Sci U S A. 2010 Nov 16;107(46):20015-20. PubMed PMID: 21041625. Pubmed Central PMCID: 2993415. Epub 2010/11/03. eng.
84. Peterson BS, Vohr B, Kane MJ, Whalen DH, Schneider KC, Katz KH, et al. A functional magnetic resonance imaging study of language processing and its cognitive correlates in prematurely born children. Pediatrics. 2002 Dec;110(6):1153-62. PubMed PMID: 12456913. Epub 2002/11/29. eng.

85. Schafer RJ, Lacadie C, Vohr B, Kesler SR, Katz KH, Schneider KC, et al. Alterations in functional connectivity for language in prematurely born adolescents. Brain. 2009 Mar;132(Pt 3):661-70. PubMed PMID: 19158105. Pubmed Central PMCID: 2664451. Epub 2009/01/23. eng.
86. Ment LR, Peterson BS, Vohr B, Allan W, Schneider KC, Lacadie C, et al. Cortical recruitment patterns in children born prematurely compared with control subjects during a passive listening functional magnetic resonance imaging task. J Pediatr. 2006 Oct;149(4):490-8. PubMed PMID: 17011320. Pubmed Central PMCID: 2386989. Epub 2006/10/03. eng.
87. Gozzo Y, Vohr B, Lacadie C, Hampson M, Katz KH, Maller-Kesselman J, et al. Alterations in neural connectivity in preterm children at school age. Neuroimage. 2009 Nov 1;48(2):458-63. PubMed PMID: 19560547. Pubmed Central PMCID: 2775072. Epub 2009/06/30. eng.
88. Mullen KM, Vohr BR, Katz KH, Schneider KC, Lacadie C, Hampson M, et al. Preterm birth results in alterations in neural connectivity at age 16 years. Neuroimage. 2011 Feb 14;54(4):2563-70. PubMed PMID: 21073965. Pubmed Central PMCID: 3020252. Epub 2010/11/16. eng.
89. Nosarti C, Rubia K, Smith AB, Frearson S, Williams SC, Rifkin L, et al. Altered functional neuroanatomy of response inhibition in adolescent males who were born very preterm. Dev Med Child Neurol. 2006 Apr;48(4):265-71. PubMed PMID: 16542513. Epub 2006/03/18. eng.
90. Leversen KT, Sommerfelt K, Elgen IB, Eide GE, Irgens LM, Juliusson PB, et al. Prediction of outcome at 5 years from assessments at 2 years among extremely preterm children: a Norwegian national cohort study. Acta Paediatr. 2012 Mar;101(3):264-70. PubMed PMID: 22026562. Epub 2011/10/27. eng.
91. Sommerfelt K, Ellertsen B, Markestad T. Parental factors in cognitive outcome of non-handicapped low birthweight infants. Arch Dis Child Fetal Neonatal Ed. 1995 Nov;73(3):F135-42. PubMed PMID: 8535868. Pubmed Central PMCID: 2528464. Epub 1995/11/01. eng.
92. Farooqi A, Hagglof B, Serenius F. Behaviours related to executive functions and learning skills at 11 years of age after extremely preterm birth: a Swedish national prospective follow-up study. Acta Paediatr. 2013 Mar 4. PubMed PMID: 23458380. Epub 2013/03/06. Eng.
93. Anderson PJ, De Luca CR, Hutchinson E, Spencer-Smith MM, Roberts G, Doyle LW, et al. Attention problems in a representative sample of extremely preterm/extremely low birth weight children. Dev Neuropsychol. 2011;36(1):57-73. PubMed PMID: 21253991. Epub 2011/01/22. eng.
94. Viken OG. En fMRI-studie av eksekutive funksjoner studert ved en Stroop n-back oppgave. 2007. Masteroppgave, Norges Teknisk - Naturvitenskapelige Universitet, Medisinsk fakultet, Institutt for Sirkulasjon og Bildediagnostikk, Universitetet i Bergen Psykologisk fakultet, Institutt for biologisk og medisinsk psykologi.
95. Baddeley A. Working memory and language: an overview. J Commun Disord. 2003 May-Jun;36(3):189-208. PubMed PMID: 12742667. Epub 2003/05/14. eng.
