INFLUENCE OF BIRTH WEIGHT ON THE RISK AND CLINICAL PRESENTATION OF SCHIZOPHRENIA

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ACADEMIC DISSERTATION

To be presented, with the permission of the Faculty of Medicine of the University of Helsinki, for public examination in the Christian Sibelius Auditorium, Psychiatric Centre, on April 28th 2017, at 12 noon.

Helsinki 2017
To friends and family
ABSTRACT

Pre- and perinatal environmental factors have been associated with increased schizophrenia risk, particularly in combination with genetic liability. Both low and high birth weight have been associated with an increased risk of autism spectrum disorders and schizophrenia. The interaction of specific schizophrenia susceptibility genes and specified pre- and perinatal environmental factors have recently been described in relation to augmented schizophrenia risk.

In this thesis, the relationship between birth weight and schizophrenia risk was investigated as part of a large Finnish schizophrenia family study sample based on the “Genetic Epidemiology and Molecular Genetics of Schizophrenia in Finland” study, in which genetic susceptibility for schizophrenia is known to be elevated relative to the general population (Study I). The study sample consisted of two subsamples having at least one sibling with a diagnosis of schizophrenia. The associations between birth weight and symptom severity of schizophrenia and psychotic disorders (Study II) and cognitive functioning in schizophrenia (Study III) were also characterized. A gene-environment interaction, focusing on birth weight and specific genes from the DISC1 network (Study IV), which had previously been found to be associated with increased schizophrenia risk in the same cohort, was also investigated in relation to schizophrenia risk.

A 1.68-fold increase in schizophrenia risk was observed in subjects presenting with a high birth weight (>4000 g) relative to subjects with an intermediate birth weight of 3000-4000 g (Study I). In particular, schizophrenia risk was elevated among high birth weight individuals in combination with specific variants of the NDE1 gene (Study IV). Both low and high birth weight were found to be associated with increased severity of disorganized and negative symptom dimensions, whereas birth weight was not associated with symptoms of reality distortion (Study II). Both low and high birth weight, compared with the intermediate birth weight range, were associated with a slight decrease in cognitive performance among both subjects with schizophrenia and their unaffected first-degree relatives (Study III).

The observations in the thesis corroborate existing findings describing an association between high birth weight and increased schizophrenia risk. An association between low birth weight and increased schizophrenia risk, a finding widely documented in the literature, was not seen here. Birth weight was found to influence both symptom severity and cognitive performance in schizophrenia. The findings also suggest that the functions of NDE1 during the
early stages of neurodevelopment are vulnerable to the influence of pre- and perinatal environmental factors associated with high birth weight, with the propensity to augment subsequent schizophrenia susceptibility among offspring. The mechanisms underlying the association between high birth weight and schizophrenia risk are speculative, but may involve factors such as pre- or perinatal hypoxia, maternal metabolic and immunological mechanisms during pregnancy.
TIIVISTELMÄ

Raskauden- ja synnytyksenaikaisten komplikaatioiden on todettu lisäävän skitsofreniaan sairastumisen riskiä, etenkin geneettiseen alttiuteen yhdistettynä. Sekä alhainen että suuri syntymäpaino on todettu yhdistyvän kohonneeseen riskiin sairastua skitsofreniaan ja myös autismiryhmän häiriöihin. Tiettyjen skitsofrenialle altistavien geenien on todettu lisäävän skitsofrenia alttiutta sikiö- ja synnytyksenaikaisin komplikaatioihin yhdistettynä.

Tässä väitöskirjatyössä tutkittiin syntymäpainon yhteyttä skitsofreniariskiin (osajulkaisu I), psykoosioireiden vaikeusasteeseen (osajulkaisu II), ja kognitiiviseen suorituskykyyn (osajulkaisu III) osana ”Vakavien mielenterveyshäiriöiden geneettinen epidemiologia ja molekyyligeneettinen perusta” –projektia varten kerättyä perheaineistossa, jossa skitsofrenian geneettisen alttiuden tiedetään olevan muuta väestöä suurempi. Aineisto koostui kahdesta osa-aineistosta, joissa kussakin perheyksikössä oli vähintään yksi skitsofreniaan sairastunut jälkeläinen. Työssä tutkittiin myös tiettyjen DISC1-polun geenien ja syntymäpainon yhteisvaikutuksia skitsofrenia-alttiuteen (osajulkaisu IV). Analyyseissä tarkastelun kohteena olevien DISC1-polun geeni varianttien on aikaisemmissa tutkimuksissa osoitettu lisäävän skitsofreniariskiä samassa perheaineistossa.

Skitsofreniariskin havaittiin olevan 1.68-kertaisesti suurempi yksilöillä yksilöillä, joiden syntymäpaino oli suuri (>4000 g), verrattuna yksilöillä, joiden syntymäpaino oli 3000 g – 4000 g (osajulkaisu I). Skitsofrenia-alttiuden havaittiin olevan korkeampi etenkin niiden yksilöiden joukossa, jotka kantoivat NDE1 geenin tiettyjä variantteja ja joilla oli suuri syntymäpaino (osajulkaisu IV). Sekä alhainen että suuren syntymäpainon havaittiin yhdistyvän hajanaisten ja negatiivisten oireiden vaikeusasteeseen, kun taas syntymäpainolla ei todettu olevan yhteyttä aistiharhojen ja harhaluulojen vaikeusasteeseen (osajulkaisu II). Sekä alhainen että suuren syntymäpainon havaittiin yhdistyvän alempaan kognitiiviseen suorituskykyyn sekä skitsofreniapotilailla ja heidän ensimmäisen asteen sukulaisilla (osajulkaisu III).

Tämän väitöskirjatyön havainnot tukevat aikaisempia havaintoja siitä, että suuri syntymäpaino saattaa lisätä skitsofreniaan sairastumisen todennäköisyyttä. Toisinaan kuin aikaisemmissa tutkimuksissa alhainen syntymäpaino ei lisännyt sairastumisriskiä. Tutkimuksen havainnot tukevat myös käsitystä siitä, että NDE1-geenin toiminta keskushermoston kehityksessä on altiin syntymäpainoon liittyvien raskauden- ja synnytyksenaikaisten komplikaatioiden vaikutukselle. Havaintojen taustalla
olevat patofysiologiset mekanismit jäävät todentamatta, mutta niiden arvioidaan mahdollisesti liittyyvän sikiön raskauden- tai synnytyksenaikeeseen hapenpuutteeseen, äidin raskaudenaikaisiin metabolisiin tai immunologisiin tekijöihin.
ACKNOWLEDGEMENTS

This study was conducted at the Mental Health Unit of the National Institute for Health and Welfare (THL), formerly the National Public Health Institute (KTL), in 2008-2016. I wish to thank Professor Juhani Eskola and Professor Pekka Puska, who have been the Director Generals of THL during this work, for providing such a great place to do research. I sincerely thank the former and the current Heads of the Mental Health Unit, Professor Jouko Lönnqvist, Professor Mauri Marttunen and Professor Jaana Suvisaari, for providing such excellent research facilities. Professor Antti Mäkitie, Head of the Doctoral Program in Clinical Research, I thank for the opportunity to study in the graduate school.

I warmly thank all patients and their families for participating in the study.

My deepest gratitude is owed to my supervisors for their commitment and support throughout this process. I am very grateful to Professor Tiina Paunio for her pragmatic and constructive comments every step of the way. Her vision and encouragement have propelled my thesis forward, especially during the times when momentum was required. I am indebted to Professor Jaana Suvisaari for her exceptional vision, dedication and continuing guidance throughout the making of this thesis. The level of commitment and support that I received from Jaana is immense; this together with her empathetic nature form a combination of qualities that I have never before encountered to such a high degree in any professional setting during the course of my career. I can honestly say that it has been honour to have had you as my supervisor. I also extend my gratitude to Professor Annamari Tuulio-Henriksson for her active and constructive role during the initial part of the project. I especially thank her for the enjoyable discussions on matters related to cognition.

I owe a debt of gratitude to both the late Professor Leena Peltonen-Palotie and Professor Jouko Lönnqvist for their central roles in the establishment of the Finnish schizophrenia family study sample. I am very grateful to have had the opportunity to collaborate with Professor Jouko Lönnqvist during this project. His vision and enthusiasm towards the field of psychiatry are truly inspiring. He is a formidable and highly respected figure in psychiatry, both nationally and internationally, yet he is exceedingly gracious and respectful with all of his colleagues irrespective of their level of proficiency. I am also very grateful to have had the opportunity to meet the late Professor Peltonen-Palotie during the initial phase of the project. Her spirit and dedication towards research live on in those with whom she came into contact.
I am grateful to have collaborated with Docent William Hennah. Thank you Will for numerous insightful discussions on the genetics of neurodevelopment. These conversations have been most inspiring and motivating.

Maiju Pankakoski, I warmly thank for statistical expertise during the course of this project. Maiju was always patient and took the time to explain in detail topics related to statistics. This thesis would not have been written without your huge contribution.

I warmly thank Minna Torniainen-Holm, the first author of the third article, for numerous fruitful discussions on statistics and cognition. I also thank Marjut Grainger for her active role in the management of data.

This thesis has been written in a fairly solitary fashion, in parallel with my ongoing clinical work. I warmly thank my other co-authors for their valuable contributions: Docent Jaana Suokas, Docent Jari Haukka, Ulriika Lehto, Laura Häkkinen, and Liisa Tomppo. I am also grateful to all fellow researchers at THL, especially those whom I have had the opportunity to discuss matters related to the project: Laura Auvinen-Lintunen, Teija Kasteenpohja, Nina Markkula and Satu Viertio. Thank you both Niina and Laura for supportive discussions that motivated me to complete this thesis.

I thank all those who participated in the collection of the Finnish schizophrenia study sample, particularly Professor Jesper Ekelund and Ritva Arajärvi, with whom I had the good fortune to have discussed matters related to the study sample.

I am sincerely grateful to the reviewers of this thesis, Professor Soili Lehto and Professor Christina Dalman, for their constructive comments and valuable suggestions. The manuscript has been greatly improved by these comments. I also warmly thank Professor Jyrki Korkeila for accepting the role of Opponent and Professor Tiina Paunio for accepting the role of Custos in the defence of my thesis.

Carol Ann Pelli is thanked for editing the language of this manuscript.

I thank my former and current superiors and former clinical supervisors during my years in HUS Psychiatry. Docent Matti Holi, Docent Tuula Kieseppä, Docent Samuli Saarni, Docent Tanja Svirskis and Docent Kaisla Joutsenniemi I thank for being very supportive of research in HUS. Matti Holi and Tuula Kieseppä have been my long-standing supervisors during my training at Peijas Hospital, and I warmly thank them for their support, mentorship and enthusiasm towards the field of psychiatry, which have greatly influenced the way I view and practice psychiatry today. I also thank all my colleagues at HUS Psychiatry with whom I have had the good fortune to work
and discuss matters related to clinical psychiatry with: Kristiina Golan, Anniina Koski, Max Franzén, Anne Meriluoto, Marcelo Chirambarro, Maxim Shein, Elena Chukhina, Evgeny Chukhin, Juhana Santti, Jorma Oksanen, Allan Seppänen, Docent Katinka Tuisku, Docent Pekka Tani and Professor Nina Lindberg.

I warmly thank all of my friends, many of whom I have known since elementary school. I am especially grateful to Niko, Janne and Mathew for many enjoyable moments and laughs over the years.

Last, but not least, I thank my parents, Sakari and Marita, for providing me with the tools needed to lead an enjoyable and fulfilling life. My mother I thank for giving me a strong sense of empathy. My father I thank for his energetic and pragmatic approach to life, teaching me to be forever active and inquisitive about the world around me and also to appreciate the small everyday things in life, which can so easily be taken for granted. My brothers, Vesa and Juha, I thank for brotherly support manifesting as time spent together at the cinema, ice hockey matches and enjoying big steaks.

Without a doubt, the greatest support group has been by family. Thank you Marika for this ongoing journey and for all that we have shared. Thank you for always encouraging me to do what I feel is right, even if this has sometimes resulted in a bumpier road for you. I cannot comprehend my life without you. Mikael and Emilia, thank you for always being so happy to see me as soon as I walk in the door. Thank you for making every day fun and special. The future is so bright for both of you.

Financial support from the Academy of Finland, the Sigrid Juselius Foundation, the Finnish Medical Foundation, the Finnish Foundation for Psychiatric Research and the Jalmari and Rauha Ahokas Foundation is gratefully acknowledged.

Asko Wegelius
Helsinki, March 2017
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<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
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<tbody>
<tr>
<td>AKT1</td>
<td>Serine-Threonine Protein Kinase</td>
</tr>
<tr>
<td>APP</td>
<td>Amyloid Precursor Protein</td>
</tr>
<tr>
<td>ASD</td>
<td>Autism Spectrum Disorder</td>
</tr>
<tr>
<td>BDNF</td>
<td>Brain-Derived Neurotrophic Factor</td>
</tr>
<tr>
<td>cAMP</td>
<td>Cyclic Adenosine Monophosphate</td>
</tr>
<tr>
<td>CHRNA7</td>
<td>Cholinergic Receptor Nicotinic Alpha 7 Subunit</td>
</tr>
<tr>
<td>CMV</td>
<td>Cytomemalovirus</td>
</tr>
<tr>
<td>CNR1</td>
<td>Cannabinoid Receptor 1</td>
</tr>
<tr>
<td>CNS</td>
<td>Central Nervous System</td>
</tr>
<tr>
<td>CNV</td>
<td>Copy Number Variant</td>
</tr>
<tr>
<td>COMT</td>
<td>Catecholamine-O-Methyl-Transferase</td>
</tr>
<tr>
<td>DAT1</td>
<td>Dopamine Transporter 1</td>
</tr>
<tr>
<td>DISC</td>
<td>Disrupted in Schizophrenia</td>
</tr>
<tr>
<td>DIXDC1</td>
<td>DIX domain containing 1</td>
</tr>
<tr>
<td>DRD2</td>
<td>D2-Dopamine Receptor</td>
</tr>
<tr>
<td>DRD4</td>
<td>D4-Dopamine Receptor</td>
</tr>
<tr>
<td>DSM</td>
<td>Diagnostic and Statistical Manual of Mental Disorders</td>
</tr>
<tr>
<td>DTNBP1</td>
<td>Dystrobrevin Binding Protein 1</td>
</tr>
<tr>
<td>FEZ1</td>
<td>Fasciculation and Elongation Protein Zeta 1</td>
</tr>
<tr>
<td>FK506</td>
<td>FK506 Binding Protein</td>
</tr>
<tr>
<td>FOXP2</td>
<td>Forkhead Box P2</td>
</tr>
<tr>
<td>FTD</td>
<td>Formal Thought Disorder</td>
</tr>
<tr>
<td>GAF</td>
<td>Global Assessment of Functioning</td>
</tr>
<tr>
<td>GEE</td>
<td>General Estimating Equation</td>
</tr>
<tr>
<td>GSK3β</td>
<td>Glycogen Synthase Kinase 3 Beta</td>
</tr>
<tr>
<td>GRIN</td>
<td>Glutamate Receptor, Ionotropic, N-Methyl D-Aspartate</td>
</tr>
<tr>
<td>GRM</td>
<td>Glutamate Metabotropic Receptor</td>
</tr>
<tr>
<td>GWAS</td>
<td>Genome-Wide Association Study</td>
</tr>
<tr>
<td>HBW</td>
<td>High Birth Weight</td>
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<tr>
<td>HLA</td>
<td>Human Leukocyte Antigen</td>
</tr>
<tr>
<td>HRR</td>
<td>Hazard Rate Ratio</td>
</tr>
<tr>
<td>HSV</td>
<td>Herpes Simplex Virus</td>
</tr>
<tr>
<td>HTTLPR</td>
<td>Serotonin-transporter-linked polymorphic region</td>
</tr>
<tr>
<td>ICD</td>
<td>International Classification of Diseases</td>
</tr>
<tr>
<td>IGF</td>
<td>Insulin-like factor</td>
</tr>
<tr>
<td>IRR</td>
<td>Incidence Rate Ratio</td>
</tr>
<tr>
<td>LBW</td>
<td>Low Birth Weight</td>
</tr>
<tr>
<td>LD</td>
<td>Linkage disequilibrium</td>
</tr>
<tr>
<td>LIS1</td>
<td>Lissencephaly-1</td>
</tr>
<tr>
<td>Acronym</td>
<td>Full Form</td>
</tr>
<tr>
<td>---------</td>
<td>-----------</td>
</tr>
<tr>
<td>MATRICS</td>
<td>Measurement and Treatment Research to Improve Cognition in Schizophrenia</td>
</tr>
<tr>
<td>MET</td>
<td>Methionine</td>
</tr>
<tr>
<td>MTHFR</td>
<td>Methylene tetrahydrofolate reductase</td>
</tr>
<tr>
<td>NDE1</td>
<td>Nuclear Distribution Factor E</td>
</tr>
<tr>
<td>NDE1</td>
<td>Nuclear Distribution Factor E-like Factor</td>
</tr>
<tr>
<td>NFBC</td>
<td>Northern Finland 1966 Birth Cohort</td>
</tr>
<tr>
<td>NICE</td>
<td>National Institute of Health and Care Excellence</td>
</tr>
<tr>
<td>NMDA</td>
<td>N-Methyl-D-Aspartate</td>
</tr>
<tr>
<td>NOSA1P</td>
<td>Nitric Oxide Synthase 1 [Neuronal] Adaptor Protein</td>
</tr>
<tr>
<td>NOTCH4</td>
<td>Neurogenic Locus Notch Homologue</td>
</tr>
<tr>
<td>NRG1</td>
<td>Neuregulin 1</td>
</tr>
<tr>
<td>OR</td>
<td>Odds Ratio</td>
</tr>
<tr>
<td>PANSS</td>
<td>Positive and Negative Syndrome Scale</td>
</tr>
<tr>
<td>PDE</td>
<td>Phosphodiesterase</td>
</tr>
<tr>
<td>PIS</td>
<td>Population Information System</td>
</tr>
<tr>
<td>PPD</td>
<td>Primary Psychotic Disorder</td>
</tr>
<tr>
<td>PRODH</td>
<td>Proline Dehydrogenase</td>
</tr>
<tr>
<td>SANS</td>
<td>Scale for Assessment of Negative Symptoms</td>
</tr>
<tr>
<td>SAPS</td>
<td>Scale for Assessment of Positive Symptoms</td>
</tr>
<tr>
<td>SNP</td>
<td>Single-Nucleotide Polymorphism</td>
</tr>
<tr>
<td>SNV</td>
<td>Single-Nucleotide Variation</td>
</tr>
<tr>
<td>SSD</td>
<td>Schizophrenia Spectrum Disorder</td>
</tr>
<tr>
<td>ST8SIA2</td>
<td>ST8 Alpha-N-acetyl-Neuraminide Alpha-2,8-Sialyltransferase 2</td>
</tr>
<tr>
<td>T2DM</td>
<td>Type II Diabetes Mellitus</td>
</tr>
<tr>
<td>TNF</td>
<td>Tumour Necrosis Factor</td>
</tr>
<tr>
<td>TRAK1</td>
<td>Trafficking Kinesin Protein 1</td>
</tr>
<tr>
<td>RGS4</td>
<td>Regulator of G-Protein Signalling 4</td>
</tr>
<tr>
<td>ROS</td>
<td>Reactive oxygen species</td>
</tr>
<tr>
<td>TPH</td>
<td>Tryptophan Hydroxylase</td>
</tr>
<tr>
<td>VAL</td>
<td>Valine</td>
</tr>
<tr>
<td>VLBW</td>
<td>Very low birth weight</td>
</tr>
<tr>
<td>ZEB1</td>
<td>Zinc Finger E-Box Binding Homeobox 1</td>
</tr>
</tbody>
</table>
1. INTRODUCTION

Accumulating evidence suggests that the aetiology of schizophrenia is multifactorial, involving the interaction of genetic risk variations and environmental factors, and its pathogenesis has been proposed to originate long before the onset of psychotic symptoms (Lichtenstein et al., 2009; Insel, 2010; Van Os et al., 2010; Dick, 2011; Sullivan et al., 2012). Although psychotic symptoms are central to the diagnosis of schizophrenia, cognitive impairment and negative symptom severity dictate outcome-related measures (Karow et al., 2014; Kirkpatrick, 2014). Many schizophrenia-associated risk factors have been characterized, but less is known about the impact of these risk factors on symptom severity and clinical presentation of the disorder.

Pre- and perinatal factors represent a widely replicated group of environmental factors associated with schizophrenia risk (Cannon et al., 2002a; Byrne et al., 2007; Rapoport et al., 2012). The association between low birth weight (LBW) and schizophrenia is a consistent finding in the literature (Cannon et al., 2002a; Abel et al., 2010; Byars et al., 2014). Birth weight has been speculated to represent a proxy variable reflecting the influence of various environmental factors acting on the developing foetus (Fineberg et al., 2013; Galjaard et al., 2013; O'Connor et al., 2013). Evidence has suggested that high birth weight (HBW) may also increase schizophrenia risk (Hultman et al., 1997; Gunnell et al., 2003; Bersani et al., 2007; Moilanen et al., 2010; Keskinen et al., 2013).

A recent genome-wide association study (GWAS) has supported previous findings implicating a role for genes involved in immunological functioning, dopaminergic and glutamatergic neurotransmission, calcium signalling, cell adhesion, neuronal plasticity, and neurotransmitter release in schizophrenia-associated pathogenesis (Schizophrenia Genomics Consortium, 2014).

Genes of the Disrupted in Schizophrenia 1 (DISC1) network have been established to be associated with neurodevelopment (Devine et al., 2016). An association between schizophrenia and variants of DISC1, as well as other DISC1-associated gene variants, including nuclear distribution factor E (NDE1), nuclear distribution factor E-like factor (NDEL1), phosphodiesterase 4B (PDE4B) and phosphodiesterase 4D (PDE4D), have been found in the Finnish schizophrenia family sample (Ekelund et al., 2001; Hennah et al., 2003; Hennah et al., 2004; Hennah et al., 2007; Tomppo et al., 2009). Evidence suggests that DISC1 may also be associated with intermediate
neurocognitive phenotypes in schizophrenia and other psychiatric disorders (Hennah et al., 2005; Carless et al., 2011).

The aim of this thesis was to investigate the relationship between birth weight and both schizophrenia risk and the clinical presentation of schizophrenia, with respect to symptom severity and cognitive functioning, in a Finnish schizophrenia family study sample with high genetic loading for schizophrenia (Hovatta et al., 1997; Hovatta et al., 1999; Ekelund et al., 2000; Paunio et al., 2001). The interaction between birth weight and genetic variants from the DISC1 network (Hennah et al., 2007) was also evaluated.

It was hypothesized that the association between birth weight and schizophrenia risk would be augmented in a cohort presenting with increased genetic loading for schizophrenia. It was also hypothesized that deviations of birth weight may influence symptom severity and cognitive functioning in schizophrenia. Genes of the DISC1 network were postulated to interact with birth weight in augmenting schizophrenia risk.
2. REVIEW OF THE LITERATURE

Schizophrenia is one of the most severe psychiatric disorders, characterized by profound disturbances in fundamental processes governing human behaviour (Insel, 2010). The disorder typically manifests with marked social and occupational impairment and reduced life expectancy (McEvoy et al., 2005; Lauronen et al., 2007; Perälä et al., 2007; Saha et al., 2007; Kiviniemi et al., 2010; Wildgust et al., 2010; Laursen et al., 2012; Viertiö et al., 2012; Crump et al., 2013; Nordentoft et al., 2013; Suvisaari et al., 2013; Ringen et al., 2014). In addition to elevated morbidity, the financial burden of schizophrenia on society is formidable (Gustavsson et al., 2011).

