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Vorwort

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Eidesstattliche Erklärung

Ich erkläre hiermit an Eidesstatt, dass ich die vorliegende Diplomarbeit selbständig verfasst, andere als die angegebenen Quellen und Hilfsmittel nicht benutzt und mich auch sonst keiner unerlaubten Hilfe bedient habe. Ich versichere weiters, dass ich diese Diplomarbeit bisher weder im In- noch im Ausland in irgendeiner Form als Prüfungsarbeit vorgelegt habe.

Wien, 30. Dezember 2016 Andrea Carausu

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Abbreviations

*R	repeat allele
´3UTR	´3 untranslated region
ADHD	Attention Deficit Hyperactivity Disorder
Ag/AgCl	silver chloride
ANOVA	analysis of variance
ANCOVA	analysis of covariance
ASR	acoustic startle response
DA	dopamine
DAT	dopamine transporter
DAT1	SLC6A3; dopamine transporter gene
DAT KO- mice	dopamine transporter knockout- mice
dB	dezibel
DSM	Diagnostic and Statistical Manual
e.g.	for example
EMG	Electromyogram / electromyographic
EMG-L	left eye EMG channel
EMG-R	right eye EMG channel
HD	Huntington's Disease
i.e.	that is
INPP	Institute of Normal and Pathological Physiology
ISI	interstimulus interval
ITI	intertrial interval
ms	milliseconds
MHz	MegaHertz
N	sample size
NEO-FFI	NEO Five Factor Inventory
OCD	Obsessive-Compulsive Disorder
PA	pulse-alone
PP	prepulse-pulse
PPA	prepulse-alone
PPF	prepulse facilitation
PPI	prepulse inhibition
PTSD	Post-Traumatic Stress Disorder
SD	standard deviation
SPQ	Schizotypal Personality Questionnaire
STPD	Schizotypal Personality Disorder
TS	Tourette Syndrome

Abstract

Prepulse inhibition (PPI) of the acoustic startle reflex (ASR) is an operational measure of sensorimotor gating. Gating refers to an automatic cognitive filter mechanism which screens irrelevant sensory stimuli out of awareness. Reduced PPI has been found in schizophrenia (SCZ) patients and may index an inhibitory failure and information processing deficits which may lead to sensory flooding and cognitive fragmentation in these patients. Additionally, studies suggest that some of the clinical signs and reduced PPI result from the same underlying neural dysfunction. Moreover, PPI deficits have not only been found in SCZ patients but also in schizotypal personality disorder and healthy individuals scoring high on schizotypal traits, as well as in clinically unaffected relatives of schizophrenia patients, which provides the use of PPI as an endophenotype in genetic studies of SCZ. The gene SLC6A3, encoding for the dopamine transporter (DAT), is of great interest, as it plays a crucial role in dopamine (DA) neurotransmission which is disturbed in SCZ.

The primary goal of the present work was to examine the potential functional effects of SLC6A3 genotype in healthy individuals on PPI and prepulse facilitation (PPF) of the ASR at several prepulse-lead-intervals (ISIs: 30ms, 60ms, 120ms, 2000ms, 4000ms). Healthy careers of risk alleles in polymorphisms in 3'UTR and intron 8 were expected having lower PPI. Schizotypal and personality traits were assessed using questionnaires and included in the analyses. Our data showed no significant main effect of SLC6A3 genotype on PPI/PPF.. However, we found inverse relationships between PPI of ISI of 30ms and the personality traits of "Neuroticism" and "Unusual Perceptual Experiences". Our analysis also showed that neuroticism could influence the effect of 3'UTR genotype on PPI.