96. Gevins A, Cutillo B. Spatiotemporal dynamics of component processes in human working memory. Electroencephalogr Clin Neurophysiol. 1993 Sep;87(3):128-43. PubMed PMID: 7691540. Epub 1993/09/01. eng.
97. Hugdahl K, Rund BR, Lund A, Asbjornsen A, Egeland J, Ersland L, et al. Brain activation measured with fMRI during a mental arithmetic task in schizophrenia and major depression. Am J Psychiatry. 2004 Feb;161(2):286-93. PubMed PMID: 14754778. Epub 2004/02/03. eng.
98. Owen AM, McMillan KM, Laird AR, Bullmore E. N-back working memory paradigm: a meta-analysis of normative functional neuroimaging studies. Hum Brain Mapp. 2005 May;25(1):46-59. PubMed PMID: 15846822. Epub 2005/04/23. eng.
99. Schleepen TM, Jonkman LM. The development of non-spatial working memory capacity during childhood and adolescence and the role of interference control: an N-Back task study. Dev Neuropsychol. 2010 Jan;35(1):37-56. PubMed PMID: 20390591. Epub 2010/04/15. eng.
100. MacLeod CM, MacDonald PA. Interdimensional interference in the Stroop effect: uncovering the cognitive and neural anatomy of attention. Trends Cogn Sci. 2000 Oct 1;4(10):383-91. PubMed PMID: 11025281. Epub 2000/10/12. Eng.

101. Harrison BJ, Shaw M, Yucel M, Purcell R, Brewer WJ, Strother SC, et al. Functional connectivity during Stroop task performance. Neuroimage. 2005 Jan 1;24(1):181-91. PubMed PMID: 15588609. Epub 2004/12/14. eng.
102. Laird AR, McMillan KM, Lancaster JL, Kochunov P, Turkeltaub PE, Pardo JV, et al. A comparison of label-based review and ALE meta-analysis in the Stroop task. Hum Brain Mapp. 2005 May;25(1):6-21. PubMed PMID: 15846823. Epub 2005/04/23. eng.
103. Peterson BS, Skudlarski P, Gatenby JC, Zhang H, Anderson AW, Gore JC. An fMRI study of Stroop word-color interference: evidence for cingulate subregions subserving multiple distributed attentional systems. Biol Psychiatry. 1999 May 15;45(10):1237-58. PubMed PMID: 10349031. Epub 1999/06/01. eng.
104. Pardo JV, Pardo PJ, Janer KW, Raichle ME. The anterior cingulate cortex mediates processing selection in the Stroop attentional conflict paradigm. Proc Natl Acad Sci U S A. 1990 Jan;87(1):256-9. PubMed PMID: 2296583. Pubmed Central PMCID: 53241. Epub 1990/01/01. eng.
105. Markestad T, Kaaresen PI, Ronnestad A, Reigstad H, Lossius K, Medbo S, et al. Early death, morbidity, and need of treatment among extremely premature infants. Pediatrics. 2005 May;115(5):1289-98. PubMed PMID: 15867037. Epub 2005/05/04. eng.
106. Luu TM, Vohr BR, Allan W, Schneider KC, Ment LR. Evidence for catch-up in cognition and receptive vocabulary among adolescents born very preterm. Pediatrics. 2011 Aug;128(2):313-22. PubMed PMID: 21768322. Pubmed Central PMCID: 3146356.
107. Wang LW, Wang ST, Huang CC. Preterm infants of educated mothers have better outcome. Acta Paediatr. 2008 May;97(5):568-73. PubMed PMID: 18394101.
108. Sommerfelt K. Long-term outcome for non-handicapped low birth weight infants--is the fog clearing? Eur J Pediatr. 1998 Jan;157(1):1-3. PubMed PMID: 9461353. Epub 1998/02/14. eng.
109. Serenius F, Kallen K, Blennow M, Ewald U, Fellman V, Holmstrom G, et al. Neurodevelopmental outcome in extremely preterm infants at 2.5 years after active perinatal care in Sweden. JAMA. 2013 May 1;309(17):1810-20. PubMed PMID: 23632725.