2.1. HISTORICAL PERSPECTIVE

In the late nineteenth century, the German psychiatrist Emil Kraepelin introduced the term *Dementia praecox* to describe a form of premature dementia that (1) had an onset in late adolescence and (2) was proposed to have a deteriorating course. Kraepelin described *Dementia praecox* as a disorder distinct from the remitting and non-deteriorating course seen in individuals presenting with manic-depressive illness (Insel, 2010).

The term *schizophrenia* was introduced in 1908 by the Swiss psychiatrist Eugen Bleuler, who emphasized the construct as being highly heterogeneous with respect to both clinical presentation and prognosis, as echoed by Bleuler’s use of the term “a group of schizophrenias” (Insel, 2010). Bleuler’s focus was on delineating a core unifying symptom underlying the disorder, which he concluded as being “the breaking up or splitting of psychic functioning”. Bleuler described the terms *Affect, Autism, loosening of Associations, Ambivalence* and deficits in *Attention* to describe core manifestations of schizophrenia (Bleuler, 1911, translated by Zinkin, 1950; Kuhn, 2004; Hahn et al., 2012). Bleuler’s description of the disorder and use of the Greek term *phren* – mind – has been suggested to emphasize *impaired cognition* as a defining core feature of the original definition of schizophrenia (Andreasen, 1999).

The diagnosis of schizophrenia was refined by the German psychiatrist Kurt Schneider, who set out to differentiate schizophrenia from other psychotic disorders by attempting to describe symptoms specific to schizophrenia (Cutting, 2015). Schneider’s eleven *first rank symptoms* audible thoughts, voices arguing and/or discussing, voices commenting, somatic passivity
experiences, thought withdrawal, thought insertion, thought broadcasting, made volition, made affect, made impulse and delusional perception have remained central to the diagnostic criteria of schizophrenia (Carpenter, 1974; Nordgaard et al., 2008; Tandon et al., 2013; American Psychiatric Association, 2014). Schneider emphasized these symptoms of reality distortion as being pathognomonic to schizophrenia, which has been suggested to have led to a relative neglect in the development of treatments for other core symptoms underlying the disorder (Carpenter et al., 2004).

2.2. DIAGNOSIS OF SCHIZOPHRENIA

Despite recent advances in the fields of genetics and brain imaging, the diagnosis of schizophrenia is still purely descriptive, based on clinical observation and patient interviews (World Health Organization, 1993; American Psychiatric Association, 2014). Kraepelin, Bleuler and Schneider’s conceptualization of schizophrenia reflect the roots of today’s diagnostic classification of schizophrenia (Tandon et al., 2013). The diagnostic criteria of schizophrenia with respect to the International Classification of Diseases, tenth revision (ICD-10) (World Health Organization, 1993), the Diagnostic and Statistical Manual of Mental Disorders, fourth edition, text revision (DSM-IV) (American Psychiatric Association, 2000) and the Diagnostic and Statistical Manual of Mental Disorders, fifth edition (DSM-5) (American Psychiatric Association, 2014) are presented in Tables 1, 2 and 3, respectively.

DSM-IV includes the criterion that symptoms must result in functional impairment. ICD-10 does not include a criterion of functional impairment. DSM-IV also requires that continuous signs of the disturbance in some form have persisted for a period of at least 6 months, in comparison to ICD-10, which requires that symptoms have been present for one month. Both the ICD-10 and DSM-IV differentiate between different subtypes of schizophrenia.

In DSM-5 (American Psychiatric Association, 2014), the diagnosis of schizophrenia has been further refined relative to DSM-IV. In DSM-5, the special emphasis on Schneiderian first-rank symptoms has been eliminated. In DSM-5 two or more of the following symptoms must be present for a significant proportion of time over a one-month period: delusions, hallucinations, disorganized speech, grossly disorganized or catatonic behaviour and negative symptoms, and one of the symptoms must be delusions, hallucinations or disorganized speech (Tandon et al., 2013). DSM-5 no longer distinguishes different subtypes of schizophrenia, and catatonia can be used as a descriptive specifier to describe the phenotype of any psychiatric disorder.
<table>
<thead>
<tr>
<th>Table 1.</th>
<th>ICD-10 diagnostic criteria of schizophrenia.</th>
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<tbody>
<tr>
<td>G1.1 <em>Either at least one of the syndromes, symptoms and signs A1-A4 should be present for most of the time for at least one month.</em></td>
<td>A1. Thought echo, thought insertion, thought withdrawal, thought broadcasting.</td>
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<td></td>
<td>A2. Delusions of control, influence or passivity, clearly referring to body or limb movements or specific thoughts, actions or sensations; delusional perception.</td>
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<td></td>
<td>A3. Hallucinatory voices giving a running commentary on the patient's behaviour, or discussing him between themselves, or other types of hallucinatory voices coming from some part of the body.</td>
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<td></td>
<td>A4. Persistent delusions of other kinds that are culturally inappropriate and completely impossible</td>
</tr>
<tr>
<td>G1.2 <em>Or at least two of the syndromes, symptoms and signs B1-B4 should be present for most of the time for at least one month.</em></td>
<td>B1. Persistent hallucinations in any modality, when accompanied by delusions, without clear affective content, or when accompanied by persistent over-valued ideas.</td>
</tr>
<tr>
<td></td>
<td>B2. Neologisms, breaks or interpolations in the train of thought, resulting in incoherence or irrelevant speech.</td>
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<td></td>
<td>B3. Catatonic behaviour, such as excitement, posturing or waxy flexibility, negativism, mutism and stupor.</td>
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<tr>
<td></td>
<td>B4. Negative symptoms such as marked apathy, paucity of speech and blunting or incongruity of emotional responses.</td>
</tr>
<tr>
<td>G2. Most commonly used exclusion criteria:</td>
<td>If the patient also meets criteria for manic episode (F30) or depressive episode (F32), the criteria listed under G1.1 and G1.2 above must have been met before the disturbance of mood developed.</td>
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<td></td>
<td>The disorder is not attributable to organic brain disease (F0X) or to alcohol- or drug-related intoxication, dependence or withdrawal.</td>
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Table 2. DSM-IV diagnostic criteria of schizophrenia.

| A. Two (or more) of the symptoms A1-A5, are present for a significant portion of time during a 6-month period (or less if successfully treated) | (A1) Delusion  
(A2) Hallucinations  
(A3) Disorganized speech  
(A4) Grossly disorganized or catatonic behaviour  
(A5) Negative symptoms  
Only one Criterion is required if delusions are bizarre or hallucinations consist of:  
(I) a voice keeping up a running commentary on the person's behaviour or thoughts, or  
(II) two or more voices conversing with each other. |
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<td>B. Social/occupational dysfunction</td>
<td>For a significant portion of the time since the onset of the disturbance, one or more major areas of functioning such as work, interpersonal relations or self-care are markedly below the level achieved prior to the onset (or when the onset is in childhood or adolescence, failure to achieve an expected level of interpersonal, academic or occupational achievement).</td>
</tr>
</tbody>
</table>
| C. Duration | Continuous signs of the disturbance persist for at least 6 months. This 6-month period must include at least one month of symptoms (or less if successfully treated) that meet Criterion A (i.e., active-phase symptoms) and may include periods of prodromal or residual symptoms.  
During these prodromal or residual periods, the signs of the disturbance may be manifested by only negative symptoms or two or more symptoms listed in Criterion A present in an attenuated form. |
| D. Schizoaffective and Mood Disorder exclusion | Schizoaffective Disorder and Mood Disorder With Psychotic Features have been ruled out because either:  
(1) no Major Depressive Episode, Manic Episode or Mixed Episode have occurred concurrently with the active-phase symptoms; or  
(2) if mood episodes have occurred during active-phase symptoms, their total duration has been brief relative to the duration of the active and residual periods. |
| E. Substance/general medical condition exclusion | The disturbance is not due to the direct physiological effects of a substance or a general medical condition. |
| F. Relationship to a Pervasive Developmental Disorder | If there is a history of Autistic Disorder or another Pervasive Developmental Disorder, the additional diagnosis of Schizophrenia is made only if prominent delusions or hallucinations are also present for at least one month (or less if successfully treated). |
Table 3. DSM-5 diagnostic criteria of schizophrenia.

| A. Two (or more) of the following symptoms. Each present for a portion of time during a one-month period (or less if successfully treated). At least one of these must be (A1), (A2) or (A3). | (A1) Delusions  
(A2) Hallucinations  
(A3) Disorganized speech  
(A4) Disorganized behaviour or catatonic behaviour  
(A5) Negative symptoms |
|---|---|
| B. For a significant portion of time since the onset of the disturbance the level of functioning in one or more major areas of the following is markedly below the level achieved prior to the onset: | Work, interpersonal relations, self-care.  
Or when the onset is in childhood or adolescence, there is failure to achieve expected level of interpersonal, academic, occupational functioning. |
| C. Continuous signs of the disturbance persist for at least 6 mos. This 6-month period must include at least one month of symptoms (less if successfully treated) that meet Criterion A | Active-phase symptoms may include periods of prodromal symptoms  
Residual symptoms. During these prodromal or residual period the signs of the disturbance may be manifested by only negative symptoms or by two or more symptoms listed in Criterion A present in an attenuated form, e.g. odd beliefs, unusual perceptual experiences). |
| D. Schizoaffective disorder, depressive or bipolar disorder with psychotic features have been ruled out because either | (1) No major depressive or manic episodes has occurred concurrently with active-phase symptoms.  
(2) Or they have been present for a minority of the total duration of the active and residual periods of the illness. |
| E. The disturbance is not attributable to the physiological effects of a substance | A drug of abuse, a medication or a medical condition. |
| F. If there is a history of autism spectrum disorder or a communication disorder of childhood onset. | The additional diagnosis of schizophrenia is made only if prominent delusions or hallucinations, in addition to other required symptoms of schizophrenia, are also present for at least one month. |
2.3. SYMPTOMS OF SCHIZOPHRENIA

Although the diagnosis of schizophrenia is categorical, psychotic symptoms in the general population have been suggested to reflect a dimensional continuum (Linscott and Van Os, 2010). The schizophrenia phenotype has been suggested to reflect an extreme phenotype of this continuum (Van Os et al., 2010). Dissection of the schizophrenia phenotype into clinically relevant symptom dimensions has been proposed to both facilitate our understanding of the aetiological mechanisms underlying the disorder and increase the efficacy of treatments targeting specific symptom dimensions (Derks et al., 2010; Wykes et al., 2011).

Symptoms have broadly been found to cluster into two clearly discernible domains, historically referred to as positive and negative symptoms (Andreasen et al., 1995). Positive symptoms have been suggested to reflect a state presenting with “an excess of vital properties” (the presence of something that should be absent), and negative symptoms reflecting “the loss of vital properties” (the absence of something that should be present) (Andreasen, 1995; Messinger et al., 2011).

Structured symptom rating scales have been developed to aid goal-directed treatment of psychotic disorders. Commonly used scales include (1) the Scale for Assessment of Positive Symptoms (SAPS) (Andreasen, 1984), (2) the Scale for Assessment of Negative Symptoms (SANS) (Andreasen, 1983), (3) the Positive and Negative Syndrome Scale (PANSS) (Kay et al., 1987), (4) the Brief Psychiatric Rating Scale (Overall and Gorham, 1962). The clinical reliability and validity of symptom rating scales is well established (Klimidis et al., 1993; Andreasen et al., 1995).

2.3.1. POSITIVE SYMPTOMS

Positive symptoms are longitudinally more fluctuating than negative symptoms, which are more stable and enduring (Andreasen et al., 1995; Kirkpatrick et al., 2006; Kirkpatrick, 2014; Austin et al., 2015) (Table 4).

Positive symptoms in psychotic disorders segregate into two clinically distinguishable domains: (1) symptoms of reality distortion and (2) disorganized symptoms, which differ with respect to clinical presentation, symptom stability and outcome measures (Ventura et al., 2010). Symptoms of reality distortion consist of (1) delusions and (2) hallucinations (Andreasen, 1986). Disorganized symptoms are typically divided into (1) disorganized
thought, referring to the terms conceptual disorganization and positive formal thought disorder and (2) disorganized behaviour (Ventura et al., 2010) (Figure 1).

Disorganized symptoms have been found to be associated with greater cognitive impairment (Ventura et al., 2010; Ventura et al., 2013; Minor and Lysaker, 2014) and greater impairment in long-term functioning than symptoms of reality distortion (Ventura et al., 2010). Formal thought disorder has been found to moderate the relationship between neurocognition, social cognition and metacognition, suggesting that the presence of disorganized symptoms in schizophrenia limits the effective utilization of the neurocognitive abilities necessary for performing social cognitive or metacognitive tasks (Minor and Lysaker, 2014; Minor et al., 2015).
Table 4. Scale for Assessment of Positive Symptoms (SAPS) (Andreasen, 1984).

<table>
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<tr>
<th>Hallucinations</th>
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<tbody>
<tr>
<td>• 1. Auditory hallucinations</td>
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<td>• 2. Voices commenting</td>
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<tr>
<td>• 3. Voices conversing</td>
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<tr>
<td>• 4. Somatic hallucinations</td>
</tr>
<tr>
<td>• 5. Olfactory hallucinations</td>
</tr>
<tr>
<td>• 6. Visual hallucinations</td>
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<td>• 7. <strong>Global rating of hallucinations</strong></td>
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<th>Delusions</th>
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<tr>
<td>• 8. Persecutory delusions</td>
</tr>
<tr>
<td>• 9. Delusions of jealousy</td>
</tr>
<tr>
<td>• 10. Delusions of guilt/sin</td>
</tr>
<tr>
<td>• 11. Grandiose delusions</td>
</tr>
<tr>
<td>• 12. Religious delusions</td>
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<tr>
<td>• 13. Somatic delusions</td>
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<tr>
<td>• 14. Delusions of reference</td>
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<tr>
<td>• 15. Delusions of being controlled</td>
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<tr>
<td>• 16. Delusions of mind reading</td>
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<tr>
<td>• 17. Thought broadcasting</td>
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<td>• 18. Thought insertion</td>
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<td>• 19. Thought withdrawal</td>
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<tr>
<td>• 20. <strong>Global rating of delusions</strong></td>
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<th>Bizarre behavior</th>
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<tr>
<td>• 21. Clothing and appearance</td>
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<tr>
<td>• 22. Social and sexual behaviour</td>
</tr>
<tr>
<td>• 23. Aggressive and agitated behaviour</td>
</tr>
<tr>
<td>• 24. Repetitive or stereotyped behaviour</td>
</tr>
<tr>
<td>• 25. <strong>Global rating of bizarre behaviour</strong></td>
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<tr>
<th>Positive formal thought disorder</th>
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<tbody>
<tr>
<td>• 26. Derailment</td>
</tr>
<tr>
<td>• 27. Tangentiality</td>
</tr>
<tr>
<td>• 28. Incoherence</td>
</tr>
<tr>
<td>• 29. Illogicality</td>
</tr>
<tr>
<td>• 30. Circumstantiality</td>
</tr>
<tr>
<td>• 31. Pressure of speech</td>
</tr>
<tr>
<td>• 32. Distractible speech</td>
</tr>
<tr>
<td>• 33. Changing</td>
</tr>
<tr>
<td>• 34. <strong>Global rating of formal thought disorder</strong></td>
</tr>
</tbody>
</table>
2.3.2. NEGATIVE SYMPTOMS

In the 1970s and 1980s, negative symptoms were described as being separable from other symptoms of schizophrenia (Carpenter et al., 1988; Strauss et al., 2013). Negative symptoms are divided into primary and secondary negative symptoms with respect to aetiological considerations (Carpenter et al., 1988; Kirkpatrick, 2014). Primary negative symptoms, also referred to as deficit symptoms, are suggested to be more proximal to the biological core of the disorder and are associated with increased cognitive, social and functional impairment (Kirkpatrick, 2014). Secondary negative symptoms are proposed to reflect the manifestations of the side-effects of medications, affective symptoms and limited social stimulation (Kirkpatrick, 2014).

Negative symptoms are associated with cognitive, social and functional impairment in schizophrenia (Bottlender et al., 2010; Fulford et al., 2013; Kirkpatrick, 2014). Negative symptoms are enduring and more refractory to antipsychotic treatment than positive symptoms (Kirkpatrick et al., 2006; Levine and Leucht, 2013; Ventura et al., 2015).

Negative symptoms have been suggested to be multidimensional and have been dissected into subgroups according to clinical characteristics. In the SANS, negative symptoms are divided into the five following dimensions: affective flattening, alogia, avolition-apathy, anhedonia-asociality and attention (Andreasen, 1983) (Table 5). Based on this division, negative symptoms have been found to coalesce into at least two separable dimensions (1) an expressive factor consisting of affective flattening and alogia and (2) an avolition factor consisting of avolition, anhedonia and asociality (Blanchard and Cohen, 2006; Kirkpatrick, 2014) (Figure 1).
**Table 5.** Scale for Assessment of Negative Symptoms (SANS) (Andreasen, 1983).

<table>
<thead>
<tr>
<th>Category</th>
<th>Items</th>
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</table>
| Affective flattening    | • 1. Facial expression  
                          • 2. Spontaneous movements  
                          • 3. Expressive gestures  
                          • 4. Eye contact  
                          • 5. Affective non-responsivity  
                          • 6. Vocal inflections  
                          • 7. Inappropriate affect  
                          • 8. Global rating of affective flattening |
| Alogia                  | • 9. Poverty of speech  
                          • 10. Poverty of content  
                          • 11. Blocking  
                          • 12. Latency of response  
                          • 13. Global rating of alogia |
| Avolition–apathy        | • 14. Grooming and hygiene  
                          • 15. Impersistence at work  
                          • 16. Physical anergia  
                          • 17. Global rating of avolition–apathy |
| Anhedonia–asociality    | • 18. Recreational interests  
                          • 19. Sexual interest  
                          • 20. Intimacy and closeness  
                          • 21. Relationships with friends  
                          • 22. Global rating of anhedonia–asociality |
| Attention               | • 23. Social inattentiveness  
                          • 24. Inattentiveness during testing  
                          • 25. Global rating of attention |
2.4. COGNITIVE IMPAIRMENT IN SCHIZOPHRENIA

Patients with schizophrenia typically present with cognitive impairment (Keefe and Reichenberg, 2014). Evidence suggests that cognitive performance is typically 1-2 standard deviations lower in individuals with schizophrenia than in age-matched nonaffected peers. Subclinical cognitive impairment has also been observed among a proportion of first-degree relatives of subjects with schizophrenia, suggesting that cognitive impairment is associated with genetic liability (Faraone et al., 1999; Tuulio-Henriksson et al., 2003; Kuha et al., 2007).
Cognitive impairment typically manifests prior to the onset of first psychotic symptoms and remains relatively stable after the remission of positive symptoms (Fioravanti et al., 2005; Lewandowski et al., 2011; Rapoport et al., 2012; Nuechterlein et al., 2014). Evaluation of cognitive impairment is used in clinical practice both as a predictor of functional outcome and to aid the planning of rehabilitation interventions (Green et al., 2000; Lepage et al., 2014; Bechi et al., 2015).

Cognitive impairment in schizophrenia is relatively unspecific and has been found to be associated with various domains of cognitive functioning, including language functions, executive functioning (reasoning, problem solving), speed of processing, attention, working memory, verbal learning and memory, visual learning and memory, verbal comprehension, social cognition and motor skills (Tuulio-Henriksson et al., 2003; Paunio et al., 2004; Fioravanti et al., 2005; Hennah et al., 2005; Mesholam-Gately et al., 2009; Gur et al., 2011; Lesh., 2011; Barch et al., 2012; Catts et al., 2013; Schaefer et al., 2013).

Recently, the National Institute of Mental Health (NIMH) launched the Measurement and Treatment Research to Improve Cognition in Schizophrenia (MATRICS) initiative to validate specific clinically relevant areas of cognition in order to standardize the assessment of treatment efficacy on cognition in clinical trials (Green and Nuechterlein, 2004; Nuechterlein et al., 2014). The MATRICS panel suggested that seven cognitive domains be assessed when evaluating treatment efficacy: (1) processing speed, (2) attention/vigilance, (3) working memory, (4) verbal learning, (5) visual learning, (6) reasoning and problem solving and (7) social cognition.

Both negative and disorganized symptoms have been found to be associated with impairment in several neurocognitive domains. Although impairment is relatively generalized, some areas of cognition have been studied more than others. Disorganized symptoms have been associated with deficits involving intelligence, attention span, sensory motor function and poor concept attainment (Basso et al., 1998; O’Leary et al., 2000). Negative symptoms have been associated with deficits involving verbal learning and memory, verbal fluency, visual memory, visual motor sequencing, intelligence, executive function, sustained attention and sensory motor function (Basso et al., 1998; O’Leary et al., 2000).
2.5. EARLY PHASES OF SCHIZOPHRENIA

Characterization of the longitudinal course of schizophrenia has found that symptoms and functional impairment typically emerge years before the onset of overt psychotic symptoms (Häfner and Maurer, 2006).

Subtle developmental abnormalities during childhood (neurological, emotional, social and cognitive) have been reported in prospective studies among subjects who later develop schizophrenia (Isohanni et al., 2000; Mäki et al., 2005; Niemi et al., 2005; Insel, 2010; Rapoport et al., 2012).

The prodromal phase of schizophrenia has been defined as the period between the onset of behavioural change in a person and the onset of psychotic symptoms (Yung et al., 1996). A constellation of symptoms, including depressive symptoms, anxiety, dysphoria, negative symptoms and non-specific subjective experiences, also typically increasingly manifest during progression of the disorder (Häfner et al., 2003). Depressive symptoms are particularly common among individuals who later develop schizophrenia, with approximately 85% of patients presenting with depressive symptoms during the early course of the disorder and 25% presenting with depressive symptoms upon first admission (Häfner and Maurer, 2006). Increasing symptom severity typically presents with increasing functional (social, occupational and academic) impairment (Häfner et al., 2003). Some authors have dissected the prodromal phase into two phases with respect to the presentation of overt psychotic symptoms: a prepsychotic prodromal stage (mean duration 4.8 years) and a psychotic prephase (mean duration 1.3 years) (Häfner and Maurer, 2006). The psychotic prephase is characterized by the presentation of brief and attenuated psychotic symptoms, which eventually give way to overt psychotic symptoms. The duration of untreated psychosis is inversely associated with functional outcome, emphasizing the role of early intervention as a means to improve treatment outcome (Penttilä et al., 2014).