Keywords: prepulse inhibition, prepulse facilitation, sensorimotor gating, schizophrenia, SLC6A3, 3´untranslated region, intron8

Zusammenfassung (Abstract deutsch)

Die Prä-Puls-Hemmung (PPI) des akustischen Schreckreflexes ist eine operationale Messung von Sensomotorischem Gating. Gating bezeichnet einen automatischen Filtermechanismus des Zentralnervensystems, der die Ausblendung irrelevanter sensorischer Reizinformationen bewerkstelligt. Niedrige PPI- Werte wurden bei Schizophrenie (SCZ)-Patienten nachgewiesen und deuten auf eine fehlerhafte Informationsverarbeitung sowie eine Störung hin, sensorische Reize zu hemmen, was bei diesen Patienten zu Reizüberflutung und kognitiver Fragmentierung führen kann. Des Weiteren suggerieren Studien, dass sowohl einige klinischen Symptome als auch eine niedrige PPI demselben neuronalen Defizit unterliegen würden. Zudem wurden Defizite in der PPI nicht nur bei Schizophrenen, sondern auch im Zusammenhang mit schizotypischer Persönlichkeitsstörung nachgewiesen, sowie bei gesunden Individuen, deren schizotype Persönlichkeitseigenschaften stark ausgeprägt sind, als auch bei klinisch unauffälligen Verwandten von SCZ- Patienten, was die Benutzung von der PPI als Endophenotyp in genetischen Studien gewährleistet. Das Gen SLC6A3, welches den Dopamintransporter (DAT) kodiert, ist hier von Bedeutung, als dass es eine wichtige Rolle bei der dopaminergen Neurotransmission einnimmt, welche bei Schizophrenen dysfunktional ist.

Das primäre Ziel der vorliegenden Arbeit war es, den potentiell funktionellen Einfluss des SLC6A3 Genotyps auf die PPI und die Prä-Puls-Erhöhung (PPF) anhand mehrerer Prepulse-Leitintervalle (ISIs: 30ms, 60ms, 120ms, 2000ms, 4000ms) in gesunden Individuen zu untersuchen. Die Erwartung war, dass gesunde Individuen, die im Vorhinein als Träger von Risiko- Allelen klassifiziert wurden, eine kleinere PPI aufweisen würden. Des Weiteren wurden schizotype sowie andere Persönlichkeitseigenschaften anhand von Persönlichkeitsfragebögen erhoben und in die Analysen miteinbezogen. Unsere Daten zeigten keinen signifikanten Haupteffekt des SLC6A3 auf die PPI/PPF. Jedoch zeigten sich negative Zusammenhänge zwischen der PPI in einem ISI von 30ms und den Persönlichkeitseigenschaften „Neurotizismus“ und „Ungewöhnliche perzeptuelle Erfahrungen“. Unsere Analyse zeigte auch, dass Neurotizismus einen Einfluss auf den Effekt des 3´UTR Genotypen auf PPI haben könnte.

Schlagwörter: Prä-Puls-Hemmung, Prä-Puls-Erhöhung, sensomotorisches Gating, Schizophrenie, SLC6A3, 3´untranslated region, intron8

1. Introduction

1.1 Sensorimotor gating (PPI)

Gating represents a basic neurocognitive process, by which irrelevant stimuli are filtered or "gated out" of awareness, so that attention can be focused on the most salient aspects of the stimulus-laden environment (Braff & Geyer, 1990; Braff et al., 2001). Under natural circumstances gating is a continuously active process, which contributes to our ability to modulate a continuous stream of sensory and cognitive information (Light & Braff, 1999). Effective gating helps to prevent sensory overload, whereas when it fails, it may cause sensory inundation, cognitive fragmentation, disorganized thinking and possibly other psychotic symptoms (Braff & Geyer, 1990; Light & Braff, 1999).

There are two frequently used operational measures of assessing gating of sensory stimuli: suppression of the P50 wave of auditory evoked potentials, which is referred to as sensory gating and prepulse inhibition (PPI) of the acoustic startle response (ASR), usually termed sensorimotor gating (Braff & Light, 2005).