110. Costeloe K, Hennessy E, Gibson AT, Marlow N, Wilkinson AR. The EPICure study: outcomes to discharge from hospital for infants born at the threshold of viability. Pediatrics. 2000 Oct;106(4):659-71. PubMed PMID: 11015506.
111. Braver TS, Cohen JD, Nystrom LE, Jonides J, Smith EE, Noll DC. A parametric study of prefrontal cortex involvement in human working memory. Neuroimage. 1997 Jan;5(1):49-62. PubMed PMID: 9038284. Epub 1997/01/01. eng.
112. Elgen I, Sommerfelt K. Low birthweight children: coping in school? Acta Paediatr. 2002;91(8):939-45. PubMed PMID: 12222719. Epub 2002/09/12. eng.
113. Nichols TE, Holmes AP. Nonparametric permutation tests for functional neuroimaging: a primer with examples. Hum Brain Mapp. 2002 Jan;15(1):1-25. PubMed PMID: 11747097. Epub 2001/12/18. eng.
114. Levene H. Robust test for equality of variances. Contributions to probability and statistics: essays in honour of Harold Hoteling. Stanford University Press1960. p. 278-92.
115. Aukland SM, Odberg MD, Gunny R, Chong WK, Eide GE, Rosendahl K. Assessing ventricular size: is subjective evaluation accurate enough? New MRI-based normative standards for 19-year-olds. Neuroradiology. 2008 Dec;50(12):1005-11. PubMed PMID: 18622601. Epub 2008/07/16. eng.
116. Rushe TM, Temple CM, Rifkin L, Woodruff PW, Bullmore ET, Stewart AL, et al. Lateralisation of language function in young adults born very preterm. Arch Dis Child Fetal Neonatal Ed. 2004 Mar;89(2):F112-8. PubMed PMID: 14977893. Pubmed Central PMCID: 1756037. Epub 2004/02/24. eng.
117. Gaillard WD, Grandin CB, Xu B. Developmental aspects of pediatric fMRI: considerations for image acquisition, analysis, and interpretation. Neuroimage. 2001 Feb;13(2):239-49. PubMed PMID: 11162265.
118. Gaillard WD, Hertz-Pannier L, Mott SH, Barnett AS, LeBihan D, Theodore WH. Functional anatomy of cognitive development: fMRI of verbal fluency in children and adults. Neurology. 2000 Jan 11;54(1):180-5. PubMed PMID: 10636145.

119. Beneventi H. Neuronal correlates of working memory in dyslexia. Bergen: University of Bergen, Norway; 2010.
120. Liotti M, Pliszka SR, Perez R, Kothmann D, Woldorff MG. Abnormal brain activity related to performance monitoring and error detection in children with ADHD. Cortex. 2005 Jun;41(3):377-88. PubMed PMID: 15871602. Epub 2005/05/06. eng.
121. Agam Y, Joseph RM, Barton JJ, Manoach DS. Reduced cognitive control of response inhibition by the anterior cingulate cortex in autism spectrum disorders. Neuroimage. 2010 Aug 1;52(1):336-47. PubMed PMID: 20394829. Pubmed Central PMCID: 2883672. Epub 2010/04/17. eng.
122. Cavanna AE, Trimble MR. The precuneus: a review of its functional anatomy and behavioural correlates. Brain. 2006 Mar;129(Pt 3):564-83. PubMed PMID: 16399806. Epub 2006/01/10. eng.
123. Hervey AS, Epstein JN, Curry JF, Tonev S, Eugene Arnold L, Keith Conners C, et al. Reaction time distribution analysis of neuropsychological performance in an ADHD sample. Child Neuropsychol. 2006 Apr;12(2):125-40. PubMed PMID: 16754533. Epub 2006/06/07. eng.
124. Epstein JN, Langberg JM, Rosen PJ, Graham A, Narad ME, Antonini TN, et al. Evidence for higher reaction time variability for children with ADHD on a range of cognitive tasks including reward and event rate manipulations. Neuropsychology. 2011 Jul;25(4):427-41. PubMed PMID: 21463041. Epub 2011/04/06. eng.