Early intervention strategies have been developed for the detection of subjects presenting with attenuated or brief psychotic symptoms and those with an elevated risk of psychosis (Häfner et al., 1992; McGlashan, 1998; Miller et al., 2003).
2.6. AETIOLOGICAL MODELS OF SCHIZOPHRENIA

Individuals with schizophrenia have been found to present with a small but significant reduction in intracranial volume and a reduction in total brain volume, with respect to both total grey and white matter (Fusar-Poli et al., 2013; Haijma et al., 2013). Although widely investigated, the clinical utility of brain imaging findings remains limited. Several aetiological models have been put forth to describe and integrate the postulated biological and psychological mechanisms underlying the disorder (Linscott and Van Os, 2010; Howes and Murray, 2014; Howes et al., 2015).

From a historical perspective, schizophrenia was once considered to be a neurodegenerative disorder, with a deteriorating course, the biological onset of which was thought to correlate with the manifestation of symptoms, typically during early adulthood (Weinberger, 1995). The neurodevelopmental model of schizophrenia was based on observations describing an association between obstetric complications and increased schizophrenia risk and evidence suggesting that brain imaging findings did not show significant progression after disease onset (Lewis and Murray, 1987; Murray and Lewis, 1987; Weinberger, 1987). However, today some research indicates that extended periods of symptom relapse may be associated with reductions in brain volume (Andreasen et al., 2013) and that reductions in brain volume may even be associated with the use of antipsychotic medications (Guo et al., 2015). Findings describing the interaction between genetic liability and obstetric complications in relation to increased schizophrenia risk and the presence of subtle deviations in emotional, cognitive and motor development in children prior to the onset of schizophrenia have complemented the model (Cannon et al., 1989; Howes and Murray, 2014). A reductionistic “two hit” model was later put forth to exemplify the longitudinal relationship between cumulative environmental insults with the potential to impact the neurodevelopmental process (Keshavan et al., 2011). In this model, genetic susceptibility, in combination with pre- or perinatal factors, was suggested to exert a subtle disruptive effect on the neurodevelopmental process upon birth, decreasing the threshold for developing the disorder upon encounter with future environmental adversities.

The dopamine hypothesis was based on findings demonstrating that the administration of drugs that increase dopamine transmission (e.g. amphetamine) cause schizophrenia-like symptoms, whereas the administration of drugs that decrease dopamine transmission (e.g. reserpine) reduce schizophrenia-like symptoms (Howes et al., 2015). Further evidence of the central role of dopamine in schizophrenia came from widely replicated findings showing that antipsychotics reduce psychotic symptoms by blocking
striatal dopamine D2/3 receptors (Carlsson, 1977; Howes et al., 2012; Dragicevic et al., 2015; Howes et al., 2015; Sarpal et al., 2016). An association between the D2-receptor gene and schizophrenia was also found in the most recent GWAS (Schizophrenia Genomics Consortium, 2014). However, in a meta-analysis of more than 50 brain imaging studies focusing on the striatal dopamine system in schizophrenia the alterations in postsynaptic D2/3 receptor availability were small and inconsistent and the presynaptic dopamine transporter availability did not differ among cases and controls (Howes et al., 2012). The meta-analysis of Howes et al. (2012) corroborated previous evidence suggesting that schizophrenia was associated with increased presynaptic dopamine synthesis capacity, dopamine release and increased baseline synaptic dopamine concentrations.

The presynaptic dysregulation of dopamine has been suggested to be susceptible to the influence of environmental factors, including pre- and perinatal factors and stressful lifetime events during childhood (Howes and Murray, 2014; Howes et al., 2015). In animal studies, prenatal infection, Caesarean section and mild prenatal/perinatal hypoxia have been found to be associated with elevated dopamine synthesis and sensitization of the dopaminergic system (Howes and Murray, 2014). Subjects with a history of low maternal care during childhood have shown increased striatal dopamine release upon stressful events, suggesting sensitization of the dopaminergic system to early psychosocial stress (Howes and Murray, 2014). Preclinical findings indicate that aberrant neurodevelopment may influence the dopamine system. For example, in an animal model the transient knockdown of DISC1 expression during the pre- and perinatal stages of development has been demonstrated to be associated with abnormalities in postnatal mesocortical dopaminergic transmission and behavioural abnormalities after puberty (Niwa et al., 2010).

Glutamate is the primary excitatory neurotransmitter of the central nervous system (Kim et al., 1980). The glutamate hypothesis suggests that N-methyl-D-aspartic acid (NMDA) receptor dysfunction (hypofunction) is involved in the pathogenesis of schizophrenia (Stone et al., 2011). The role of glutamatergic transmission in schizophrenia is based on observations describing positive, negative and cognitive symptoms elicited upon the administration of NMDA receptor antagonists ketamine, dizocilpine and phencyclidine respectively (Howes et al., 2015). Some studies have suggested that agonists of the NMDA receptor (glycine or D-serine), acting at the glycine modulatory site on the NMDA receptor, are associated with improvement in residual positive and negative symptoms in patients with schizophrenia when co-administered with antipsychotic treatment (Tsai and Lin, 2010). NMDA receptor hypofunction in prefrontal gamma-aminobutyric acid (GABA) interneurons has been suggested to be associated with a reduction in the
inhibitory regulatory capacity of the prefrontal cortex over dopaminergic mesolimbic structures, resulting in hyperactive mesolimbic dopamine D2 transmission and psychotic symptoms (Nakazawa et al., 2012). Dopamine and glutamate theories have been suggested to converge in relating prefrontal glutamatergic hypofunction with subcortical hyperdopaminergic transmission (Howes et al., 2015). An association between several glutamate receptor genes and schizophrenia was also replicated in the most recent GWAS (Schizophrenia Genomics Consortium, 2014). One of the genes identified in the study, the metabotropic glutamate receptor 3 (GRM3), has also been found to interact with severe obstetric complications, with a concomitant decrease in hippocampal volume (Haukvik et al., 2010). GRM3 mRNA has also been observed to be downregulated in response to ischaemia (Raghavendra Rao et al., 2002; Lu et al., 2004).

2.7. RISK FACTORS

Both genetic and environmental factors mediate schizophrenia risk (Sullivan et al., 2012). Schizophrenia has been associated with heritability estimates of up to 81%, compared with heritability estimates of 75% for bipolar disorder and 37% for major depressive disorder, suggesting that schizophrenia is associated with a strong genetic component, equivalent to that of autism spectrum disorders (ASDs) (80%) (Sullivan et al., 2012). Environmental factors play an important role in the pathophysiology of schizophrenia as well (Brown, 2011a), and the role of obstetric complications is widely acknowledged (Cannon et al., 2002a).

2.7.1. GENETIC RISK FACTORS

The genome consists of approximately 3 billion base pairs, the vast majority of which (>98%) comprise of non-coding sequence, containing regulatory elements and sequences of yet unknown function, the remainder (<2%) being protein coding sequence (www.genome.gov). Any two individuals are genetically 99.9% identical. Identification of the genetic variation in the remaining 0.1% of the genome, which is associated with morbidity, is thus challenging and requires increasingly larger samples to detect the subtle differences in genetic variation conferring genetic risk in a population. Most of the variation in the genome is associated with sequences of a small individual effect on morbidity. Of all human proteins (n=19 692), 67% are expressed in the brain (www.humanproteinatlas.org). The variation of single nucleotides in the genome is arbitrarily considered common if it occurs in >1%
of the population, referred to as a single-nucleotide polymorphism (SNP), and rare if it occurs in <1% of the population, referred to as a single-nucleotide variation (SNV). Larger deletions and duplications in the genome, termed copy number variants (CNVs), are typically of larger effect size, i.e. exert a more substantial effect on phenotype (Sullivan et al., 2012). Genetic variation is transmitted though generations or it can occur as a result of spontaneous (de novo) mutations (Sanders et al., 2012; Fromer et al., 2014).

Compared with most populations in Europe, the Finnish population exhibits less genetic diversity and more linkage disequilibrium (LD) (Sajantila et al., 1996; Varilo et al., 2003; Jakkula et al., 2008). One consequence of this is the Finnish Disease Heritage, a group of 36 rare hereditary, monogenic diseases that are more prevalent in Finland than elsewhere in the world (http://www.findis.org/heritage.html). These diseases have been found to cause mental retardation, visual impairment, congenital malformations, bone disorders, hearing loss, metabolic disturbances, epileptic or deteriorating neurological diseases, blood disorders and multisystemic syndromes (Norio, 2003).

Isolates, founded by a small group of settlers experiencing significant population growth, present with elevated levels of LD and exhibit fewer regions of very low LD relative to outbred populations (Service et al., 2006). Population isolates, which exhibit both genetic and environmental homogeneity, have been utilized in the identification of rare genetic variants enriched in specific subpopulations segregating with complex genetic disorders including schizophrenia (Hovatta et al., 1999; Myles-Worsley et al., 1999; Peltonen et al., 2000; DeLisi et al., 2002; Jakkula et al., 2008; Åberg et al., 2008; Paunio et al., 2009; Myles-Worsley et al., 2011; Peltola et al., 2016). The Finnish population contains several isolates (Jakkula et al., 2008).

The Finnish Internal Isolate in Northeastern Finland was originally founded in the 17th century by 34 families (Hovatta et al., 1997). Until World War II, the population of the internal Isolate remained relatively isolated. In the Isolate, people have predominantly found spouses from the same village, and even marriages between people living in northern and southern parts of the same municipality have been rare. The estimated kinship coefficient is 1.43 times higher than the expected value (Paunio et al., 2009). After World War II, emigration from the municipality to southern Finland increased, whereas immigration into the municipality remained limited (Hovatta et al., 1999). Due to the small number of founders and rapid expansion, the population in the region is genetically more homogeneous and LD is higher than in the rest of Finland or in other isolates investigated throughout the world (Varilo et al., 2003; Service et al., 2006; Jakkula et al., 2008). The lifetime risk of schizophrenia in the Isolate region is elevated (3.2%) relative to the rest of
Finland and other populations (Hovatta et al., 1997; Saha et al., 2005; McGrath et al., 2008). Some differences between the Isolate and the rest of Finland have been reported with respect to both susceptibility loci and frequency of specific risk alleles (Paunio et al., 2001; Hennah et al., 2003; Paunio et al., 2004; Wedenoja et al., 2008; Wessman et al., 2009; Wedenoja et al., 2010; Stoll et al., 2013; Peltola et al., 2016). For example, an intragenic short tandem repeat allele within the Reelin gene, which has been found to be associated with moderate cognitive impairment in the rest of Finland, is almost absent in individuals originating from the Isolate (Wedenoja et al., 2008; Wedenoja et al., 2010). Conversely, a deletion encompassing the the topoisomerase 3β (TOP3β) gene, associated with a 2-fold risk of schizophrenia, is enriched in the internal Isolate (Stoll et al. 2013).

Investigation of symptom dimensions between individuals with schizophrenia in relation to the Isolate region and the rest of Finland found that individuals with schizophrenia descending from the Isolate presented with less positive symptoms (delusions and hallucinations) than individuals with schizophrenia deriving from the rest of Finland, which was suggested to be related to the genetic homogeneity of the Isolate (Arajärvi et al., 2006).

GENOME-WIDE ASSOCIATION STUDIES

Recent GWASs investigating the association of common genetic variability of small effect size on morbidity in large multinational population-based samples have shed increasing light on common gene variants associated with schizophrenia (Schizophrenia Genomics Consortium, 2014).

In the most recent GWAS to date, encompassing 36,989 subjects with schizophrenia, 108 loci meeting genome-wide significance were identified (Schizophrenia Genomics Consortium, 2014). Of these loci, 75% were found to associate with protein-coding regions, and 8% were found to be within close proximity (<20 kb) to protein coding regions. The study corroborated previous findings describing variants associated with pre-existing biologically relevant aetiological determinants of schizophrenia, including genes involved in (1) dopamine signalling (D2-dopamine receptor), (2) glutamate signalling, (3) calcium signalling, (4) synaptic functioning and plasticity, (5) potassium and cholinergic signalling, and (5) neurodevelopment (Table 6).
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<td><strong>Glutamate signalling</strong></td>
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<td>• AMPA type subunit 1 (GRIA1)</td>
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<td>• Serine racemase (SRR)</td>
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<td>• Chloride voltage-gated channel 3 (CLCN3)</td>
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<td>• Solute carrier family 38 member 7 (SLC38A7)</td>
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<td><strong>Calcium signalling</strong></td>
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<td>• auxiliary subunit beta 2 (CACNB2)</td>
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<td>• Regulating synaptic membrane exocytosis 1 (RIMS1)</td>
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<td>• Calcium/calmodulin-dependent protein kinase kinase 2 (CAMKK2)</td>
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<td>• Neurogranin (NRGN)</td>
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<td><strong>Synaptic function and plasticity</strong></td>
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<td>• Potassium channel tetramerization domain containing 1 (KCTD)</td>
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<td>• Neurogin 4 (NLGN4X)</td>
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<td>• Contactin 4 (CNTN4)</td>
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<td>• Immunoglobulin superfamily member 9B (IGSF9B)</td>
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<td>• Myocyte enhancer factor 2C (MEF2C)</td>
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<td>• Pleiotrophin (PTN)</td>
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<td>• Connector enhancer of kinase suppressor of Ras 2 (CNKSR2)</td>
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<td>• p21 (RAC1) activated kinase 6 (PAK6)</td>
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<td>• Synaptosome-associated protein 91 (SNAP91)</td>
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<td><strong>Other ion channels</strong></td>
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<td>• Potassium voltage-gated channel subfamily B member 1 (KCNB1)</td>
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<td>• Hyperpolarization activated cyclic nucleotide-gated K⁺ channel 1 (HCN1)</td>
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<td>• Cholinergic receptor nicotinic</td>
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<td>• alpha 3 subunit (CHRNA3)</td>
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<td>• alpha 5 subunit (CHRNA5)</td>
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<td>• beta 4 subunit (CHRNB4)</td>
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<td><strong>Neurodevelopment</strong></td>
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<td>• FMR1 autosomal homologue 1 (FXR1)</td>
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<td>• SATB homeobox 1 (SATB)</td>
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COPY NUMBER VARIATIONS

CNVs are large deletions, duplications or rearrangements in the genome, typically >1 kb in size (Heyes et al., 2015). Genetic research on neurodevelopmental disorders has focused on the role of CNVs of large effect size (Purcell et al., 2014; Sanders et al., 2015). Large CNVs are found in 3.3% of individuals with ASDs and in 2.4% of individuals with schizophrenia, compared with 0.5% of individuals without these disorders (Kirov, 2015). CNVs have been reported to be associated with a 2- to 50-fold increase in schizophrenia risk (Sullivan et al., 2012; Kirov, 2015). At least 11 CNVs have been shown to increase schizophrenia risk, including deletions at 1q21.1, 3q29, 15q11.2, 15q13.3 and 22q11.2 and duplications at 1q21.1, 7q11.23, 15q11.2-q13.1, 16p13.1 and 16p11.2 (Kirov, 2015). Many of the identified CNVs are pleiotropic in that they associate with several neurodevelopmental outcomes, including cognitive deficits, mental retardation, epilepsy, ASDs and schizophrenia (Szatkiewicz et al., 2014). Interestingly, female sex has been associated with an elevated mutation burden, implying that larger CNVs are required for clinical manifestations to take effect in females than in males, sometimes referred to as the ‘female protective model’ (Jacquemont et al., 2014). However, in a proportion of individuals carrying CNVs only a modest effect on cognitive traits has been observed (Stefansson et al., 2014).

The mechanisms underlying the relationship between neurodevelopment and dopaminergic dysfunction in relation to psychosis have been investigated among individuals presenting with the 22q11.2 deletion syndrome, in which a large deletion on chromosome 22q11.2 has been found to be associated with both neurodevelopmental outcomes and a 25-fold increased risk of developing schizophrenia (Karayiorgou et al., 2010; Rees et al., 2014; Schneider et al., 2014; Vasa et al., 2015). Up to 40% of 22q11.2 deletion subjects have been reported to develop a psychotic disorder by adulthood (Schneider et al., 2014). There is some evidence that duplications in the 22q11.2 region may reduce schizophrenia risk, suggestive of a genetically conferred protective function (Rees et al., 2014). Longitudinal investigation of the premorbid phase among 22q11.2 deletion subjects has found that psychotic symptoms are preceded by cognitive decline (Vorstman et al., 2015). Most of the genes within 22q11.2 are expressed in the brain, of which the catecholamine-O-methyl-transfrease (COMT) gene has been widely investigated in relation to schizophrenia risk (Casp et al., 2005; Karayiorgou et al., 2010). Recently, a deletion in 22q11.22, encompassing the TOP3β gene, was found to be associated with cognitive impairment and schizophrenia in a Finnish study sample (Stoll et al., 2013).
The gene DISC1 was originally identified in a large Scottish pedigree, in which a chromosomal translocation was found to segregate with severe mental illness (St Clair et al., 1990; Blackwood et al., 2001). Identification of a breakpoint on chromosome 1q42 led to the identification of two genes directly disrupted by the translocation, which were named Disrupted in Schizophrenia 1 and 2 (DISC1 and DISC2), respectively (Millar et al., 2000). Evidence of linkage and association at 1q42 and in particular DISC1 in relation to schizophrenia susceptibility has subsequently been replicated in the Finnish schizophrenia family cohort (Ekelund et al., 2001; Hennah et al., 2003; Ekelund et al., 2004; Hennah et al., 2004). Various proteins, including the amyloid precursor protein (APP), DIX domain containing 1 (Dixdc1), lissencephaly-1 (LIS1), NDE1, NDEL1, PDE4B and PDE4D, which have been found to interact with DISC1, have also been associated with neurodevelopmental outcomes and increased schizophrenia risk (Kamiya et al., 2006; Hennah and Porteous, 2009; Bradshaw and Porteous, 2012; Soda et al., 2013). Of these genes, NDE1, NDEL1, PDE4B and PDE4D have been associated with schizophrenia in the Finnish schizophrenia family cohort (Hennah et al., 2007; Tomppo et al., 2009).

Since its discovery by Millar et al. (2000), many researchers have investigated the functions of DISC1, which has since been found to play a role in neuronal proliferation and migration, neurite outgrowth, maintenance of synapse composition and glutamatergic and dopaminergic neurotransmission (Millar et al., 2003; Higginbotham and Gleeson, 2007; Niwa et al., 2010; Brandon and Sawa, 2011; Ishizuka et al., 2011; Lee et al., 2011; Eykelenboom et al., 2012; Lepagnol-Bestel et al., 2013; Steinecke et al., 2014; Devine et al., 2016; Tang et al., 2016).

Emerging evidence suggests that DISC1 is involved in the regulation of neuronal cell division and the maintenance of neuronal and synaptic morphology by mediating the intracellular trafficking of proteins, protein complexes, organelles, vesicles, mitochondria, neurotransmitters and ion channels within neurons (Devine et al., 2016). DISC1 has been found to localize to different intracellular compartments including mitochondria, the nucleus, the centrosome, the Golgi apparatus and the endoplasmic reticulum (Millar et al., 2005a; Higginbotham and Gleeson, 2007; Lepagnol-Bestel et al., 2013; Park et al., 2015), and its intracellular localization has been postulated to be disrupted upon truncation (Devine et al., 2016). DISC1-mediated intracellular transport has been proposed to occur via its interaction with a number of specific binding partners, adaptor and motor proteins such as microtubules, actin and kinesin-associated proteins, fasciculation and
elongation protein zeta 1 (FEZ1), β4-spectrin, dynactin, intersectins, inesins, glycogen synthase kinase 3 beta (GSK3β), NDE1 and NDEL1 (Devine et al., 2016). It has been proposed that DISC1 may be involved in mediating antero- and retrograde axonal transport within the cell (Devine et al., 2016).

To date, over 50 DISC1 mRNA splice variants have been identified, with different variants being differentially expressed during different periods in neurodevelopment, with the expression of some variants peaking during distinct periods of foetal life (Nakata et al., 2009). This is reflective of the complexity of transcriptional and translational regulatory processes involved in development.

**NDE1 and NDEL1**

NDE1 and NDEL1 are coiled-coil proteins that bind to and form complexes with LIS1 (Derewenda et al., 2007). NDEL1 and NDE1 also interact with the cytoskeletal motor protein dynein, which forms a functional link between both proteins and microtubules, in the regulation of mitosis, cell migration, and microtubule-associated intracellular transport processes (McKenney et al., 2010; Bradshaw, 2016). Both NDEL1 and NDE1 have been found to interact directly with DISC1 (Gadelha et al., 2013; Bradshaw, 2016). Both are critical for cortical development (Shu et al., 2004; Pei et al., 2014).

The *NDEL1* gene, comprising 10 exons, is situated in the 17q13.1 region (Hayashi et al., 2005). The interaction of DISC1 and NDEL1 has been suggested to regulate neuronal morphogenesis and neuronal positioning during neuronal integration (Duan et al., 2007). *NDEL1* has been associated with increased schizophrenia risk in the Finnish schizophrenia family study sample (Tomppo et al., 2009). The enzyme activity of NDEL1 has been reported to be reduced in the plasma of schizophrenia patients (Gadelha et al., 2013).

The *NDE1* gene is situated in the 16p13.11 region. Deletions in 16p13.11 have been associated with autism, epilepsy and intellectual disability (Ullmann et al., 2007; Hannes et al., 2009; de Kovel et al., 2010; Heinzen et al., 2010; Mefford et al., 2010; Mullen et al., 2013). Functional mutations and deletions in the *NDE1* gene have been associated with marked neurodevelopmental outcomes, including failure of neurogenesis and deficient cortical lamination (Alkuraya et al., 2011; Bakircioglu et al., 2011; Paciorkowski et al., 2013). DISC1 has been found to moderate the association of NDE1 and GSK3β with the trafficking kinesin protein 1 (TRAK1) in the cyclic adenosine monophosphate (cAMP)-mediated regulation of axonal mitochondrial transport (Ogawa et al., 2016). In a mouse model, NDE1 has been found to be
expressed in the subventricular zone of the forebrain and the subgranular zone of the hippocampus (Pei et al., 2014). Knockdown of NDE1 in zebrafish embryos has been found to result in the suppression of neuronal cell division, leading to left–right patterning defects, implicating NDE1 in the lateralization of the central nervous system (Kim et al., 2011). Variants of NDE1 have been associated with increased schizophrenia risk (Hennah et al., 2007; Moens et al., 2011; Kimura et al., 2015).

**PDE4B and PDE4D**

PDEs constitute a family of 11 different enzymes (PDE1–PDE11) encoded by 21 genes, the majority of which are expressed as multiple variants of up to 100 individual proteins (Ricciarelli and Fedele, 2015). The intracellular inactivation of the second messenger cAMP is controlled by PDE4 by degradation of cAMP to inactive AMP, attenuating cAMP-mediated intracellular signalling. PDE4D has been found to associate with memory functions (Ricciarelli and Fedele, 2015). PDE4B is known to be a target of antidepressant rolipram (Millar et al., 2005b). PDE4B and PDE4D have been associated with increased schizophrenia risk (Tomppo et al. 2009). The interaction of DISC1 and PDE4 has been shown to play an important role in the functional activity of NDE1 (Bradshaw et al., 2011).
2.7.2. ENVIRONMENTAL RISK FACTORS

Environmental risk factors associated with schizophrenia risk can broadly be divided into (1) pre- and perinatal and (2) postnatal (premorbid) risk factors (Brown, 2011a; Rapoport et al., 2012).