Sensorimotor gating describes the regulation of sensory information and its transmission to motor output systems. Prior to transduction to the motor output system, sensory information requires filtering processes (Powell et al., 2012). PPI, as a laboratory paradigm, is a form of modulation or plasticity of the startle reflex (Graham, 1975). The startle reflex consists of a contraction of the skeletal and facial muscles and is a universal, primitive and protective body response to sudden and intense stimuli (Light & Braff, 1999; Braff et al., 2001). In the PPI paradigm, a relatively weak sensory event (the prepulse) is presented 30–500ms before a strong startle-eliciting stimulus (the pulse) which reduces the magnitude of the startle response compared with the response elicited in the absence of a prepulse (Graham, 1975; Braff et al., 2001; Braff & Light, 2005; Braff, 2010). The protection of processing hypothesis states that the prepulse activates preattentional mechanisms which reduce the impact of any further stimulation until the processing of the prepulse is complete. Thus, PPI is believed to reflect the ability to selectively protect the processing of relevant sensory information without interference from subsequent strong or disrupting stimuli for the purpose of

preventing an inundation of information (Braff et al., 2001; Kumari et al., 2008; Braff, 2010, Kohl et al., 2013).

In contrast to the short lead-interval prepulse inhibition effect, the amplitude of the startle response is enhanced when the interval between the prepulse and the pulse (i.e. the prepulse- lead interval or interstimulus interval, ISI) exceeds 1000ms, therefore the term prepulse facilitation (PPF) is used (Dawson et al., 1993). PPI and PPF are thought to represent different neurocognitive processes (Graham, 1975; Dawson et al., 2000; Wynn et al., 2004). PPF, which is relatively less well-studied, is supposed to be caused by a combination of arousal and sustained or selective attention elicited by the prepulse and reflect a later stage of generalized alerting or orienting (Dawson et al., 2000; Wynn et al., 2005).

PPI and PPF are elicited stimuli of any modality, including visual, acoustic, tactile or olfactory. Acoustic stimuli are used most often for experiments, and the majority of human studies measure the eyeblink component of startle using electromyography (EMG) of the orbicularis oculi muscle (Graham 1975; Blumenthal et al., 2005). In experiments, PPI is defined as percentage reduction in the amplitude of the ASR in prepulse-pulse (PP) -trials in contrast to pulse-alone (PA) -trials. The amount of inhibition depends on prepulse and pulse characteristics, such as the pulse and prepulse intensity or prepulse to pulse interval (Blumenthal et al., 1996). Prepulse-to-pulse intervals (ISIs) of 30-240ms are generally used, whereby maximal PPI is observed at ISI of 120ms, with relatively weaker PPI observed at shorter and longer intervals (Braff et al., 2001).

At ISIs of 30 or 60ms, which are considered to be too short to be substantially influenced by a volitional distribution of attentional resources, PPI is thought to index automatic preattentive processing of the prepulse (Dawson et al., 1993; Swerdlow et al., 2008). At ISIs between 120 and 240ms, attentional modulation of PPI is possible by instructing subjects to focus on the prepulse (Dawson et al., 1993 & 2000; Wynn et al., 2003 & 2004; Kohl et al, 2013). Accordingly, the interval of 120ms is “in-between” preconscious and conscious information processing, based on perceptual detection thresholds (Swerdlow et al., 2008). Above ISIs of 120ms, PPI may measure in addition to automatic processing also a rapid, volitional attentional modulation of early sensory processing (Dawson et al., 1993; Bitsios et al., 2005). All in all, PPI may be ideal for the

study of pathological groups for whom attentional dysfunctions are presumed (Dawson et al., 2000).

1.2 PPI in schizophrenia

It has been demonstrated that PPI is reduced in people suffering from schizophrenia (SCZ) and it is considered to be one of the most consistent findings in SCZ research (Braff et al., 2001 & 2007; Braff, 2010; Takashi et al., 2011; Powell et al., 2016). In these patients prepulses did not generate full levels of inhibition compared to healthy comparison subjects. In addition to PPI, also PPF has been proved to be impaired in SCZ patients (Campanella et al., 2009; Dawson et al., 2003 & 2004).