2.7.2.1. PRE- AND PERINATAL ENVIRONMENTAL RISK FACTORS

Obstetric complications have collectively been associated with a 2-fold increase in the risk of developing schizophrenia, highlighting the relationship between neurodevelopment and schizophrenia (Murray and Lewis, 1987; Weinberger et al., 1987; Geddes and Lawrie, 1995; Cannon et al., 2002a; Clarke et al., 2006; Brown, 2011a; Rapoport et al., 2012). Obstetric complications have also been associated with a variety of psychiatric disorders, for instance LBW has been associated with an increased risk of depression (Räikkonen et al., 2008), eating disorders (Favaro et al., 2006), attention deficit disorders (Banerjee et al., 2007) and autism (Schendel and Bhasin, 2008). Obstetric complications are thus not risk factors specific to schizophrenia.

In a meta-analysis on the association of obstetric complications and schizophrenia (Cannon et al., 2002a), consisting of eight prospective population-based studies, the following 10 obstetric complications were found to associate significantly with schizophrenia risk: diabetes in pregnancy (OR 7.76, 95%CI 1.37-43.90), birth weight <2000 g (OR 3.89, 95%CI), emergency Caesarean section (OR 3.24, 95%CI 1.40-7.50), congenital malformations (OR 2.35, 95%CI 1.21-4.57), uterine atony (OR 2.29, 95%CI 1.51-3.50), rhesus variable (OR 2, 95%CI 1.01-3.96), asphyxia (OR 1.74, 95%CI 1.15-2.62), bleeding in pregnancy (OR 1.69, 95%CI 1.14-2.52) and birth weight <2500 g (OR 1.67, 95%CI 1.22-2.29).

In a Danish population-based nested case-control study consisting of 1039 individuals with schizophrenia and 24 826 matched controls, Byrne et al. (2007) described an association between schizophrenia and maternal influenza (IRR 8.2, 95%CI 1.4-48.8), preeclampsia (IRR 2.72, 95%CI 1.0-7.3), haemorrhage during delivery (IRR 2.43, 95%CI 1.1-5.6), maternal sepsis of childbirth and the puerperium (IRR 2.91, 95%CI 1.1-7.9), threatened premature delivery (IRR 2.39, 95%CI 1.4-4.1), manual extraction of the baby (IRR 2.15, 95%CI 1.1-4.4), non-attendance of antenatal appointments (IRR 2.08, 95%CI: 1.0-4.4) and gestational age of 37 weeks or below (IRR 1.51, 95%CI 1.0-2.2).
Pre- and perinatal factors associated with schizophrenia can be grouped into the following categories: (1) prenatal immunological factors, (2) prenatal maternal stress, (3) pre- and perinatal hypoxia, (4) maternal prenatal nutrition, and (5) factors related to foetal growth (Clarke et al., 2006). The categorical division of environmental factors is acknowledged to be reductionistic in that different environmental exposures co-precipitate and influence one another. For instance, gestational diabetes has been found to be associated with immune system activation, increased oxidative stress, foetal hypoxia and deviant intrauterine growth, and thus, causal inferences are potentially confounded by complexity (Van Lieshout and Voruganti, 2008). Moreover, the categorical division of environmental factors with respect to the foetomaternal boundary often neglects the central role occupied by the placenta in relation to foetal growth (Patterson, 2007). In addition to regulating the selective permeability of nutrients, oxygen and antibodies to the foetus, it constitutes an immunological interface between the mother and the foetus, promoting maternal tolerance to the developing foetus (Hsiao and Patterson, 2012).

The relationship between prenatal maternal infection and schizophrenia gained momentum when Mednick et al. (1988) described an association between schizophrenia and prenatal exposure to influenza among subjects born in Helsinki during the 1957 epidemic. Other prenatal viral and bacterial microbial infections (Rubella, toxoplasma, herpes simplex virus type 2) have since been found to be associated with increased schizophrenia risk (Brown and Derkits, 2010). Prenatal infection has been found to interact with genetic liability in increasing schizophrenia risk (Clarke et al., 2009; Blomström et al., 2016). Maternal infections during pregnancy requiring hospitalization in combination with both (1) autoimmune disorders and (2) anaemia during pregnancy have been found to exert additive effects on schizophrenia risk among offspring (Benros et al., 2011; Nielsen et al., 2016). Elevated maternal C-reactive protein levels during pregnancy have been associated with an increased risk of both ASDs and schizophrenia among offspring (Canetta et al., 2014; Knuesel et al., 2014). Interestingly, however, individuals who develop non-affective psychoses have also been found to have decreased levels of certain acute-phase proteins at birth (Gardner et al., 2013; Blomström et al., 2015). The potential molecular mechanisms underlying the association between prenatal infection and schizophrenia remain unsubstantiated, but it has been suggested that the activation of the immune system is in itself a central mediator of schizophrenia risk (Muller et al., 2015). A proposed mechanism suggests that an increase in the permeability of the blood-brain barrier may upon infection/inflammation result in an influx of proinflammatory cytokines into the central nervous system of the foetus, with implications on neurodevelopment (Brown, 2011b).
Rhesus (Rh) incompatibility during pregnancy, an example of maternal-foetal genotype incompatibility, has been associated with increased schizophrenia risk, after a previous Rh-incompatible pregnancy (Kraft et al., 2004). Rh incompatibility disease has been associated with foetal hypoxia and increased levels of unconjugated bilirubin, which is known to exert neurotoxic effects (Morioka et al., 2015). Interestingly, evidence also suggests that the Human Leukocyte Antigen-B (HLA-B) locus increases schizophrenia risk among female offspring when mothers and their daughters match and are "too alike" with respect to alleles at the HLA-B locus (Palmer et al., 2006).

In 1978, Huttunen and Niskanen described an association between paternal death, a proxy for maternal stress, during pregnancy and increased schizophrenia risk among offspring. The association between maternal stress during pregnancy and schizophrenia has since been replicated. Pregnancies during the invasion of the Netherlands by Germany in World War II and during the Israeli Six Day War were found to be associated with an increased schizophrenia risk among offspring (van Os and Selten, 1998; Malaspina et al., 2008). In a large population cohort consisting of 1.38 million Danish births from 1973 to 1995, Khashan et al. (2008) described a 1.67-fold increase in schizophrenia risk among offspring when mothers had been exposed to death of a relative during the first trimester. Additionally, increased schizophrenia risk has been described among offspring born to mothers who report their pregnancies as being unwanted (Myhrman et al., 1996; Herman et al., 2006). However, in a recent study by Abel et al. (2014) comprising 1,045,336 Swedish births between the years 1973 and 1985 an association was described between postnatal but not prenatal maternal bereavement stress and an increased risk of psychosis among offspring. The observed association was not mediated by family history of psychotic disorders.

It has been suggested that the biological mechanisms underlying the association between maternal stress during pregnancy and increased schizophrenia risk among offspring may involve foetal exposure to glucocorticoids, inflammation, placental factors or factors related to maternal health (Brown, 2011b). Stressful life events during pregnancy have been associated with LBW (Khashan et al., 2008), which is also a schizophrenia-associated risk factor (Cannon et al., 2002a).

Pre- and perinatal hypoxia has been suggested to interact with genetic liability, increasing schizophrenia risk (McNeil et al., 1999; Stefanis et al., 1999; Rosso et al., 2000; Cannon et al., 2002b; Van Erp et al., 2002; Schulze et al., 2003; Ebner et al., 2008; Haukvik et al., 2010). Pre- and perinatal hypoxia has also been observed to be associated with cognitive and behavioural outcomes during childhood (Goldstein et al., 2000). Chronic prenatal and acute perinatal hypoxia have been suggested to exert a differential effect on
neurodevelopment (Anastario et al., 2012). Maternal smoking during pregnancy, which is associated with chronic foetal hypoxia, has also been found to associate with increased schizophrenia risk (Niemelä et al., 2016) and greater severity of negative symptoms among offspring who develop schizophrenia (Stathopoulou et al., 2013). The frequency of acute perinatal hypoxia, as indicated by lower Apgar 5 min scores, has been suggested to be elevated in at-risk mental state subjects relative to healthy controls (Kotlicka-Antczak et al., 2014). Evidence suggests that disrupted neurotrophic signalling may contribute to hypoxia-related outcome measures, and perinatal hypoxia has been associated with a 20% decrease in brain-derived neurotrophic factor (BDNF) levels in neonatal cord blood samples of subjects who develop schizophrenia (Cannon et al., 2008).

Preliminary evidence suggesting an association between prenatal malnutrition and schizophrenia came from studies describing the effects of in utero exposure to famine in the Dutch Hunger Winter of 1944-1945 (Brown and Susser, 2008). A direct association between nutritional deprivation and schizophrenia in famine was difficult to decipher since famine is also associated with other environmental factors, including infection and stress, both of which have been associated with schizophrenia (Brown, 2011a). In animal models, prenatal protein deprivation has been associated with structural central nervous system (CNS) manifestations and cognitive outcomes among offspring (Meyer and Feldon, 2010).

Both decreased and increased concentrations of neonatal vitamin D have been associated with increased schizophrenia risk (McGrath et al., 2010), and decreased maternal concentrations of 25(OH)D (a metabolite of vitamin D) have been associated with foetal growth restriction, increased risk of preterm birth and small size for gestational age at birth (Miliku et al., 2016).

Maternal iron deficiency during pregnancy has been associated with neurocognitive outcomes and an increased risk of schizophrenia spectrum disorders (SSDs) among offspring (Insel et al., 2008; Radlowski and Johnson, 2013). Iron is essential for brain development and functioning, and iron deficiency has been suggested to be related to nutritional deprivation. In a study comprising 6872 offspring, Insel et al. (2008) described an inverse relationship between decreasing maternal haemoglobin concentration and increasing SSD susceptibility among offspring. In the study, a mean maternal haemoglobin concentration of $< 10.0 \text{ g/dL}$ was found to be associated with a 4-fold increase in SSD risk.

Maternal diabetes during pregnancy has been associated with increased schizophrenia risk in a meta-analysis (Cannon et al., 2002a). This finding was not replicated in a subsequent population-based study in the Danish
population (Byrne et al., 2007). The biological mechanisms underlying the association of maternal diabetes and schizophrenia have been suggested to involve factors related to immune activation, oxidative stress, hypoxia and deviant birth weight (Van Lieshout and Voruganti, 2008). To date, no study has investigated the difference in the relationship between gestational diabetes and maternal diabetes prior to pregnancy in relation to subsequent schizophrenia risk.

**BIRTH WEIGHT AND SCHIZOPHRENIA**

Birth weight is mediated by two major factors: (1) duration of gestation and (2) intrauterine growth rate. LBW can occur as the result of either a shortened period of gestation or retarded intrauterine growth, and sometimes a combination of both (Kramer, 1987). Prematurity is defined as a gestational age of less than 37 weeks. Birth weight is physiologically related to gestational age, and prematurity and postmaturity are commonly associated with decreasing and increasing birth weight, respectively (Stillerman et al., 2008). Deviations of birth weight, both decreasing and increasing, and pre- and postmaturity have been associated with increased perinatal mortality and morbidity among offspring (Battaglia et al., 1966; Chase et al., 1969; Basso et al., 2006; Henriksen, 2008; Stillerman et al., 2008). The World Health Organization has defined LBW as a birth weight of <2500 g, based upon findings demonstrating that the mortality of newborns with a birth weight under this cut-off value is significantly increased (Kramer, 1987). The term intrauterine growth restriction is used to describe neonates unable to reach their genetically determined potential size as a result of reduced foetal growth due to environmental factors (Stillerman et al., 2008). Although a unitary definition of HBW has not been assigned, a birth weight of >4000 g or above the 90th percentile, has frequently been used in the literature to denote HBW, as elevated neonatal morbidity has been documented above this cut-off point (Battaglia et al., 1966; Jolly et al., 2003; Henriksen, 2008).

In a large meta-analysis comprising 895 articles published in 1970-1984, the determinants of LBW were categorized into the following groups: (1) genetic and constitutional factors, (2) demographic and psychosocial factors, (3) obstetric factors, (4) nutritional factors, (5) maternal morbidity during pregnancy, (6) toxic exposures and (7) antenatal care (Table 7) (Kramer, 1987).

1. Genetic and constitutional factors
   - Infant sex
   - Racial/ethnic origin
   - Maternal height, pre-pregnancy weight and maternal haemodynamic factors
   - Paternal height and weight

2. Demographic and psychosocial factors
   - Maternal age
   - Socioeconomic status
   - Marital status
   - Parental psychiatric factors

3. Obstetric factors
   - Parity
   - Pregnancy interval
   - Birth weight in prior pregnancies
   - Prior infertility, spontaneous/induced abortion, or stillbirth/neonatal death

4. Nutritional factors
   - Gestational weight gain
   - Caloric intake and energy expenditure (protein and iron status)
   - Vitamin B6/B12, vitamin D and folic acid
   - Trace elements (zinc, copper, calcium, phosphorus)

5. Maternal morbidity during pregnancy
   - General morbidity and episodic illness
   - Infections, and genital and urinary tract infection

6. Toxic exposures
   - Cigarette smoking
   - Alcohol and caffeine consumption
   - Cannabis use and narcotic addiction

6. Antenatal care
   - Frequency and quality of antenatal care visits
In addition to genetic and constitutional factors, the determinants of HBW include: gestational age > 40 weeks, previous delivery of a macrosomic baby, pregestational diabetes, gestational diabetes, increased maternal age, increased parity, maternal pre-pregnancy weight, maternal weight gain during pregnancy (Jolly et al., 2003; Wallace and McEwan, 2007). Gestational diabetes is one of the strongest triggers of HBW (Wallace and McEwan, 2007). Maternal hyperglycaemia leads to elevated glucose levels in the foetus, resulting in foetal hyperinsulinaemia, which stimulates foetal growth (Wallace and McEwan, 2007). In a large study consisting of 350,311 singleton pregnancies in the United Kingdom born in 1988-1997, HBW was found to be associated with the following obstetric complications, many of which are associated with perinatal hypoxia: prolonged first and second stage of labour, instrumental vaginal delivery, third-degree perineal trauma, emergency Caesarean section, postpartum haemorrhage, Apgar score <4 and admission to the special care baby unit (Jolly et al., 2003).

Several meta-analyses have investigated the association between obstetric complications and schizophrenia risk (Geddes and Lawrie, 1995; Geddes et al., 1999; Cannon et al., 2002a). In the most recent meta-analysis, Cannon et al. (2002a) reported a 1.67-fold and a 3.39-fold increase in schizophrenia risk in relation to birth weights <2500 g and <2000 g, respectively. Several studies have since been published on the association between obstetric complications, including birth weight, and schizophrenia risk (Gunnell et al., 2003; Gunnell et al., 2005; Byrne et al., 2007; Abel et al., 2010; Moilanen et al., 2010; Eide et al., 2013; Keskinen et al., 2013; Byars et al., 2014). Some studies have failed to find an association between birth weight and schizophrenia (Gunnell et al., 2005; Byrne et al., 2007). In addition, two recent large population-based cohort studies have described an association between birth weight and ASDs (Abel et al., 2013; Byars et al., 2014). The central findings in relation to the association of birth weight and schizophrenia are presented in Table 8.

Gunnell et al. (2003) investigated the association between birth weight and schizophrenia in a cohort of 246,655 Swedish male conscripts, 80 of whom had been assigned a diagnosis of schizophrenia over a 3-year follow-up. A reverse J-shaped association was found between birth weight and schizophrenia in men, in which LBW (<2500 g) and HBW (>4000 g) were associated with an increase in schizophrenia risk. A subsequent investigation by Gunnell et al. (2005) of a larger cohort consisting of 719,476 Swedish males and females, 736 of whom had developed schizophrenia over a 9.9-year follow-up, failed to detect an association between birth weight and schizophrenia. However, the authors concluded that they could not rule out a small increase in schizophrenia risk among the heavier babies (>4000 g).
A 34-year follow-up of the Northern Finland 1966 birth cohort (NFBC) comprising 12,058 subjects, 111 subjects with a diagnosis of schizophrenia, found that both LBW (<2500 g) (OR 2.5) and HBW (>4500 g) (OR 2.4) were associated with increased schizophrenia risk (Moilanen et al., 2010). Further investigation of the cohort, with additional 44-year follow-up data, found that HBW, in comparison with LBW, was associated with augmented schizophrenia risk, specifically among offspring with a history of parental psychosis, indicative of increased genetic liability, whereas LBW was associated with schizophrenia among offspring without a history of parental psychosis (Keskinen et al., 2013). This observation was suggested to be reflective of a potential HBW-mediated gene-environment interaction.

In a large population-based cohort study comprising 1.49 million subjects from Sweden and Denmark, 5445 of whom had a SSD, Abel et al. (2010) observed an association between LBW (<2500 g) and an increased risk of psychotic disorders (ICD-9 diagnoses 290-319 and ICD-10 diagnoses F20-F29). In a Norwegian population-based cohort study consisting of 873,612 individuals, 2207 with schizophrenia born in 1967-1982, pre-eclampsia was proposed to mediate the association observed between LBW and schizophrenia, suggesting a role for placental dysfunction in relation to LBW-associated schizophrenia risk (Eide et al., 2013). Recently, in a Danish population-based study consisting of 1.79 million singleton births, Byars et al. (2014) found that the risk of ASDs was increased among subjects with “above average” birth weight (3691-4090 g), whereas schizophrenia risk was increased among subjects presenting with “below average” birth weight (2891-3290 g) (Byars et al., 2014). Abel et al. (2013) also investigated the association of birth weight and ASDs in a Swedish study sample consisting of 4283 subjects with ASDs and 36,588 controls. In the study conducted by Abel et al. (2013), the risk of ASDs was found to increase in relation to both decreasing and increasing birth weight, in which the greatest ASD risk was observed among individuals presenting with a birth weight of 2.0 standard deviations (SDs) below or 2.0 SDs above the mean for gestational age.
Table 8. Main findings of the association between birth weight and schizophrenia in studies not included in earlier meta-analyses. Significant/nonsignificant associations have been illustrated (green/red).

<table>
<thead>
<tr>
<th>Study type</th>
<th>Sample size</th>
<th>N(SZ or ASD)</th>
<th>Year of birth</th>
<th>Result (OR/HR)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gunnell et al. (2003)</td>
<td>Population-based cohort (Sweden)</td>
<td>246 655 male conscripts</td>
<td>80</td>
<td>1973–1980</td>
</tr>
<tr>
<td>Abel et al. (2010)</td>
<td>Population-based cohort (Sweden and Denmark)</td>
<td>1.49 million</td>
<td>5445*</td>
<td>1973–1994 (Sweden) 1979–1996 (Denmark)</td>
</tr>
<tr>
<td>Eide et al. (2013)</td>
<td>Population-based cohort study (Norway)</td>
<td>873 612</td>
<td>2207</td>
<td>1967–1982</td>
</tr>
<tr>
<td>Byars et al. (2014)</td>
<td>Population-based cohort (Denmark)</td>
<td>1.79 million</td>
<td>95345 had a SZ spectrum or an ASD</td>
<td>1978–2009</td>
</tr>
<tr>
<td>Moilanen et al. (2010)</td>
<td>Prospective birth cohort (Finland)</td>
<td>12 058</td>
<td>111</td>
<td>1966</td>
</tr>
<tr>
<td>Keskinen et al. (2013)</td>
<td>Prospective birth cohort (Finland)</td>
<td>10 526</td>
<td>150</td>
<td>1966</td>
</tr>
</tbody>
</table>

* Note that in the study by Abel et al. (2010) the association between birth weight and psychotic disorders in general was investigated (ICD-9 diagnoses 290-319, ICD-10 diagnoses F20-F29). SZ = Schizophrenia. ASD = Autism spectrum disorder.
BIRTH WEIGHT AND SCHIZOPHRENIA; POTENTIAL CAUSAL MECHANISMS

Several theories on the causal mechanisms underlying the association between birth weight and schizophrenia risk have been put forth. The aetiological mechanisms associated with LBW likely differ from those associated with HBW.

LOW BIRTH WEIGHT

LBW has been associated with an increased risk of various medical conditions later in life, including coronary heart disease, stroke, hypertension and non-insulin-dependent diabetes (Barker, 2004). The developmental origins of disease hypothesis, the Barker hypothesis, proposes that birth weight reflects maternal nutritional status during pregnancy. The hypothesis postulates that the foetus responds to prenatal malnutrition with permanent changes in the anatomy and physiology of numerous organ systems, reflected by reductions in cell numbers, alterations in organ structure and the resetting of hormonal axes (Barker et al., 1995). It has been suggested that this response is physiologically adaptive in preparing the organism for an environment in which nutrition is limited. However, when the organism later encounters an environment of nutritional excess the adaptive capacity of the genetically programmed “thrifty genotype” is limited, resulting in pathology. It has been suggested that the underlying processes are epigenetically driven (El Hajj et al., 2014).

LBW has been associated with poorer pre-morbid functioning and an earlier at age onset of schizophrenia (Rifkin et al., 1994). Decreasing birth weight has also been associated with increasing cognitive impairment in the general population and in individuals with a SSD (Tanskanen et al., 2011; Freedman et al., 2013). Very low birth weight (VLBW) (< 1500 g) has been associated with neurocognitive outcomes, including inattention and reduced psychosocial functioning during adolescence (Indredavik et al., 2010). The association between birth weight and brain structure has also been investigated. Increasing birth weight has been found to associate with increasing cortical surface area, whereas decreasing birth weight has been associated with smaller total surface area, within multiple regions (Walhovd et al., 2012; Haukvik et al., 2014).

Insulin-like factors (IGFs), in particular IGF-I and IGF-II, play a role in human growth and neurogenesis, and have been hypothesized to play a role in the
pathogenesis of schizophrenia (Gunnell and Holly, 2004). The epigenetic regulation of IGF-II expression has been proposed to associate with both cognitive functioning and birth weight (Heijmans et al., 2008; Wehkalampi et al., 2013; Iwamoto and Ouchi, 2014). Prenatal undernutrition (Heijmans et al., 2008), VLBW (Wehkalampi et al., 2013), pre-eclampsia (He et al., 2013) and decreased cognitive functioning (working memory) (Cordova-Palomera et al., 2014) have all been associated with decreased levels of IGF-II gene methylation.

Prenatal infection and pre- and perinatal hypoxia have been associated with increased schizophrenia risk, particularly in combination with genetic susceptibility (Nicodemus et al., 2008; Clarke et al., 2009). Foetal growth among individuals with genetic liability for schizophrenia has been suggested to be particularly vulnerable to the effects of prenatal exposure to infection or hypoxia (Fineberg et al., 2013). Pre-eclampsia has also been reported to mediate the association observed between LBW and schizophrenia, suggesting a role for placental dysfunction in relation to LBW-associated schizophrenia risk (Eide et al., 2013).

Exposure to stress during pregnancy has been found to activate the hypothalamic–pituitary–adrenal (HPA) axis, leading to increased maternal glucocorticoid secretion, which has been associated with decreased birth weight (Brown, 2011a).

**HIGH BIRTH WEIGHT**

HBW has been associated with increased schizophrenia risk, particularly among offspring presenting with increased genetic susceptibility for schizophrenia (Keskinen et al., 2013). Other studies have also found an association between HBW and schizophrenia (Hultman et al., 1997; Gunnell et al., 2003; Bersani et al., 2007; Moilanen et al., 2010) and ASDs (Abel et al., 2013; Byars et al., 2014). However, several studies reporting an association between LBW and schizophrenia have not detected an association between HBW and schizophrenia (Dalman et al., 1999; Dalman et al., 2001; Abel et al., 2010).

Genetic susceptibility to schizophrenia has been suggested to increase the vulnerability of the neurodevelopmental process to the effects of obstetric complications, with respect to which the association between perinatal hypoxia and genetic liability represents a particularly well-established finding (Stefanis et al., 1999; Rosso et al., 2000; Cannon et al., 2002b; Van Erp et al., 2002; Schulze et al., 2003; Ebner et al., 2008). Perinatal hypoxia has been
found to interact with variants of serine-threonine protein kinase 1 (AKT1), BDNF, dystrobrevin binding protein 1 (DTNBP1) and GRM3 in association with increased schizophrenia risk (Nicodemus et al., 2008). A variant of GRM3 has been found to interact with perinatal hypoxia in association with a reduction in hippocampal volume among offspring who develop schizophrenia (Haukvik et al., 2010). Genetic predisposition has been suggested to mediate the effect of LBW and pre- and perinatal hypoxia on school performance (Forsyth et al., 2012). Keskinen et al. (2013) proposed that the observed association between HBW and increased schizophrenia risk, in combination with increased genetic liability, could involve epigenetic mechanisms impacting the neurodevelopmental process and conferring elevated sensitivity to subsequent environmental insults.

Gestational diabetes, one of the strongest triggers of HBW, has been suggested to be associated with increased prenatal oxidative stress, immune system disturbances and chronic foetal hypoxia, factors which have been suggested to represent potential schizophrenia-associated risk factors (Wallace and McEwan, 2007; Reece et al., 2009; Benros et al., 2011; Ornoy, 2011; Vambergue and Fajardy, 2011; Muller et al., 2015). Although an association between maternal diabetes and schizophrenia has been documented (Cannon et al., 2002a), this association has not been extensively replicated. The meta-analysis conducted by Cannon et al. (2002a) was based on two studies in which an association between diabetes in pregnancy and schizophrenia risk among offspring was described (Jones et al., 1998; Hultman et al., 1999); neither study specified type of diabetes (type I vs. type II vs. gestational diabetes mellitus). The foundation for this association is based on the observation that three individuals, who were exposed to diabetes during pregnancy, developed schizophrenia (n total=237). Maternal obesity, which is associated with both gestational diabetes and HBW, has been associated with increased schizophrenia risk among offspring (Khandaker et al., 2012). Although the original developmental origins of disease hypothesis described by Barker et al. (1995) focused on the investigation of LBW in relation to future morbidity among offspring, HBW has also been reported to be associated with increased diabetes mellitus type II (T2DM) susceptibility (McCance et al., 1994). Interestingly, T2DM has been found to be more prevalent among parents of individuals with non-affective psychotic disorders, suggestive of shared environmental or genetic risk factors (Miller et al., 2016). Gestational diabetes has also been found to be associated with various HBW independent pre- and perinatal factors, including placental abnormalities, pre-eclampsia, prenatal hypoxia, decreased birth weight, lower Apgar scores, neonatal hypoglycaemia and Caesarean section, many of which have been independently associated with schizophrenia (Cannon et al., 2002a; Wadsack et al., 2012).
The mechanisms underlying the potential association between maternal diabetes during pregnancy and increased schizophrenia risk among offspring remain unsubstantiated (Cannon et al., 2002a). Maternal hyperglycaemia causes foetal hyperglycaemia and foetal hyperinsulinaemia, increasing foetal metabolism and oxygen consumption and resulting in foetal hypoxia (Cvitic et al., 2014). The foetus responds to hypoxia by increasing red blood cell production, increasing iron demand of the foetus (Teramo, 2014). Foetal erythropoiesis is driven by the mobilization of iron reserves from tissues/organs, including the brain (Petry et al., 1992), with potential neurodevelopmental consequences (Radlowski and Johnson, 2013), which from a theoretical standpoint has been postulated to elevate subsequent schizophrenia risk (Van Lieshout and Voruganti, 2008). Diabetes during pregnancy is also associated with an increase in the production of proinflammatory cytokines, e.g. interleukin 6 and tumour necrosis factor alpha (TNF-α) (Cvitic et al., 2014), which have been found to be elevated in psychosis (Khandaker et al., 2015). Elevated levels of proinflammatory cytokines during pregnancy have been associated with increased cognitive impairment and aberrant dopaminergic functioning among offspring (Girgis et al., 2014) and have thus been suggested to exhibit the potential to influence the neurodevelopmental process in relation to increased schizophrenia risk (Van Lieshout and Voruganti, 2008). Maternal diabetes during pregnancy is also associated with increased oxidative stress, resulting from a dysbalance between the levels of reactive oxygen species (ROS) and antioxidant defences (Shang et al., 2015). ROS have the propensity to damage DNA, protein and lipids, thus influencing cellular functioning. It has been proposed that the brain is particularly susceptible to the disruptive effects of oxygen radicals due to its high oxygen consumption (Van Lieshout and Voruganti, 2008).

Interestingly, HBW has also been associated with an increased risk of immunological disorders among offspring, including systemic lupus erythematosus, rheumatoid arthritis and some childhood leukaemias (Simard et al., 2008; Caughey et al., 2009; Mandl et al., 2009).

Maternal schizophrenia has been associated with increased risk of obstetric complications, including LBW among offspring (Jablensky et al., 2005; Schneid-Kofman et al., 2008). General maternal psychiatric morbidity during pregnancy has been associated with increased perinatal morbidity and mortality among offspring, and women with schizophrenia are more likely to receive inadequate prenatal care (Lin et al., 2009). In a study on pregnancy-related mortality, Schneid-Kofman et al. (2008) found psychiatric patients to be older and to have a higher prevalence of diabetes and hypertensive disorders during pregnancy, associated with an elevated perinatal mortality rate, congenital malformations, low Apgar scores and LBW (<2500 g).
2.7.2.2. POSTNATAL RISK FACTORS

An association between urbanicity and increased schizophrenia risk has been described (Vassos et al., 2012). The observed association has been suggested to be "dose-dependent", in that both rising levels of urbanicity and increasing duration of time spent in an urban area have been proposed to be associated with a linear increase schizophrenia risk (Pedersen and Mortensen, 2001). Proposed causal factors include selective migration, social fragmentation and social deprivation, nutritional effects and exposure to various environmental factors (infections, stress, pollutants) (Pedersen and Mortensen, 2006; Zammit et al., 2010; Lederbogen et al., 2011; Vassos et al., 2012).

Infection and inflammation during childhood and adolescence have been associated with an increased risk of psychotic disorders. Childhood CNS viral infections have been found to increase the risk of psychotic disorders (Khandaker et al., 2012). Severe bacterial infections during childhood have also been found to increase the risk for psychotic disorders (Blomström et al., 2014). Possible mechanisms include both the direct effect of the pathogen, and the effect of the inflammatory response on the developing brain (Khandaker et al., 2012). Evidence suggests that autoimmune diseases may increase schizophrenia risk (Benros et al., 2011). An association has also been reported between first hospital contact due to infection and increased schizophrenia risk (Nielsen et al., 2014). The role of the immune system in relation to the pathogenesis of schizophrenia has recently gained further insight upon GWAS findings, implicating an association between the gene coding for complement component 4 and schizophrenia (Schizophrenia Genetics Consortium, 2014; Sekar et al., 2016). The classic complement system has been observed to mediate synapse elimination in a mouse model (Stevens et al., 2007).

Various social and psychological factors during childhood, reflecting childhood adversity, such as loss of a parent, childhood maltreatment and socioeconomic adversity, have been associated with increased risk of schizophrenia and other psychotic disorders (Wicks et al., 2005; Brown, 2011a; Varese et al., 2012).

Migration has been found to be associated with increased schizophrenia risk (McGrath et al., 2004, Saha et al., 2005, Fearon et al., 2006, Bourque et al., 2011). Schizophrenia risk has been found to be elevated among both first-generation and second-generation migrants (Bourque et al., 2011). Moreover, refugee migrants have been found to present with a higher risk of non-affective psychotic disorders than non-refugee migrants (Hollander et al., 2016). Several biological and social factors have been suggested to explain these observations, including viral infections, vitamin D deficiency, social
discrimination and social defeat (Cantor-Graae and Selten, 2005; Bourque et al., 2011).

Cannabis use has been associated with a 2.1-fold increase in schizophrenia risk (Henquet et al., 2008). Cannabis has also been associated with earlier age at onset of psychotic disorders (Decoster et al., 2011). A dose-related effect has been observed, with heavy use being associated with a 3.9-fold increase in schizophrenia risk, in comparison to the general population (Marconi et al., 2016).
2.7.3. GENE-ENVIRONMENT INTERACTIONS

A proportion of the high heritability seen in schizophrenia has been suggested to be accounted for by gene-environment interactions (Howes and Murray, 2014). A gene-environment interaction posits that the effect of a specific environmental exposure on disease risk is different among individuals with different genotypes, or that the effect of a genotype on disease risk is different among individuals with different environmental exposures (Ottman, 1996). Pre- and perinatal factors have been noted to increase schizophrenia risk, particularly among subjects with increased familial liability (Jablensky et al., 2005; Clarke et al., 2009; Keskinen et al., 2013). For instance, one study found that the effect of prenatal exposure to pyelonephritis on psychosis risk was five times greater among individuals with a family history of psychosis than individuals without a family history (Clarke et al., 2009). Recently, the interaction of specific gene variants and defined environmental exposures has been investigated in relation to psychiatric morbidity (Caspi et al., 2002; Caspi et al., 2003; Dick et al., 2011; Uher, 2014), including schizophrenia risk (Caspi et al., 2005). Many gene-environment interactions have been described in relation to psychotic symptoms (Modinos et al., 2013) and can be categorized according to developmental phase: (1) pre- and perinatal and (2) postnatal-premorbid.

2.7.3.1. PRE- AND PERINATAL PERIODS

Investigation of potential gene-environment interactions during the peri- and neonatal periods has focused on environmental exposures related to: (1) perinatal hypoxia (Schmidt-Kastner et al., 2012), (2) maternal infection during pregnancy (Demontis et al., 2011; Borglum et al., 2014) and (3) season of birth (Narita et al., 2000; Chotai et al., 2003) (see Table 9).

Perinatal hypoxia has been found to interact with variants of AKT1, BDNF, DTNBP1 and GRM3 in association with increased schizophrenia risk (Nicodemus et al., 2008). A variant of GRM3 has been found to interact with perinatal hypoxia in association with a reduction in hippocampal volume among offspring who develop schizophrenia (Haukvik et al., 2010).

Evidence suggests an interaction between variants of NMDA-receptor genes (GRIN2A and GRIN2B) and maternal Herpes Simplex Virus – type 2 (HSV-2) infection, denoted by seropositivity during pregnancy, in relation to increased schizophrenia risk among offspring (Demontis et al., 2011). An interaction between the gene zinc finger E-box binding homeobox 1 (ZEB1) and maternal
cytomemalovirus (CMV) infection during pregnancy in relation to schizophrenia risk has also been reported (Borglum et al., 2014).

An interaction between season of birth and genetic variants of the dopamine receptor 4 (DRD4) (Chotai et al., 2003), the human leukocyte antigen -antigen D related receptor 1 (HLA-DR1) (Narita et al., 2000) and the serotonin transporter-linked polymorphic region (5-HTTLPR) (Chotai et al., 2003) have been reported in relation to elevated schizophrenia risk.
Table 9. Pre- and perinatal gene-environment interactions and psychosis. The interactions involving hypoxia are depicted in light blue, infection during pregnancy in yellow, and season of birth in light red. Significant / nonsignificant associations are indicated in green/red. Modification of the table presented by Modinos et al. (2013).

<table>
<thead>
<tr>
<th>GENE</th>
<th>AUTHOR</th>
<th>SNP</th>
<th>ENVIRONMENTAL EXPOSURE</th>
<th>OUTCOME VARIABLE</th>
<th>RESULT</th>
</tr>
</thead>
<tbody>
<tr>
<td>AKT1</td>
<td>Nicodemus et al. (2008)</td>
<td>rs2494735, rs3803300, rs1130233</td>
<td>Perinatal hypoxia</td>
<td>Schizophrenia</td>
<td>+ (+)</td>
</tr>
<tr>
<td>BDNF</td>
<td>Nicodemus et al. (2008)</td>
<td>rs2049046, ss76882600</td>
<td>Perinatal hypoxia</td>
<td>Schizophrenia</td>
<td>+ (+)</td>
</tr>
<tr>
<td></td>
<td>Haukvik et al. (2010)</td>
<td></td>
<td>Perinatal hypoxia</td>
<td>Hippocampal volume</td>
<td>-</td>
</tr>
<tr>
<td>CHRNA7</td>
<td>Nicodemus et al. (2008)</td>
<td></td>
<td>Perinatal hypoxia</td>
<td>Schizophrenia</td>
<td>-</td>
</tr>
<tr>
<td>DRD4</td>
<td>Chotai et al. (2003)</td>
<td>DRD4</td>
<td>Seasonality of birth</td>
<td>Schizophrenia</td>
<td>+ (women)</td>
</tr>
<tr>
<td>DTNBP1</td>
<td>Nicodemus et al. (2008)</td>
<td>rs875462</td>
<td>Perinatal hypoxia</td>
<td>Schizophrenia</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Haukvik et al. (2010)</td>
<td>11 SNPs</td>
<td>Perinatal hypoxia</td>
<td>Hippocampal volume</td>
<td>-</td>
</tr>
<tr>
<td>GRM3</td>
<td>Nicodemus et al. (2008)</td>
<td>rs7808623</td>
<td>Perinatal hypoxia</td>
<td>Schizophrenia</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Haukvik et al. (2010)</td>
<td>rs13242038</td>
<td>Perinatal hypoxia</td>
<td>Hippocampal volume</td>
<td>+</td>
</tr>
<tr>
<td>HLA</td>
<td>Tochigi et al. (2002)</td>
<td>HLA-A24, HLA-A26</td>
<td>Seasonality of birth</td>
<td>Schizophrenia</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Narita et al. (2000)</td>
<td>HLA-DR1</td>
<td>Seasonality of birth</td>
<td>Schizophrenia</td>
<td>+</td>
</tr>
<tr>
<td>5-HTTLPR</td>
<td>Chotai et al. (2003)</td>
<td>5-HTTLPR</td>
<td>Seasonality of birth</td>
<td>Schizophrenia</td>
<td>+ (women)</td>
</tr>
<tr>
<td>MTHFR</td>
<td>Muntjewerff et al. (2011)</td>
<td>rs1801133</td>
<td>Seasonality of birth</td>
<td>Schizophrenia</td>
<td>-</td>
</tr>
<tr>
<td>NMDA</td>
<td>Demontis et al. (2011)</td>
<td>GRIN2A, GRIN2B</td>
<td>Maternal HSV-2 seropositivity during pregnancy</td>
<td>Schizophrenia</td>
<td>+</td>
</tr>
<tr>
<td>NOTCH4</td>
<td>Nicodemus et al. (2008)</td>
<td></td>
<td>Perinatal hypoxia</td>
<td>Schizophrenia</td>
<td>-</td>
</tr>
<tr>
<td>NRG1</td>
<td>Nicodemus et al. (2008)</td>
<td></td>
<td>Perinatal hypoxia</td>
<td>Schizophrenia</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Haukvik et al. (2010)</td>
<td>12 SNPs</td>
<td>Perinatal hypoxia</td>
<td>Hippocampal volume</td>
<td>-</td>
</tr>
<tr>
<td>PRODH</td>
<td>Nicodemus et al. (2008)</td>
<td></td>
<td>Perinatal hypoxia</td>
<td>Schizophrenia</td>
<td>-</td>
</tr>
<tr>
<td>RGS4</td>
<td>Nicodemus et al. (2008)</td>
<td></td>
<td>Perinatal hypoxia</td>
<td>Schizophrenia</td>
<td>-</td>
</tr>
<tr>
<td>TNF-α</td>
<td>Nicodemus et al. (2008)</td>
<td></td>
<td>Perinatal hypoxia</td>
<td>Schizophrenia</td>
<td>-</td>
</tr>
<tr>
<td>TPH</td>
<td>Chotai et al. (2003)</td>
<td>rs1800532</td>
<td>Seasonality of birth</td>
<td>Schizophrenia</td>
<td>-</td>
</tr>
<tr>
<td>ZEB1</td>
<td>Borglum et al. (2014)</td>
<td>rs7902091</td>
<td>Maternal CMV infection during pregnancy</td>
<td>Schizophrenia</td>
<td>+</td>
</tr>
</tbody>
</table>

AKT1 (serine-threonine protein kinase), BDNF (brain-derived neurotrophic factor), CHRNA7 (cholinergic receptor nicotinic alpha 7 subunit), DRD4 (dopamine receptor 4), DTNBP1 (dystrobrevin binding protein 1), GRM3 (glutamate metabotropic receptor 3), HLA (Human Leukocyte Antigen), 5-HTTLPR (Serotonin-transporter-linked polymorphic region), MTHFR (methylene tetrahydrofolate reductase), NMDA (N-methyl-D-aspartate), NOTCH4 (neurogenic...
locus notch homologue 4), NRG1 (neuregulin 1), PRODH (proline dehydrogenase), RGS4 (regulator of G-protein signalling 4), TNF-α (tumour necrosis factor alpha), TPH (tryptophan hydroxylase), ZEB1 (zinc finger E-box binding homeobox 1).

### 2.7.3.2. POSTNATAL-PREMORBID PERIOD

The most widely investigated gene-environment interactions relative to the postnatal-premorbid period has focused on environmental exposures related to: (1) cannabis use, (2) childhood trauma and (3) stressful life events (Table 10).

An interaction has been described between COMT Val158Met and cannabis use in relation to (1) psychotic experiences (Henquet et al., 2006; Henquet et al., 2009), (2) increased susceptibility to schizophreniform disorder (Caspi et al., 2005) and (3) increased symptom severity in schizophrenia (Nieman et al., 2016). Some studies have failed to detect an interaction between COMT and cannabis use in psychosis (Zammit et al., 2007). COMT is involved in the synaptic metabolism of dopamine. The substitution of valine (Val) to methionine (Met) at codon 158, denoted COMT Val158Met, is the result of a missense G to A mutation, resulting in the translation of an enzyme, which is slower at breaking down dopamine (Lachman et al., 1996). Individuals carrying the Met/Met genotype have the lowest and Val/Val the highest enzymatic activity, and heterozygotes present with intermediate enzymatic activity (Mannistö et al., 1999). The Val/Val genotype has been associated with an increased risk of psychosis (Caspi et al., 2005).

An interaction between AKT1 and cannabis use has been found in relation to increased risk of (1) psychotic experiences (Bhattacharyya et al., 2012) or psychosis (Di Forti et al., 2012) and (2) earlier age at onset of schizophrenia (Decoster et al., 2011). Some studies have failed to detect an interaction between AKT1 and cannabis in psychosis (Zammit et al., 2007). AKT1 is a protein serine/threonine kinase involved in various intracellular functions, including the regulation of cell-cycle progression, cell size, cell metabolism and cell survival (apoptosis) (Freyberg et al., 2010). It has also been found to be an important signalling molecule downstream of the dopamine D2 receptor (DRD2) (Arguello and Gogos, 2008).

The interaction of receptors mediating dopamine transmission has also been investigated in relation to cannabis use in psychosis. The dopamine transporter gene (DAT1) has been found to interact with cannabis in association with both psychotic experiences and regional brain activation (Bhattacharyya et al., 2012). An interaction between DRD2 and cannabis use
has also been documented in relation to psychotic symptoms (Colizzi et al., 2015).

Evidence suggests an interaction between the cannabinoid receptor (CNR1) and cannabis use in relation to both reduced white matter volume and increased neurocognitive impairment (Ho et al., 2011). The simultaneous analysis of the interaction of multiple schizophrenia-associated genes, 108 loci identified by the Schizophrenia Genetics Consortium (2014) and cannabis use has revealed an interaction between polygenic risk and cannabis use in relation to cortical thickness, particularly among males (French et al., 2015).

Variants of BDNF and childhood trauma have been found to interact, in association with an increase in psychotic experiences (Alemany et al., 2011). Aas et al. (2014) recently described an interaction between a Val66Met variant of BDNF and childhood trauma in relation to reduced BDNF mRNA concentration, reduced hippocampal volume and increased schizophrenia risk.

Other genes, including the FK506-binding protein gene (Fk506), the forkhead box P2 gene (FOXP2) and the nitric oxide synthase 1 [neuronal] adaptor protein gene (NOSA1P), have been proposed to interact with childhood trauma, in relation to increased cognitive impairment in schizophrenia (Fk506), auditory verbal hallucinations (FOXP2) and increased schizophrenia risk (NOSA1P), respectively (Husted et al., 2012; McCarthy-Jones et al., 2014; Green et al., 2015).

Several studies have reported an interaction between the COMT Val158Met variant and stress in relation to both increased psychotic experiences and symptoms (Stefanis et al., 2007; van Winkel et al., 2008; Simons et al., 2009; Peerbooms et al., 2012). Psychotic experiences in relation to the interaction of stress and COMT Val158Met have been suggested to be modulated by a variant of the methylenetetrahydrofolate reductase (MTHFR) gene (Peerbooms et al., 2012). A variant of the neuregulin 1 (NRG1) gene has also been implicated to interact with psychosocial stress in relation to unusual thought content (Keri et al., 2009).
Table 10. Postnatal-premorbid gene-environment interactions and psychosis. The interactions involving cannabis are depicted in light green and childhood trauma / stress in grey. Significant / nonsignificant associations are indicated in green/red. Modification of the table presented by Modinos et al. (2013).