However, PPI deficits are not specific to SCZ and can also be observed in the Tourette syndrome (TS), Huntington's disease (HD) and in obsessive-compulsive disorder (OCD). Inhibitory failures have also been found in patients with post-traumatic stress disorder (PTSD), panic disorder, manic bipolar disorder, Fragile X syndrome, nocturnal enuresis and adults with autism, but evidence is still poor (Swerdlow et al., 1995; Braff et al., 2001; Powell et al., 2012; Kohl et al., 2013). Although the core symptoms of these disorders are diverse, they might be classified as “family of gating disorders”, because they all display a common feature - a loss in the normal ability to suppress irrelevant information, with a gating deficit predominating in the cognitive domain in some disorders and in the sensory or motor domains in others (Geyer, 2006; Braff, 2010; Powell et al., 2012).

It has been hypothesized that both clinical signs and reduced PPI in several disorders result from the same underlying neural deficit within limbic and basal ganglia neural substrates (Swerdlow et al., 1998). The prepulse has inhibitory influences that can be regulated by connections between limbic cortico-striato-pallido-pontine and related cortico-striato-pallido-thalamic circuitry, and the pontine tegmentum, where the primary startle circuit is organized. More specifically, the amount of PPI exhibited by any organism at any given moment reflects activity at many different levels of forebrain and other related neural structures and its output via the pons (Braff et al., 2001; Franklin et al., 2009). Extensive research has shown that the above mentioned

forebrain neural circuitry is not only implicated in neurophysiological deficits but also related to the neurobiology of SCZ (Braff, 2010). Thus, PPI may be a valuable model for the study of the neural substrates of SCZ, because of the relevance of the gating "anatomy" to the pathophysiology of this disorder (Swerdlow et al., 1998). Accordingly, impaired sensory gating is recognized within the Diagnostic and Statistical Manual (DSM, American Psychiatric Association, 1994) as clinically important feature associated with SCZ, which could theoretically lead to a variety of severe dysfunctions in perception, attention and thinking (Dawson et al., 2000). Particularly, psychotic patients are considered to have deficits in sensory registration and the early stages of information processing which, in turn, leads to a cascade of "downstream" deficits in cognitive integration, and ultimately to clinical symptoms and functional impairment (Braff et al., 2001). It has been suggested that the resulting "sensory flooding" is responsible for increased distractibility, misperceptions and disorganized and fragmented thinking in these individuals (see figure 1; Braff, 2010).

Low PPI has not only been reported in schizophrenia patients but also in individuals across the "SCZ spectrum" of disorders, who do not show overt psychosis. Thus, the decreased PPI has been found in individuals with schizotypal personality disorder (STPD) (Cadenhead, 1993), clinically unaffected relatives of SCZ patients (Cadenhead, 2000; Takashi et al., 2010) and individuals scoring high on psychometric measures of psychosis-proneness (Kumari et al., 2008). In addition, PPF deficits have been found in SCZ patients and their siblings (Dawson et al., 2003 & 2004). The findings that PPI and PPF are heritable, support the use of startle response as a potential biological marker of deficient gating mechanisms in SCZ spectrum disorders, and moreover, make it valid candidates as endophenotypic markers in genetic studies of SCZ spectrum disorders (Braff & Freedman, 2002; Braff, 2010 & 2011; Campanella et al., 2009). All common complex mental disorders like SCZ are multifactorial (genetic, environmental and social), -and polygenetic, which makes identification of disease-promoting genes difficult. Therefore, the endophenotype approach was proposed to facilitate the identification of susceptibility genes. Endophenotypes are objectively measurable characteristics which are thought to be closer to the biological processes and especially to the genetic background as they may depend on variation of fewer genes than the more complex disease phenotype, which is based on qualitative and

subjective clinical symptoms (Braff & Light, 2005; Braff et al., 2007; Cannon & Keller, 2006).