<table>
<thead>
<tr>
<th>GENE</th>
<th>AUTHOR</th>
<th>SNP</th>
<th>ENVIRONMENTAL EXPOSURE</th>
<th>OUTCOME VARIABLE</th>
<th>RESULT</th>
</tr>
</thead>
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<tr>
<td>AKT1</td>
<td>Decoster et al. (2011)</td>
<td>rs6265</td>
<td>Cannabis</td>
<td>Age at onset of schizophrenia</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Di Forti et al. (2012)</td>
<td>rs2494732</td>
<td>Cannabis (1) lifetime history and (2) frequency of use</td>
<td>Psychotic disorder</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Bhattacharya et al. (2012)</td>
<td>rs1130233</td>
<td>Cannabis (delta-9-THC)</td>
<td>Psychotic experiences, regional brain activation</td>
<td>+</td>
</tr>
<tr>
<td>BDNF</td>
<td>Alemany et al. (2011)</td>
<td>rs6265</td>
<td>Childhood abuse</td>
<td>Psychotic experiences</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Aas et al. (2014)</td>
<td>Val66met</td>
<td>Childhood trauma</td>
<td>BDNF mRNA, Hippocampal volume, Schizophrenia risk</td>
<td>+</td>
</tr>
<tr>
<td>CNR1</td>
<td>Zammit et al. (2007)</td>
<td>rs1049353</td>
<td>Cannabis</td>
<td>Psychotic disorder</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Ho et al. (2011)</td>
<td>rs12720071</td>
<td>Cannabis</td>
<td>WM volumes and neurocognitive impairment</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Caspi et al. (2005)</td>
<td>rs4680</td>
<td>Cannabis</td>
<td>Schizophrenia disorder</td>
<td>+</td>
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<tr>
<td></td>
<td>Henquet et al. (2006)</td>
<td>rs4680</td>
<td>Cannabis (delta-9-THC)</td>
<td>Psychotic experiences</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Zammit et al. (2007)</td>
<td>rs4680 rs737865 rs165599</td>
<td>Cannabis</td>
<td>Psychotic disorder</td>
<td>-</td>
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<td>Henquet et al. (2009)</td>
<td>rs4680</td>
<td>Cannabis</td>
<td>Psychotic experiences</td>
<td>+</td>
</tr>
<tr>
<td>COMT</td>
<td>Nieman et al. (2016)</td>
<td>rs4680</td>
<td>Cannabis</td>
<td>Symptom severity, schizophrenia</td>
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<td></td>
<td>Stefanis et al. (2007)</td>
<td>rs4680</td>
<td>Stress</td>
<td>Psychotic symptoms</td>
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<td>Stress</td>
<td>Psychotic experiences</td>
<td>+</td>
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<td>Simons et al. (2009)</td>
<td>rs4680</td>
<td>Stress</td>
<td>Feelings of paranoia</td>
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</tr>
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<td>Collip et al. (2011)</td>
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<td>Stress</td>
<td>Psychotic experiences</td>
<td>+</td>
</tr>
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<td>Peerbooms et al. (2012)</td>
<td>rs4680</td>
<td>Stress</td>
<td>Psychotic experiences</td>
<td>+</td>
</tr>
<tr>
<td>COMT - MTHFR</td>
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<td>rs1801133</td>
<td>Stress</td>
<td>Psychotic experiences</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Peerbooms et al. (2012)</td>
<td>rs1801131</td>
<td>Stress</td>
<td>Psychotic experiences</td>
<td>-</td>
</tr>
<tr>
<td>Gene</td>
<td>Reference</td>
<td>SNP(s)</td>
<td>Condition</td>
<td>Phenotype</td>
<td>Significance</td>
</tr>
<tr>
<td>-------</td>
<td>-----------</td>
<td>--------</td>
<td>------------------------------------</td>
<td>------------------------------------</td>
<td>--------------</td>
</tr>
<tr>
<td>DAT1</td>
<td>Bhattacharyya et al. (2012)</td>
<td>3’UTR VNTR</td>
<td>Cannabis (delta-9-THC)</td>
<td>Psychotic experiences and regional brain activation</td>
<td>+</td>
</tr>
<tr>
<td>DRD2</td>
<td>Colizzi et al. (2015)</td>
<td>rs1076560</td>
<td>Cannabis</td>
<td>Psychotic symptoms</td>
<td>+</td>
</tr>
<tr>
<td>FK506</td>
<td>Green et al. (2015)</td>
<td>rs1360780, rs9470080, rs4713902, rs9394309</td>
<td>Childhood maltreatment</td>
<td>Cognition in schizophrenia</td>
<td>+</td>
</tr>
<tr>
<td>FOXP2</td>
<td>McCarthy-Jones et al. (2014)</td>
<td>rs1456031</td>
<td>Childhood parental emotional abuse</td>
<td>Auditory verbal hallucinations</td>
<td>+</td>
</tr>
<tr>
<td>NOSA1P</td>
<td>Husted et al. (2012)</td>
<td>rs6994992, rs10954867, rs7005288</td>
<td>Cumulative adversity index: childhood illness, family instability</td>
<td>Schizophrenia</td>
<td>+</td>
</tr>
<tr>
<td>NRG1</td>
<td>Keri et al. (2009)</td>
<td>rs6994992, rs10954867, rs7005288</td>
<td>Psychosocial stress</td>
<td>Unusual thoughts</td>
<td>+</td>
</tr>
<tr>
<td>ST8SIA2</td>
<td>Mandelli et al. (2016)</td>
<td>rs3759917, rs11632521, rs3784722, rs4777989, rs2290492, rs8035760, rs11853992, rs17522085</td>
<td>Stressful life events</td>
<td>Schizophrenia spectrum disorder risk</td>
<td>-</td>
</tr>
<tr>
<td>POLYGENIC</td>
<td>Bani-Fatemi et al. (2016)</td>
<td>rs7897059</td>
<td>Childhood trauma</td>
<td>Schizophrenia - suicide attempt</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>van Winkel and the Genetic Risk Outcome Psychosis Group (2011)</td>
<td>152 SNPs in 42 genes</td>
<td>Cannabis</td>
<td>Psychotic disorder</td>
<td>+</td>
</tr>
</tbody>
</table>

AKT1 (serine-threonine protein kinase), BDNF (brain-derived neurotrophic factor), CNR1 (cannabinoid receptor 1), COMT (catecholamine-O-methyl-transferase), MTHFR (methylene-tetrahydrofolate reductase), DAT1 (dopamine transporter 1), DRD2 (dopamine receptor 2), FK506 (FK506-binding protein), FOXP2 (forkhead box P2), NOSA1P (nitric oxide synthase 1 [neuronal] adaptor protein), NRG1 (neuregulin 1), ST8SIA2 (ST8 alpha-N-acetyl-neuraminide alpha-2,8-sialyltransferase 2).
The treatment of schizophrenia is based on the combination of antipsychotic medication and psychosocial rehabilitation (McGurk et al., 2007; Leucht et al., 2009; Dixon et al., 2010; Grant et al., 2012; Roberts et al., 2014). Up to 60% of subjects with schizophrenia have been found to respond to antipsychotic medication (Sinclair and Adams, 2014), which is relatively effective for the treatment of positive symptoms (Leucht et al., 2009). However, antipsychotic medications have proven less effective for the treatment of negative and cognitive symptoms, the two symptom dimensions demonstrating the most robust associations with functional outcome (Karow et al., 2014; Kirkpatrick, 2014). There is some evidence to suggest that some specific psychosocial interventions, in combination with antipsychotic medication, may alleviate negative and cognitive symptoms in schizophrenia (Wykes et al., 2011; Jauhar et al., 2014; Roberts et al., 2014; Aleman et al., 2016).

In 2005, the Remission in Schizophrenia Working Group established a definition for symptomatic remission to be used as a clinically relevant treatment goal (Andreasen et al., 2005; AlAqeel and Margolese, 2012). Symptomatic remission was defined as follows: “A state in which patients have experienced an improvement in symptoms to the extent that any of the remaining symptoms are of such low intensity that they no longer significantly interfere with behaviour and are below the threshold typically utilized in justifying a diagnosis of schizophrenia” (Andreasen et al., 2005). The term functional recovery has been used to broaden treatment goals beyond symptomatic remission to include the attainment of meaningful roles in the community (Lauronen et al., 2005; Liberman and Kopelowicz, 2006; Jääskeläinen et al., 2013; Valencia et al., 2015). Quality of life has also gained increasing attention as an important outcome measure (Karow et al., 2014).
3. AIMS OF THE STUDY

The thesis investigates the relationship between birth weight and schizophrenia risk in a study sample with elevated genetic susceptibility for schizophrenia. The influence of birth weight on symptom severity and cognitive functioning was also assessed. Moreover, the interaction between birth weight and specific schizophrenia susceptibility genes involved in neurodevelopment was investigated.

1. In the first study, the association between birth weight and schizophrenia risk was investigated. The association between birth weight and schizophrenia risk was hypothesized to be augmented and thus highlighted in a cohort presenting with increased genetic loading for schizophrenia. Based on preliminary findings, the association between maternal diabetes, as a proxy for potential gestational diabetes, and schizophrenia risk was also investigated.

2. In the second study, the association between birth weight and symptom severity, separately for positive, disorganized and negative symptoms, in schizophrenia and other non-affective psychotic disorders was characterized. It was hypothesized that deviations of birth weight, both increasing and decreasing birth weight, would be associated with increasing symptom severity.

3. In the third study, an association between birth weight and cognitive performance was investigated among subjects with schizophrenia and their unaffected first-degree relatives. It was hypothesized that increased and decreased birth weight, relative to intermediate birth weight, would be associated with cognitive impairment among both subjects with schizophrenia and their first-degree relatives.

4. In the fourth study, an interaction between birth weight and specific genes of the DISC1 network, which have previously been associated with increased schizophrenia risk in the same cohort, was examined in relation to schizophrenia risk. It was hypothesized that genes of the DISC1 network could interact with birth weight in augmenting schizophrenia susceptibility.
4. METHODS

4.1. THE FINNISH SCHIZOPHRENIA FAMILY STUDY SAMPLE

The Genetic Epidemiology and Molecular Genetics of Schizophrenia in Finland study, hereafter called the Finnish schizophrenia family study, has been used for the characterization of genes associated with both schizophrenia risk and its clinical manifestations, including cognitive impairment and symptom severity (Hovatta et al., 1994; Hovatta et al., 1997; Hovatta et al., 1999; Ekelund et al., 2000; Ekelund et al., 2001; Paunio et al., 2001; Tuulio-Hendriksson et al., 2002; Hennah et al., 2003; Tuulio-Hendriksson et al., 2003; Arajärvi et al., 2004; Ekelund et al., 2004; Paunio et al., 2004; Tuulio-Hendriksson et al., 2004; Arajärvi et al., 2006; Hennah et al., 2007; Turunen et al., 2007; Wedenoja et al., 2008; Pietiläinen et al., 2009; Tomppo et al., 2009).

The collection of the study sample was initiated in 1988 by the Finnish National Institute of Public Health (since 2009, the National Institute for Health and Welfare). Affected subjects were originally ascertained using three nationwide registers: (1) the Finnish Hospital Discharge Register (hospital admissions) (maintained by the National Institute of Health and Welfare), (2) the Finnish Pension Register (disability pensions) (maintained by the Finnish Social Insurance Institution) and (3) the Finnish Medication Reimbursement Register (entitlement to subsidized outpatient medication) (maintained by the National Social Insurance Institution). The family members of affected individuals were subsequently identified from the Finnish Population Information System (PIS) (maintained by the Population Register Centre) (Hovatta et al., 1997; Arajärvi et al., 2004). All four registers have been computerized from the year 1968 onwards. Information concerning hospitalization was collected between 1969 and 1998, and information on both pensions and entitlement to free medications was gathered between 1969 and 1991.

Individuals born in 1940-1976, with a diagnosis of schizophrenia, schizoaffective disorder or schizophreniform disorder, who (1) had been hospitalized, (2) had received disability pension or (3) had been granted entitlement to subsidized antipsychotic medication were identified (Figure 2). Of the 33 731 subjects identified, 6079 were excluded due to lack of information concerning family members, leaving 27 652 probands potentially eligible for contact. Register diagnoses were based on different diagnostic
classification systems over time. Before 1987, diagnoses were based on the International Classification of Diseases, Eighth revision (ICD-8) (World Health Organization, 1967). In 1987-1995 psychiatric diagnoses in Finland were assigned based on the diagnostic criteria of the Diagnostic and Statistical Manual of Mental Disorders, third edition, text revision (DSM-III-R) (American Psychiatric Association, 1987). However, during this period psychiatric diagnoses in Finland were documented using codes based on the International Classification of Diseases, Ninth revision (ICD-9) (World Health Organization, 1977). Since 1996, diagnoses have been based on ICD-10 (World Health Organization, 1993). Finnish register diagnoses regarding schizophrenia have generally been found to be accurate (Pakaslahti, 1987; Isohanni et al., 1997; Cannon et al., 1998; Mäkikyrö et al., 1998).

First-degree family members of probands (parents and siblings) were identified from the PIS. The acquisition of information concerning first-degree relatives enabled the construction of pedigrees. The psychiatric diagnoses of parents and siblings were obtained from the same three health care registers described above with respect to probands: (1) the Hospital Discharge Register, (2) the Pension Register and (3) the Medication Reimbursement Register.

Two subsamples of families, both with elevated genetic liability for schizophrenia, were drawn with respect to origin of birth: (1) nuclear families originating from an internal isolate (IS) region in northeastern Finland, having at least one sibling with schizophrenia and at least two grandparents born in the IS region (Arajärvi et al., 2004). The lifetime risk of developing schizophrenia in the IS region has been established to be 3.2% (Hovatta et al., 1997). The IS region was originally founded by 36 settlers at the end of the 17th century, and precise details of the subsequent births, deaths, marriages and movements of individuals in the IS have been well documented and archived (Hovatta et al., 1999). Genealogical links between most individuals with schizophrenia and corresponding family structures from the IS region have been traced and documented (Hovatta et al. 1999). From the rest of Finland, the “all Finland (AF) sample”, families with at least two siblings with schizophrenia were drawn (Ekelund et al., 2001; Paunio et al., 2001).

Probands with a register-based diagnosis of schizophrenia, schizoaffective disorder or schizophreniform disorder were contacted through their treating physician. Permission to contact family members was requested from the proband. A research nurse met the probands and the family members and drew blood samples after the family had consented to participate in the study. For diagnostic assessment, medical records were collected from all mental health treatment contacts. The lifetime diagnoses according to DSM-IV (American Psychiatric Association, 1994) criteria were evaluated independently by two psychiatrists, who were blind to register diagnoses and
family structure. In case of disagreement, a third psychiatrist was assigned to assess the lifetime diagnosis. One of the psychiatrists completed the Operational Criteria Checklist (OPCRIT) based on lifetime review of symptoms (McGuffin et al., 1991).

In the second phase of the study, all subjects from families from the IS region and a random sample of families from the AF sample were asked to participate in a clinical assessment protocol (Figure 2). All interviewers, who were psychiatric nurses, psychologists or residents in psychiatry, received extensive training in the use of the instruments. All participants gave blood samples, and medical records from all treatment contacts were collected from subjects with any known mental health care contacts. All patients provided written informed consent after the procedure had been explained to them. The ethics review board of the National Public Health Institute and the Hospital District of Helsinki and Uusimaa approved the study. The assessment protocol consisted of the following parts:

- The psychiatric diagnoses of patients and family members were re-evaluated according to DSM-IV (American Psychiatric Association, 1994) criteria. Both patients and their family members were interviewed using the Structured Clinical Interview for DSM-IV (SCID-I and SCID-II) (First et al., 1997). The final DSM-IV consensus diagnoses were based on information gathered from the case records, the OPCRIT ratings and the SCID interviews.

- Assessment of symptom severity was performed using SAPS (Andreasen, 1984) and SANS (Andreasen, 1983). The interview included SAPS for affected individuals and SANS for both affected individuals and their non-affected family members. SAPS and SANS scores were based on the greatest lifetime symptom severity, using all information available, including discussions with health care personnel and family members. SANS and SAPS ratings were reviewed by a senior psychiatrist together with the interviewer.

- The level of psychological, social and occupational functioning was assessed using the Global Assessment of Functioning (GAF) scale (American Psychiatric Association, 2000). Age at illness onset was defined as the age at which the first contact with a health care provider was sought for psychiatric reasons or the age at which psychiatric symptoms began to cause subjective distress or impaired functioning.

- Both patients and their family members were asked to participate in a neuropsychological evaluation covering central cognitive functions
associated with schizophrenia (Heinrichs et al., 2008). The tests were administered in a fixed order by an experienced psychologist or a trained psychiatric nurse. Test scoring was performed by an experienced psychologist.

**Figure 2.** The Finnish schizophrenia family study sample. PPD = Primary psychotic disorder.
4.1.2 PARTICIPANTS

SUBJECTS IN STUDY I

The total study sample consisted of 1051 subjects, 484 males (46.1%) and 567 females (53.9%), with information concerning birth weight. The mean birth weight of the total sample was 3430 g, with birth weight ranging from 1100 g to 5500 g. The average birth weight for females was 3360 g and for males 3490 g.

The sample consisted of 318 subjects with a primary psychotic disorder (PPD) diagnosis, which corresponded to a lifetime history of any psychotic disorder, excluding psychotic disorders caused by substance use or a general medical condition according to DSM-IV (American Psychiatric Association, 1994). The schizophrenia subgroup (n=197) resided within the PPD group. Altogether 860 subjects originated from the IS region and 191 subjects from the AF region. Of these subjects, 589 (56%) were interviewed using the Structured Clinical Interview for DSM-IV Axis I Diagnosis (SCID-CV) (First et al., 1996).


A diagnosis of gestational diabetes was not routinely in use prior to the 1970s (Golden et al., 2009), and thus, a diagnosis of maternal diabetes at the time of data acquisition was used as a proxy for gestational diabetes. Information concerning maternal diabetes was obtained from both the patient interviews and information acquired from the Medication Reimbursement Register. Altogether 22 mothers (7%) of 69 offspring in the study group were assigned a diagnosis of maternal diabetes at the time of the interview.
A total of 282 PPD subjects, from 204 families, consisting of 103 females (37%) and 179 males (63%), with both birth weight data and information on symptom severity, as measured by SAPS (Andreasen, 1984) and SANS (Andreasen, 1983) were identified. The schizophrenia subgroup (n=178) resided within the PPD group. The median age of subjects at the time of interview was 44 years, ranging from 25 to 59 years. Of subjects included in the analysis, 112 (40%) originated from the IS region and 170 (60%) from the AF region. The median year of birth was 1956. Forty-one subjects (15%) were born before 1950, 157 (56%) were born between 1950 and 1960 and 84 (29%) were born after 1960. The mean birth weight of the sample was 3450 g (SD 600 g), 3430 g for females and 3470 g for males. Symptom severity was greater among subjects with schizophrenia than among subjects presenting with other PPDs (Figure 3).

**SUBJECTS IN STUDY II**

![Bar chart showing the distribution of mean global SAPS and SANS scores among subjects with a diagnosis of schizophrenia and a diagnosis of any primary psychotic disorder (PPD). The schizophrenia subgroup resided within the PPD group.](image)

**Figure 3.** Distribution of mean global SAPS and SANS scores among subjects with a diagnosis of schizophrenia and a diagnosis of any primary psychotic disorder (PPD). The schizophrenia subgroup resided within the PPD group.
SUBJECTS IN STUDY III

The following exclusion criteria was applied in the assessment of cognitive functioning: age > 70 years, severe neurological disorder, mental retardation, severe somatic illness, current alcohol or substance use disorder and untestability due to severe symptoms. Relatives were excluded if they presented with a current psychiatric disorder.

A total of 419 subjects, corresponding to 142 subjects with schizophrenia and 277 non-affected first-degree relatives, with both data on neurocognitive performance and birth weight were identified. Out of all of the subjects in the analysis, 69% were born in a hospital, 16% were born at home with professional assistance and 15% were born at home without professional assistance. Males accounted for 97 subjects (68%) in the schizophrenia group and 133 subjects (48%) in the non-affected relatives group. Males were thus overrepresented in the schizophrenia group. The mean age at onset was 23 years (SD 5 years). In the schizophrenia group, the mean age of subjects was 44 years (SD 7 years), and the mean GAF score was 37 (SD 10) at the time of interview. In the non-affected relatives’ group, the mean age was 46 years (SD 8 years) and the mean GAF score was 78 (SD 12) at the time of interview respectively. The mean birth weight of subjects in the schizophrenia group was 3519 g (SD 609 g) and 3400 g (SD 562 g) in the non-affected relatives group.

SUBJECTS IN STUDY IV

A total of 457 subjects, 204 females (44.6%) and 253 males (55.4%), with both genetic and birth weight data were identified. The sample consisted of 142 subjects (31.1%) with schizophrenia and 315 non-affected first-degree relatives (68.9%). Males accounted for 97 subjects (68.3%) in the schizophrenia group and 156 subjects (49.5%) in the non-affected relatives group. Males were thus again overrepresented in the schizophrenia group. In total, two hundred and thirty-eight subjects (52.1%) originated from the IS region and 219 (47.9%) from the AF region. Of subjects with schizophrenia, 52 (36.6%) originated from the IS region and 90 (63.4%) from the AF region. The mean birth weight in the schizophrenia and non-affected relatives groups was 3460 g (SD 620 g) and 3440 g (SD 570 g), respectively.
4.2. **BIRTH WEIGHT DATA**

The birth weight data of 1051 subjects from 315 families were obtained from birth records. The median year of birth was 1957. Subjects had been asked for permission for retrieval of birth records by letter from the year 1999 onwards. Information concerning place of birth was available for 591 individuals with birth weight data, of which 70.5% were born in hospital, 15.5% were born at home with professional (midwife) assistance and 14% were born at home without professional assistance. Those born at home without professional assistance but with birth weight data represented cases in which the midwife was alerted, but had arrived after delivery. While birth weight was documented in almost all cases, other information in the birth records was variable, e.g. birth length was not available for those born outside hospitals. Gestational age was unavailable for the majority of cases. Out of all the subjects who had given permission to collect birth records, 298 were omitted from the study for the following reasons: (1) 26 subjects declined to participate, (2) the birth records of 154 subjects were not acquired, (3) the birth records of 53 subjects did not contain data concerning birth weight and (4) for 2 subjects, the age at onset had not been recorded, a variable required for survival analysis (Study I). For the distribution of birth weight data in Studies I-IV, see Table 11.

<table>
<thead>
<tr>
<th></th>
<th>STUDY I N(%)</th>
<th>STUDY II N(%)</th>
<th>STUDY III N(%)</th>
<th>STUDY IV N(%)</th>
</tr>
</thead>
<tbody>
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<td>52 (5)</td>
<td>15 (5)</td>
<td>16 (4)</td>
<td>22 (5)</td>
</tr>
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<td>2500–2999 g</td>
<td>167 (16)</td>
<td>44 (16)</td>
<td>49 (12)</td>
<td>71 (16)</td>
</tr>
<tr>
<td>3000–4000 g</td>
<td>703 (67)</td>
<td>183 (65)</td>
<td>294 (70)</td>
<td>307 (67)</td>
</tr>
<tr>
<td>&gt;4000 g</td>
<td>129 (12)</td>
<td>40 (14)</td>
<td>60 (14)</td>
<td>57 (12)</td>
</tr>
<tr>
<td>N(TOTAL)</td>
<td>1051</td>
<td>282</td>
<td>419</td>
<td>457</td>
</tr>
</tbody>
</table>

4.3. **NEUROPSYCHOLOGICAL MEASURES**

For the assessment of cognitive functioning, parts of the Wechsler Adult Intelligence Scale – Revised (WAIS-R; Wechsler., 1981) and the Wechsler Memory Scale – Revised (WMS-R; Wechsler, 1987) were used, as well as the Trail Making Test (Reitan and Wolfson, 1985), the Stroop test (Golden., 1978; MacLeod., 1991) and the California Verbal Learning Test (CVLT; Delis et al., 1987) (Table 12).
### Table 12. Neuropsychological tests used in the study.

| Verbal ability and visuospatial reasoning | WAIS-R Vocabulary  
|                                         | WAIS-R Similarities  
|                                         | WAIS-R Block Design  |
| Processing speed and executive functions | WAIS-R Digit Symbol  
|                                         | Trail Making part A  
|                                         | Trail Making part B  
|                                         | Stroop Interference  |
| Attention and working memory            | WMS-R Digit Span Forward  
|                                         | WMS-R Digit Span Backward  
|                                         | WMS-R Visual Span Forward  
|                                         | WMS-R Visual Span Backward  |
| Verbal learning                         | CVLT Immediate recall  
|                                         | CVLT Short delay recall  
|                                         | CVLT Long delay recall  |

### 4.4. GENOTYPES

Twenty genetic markers, representing SNPs or haplotypes, covering five genes (*DISC1, NDE1, NDEL1, PDE4B, PDE4D*) of the DISC1 network, which had previously demonstrated significant association with schizophrenia risk in the cohort, were evaluated for analysis (Hennah *et al.*, 2003; Ekelund *et al.*, 2004; Hennah *et al.*, 2007; Tomppo *et al.*, 2009). Due to the limited size of the study sample in Study IV, comprising 457 subjects with both genetic and birth weight data, an allele frequency threshold was used to ensure the statistical feasibility of the interaction model. The threshold frequency for minor allele homozygotes was set at >10%. Of the 20 markers, seven met this criterion: five variants of the *NDE1* gene (*NDE1* haplotype, rs4781678, rs2242549, rs881803, rs2075512) and one variant of both *PDE4B* (rs7412571) and *PDE4D* (PDE4D haplotype) genes. The *NDE1* haplotype, corresponding to a specific CGCC allele, comprised of the combination of the following individual *NDE1* SNPs: rs4781678, rs2242549, rs881803 and rs2075512.
4.5. **STATISTICAL ANALYSIS**

**STUDY I**

A multivariate COX frailty model was used to investigate the association between both (1) birth weight and (2) maternal diabetes in relation to subsequent schizophrenia/PPD risk among offspring (Therneau and Lumley, 2008). Birth weight (<2500 g, 2500–2999 g, 3000–4000 g, >4000 g), maternal/paternal history of any psychotic illness and sex were used as explanatory variables in the evaluation of the association between birth weight and schizophrenia risk. Birth weight (<2500 g, 2500–2999 g, 3000–4000 g), sex and maternal diabetes were used as explanatory variables in the investigation of the association between maternal diabetes and schizophrenia risk. Family unit defined frailty, and stratification of subjects with respect to both temporal (decade of birth) and regional (IS region vs. AF sample) considerations was applied in both analyses.

**STUDY II**

A General Estimating Equation (GEE) model (Liang and Zeger, 1986) was used to characterize the relationship between birth weight and symptom severity among subjects who later developed schizophrenia and a PPD spectrum diagnosis. Birth weight and symptom severity were investigated as continuous variables. The data were adjusted for sex, region of birth (IS vs. AF) and decade of birth (subjects born in or before the year 1960 vs. subjects born later than 1960). A quadratic polynomial regression model was applied to detect a possible non-linear relationship between birth weight and symptom severity. Nested models with and without a quadratic effect were evaluated using the Wald test to ascertain the best-fitting model. Statistical analyses were conducted using the R-program version 2.12.1 (R Development Core Team, 2010). A probability level of p<0.05 was considered significant.

**STUDY III**

A GEE model (Liang and Zeger, 1986), using birth weight as a continuous variable, was used to investigate the association between birth weight and cognitive functioning among both subjects with schizophrenia and their non-affected relatives. Place/type of delivery (i.e. hospital delivery, home delivery with professional assistance, home delivery without professional assistance), GAF (American Psychiatric Association, 2000), number of affected siblings
per family, place of birth (IS vs. AF), age and sex were controlled for in the model. A statistically significant quadratic term, indicative of a quadratic effect, was postulated to be suggestive of a curvilinear association between birth weight and cognitive functioning. The interaction between the observed group-specific associations was investigated to determine whether the observed influence of birth weight on cognitive functioning was different with regard to the schizophrenia and the non-affected relative groups. The effect sizes of the mean differences between the schizophrenia and the relatives group were measured using Cohen's d (Cohen, 1988), in which effect sizes were denoted as small (>0.20), medium (>0.50) and large (>0.80). A probability level of p<0.05 was considered significant. Analyses were conducted using the statistical program Stata (StataCorp, Version 9.2, College Station, Texas).

**STUDY IV**

A GEE model was used to test for gene environment interactions between birth weight and specific genes variants in relation to schizophrenia risk (Liang and Zeger, 1986). Explanatory variables included (1) birth weight (categorically divided into ≤4000 g vs. >4000 g), (2) genotype and (3) the interaction of birth weight and genotype. An additive genetic effect was presumed in the model. Place of birth (IS vs. AF) and sex were adjusted for in the model. Correction for multiple testing was performed and the level of statistical significance was set at p=0.017 (0.05/3) in view of the fact that three genes, which met the imposed allele frequency criteria were taken forward for analysis. Fisher's exact test was used to pinpoint the suggested genotype-phenotype pairings driving the observed interactions. Analyses were performed using the R-program version 3.0.2 (R Development Core Team, 2013).
5. RESULTS

5.1. BIRTH WEIGHT AND SCHIZOPHRENIA RISK

BIRTH WEIGHT AND SCHIZOPHRENIA

HBW (> 4000 g) was associated with a 1.68-fold increased risk of developing schizophrenia (Table 13). An association between HBW and PPD was not observed. An association between LBW (< 2500 g) in relation to both schizophrenia and PPD risk was not observed. Intermediate birth weight (3000-4000 g) was used as a reference.

Table 13. Hazard Rate Ratios (HRRs) of the association between birth weight and schizophrenia risk, in which birth weight (<2500 g, 2500–2999 g, 3000–4000 g, >4000 g), maternal/paternal history of any psychotic illness and sex were used as explanatory variables.

<table>
<thead>
<tr>
<th>Birth Weight</th>
<th>Outcome (SZ, PPD)</th>
<th>HRR</th>
<th>95%CI</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>HBW (&gt;4000g)</td>
<td>SZ</td>
<td>1.68</td>
<td>1.13-2.50</td>
<td>0.010</td>
</tr>
<tr>
<td></td>
<td>PPD</td>
<td>1.18</td>
<td>0.84–1.65</td>
<td>0.35</td>
</tr>
<tr>
<td>LBW (&lt;2500g)</td>
<td>SZ</td>
<td>1.00</td>
<td>0.47–2.17</td>
<td>0.99</td>
</tr>
<tr>
<td></td>
<td>PPD</td>
<td>0.95</td>
<td>0.53–1.71</td>
<td>0.87</td>
</tr>
</tbody>
</table>

GESTATIONAL DIABETES AND SCHIZOPHRENIA

It was postulated that the association between HBW and schizophrenia could be driven by gestational diabetes. The association between maternal diabetes at the time of data collection, a proxy for gestational diabetes, was evaluated in relation to schizophrenia susceptibility among offspring. A statistically significant increase in schizophrenia risk was observed among offspring of mothers who developed diabetes (HRR 1.66, 95%CI 1.01-2.72, p=0.044). An association between maternal diabetes and psychotic disorders other than schizophrenia was not observed (HRR 1.48, 95%CI 0.66-3.31, p=0.34). A direct association between maternal diabetes and HBW was not observed.
5.2. BIRTH WEIGHT AND SYMPTOM SEVERITY

Both increasing and decreasing birth weight, in relation to intermediate birth weight, were found to be associated with increasing severity of both disorganized and negative symptoms. Symptoms of reality distortion (hallucinations, delusions) were not found to associate with birth weight. Decreasing birth weight was correlated with greater symptom severity than increasing birth weight in all of the statistically significant associations observed.

Birth weight and disorganized symptoms. A U-shaped association was observed between birth weight and bizarre behaviour in both schizophrenia and PPD subjects (Table 14). A linear association was observed between birth weight and formal thought disorder, with lower birth weight correlating with greater symptom severity, among both subjects with schizophrenia and PPD.

Birth weight and negative symptoms. A U-shaped association was observed between birth weight and both affective flattening and attentional impairment among subjects with PPD (Table 14). An association between birth weight and affective flattening was not observed in the schizophrenia group. The association between birth weight and attentional impairment was of borderline statistical significance in the schizophrenia group. A statistically significant association was not observed between birth weight and alogia, avolition–apathy and anhedonia–asociality.

Table 14. Significant associations observed between birth weight and symptom dimensions (SAPS/SANS scores) in relation to both schizophrenia and PPD groups. FTD = Formal thought disorder. SZ = Schizophrenia. PPD = Primary psychotic disorder.

<table>
<thead>
<tr>
<th>Symptom</th>
<th>Trend of association</th>
<th>Group</th>
<th>βLinear, βQuadratic</th>
<th>SE</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bizarre behaviour</td>
<td>U-Shaped</td>
<td>PPD</td>
<td>βLinear -3.92, βQuadratic 0.57</td>
<td>0.76</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td></td>
<td>SZ</td>
<td>βLinear -3.14, βQuadratic 0.44</td>
<td>1.13</td>
<td>0.01</td>
</tr>
<tr>
<td>FTD</td>
<td>Linear</td>
<td>PPD</td>
<td>βLinear 0.29</td>
<td>0.12</td>
<td>0.02</td>
</tr>
<tr>
<td></td>
<td></td>
<td>SZ</td>
<td>βLinear -0.28</td>
<td>0.14</td>
<td>0.04</td>
</tr>
<tr>
<td>Affective flattening</td>
<td>U-Shaped</td>
<td>PPD</td>
<td>βLinear 1.87</td>
<td>0.67</td>
<td>0.01</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>βQuadratic 0.26</td>
<td>0.10</td>
<td>0.01</td>
</tr>
<tr>
<td>Attentional impairment</td>
<td>U-Shaped</td>
<td>PPD</td>
<td>βLinear 2.72</td>
<td>0.90</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>βQuadratic 0.36</td>
<td>0.14</td>
<td>0.01</td>
</tr>
</tbody>
</table>
5.3. BIRTH WEIGHT AND COGNITIVE IMPAIRMENT

A statistically significant curvilinear association was observed between birth weight and cognitive performance with respect to the Block Design subtest, the Digit Symbol subtest, the Trail Making Test parts A and B, the Digit Span Backward test and the Visual Span Backward test among subjects with schizophrenia and their unaffected first-degree relatives. Birth weight in the range of 3500–4000 g was associated with the highest test performance. The group × birth weight interactions and the group x birth weight-squared (group=subjects with schizophrenia vs. non-affected relatives) were not found to be statistically significant. See Figure 4 for a graphic illustration of the association between birth weight and cognitive functioning (Similarities test) relative to both subjects with schizophrenia and their unaffected relatives.

Figure 4. Predicted Similarities test scores by birth weight (kg) with respect to both (a) the first-degree relatives group (red) and (b) the schizophrenia group (black).
5.4. INTERACTION OF BIRTH WEIGHT AND GENES OF THE DISC1 NETWORK

HBW was found to interact with the NDE1 haplotype ($b=1.26$, $SE=0.5$, $p=0.012$) and one of its constituent SNPs, rs4781678 ($b=1.33$, $SE=0.51$, $p=0.010$), in association with increased schizophrenia risk (Table 15). An interaction between birth weight and the other investigated gene variants, which met the imposed allele frequency threshold criteria ($PDE4B$, $PDE4D$), in relation to schizophrenia risk was not detected. In order to further characterize the genetic mechanism (major allele homozygous vs. heterozygous vs. minor allele homozygous) underlying the observed interaction, schizophrenia risk was analysed individually with respect to NDE1 variants using the Fisher’s exact test. A significant association was observed between HBW and schizophrenia among minor allele homozygotes of the NDE1 haplotype (Figure 5) and two of its constituent SNPs, rs4781678 and rs2075512. This finding suggests that a recessive genetic model was driving the interaction.

Table 15. Interaction between birth weight ($\leq 4000$ g vs. $>4000$ g) and variants of NDE1, fulfilling the imposed allele frequency criteria, in relation to schizophrenia risk. The threshold frequency for minor allele homozygotes was set at $>10\%$.

<table>
<thead>
<tr>
<th>NDE1 Variant</th>
<th>HBW x NDE1 Variant</th>
<th>$b$</th>
<th>$SE$</th>
<th>$p$</th>
</tr>
</thead>
<tbody>
<tr>
<td>NDE1 haplotype</td>
<td></td>
<td>1.26</td>
<td>0.5</td>
<td>0.012*</td>
</tr>
<tr>
<td>rs4781678</td>
<td></td>
<td>1.33</td>
<td>0.51</td>
<td>0.010*</td>
</tr>
<tr>
<td>rs881803</td>
<td></td>
<td>1.07</td>
<td>0.53</td>
<td>0.043</td>
</tr>
<tr>
<td>rs2075512</td>
<td></td>
<td>1.29</td>
<td>0.61</td>
<td>0.035</td>
</tr>
</tbody>
</table>

* $p<0.017$
Figure 5. Association between increasing allele frequency of the *NDE1* haplotype and birth weight >4000 g (dark blue) and <4000 g (light blue) in relation to schizophrenia risk.


6. DISCUSSION

The main aim of the thesis was to investigate whether deviations in birth weight, a proxy for pre- and perinatal environmental adversity, could in combination with elevated genetic liability augment schizophrenia risk, and impact both the clinical presentation and cognitive functioning of subjects who develop schizophrenia. The interaction between birth weight and specific genes involved in neurodevelopment were also analysed.

6.1. BIRTH WEIGHT AND SCHIZOPHRENIA RISK

In the first study, HBW (> 4000 g) was found to be associated with increased risk of schizophrenia. A statistically significant association between birth weight and PPDs other than schizophrenia was not detected, suggesting that individuals who later develop schizophrenia may be particularly sensitive to pre- and perinatal factors associated with birth weight. An association between LBW (< 2500 g) and schizophrenia was not detected, despite this being a widely replicated finding in the literature (Cannon et al. 2002a; Abel et al., 2010; Byars et al., 2014). This discrepancy is proposed to reflect the assumption that the risk factors associated with schizophrenia may be different among individuals presenting with elevated genetic liability for schizophrenia, in comparison to the risk factors operating in the general population (Keskinen et al., 2013). It is also possible that morbidity later in life, which has been commonly associated with LBW, has become more apparent in more recent cohorts due the decrease in mortality seen among LBW neonates in younger cohorts as a result of advances in obstetric and neonatal care (McCormick et al., 1985; Heinonen et al., 1988; Sipilä et al., 1994). The median year of the birth study sample was 1957 and thus, subjects comprising the study group were born during a period of major advancements in obstetric services in Finland. This is, for example, reflected in the number of births that took place in hospitals, which in Finland increased from 31.0% to 92.5% between the years 1940 and 1960 (Hemminki, 1983); during this period the infant mortality rate also decreased from 4% (1950-1951) to 0.5% (1991-1995) (The official statistics of Finland XI: 62, 1962; Hemminki, 1983; Piekkalala et al., 1986; Pitkänen et al., 2000). The distribution of birth weight has also changed in the Finnish population, and the overall prevalence of HBW neonates in younger cohorts has increased (Kinnunen et al., 2003; Lahti-Koski et al., 2010).
The association between HBW and schizophrenia described in the present study in conjunction with elevated genetic loading for schizophrenia supports recent findings from the NFBC suggesting that HBW increases schizophrenia risk, particularly among subjects presenting with elevated genetic liability for schizophrenia (Keskinen et al., 2013). Interestingly, in the literature HBW has been found to associate with an increased risk of neurodevelopmental disorders, including ASDs in both the Swedish (Abel et al., 2013), and the Danish (Byars et al., 2014) populations. ASDs have been associated with elevated heritability estimates (80%) similar to that of schizophrenia (Sullivan et al., 2012), supporting evidence suggesting that the interaction of genetic predisposition and pre- and perinatal factors associated with HBW may influence neurodevelopment.

This study corroborates previous reports describing an association between HBW and increased schizophrenia risk (Hultman et al., 1997; Gunnell et al., 2003; Bersani et al., 2007; Moilanen et al., 2010). However, it was not possible to assess the direct causal factors underlying this association in the study sample. The causal mechanisms underlying the association between HBW and schizophrenia risk have not to date been investigated in the literature. One of the potential causal mechanisms underlying the association between HBW and schizophrenia could be pre- and perinatal hypoxia. HBW is associated with hypoxia-related obstetric complications, including prolonged labour, instrumental vaginal delivery, emergency Caesarean section, Apgar score <4 and increased admission to a neonatal special care unit (Jolly et al., 2003; Jastrow et al., 2010). There is evidence for an association between the overall severity of obstetric complications and increased ventricular enlargement in subjects with schizophrenia (Bersani et al., 2009), and hypoxic-ischaemia-related obstetric complications have been associated with an increased risk of developing a psychotic disorder (Zornberg et al., 2000). Some evidence suggests that subjects with genetic liability for schizophrenia, may be more susceptible to decreased foetal growth in connection with prenatal adversities such as pre-eclampsia, prenatal infection and hypoxia (Eide et al., 2013; Fineberg et al., 2013). An association between LBW and schizophrenia was not detected in this study sample, presenting with high genetic loading for schizophrenia, suggesting the pre- and perinatal factors associated with elevated genetic susceptibility may be different from those operating in the general population.

The association of perinatal hypoxia and genetic liability is a well replicated finding in relation to schizophrenia risk (Stefanis et al., 1999; Rosso et al., 2000; Cannon et al., 2002b; Van Erp et al., 2002; Schulze et al., 2003; Ebner et al., 2008). It is possible that the influence of HBW on hypoxia-related perinatal morbidity may have been greater in older cohorts, where access to
specialized obstetric and neonatal care was limited. Advances in obstetric care may have reduced the frequency and severity of HBW-associated hypoxia-related obstetric complications in younger cohorts (Henriksen, 2008; Koyanagi et al., 2013). It is also probable that the risk of hypoxia-related perinatal complications associated with the delivery of a large infant would have been elevated, particularly among subjects born at home relative to subjects born in a hospital setting.

Based on the primary finding of an association between HBW and schizophrenia, it was investigated whether this association was driven by maternal diabetes. Gestational diabetes, one of the strongest triggers of HBW (Wallace and McEwan, 2007), was not routinely diagnosed prior to the 1970s (Golden et al., 2009), thus information on maternal diabetes at the time of the interview was used a proxy for a possible history of gestational diabetes. This inference was justified based on findings demonstrating that women with gestational diabetes have been found to present with a 10-fold increased risk of developing T2DM (Shen et al., 2016). In the present study, maternal diabetes was found to increase schizophrenia risk among offspring. However, the association was found to be independent of birth weight. A statistically significant association between maternal diabetes and primary PPDs other than schizophrenia was not detected, suggesting that individuals who develop schizophrenia may be sensitive to factors related to impaired maternal glucose metabolism during pregnancy. The observation is in accordance with previous findings describing an association between maternal diabetes during pregnancy and increased schizophrenia risk among offspring (Cannon et al., 2002a). However, the meta-analysis by Cannon et al. (2002a) did not differentiate between types of diabetes (type 1, type 2 or gestational diabetes) and also did not specify whether impaired maternal glucose metabolism was present before pregnancy. In the current study, the precise nature and course of maternal diabetes in relation to pregnancy were also unknown. Offspring of diabetic mothers have been found to present with HBW, LBW and normal birth weight, depending on the severity of diabetes, the degree of diabetic control and the presence of other complicating factors (Ornoy, 2011). Maternal diabetes during pregnancy is most commonly associated with HBW. However, diabetes during pregnancy in combination with such complications as nephropathy is typically associated with LBW (Kendrick et al., 2015). Diabetes during pregnancy is associated with foetal hypoxia, immune system activation, oxidative stress, placental abnormalities, pre-eclampsia, lower Apgar scores and increased requirement of Caesarean section, factors also associated with increased schizophrenia risk (Kotlicka-Antczak et al., 2001; Cannon et al., 2002a; Van Lieshout and Voruganti, 2008; Reece et al., 2009; Benros et al., 2011; Ornoy, 2011; Vambergue and Fajardy, 2011; Wadsack et al., 2012; Eide et al., 2013). These factors commonly co-precipitate and are thus not
independent of one another. Interestingly, T2DM has been found to be more prevalent among parents of individuals with psychotic disorders, suggestive of shared environmental and genetic risk factors underlying both disorders (Fernandez-Egea et al., 2008a, Fernandez-Egea et al., 2008b; Van Welie et al., 2013; Miller et al., 2016). Interestingly, specific variants of the D2-receptor gene have recently been found to associate with elevated glucose levels among subjects with schizophrenia, strengthening the evidence for possible shared risk factors underlying glucose metabolism and psychiatric morbidity (Lawford et al., 2016).

Various autoimmune diseases, including systemic lupus erythematosus, Sjogren’s syndrome and rheumatoid arthritis, and some childhood leukaemias have been associated HBW (Simard et al., 2008; Caughey et al., 2009; Mandl et al., 2009). The mechanisms underlying an association between HBW and immunological disorders remain relatively unsubstantiated. One explanation has been suggested to relate to the effects of cortisol and prenatal dysregulation of the hypothalamic–pituitary–adrenal (HPA) axis (Phillips et al., 1998). Patients with rheumatoid arthritis have been proposed to present with abnormally low levels of cortisol, which may associate with increased inflammation (Imrich and Rovensky, 2010).

### 6.2. BIRTH WEIGHT AND SYMPTOM SEVERITY

In this study both low and high birth weight were associated with increased symptom severity, particularly with respect to disorganized and negative symptom dimensions, among subjects diagnosed with a PPD diagnosis, with elevated genetic loading for schizophrenia. These findings are in line with previous reports describing an association between the increasing severity of obstetric complications and the increasing severity of both positive and negative symptoms in schizophrenia (Kotlicka-Antczak et al., 2001; Guerra et al., 2002; Ruiz-Veguilla et al., 2008), suggesting that the pre- and perinatal environmental factors mediating the association between birth weight and schizophrenia risk may also influence symptom severity.

In the study a U-shaped relationship between both increasing and decreasing birth weight and increasing symptom severity was described with respect to bizarre behaviour, affect flattening, alogia and attentional difficulties. Whereas, a linear association was observed between decreasing birth weight and increasing symptom severity of positive formal thought disorder. Interestingly, ASDs, which have also been associated with increasing birth weight (Abel et al., 2013; Byars et al., 2014), have been commonly found to present with symptoms resembling disorganized and negative symptoms
One of the central findings of the study was that birth weight was not associated with symptoms of reality distortion, in line with the hypothesis presented by some authors, suggesting that symptoms of reality distortion represent a more distal phenotypic characteristic of SSDs relative to disorganized and negative symptoms, which have been proposed to correlate more strongly with both neurodevelopment (Tsuang et al., 2000; Andreasen, 1999) and outcome measures, including social and functional impairment (Eslami et al., 2011; Moskowitz and Heim, 2011; Fulford et al., 2013).

Relatively few studies to date have investigated the relationship between the severity of obstetric complications in relation to symptom severity in schizophrenia. Individuals with a history of obstetric complications (as measured by APGAR score) have been reported to present with more prominent negative symptoms (Kotlicka-Antczak et al., 2001; Ruiz-Veguilla et al., 2008). In addition, Gallagher et al. (2014) found this association in female patients, but not in male patients. The mechanisms underlying the association have been suggested to be related to perinatal hypoxia. Maternal smoking during pregnancy, which may cause chronic foetal hypoxia, has been reported to be associated with a greater severity of negative symptoms in schizophrenia (Stathopoulou et al., 2013). Earlier age at onset of psychotic symptoms, related to poorer prognosis, has also been associated with perinatal hypoxia (Verdoux et al., 1997). However, the association between obstetric complications and negative symptoms may also be related to sociodemographic factors, including lower social class (Jones et al., 2011).