1.3 Correlations of PPI with psychiatric symptoms, neurocognitive measures and personality

Within patients with SCZ PPI is reported to show weak negative association with positive or negative symptoms using ratings from general psychiatric symptom scales and relatively stronger negative correlation with thought disorder. PPI also correlates negatively with distractibility and positively with functioning of daily life in SCZ (see Braff et al., 2001).

The occurrence of illusory perceptions, which is a common positive symptom in SCZ, is theorised to result from biases in metacognitive abilities, in particular, the disability to differentiate between self-generated and external sources of information. Projection of an internal state or a personal introspection about the reaction of another to a circumstance may arise in the form of hallucinations, especially auditory hallucinations. Accordingly, research has established a link between the frequency of auditory hallucinations and reduced suppression of irrelevant thoughts and memories. Regarding impaired PPI, it has been demonstrated that it is not related to the presence or absence of voices, but to the feeling of having no control over their occurrence and being unable to dismiss them (Kumari et al., 2008a; Kumar et al., 2009).

As already mentioned, patients diagnosed with PTSD show weaker PPI than normal individuals; and similar findings have been reported for OCD, panic disorder and in college students who scored high on the anxiety trait. These clinical observations suggest a link between emotion processing and PPI (see Franklin et al., 2009; Corr et al., 2002). Low PPI seems to be related inability to gate intrusive internal stimuli such as ruminative thoughts of guilt, failure or anxiety-provoking thoughts (see Dawson et al., 2000). Relatedly, neuroticism, which refers to a trait-like, enhanced emotional response to negative stimuli (Ettinger et al., 2004), may be especially relevant, since there is evidence that it is negatively correlated with PPI (Corr et al., 2002). In addition, neuroticism is known to be correlated with measures of schizotypy,

an association that parallels the increased levels of neuroticism observed in patients with a diagnosis of schizophrenia. Furthermore, a recent twin study demonstrated that the overlap between neuroticism and positive schizotypy has a largely genetic origin, which leads to the hypothesis that schizotypy-PPI associations are mediated by neuroticism (see Corr et al., 2002). Other studies indicated that experimentally induced emotion reduces PPI and in one study only the pure imagination of stressful situations was sufficient to reduce PPI. Accordingly, the trait of neuroticism is similarly related to the anticipation of aversive events: "The fear of not knowing what is coming next may be more salient than the event itself" (see Corr et al., 2002).

Research shows that the influence of gender on PPI may also be relevant, as women demonstrate lower inhibition than men and PPI normally fluctuates across the menstrual cycle. Furthermore, measures of personality are sensitive to gender; i.e. females generally score higher on neuroticism and lower on psychoticism (see Corr et al., 2002). In psychopathology, there is a long tradition of regarding mental illnesses as the polar opposites of normal personality continua. If the viewpoint is taken that personality is long term stability in terms of emotion, cognition and behaviour, then also the viewpoint can be adopted that illnesses of these factors are a result of dysfunction in the systems that regulate them. This perspective is important because it sheds light on the very nature of mental illnesses by studying the underlying factors that influence the system (emotion etc.) in non-clinical healthy populations, which are not variances caused by medication and chronicity (Corr et al., 2013).

1.4 State variables influencing PPI

An individual's gating processes are commonly regarded as showing behavioural plasticity and having both state and trait determinants influenced by a combination of environmental and genetic factors (Braff et al., 2001). As mentioned above (section 1.3), PPI is viewed as being trait or genetically determined and is also correlated with personality traits, like emotional characteristics. Although PPI is considered to be a stable index of individual sensorimotor gating, several factors can affect its measurement, such as medication, cigarette smoking, psychoactive drugs, fatigue,

stress, gender and hormonal status. However, these can be controlled under experimental conditions (see Swerdlow et al., 2008).

1.5 The role of dopamine in schizophrenia and PPI

Dopamine (DA) as a neurotransmitter regulates diverse aspects of brain signalling and behaviour and is involved not only in motor and neuroendocrine processes but also in cognitive processes, especially in the evaluation of the salience of environmental stimuli (Kapur, 2003). An imbalance of the dopaminergic system is associated with a variety of neurological disorders, including OCD, HD, attention deficit hyperactivity disorder (ADHD), SCZ and also with substance abuse (see Pogorelov et al., 2005).