The results of the study appear to support the view that the pre- and perinatal factors associated with both decreasing and increasing birth weight are associated with the severity of negative and disorganized symptoms in SSDs. The postulated biological mechanisms underlying the association may be linked to processes affecting neurodevelopment and cognition, and in this respect may partly overlap with the in utero environmental factors involved in the pathogenesis of ASDs.
6.3. BIRTH WEIGHT AND COGNITIVE IMPAIRMENT

Disorganized and negative symptoms, as compared with symptoms of reality distortion, have been associated with greater cognitive impairment (Jablensky, 2006; Ventura et al., 2010). In the study, both decreasing and increasing birth weight was associated with a small, but significant decrease in visuospatial reasoning, processing speed, set-shifting and verbal and visual working memory, with intermediate birth weight being associated with the highest cognitive performance. Our observation is in line with previous findings describing an association between decreasing birth weight and increasing cognitive impairment in SSDs (Rifkin et al., 1994; Hultman et al., 1999; Freedman et al., 2013). These findings are also consistent with reports describing an association between decreasing birth weight and poorer cognitive functioning in the general population (Seidman et al., 2000; Taylor et al., 2000; Bergvall et al., 2006; Tanskanen et al., 2011). Although research has predominantly described an association between LBW and cognitive functioning, particularly in relation to the VLBW range (<1500 g) (Bohm et al., 2004; Murray et al., 2014; Aanes et al., 2015), HBW has also been associated with an increased risk of neurocognitive outcomes, including externalizing symptoms and social problems in adolescents and also an increased risk of ASDs (Shenkin et al., 2004; Leonard et al., 2008; Alati et al., 2009; Buschgens et al., 2009; Van Lieshout and Boyle, 2011; Abel et al., 2013; Byars et al., 2014). Some studies have also reported an association between HBW and increased cognitive impairment in the general population (Sorensen et al., 1997; Silva et al., 2006).

The intrauterine environment has been proposed to influence cognitive functioning. Evidence suggests that maternal metabolic factors, including increased maternal body mass index (Jones et al., 1998; Schaefer et al., 2000; Kawai et al., 2004; Khandaker et al., 2012), maternal hypertension (Sorensen et al., 2003) and pre-eclampsia (Dalman et al., 1999; Eide et al., 2013), influence foetal growth and increase schizophrenia risk. Structural brain imaging findings of healthy individuals have reported an association between decreasing birth weight and a decrease in regional brain surface area (Walhovd et al., 2012; Muller et al., 2014). Prenatal factors such as undernutrition, maternal cardiovascular risk factors and HPA-axis dysregulation, have been associated with both lower birth weight and cognitive outcomes among offspring (De Rooij et al., 2010; Räikkonen et al., 2012). Epigenetic processes are speculated to be an important mechanism by which the prenatal environment alters factors related to long-term health, including cognitive functioning (Warner and Ozzane, 2010; Eriksson, 2016). The epigenetic regulation of the expression of specific genes e.g. IGFs, in particular IGF-II, may play a role in the association between birth weight,
neurodevelopment and cognitive functioning (Heijmans et al., 2008; Wehkalampi et al., 2013; Iwamoto and Ouchi, 2014). Lower methylation of the IGF-II gene has been associated with prenatal undernutrition (Heijmans et al., 2008), VLBW (Wehkalampi et al., 2013), and cognitive functioning (working memory) (Cordova-Palomera et al., 2014). However, socioeconomic factors have also been suggested to confound some of the reported associations between birth weight and cognitive functioning (Wong and Edwards, 2013).

Only a few studies to date have investigated the relationship between HBW and cognitive functioning. It is plausible that the mechanisms underlying the observed association between HBW and cognitive impairment may be related to factors associated with perinatal hypoxia, which has also been associated with cognitive impairment in the general population (Anastario et al., 2012). Lower five-minute Apgar scores, although not a direct indicator of perinatal hypoxia, have been associated with cognitive impairment in the general population (Odd et al., 2008; Ehrenstein, 2009; Stuart et al., 2011; Tweed et al., 2016). Maternal diabetes during pregnancy has also been associated with cognitive impairment among offspring (Silverman et al., 1998; Dahlquist et al., 2007; Dionne et al., 2008; Nielsen et al., 2010; Perna et al., 2015; Adane et al., 2016; Bytoft et al., 2016). Maternal pre-pregnancy obesity, maternal weight gain during pregnancy and maternal diabetes have been reported to be associated with dysregulation of the immune system (increased production of pro-inflammatory cytokines), oxidative stress, endocrinological factors (impacting cortisol, oestrogen and insulin levels) and nutritional factors (increased glucose concentration, folate deficiency), which may potentially impact neurodevelopment and increase the risk of ASDs and SSDs in offspring (Challier et al., 2008; Freeman, 2010; Van Lieshout and Boyle, 2011; Gardner et al., 2015).

### 6.4. INTERACTION BETWEEN HBW AND NDE1

Evidence suggestive of a gene-environment interaction was detected between the NDE1 gene and HBW (> 4000 g) in association with increasing schizophrenia risk. The observation corroborates previous findings describing an association between mutations in the NDE1 gene and neurodevelopmental outcomes, including congenital microencephaly (Bakircioglu et al., 2011; Paciorkowski et al., 2013), and describing a role for NDE1 in the neurodevelopmental trajectory of schizophrenia (Hennah et al., 2007). The results also concur with previous reports describing an interaction between schizophrenia susceptibility genes and pre- and perinatal environmental factors, e.g. foetal hypoxia, with a subsequent increase in schizophrenia risk (Narita et al., 2000; Chotai et al., 2003; Demontis et al., 2011; Schmidt-
Kastner et al., 2012; Borglum et al., 2014). Furthermore, the observation supports previous findings suggesting that the association of HBW and schizophrenia may be moderated by genetic predisposition (Keskinen et al., 2013).

A recent large GWAS failed to detect an association between DISC1-associated genes and schizophrenia (Schizophrenia Genomics Consortium, 2014). However, evidence suggests that the DISC1 network may be associated with intermediate neurocognitive phenotypes of schizophrenia and other psychiatric disorders (Hennah et al., 2005; Carless et al., 2011). Duplications and deletions at the 16p13.11 locus have been found to predispose to various neurodevelopmental disorders, including ASDs, attention deficit disorder, intellectual disability and schizophrenia (Tropeano et al., 2013). Large alterations of the gene NDE1, a DISC1 binding partner located within the 16p13.11 locus, have been implicated in the disruption of the neurodevelopmental process, and smaller alterations (variants) of the gene have been associated with increased schizophrenia susceptibility (Bradshaw, 2016). The interaction of HBW and NDE1, both of which have previously been identified as independent risk factors of neurodevelopmental outcomes (Abel et al., 2013; Byars et al., 2014; Bradshaw, 2016), from a hypothesis-driven perspective reflects a biologically plausible aetiological mechanism with regard to the neurodevelopmental trajectory of schizophrenia. An association between NDE1 and HBW has not previously been reported, and the finding thus needs to be replicated.

The molecular mechanisms underlying the observed interaction may be related to foetal hypoxia, which has been found to interact with variants of AKT1, BDNF, DTNBPI and GRM3 in association with increased schizophrenia risk (Nicodemus et al., 2008). Although a direct association between foetal hypoxia and NDE1 functioning has not yet been described, evidence suggests that the half-life of its binding partner DISC1 is decreased (degradation of DISC1 is increased) under hypoxic conditions (Barodia et al., 2015). Variants of NDE1, in relation to the concerted functions of the DISC1 network, may possibly be susceptible to HBW-associated perinatal hypoxia, impacting neurodevelopment and influencing subsequent schizophrenia risk.
6.5. STRENGTHS AND LIMITATIONS OF THE STUDY

General strengths of the study include the reliability of diagnostic data and the fact that information concerning birth weight was obtained from documented obstetric records, not maternal recall, minimizing bias. General limitations include not having information concerning gestational age and the socio-economic status of individual families. Gestational age, a factor reflecting foetal growth, has been found to be a predictor of future psychiatric morbidity. In particular, being small for gestational age, reflective of reduced foetal growth, has been associated with increased schizophrenia risk (Nielsen et al., 2013). The study was confined to the investigation of birth weight since other obstetric information was limited, particularly among subjects born in the 1940s and 1950s and subjects born at home. Thus, the direct evaluation of the association between other potentially relevant obstetric events, including APGAR score, infections during pregnancy, pre-eclampsia and gestational diabetes, in relation to the investigated outcome measures was not possible. A broader scope would have furthered understanding of the causal mechanisms driving the association between birth weight and the outcome measures investigated. The Finnish schizophrenia family cohort presented the opportunity to investigate the relationship between birth weight and schizophrenia in an older study sample with an elevated genetic loading for schizophrenia, in which the majority of birth weight data were from the 1940s and 1950s. Many other studies have investigated the association of obstetric complications and schizophrenia risk in younger cohorts. However, due to advances in obstetric care over the decades, it is postulated that the pre- and perinatal complications associated with neonatal mortality/morbidity may have been different in older birth cohorts. High mortality of LBW subjects in older cohorts (Rantakallio, 1969) is suggested to have hindered the ability to detect an association between LBW and schizophrenia risk in the study sample, a finding which has been extensively reported in the literature (Cannon et al., 2002a; Abel et al., 2010). Other limitations include lack of a control group with no genetic loading. Due to the high genetic loading for schizophrenia in our study sample, direct extrapolation of findings in relation to the general population cannot be made. However, Keskinen et al. (2013) found in the population-based Northern Finland 1966 cohort that high birth weight was a risk factor for schizophrenia only in families where the parent(s) had a psychotic disorder, which is consistent with our findings.
7. CONCLUSIONS

In this study, an association was observed between HBW (> 4000 g) and increased schizophrenia risk among offspring, in line with previous reports describing an association between HBW and both schizophrenia (Hultman et al., 1997; Gunnell et al., 2003; Bersani et al., 2007; Moilanen et al., 2010; Keskinen et al., 2013) and ASDs (Abel et al., 2013; Byars et al., 2014). Evidence of a putative gene-environment interaction between NDE1, a gene implicated in neurodevelopment (Bradshaw, 2016), and HBW was detected in association with increased schizophrenia risk. Furthermore, a relationship was observed between both decreasing and increasing birth weight and the increasing severity of disorganized and negative symptoms and decreasing cognitive functioning.

Schizophrenia is associated with heritability estimates of up to 81%, implying that both genetic and environmental factors moderate schizophrenia risk (Sullivan et al., 2012). The association of various independent genetic (Schizophrenia Genomics Consortium, 2014) and obstetric complications in relation to schizophrenia risk is widely acknowledged (Cannon et al., 2002a). Individuals who develop schizophrenia are speculated to be genetically more susceptible to the impact of various environmental factors, which exert their effect during the neurodevelopmental process (Insel, 2010). However, the vast majority of individuals presenting with genetic susceptibility who are exposed to environmental adversity do not develop schizophrenia. The reason for this has been suggested to be that schizophrenia susceptibility is typically not conferred by a single gene or a single environmental factor, but rather by the cumulative influence of multiple susceptibility genes interacting with multiple environmental factors over the course of time. Although many individual genetic and environmental factors have been associated with increased schizophrenia susceptibility, less is known about factors underlying resilience, which provides protection against the development of the disorder (Rutter et al., 2006). Despite increasing knowledge of both genetic and environmental risk factors associated with schizophrenia risk, fundamental questions regarding the causal pathways remain unanswered. The ultimate question is as follows: can the acquisition of this information in relation to the longitudinal accumulation of risk factors be utilized in a clinical setting to predict the probability of an individual developing schizophrenia and to aid in the discovery of preventative treatment strategies?

The neurodevelopmental trajectory of schizophrenia posits that schizophrenia risk is mediated by the interaction of genetic susceptibility and environmental
adversity occurring during the neurodevelopmental process (Insel, 2010). It has been suggested that the neurodevelopmental process represents a period of elevated susceptibility, during which the cumulative effects of various environmental factors may lead to an incrementally increasing disruptive effect on this process (Insel, 2010). The neurodevelopmental model is suggested to be corroborated by clinical observations describing schizophrenia as being associated with a longitudinal course, i.e. with the presence of notable premorbid signs, symptoms and functional impairment becoming manifest years before the onset of overt psychotic symptoms (Häfner et al., 2003). The mechanisms by which schizophrenia susceptibility genes and environmental factors interact in relation to the longitudinal course of schizophrenia remain largely hypothetical and unsubstantiated.

Individuals who develop schizophrenia have been suggested to be more vulnerable to the effects of pre- and perinatal factors, such as hypoxia and infection, than the general population (Nicodemus et al., 2008; Clarke et al., 2009). The association between HBW and increased schizophrenia risk in the present sample, presenting with elevated genetic susceptibility for schizophrenia (Hovatta et al., 1997; Hovatta et al., 1999; Ekelund et al., 2001; Hennah et al., 2003), corroborates recent reports suggesting that the association between HBW and schizophrenia may be moderated by genetic liability (Keskinen et al., 2013). The association also suggests that variants of NDE1, a gene implicated in neurodevelopment (Hennah et al., 2007; Tomppo et al., 2009), may interact with HBW with potential consequences on the neurodevelopmental trajectory. The functions of NDE1 may thus be vulnerable to the influence of environmental factors associated with HBW. Although speculative, the association between HBW and schizophrenia in the current study may be related to perinatal hypoxia upon delivery of a HBW neonate.

Patients with schizophrenia typically present with cognitive impairment (Keefe and Reichenberg, 2014). Similar, but more subtle cognitive impairment has been found in first-degree relatives of patients with schizophrenia (Tuulio-Hendriksson et al., 2003). The observation that both decreasing and increasing birth weight is associated with increased severity of disorganized and negative symptoms and decreased cognitive functioning suggests that these manifestations are proximal to the neurodevelopmental origins of the disorder (Tsuang et al., 2000), in contrast to symptoms of reality distortion, which appear to be more peripheral in relation to this trajectory.

From a clinical perspective, knowledge that pre- and perinatal factors are associated with an increased risk of future psychiatric morbidity, particularly in combination with elevated genetic susceptibility, emphasizes the need for the health care system to better recognize and address this widely replicated
association. However, identification of at-risk individuals before the advent of genetic testing can only be based on familial history of psychiatric morbidity. Evidence suggests that women with schizophrenia have fewer antenatal care visits during pregnancy (Bennedsen et al., 2001), which in itself represents a factor associated with increased schizophrenia risk among offspring (Byrne et al., 2007). The British National Institute of Health and Care Excellence (NICE) guideline regarding antenatal and postnatal mental health (Howard et al., 2014) recommends the implementation of an integrated care plan for women presenting with psychiatric disorders during pregnancy. In the plan, the roles of each of the respective health care providers are clearly defined, starting from the implementation of a monitoring schedule and designation of the provider responsible for coordination of the plan. The aim of the plan is to ensure that each participant involved acknowledges their specific responsibilities and that information concerning the patient is shared effectively. In Finland, a national guideline addressing the integrated role of different health care providers in the antenatal care of women with severe psychiatric disorders has not yet been established. Construction of such a Finnish national guideline is suggested to promote co-operation of psychiatric, antenatal and obstetric services to establish a foundation relative to which the integration of these services may be strengthened for the benefit of Finnish women with severe mental disorders and their children.
8. FUTURE AREAS OF INVESTIGATION

Although the current study has focused on the association between pre- and perinatal factors in relation to schizophrenia risk, it must be emphasized that the in utero environment has also been investigated in relation to the development of various other psychiatric disorders including depression (Räikkonen et al., 2012).

The increasing sample size of future GWASs may replicate previous findings and enable the detection of novel genes mediating schizophrenia risk, thus aiding in the formulation of novel hypothesis-driven approaches for the analysis of future gene-environment interactions.

The investigation of environmental factors in relation to gene-environment interaction analyses will require more validation with respect to the qualitative and quantitative nature of the environmental factor under consideration (Caspi et al., 2010). For instance, childhood adversity is a very broad term, encompassing a multitude of adversities of variable intensity and duration, and thus, specification of what exactly is being measured will increase the validity of findings and facilitate the replication of gene-environment interaction studies, which is a prerequisite for further large-scale investigations. The validation of environmental factors within a specific cohort will also require greater understanding of the demographic considerations of the sample and how these demographic factors differ in relation to separate populations and are subject to change over time. For instance, the epidemiological characterization of potential causal factors underlying the association between birth weight and schizophrenia is complicated by evidence suggesting that the influence of potential causal factors may change as a function of time, particularly in relation to both changes in the nutritional status of a population (Yamada et al., 2014) and advances in obstetric practice and neonatal care (The official statistics of Finland XI: 62, 1962; Hemminki, 1983; Piekkala et al., 1986; Pitkänen et al., 2000). Understanding the demographic considerations of a specific study sample may facilitate the verification of hypotheses concerning causality.

The present study corroborated previous findings implicating an association between the in utero environment and future psychiatric morbidity, in this case the associations between birth weight and schizophrenia risk, symptom severity and cognitive functioning, which are likely moderated by the interaction of genetic susceptibility and early environmental adversity. More information on pre- and perinatal factors needs to be collected in order to
elucidate the causal mechanisms. The use of serum samples to directly assess the presence of a potential biomarker in relation to schizophrenia risk has already been employed. This has resulted in the discovery of an association between the nicotine concentration of maternal serum during pregnancy and increased schizophrenia risk among offspring (Niemelä et al., 2016). A study investigating the association between maternal infection at the time of pregnancy and the risk of psychosis in offspring, reported an association between elevated maternal immunoglobulin levels against Toxoplasma gondii and cytomegalovirus and decreased levels of neonatal acute phase proteins, in relation to the increased risk of non-affective psychoses among offspring (Blomström et al., 2015). This finding was suggested to reflect an association between maternal chronic infection and deficient foetal immune system responses, in relation to increased risk of psychosis.

In addition to pre- and perinatal factors, schizophrenia risk is also mediated by postnatal factors, including psychosocial factors associated with childhood adversity, and cannabis use (Caspi et al., 2005; Alemany et al., 2011; Aas et al., 2014; Hollander et al., 2016). Prospective cohorts of increasing size are being collected to shed light on the role of parental behaviour in relation to childhood development (Nolvi et al., 2016) and will allow a more comprehensive understanding of the causal relationships in relation to the longitudinal trajectory of the disorder. Further characterization of the impact of postnatal factors and their contribution to overall schizophrenia risk in combination with the influence of both genetic and pre- and perinatal risk factors may in the future present an opportunity to define a clinically relevant polyfactorial risk score, with the potential to predict schizophrenia risk, thus having possible implications for prevention of disease progression.

The identification of genetic variation does not take into account factors related to protein expression. Finding a susceptibility gene is analogous to finding an instruction manual on “how to make a car”. Although you have gained access to the instructions and have knowledge of the endpoint, i.e. the car model you want to make, you are still far from understanding the practical considerations involved in each step of the process. One step in furthering our understanding of what lies between the gene-phenotype continuum will require the utilization of proteomics to complement genetic and epigenetic research (Nascimento et al., 2016). The study of epigenetics in psychiatric research has gained increasing momentum and will potentially allow the elucidation of the role of gene expression in psychiatric morbidity (Hannon et al., 2016). However, gene expression levels will not give substantial insight into processes of protein function, which include post-translational modification of proteins (e.g. processes related to protein glycosylation), the trafficking of proteins to appropriate intra- or extracellular destinations and
the interaction of proteins with other proteins. Moreover, proteins often function as complexes, and thus, knowledge of the functions of one component will not necessarily reveal the functions of the complex as a whole. A protein is the ultimate product of gene-environment interaction, and focusing on the biochemical, cellular, histological and neurophysiological aspects of protein functioning is necessary to gain further insight into the causal aetiological mechanisms underlying psychiatric disorders. Recently, various tissue-specific biobanks have been established, including brain biobanks, consisting of brain tissue samples in which protein localization can be investigated (Palmer-Aronsten et al., 2016). However, the use of animal models will still be required to assess protein functioning in a physiological setting (Sekar et al., 2016).

Schizophrenia represents a highly heterogeneous phenotype, and schizophrenia risk is likely to be mediated by the interaction of numerous genetic and environmental factors, each of which independently presents with small individual effect size. Genetic variants of large effect size may increase the probability of detecting a gene-environment interaction. The use of intermediate phenotypes, in combination with genetic variants with large effect sizes, may further facilitate the detection of environmental factors mediating psychiatric morbidity and shed further light on the biological mechanisms involved in neurodevelopment. It is suggested that focusing on the intermediate phenotypes associated with variants of the DISC1 network, rather than schizophrenia as an endpoint, may continue to provide valuable information on the neurodevelopmental underpinnings of psychiatric morbidity.
9. REFERENCES


Alati, R., Najman, J.M., O'Callaghan, M., Bor, W., Williams, G.M., Clavarino, A., 2009. Fetal growth and behaviour problems in early adolescence: findings


Andreasen, N.C., 1983. Scale for the Assessment of Negative Symptoms (SANS). Iowa City, University of Iowa.


Bakircioglu, M., Carvalho, O.P., Khurshid, M., Cox, J.J., Tuysuz, B., Barak, T., Yilmaz, S., Caglayan, O., Dincer, A., Nicholas, A.K., Quarrell, O., Springell, K., Karbani, G., Malik, S., Gannon, C., Sheridan, E., Crosier, M., Lisgo, S.N.,


Bradshaw, N.J., 2016. Cloning of the promoter of NDE1, a gene implicated in psychiatric and neurodevelopmental disorders through copy number variation. Neuroscience 324, 262-270.


interaction with Lis1, the causal protein of Miller-Dieker lissencephaly. Structure 15, 1467-1481.


chimeric DISC1 transcripts that encode structurally altered, deleterious mitochondrial proteins. Hum. Mol. Genet. 21, 3374-3386.


birth weight variation is related to cortical morphology across the psychosis spectrum. Schizophr. Bull. 40, 410-419.


induced effects on psychosis and cognition. Neuropsychopharmacology 31, 2748-2757.


Keefe, R.S., Reichenberg, A., 2016. Predicting Schizophrenia. JAMA Psychiatry. 73, 441-442.


pregnancy and schizophrenia in offspring: a cohort prospective study. BMC Psychiatry 8, 71.


Finland reveals susceptibility loci on chromosomes 2q and 5q. Hum. Mol. Genet. 10, 3037-3048.


Pedersen, C.B., Mortensen, P.B., 2006. Are the cause(s) responsible for urban-rural differences in schizophrenia risk rooted in families or in individuals? Am. J. Epidemiol. 163, 971-978.


Perälä, J., Suvisaari, J., Saarni, S.I., Kuoppasalmi, K., Isometsä, E., Pirkola, S., Partonen, T., Tuulio-Henriksson, A., Hintikka, J., Kieseppä, T., Harkanen,


Peltonen, L., 2007. The role of DTNBP1, NRG1, and AKT1 in the genetics of schizophrenia in Finland. Schizophr. Res. 91, 27-36.


disorder, their unaffected siblings, and healthy unrelated volunteers. Am. J. Psychiatry 159, 1514-1520.