The investigation of the dopaminergic system in the context of SCZ research lead to the DA hypothesis of schizophrenia which proposes that a hyperdopaminergic activity or an elevated sensitivity to this transmitter results in symptoms of the disease. This hypothesis was supported by the findings of positive effects of antipsychotic drugs (based on the blockade of DA receptors) on positive symptoms of SCZ. In contrast, DA agonists, such as amphetamine, could exacerbate symptoms of SCZ in patients, and could moreover provoke certain psychotic reactions in healthy individuals (see Gainetdinov et al., 2001). Additionally, the role of DA was confirmed also at the endophenotype level, because the lack of PPI in SCZ was directly related to dopaminergic activity since healthy subjects on amphetamine display PPI deficits and patients on medication show normal PPI (see Campanella et al., 2009). The activity of DA neurotransmitter system in PFC and striatum was shown to play an important role in the modulation of PPI (De Koning et al., 2014).

Interestingly, extracellular DA is strongly regulated by the dopamine transporter (DAT), which is expressed mainly in the striatum and the nucleus accumbens and is responsible for rapidly removing DA from the synaptic cleft after its release back into the presynaptic membrane (see Banaschewski et al.,2010). If the DAT is not effective, synaptic activation will persist, because DA concentration in the synaptic cleft remains elevated (Jaber et al., 1997).

The gene encoding the DAT is named DAT1 or SLC6A3. Data from animal studies suggest that mice lacking DAT1 display persistently elevated dopaminergic tone and therefore are suitable for evaluating the DA hypothesis, at least in parts (see Powell et al., 2012). Compared to wildtype controls, in homozygous DAT knockout (DAT KO) mice DA remained 100 times longer in the extracellular medium (Yang et al., 2007). These mice show hyperactive and stereotypic behaviour, increased novelty- induced activity, rearing and exploratory behaviour and sleep dysregulation. These mice also appear to have reduced anxiety levels (Carpenter et al., 2012), and additionally they represent some symptoms of SCZ, such as impaired sensorimotor gating (PPI), spatial learning and working memory (see Gainetdinov et al., 2001; Wong et al., 2012). PPI deficits in DAT KO mice have strengthened the hypothesis that dopamine hyperfunction plays an important role in sensorimotor gating and potentially the pathophysiology of schizophrenia (Powell et al., 2012).

1.6 VNTRs of DAT gene

The central role in dopamine neurotransmission and some evidence of activity alteration in schizophrenic patients (Laasko et al., 2000) make DAT1 of great interest as a candidate gene for SCZ (see section 1.5). As well as identifying a candidate gene, it is necessary to identify a polymorphism within this gene – i.e. a region which can exist in multiple forms (alleles) and ideally represents a physical and chemical change to this stretch of DNA sequence. More precisely, the alleles should be functional, so that different alleles confer corresponding differences in biological function. Genetic variation at this locus should, therefore, confer biological individual differences, which in turn should result in behavioural (phenotype) differences between people (see Corr et al., 2013). Polymorphisms in DAT1 genotype may be responsible for the efficiency of DA reuptake.

In DAT1, a polymorphism has been identified involving a variable number of tandem repeats (VNTR) in the 3’ untranslated region (3' UTR). Since it has been suggested to regulate gene expression –this polymorphism may affect DAT activity

(Vanderbergh et al., 1992; Mill et al., 2002). A VNTR is a location in a genome where a short nucleotide sequence is organized as repetitive repeats. They can be present in many genes, and often show variations in length between individuals (Guindalini et al., 2006). The VNTR in the 3´UTR of DAT1 occurs with repeat numbers ranging from 3-11, but the most common alleles contain 9 or 10 repeats of a 40-base-pair sequence (Gadow et al., 2008).

Studies indicate that the DAT1 VNTR is a functional polymorphism, with significantly lower levels of DAT produced from the 10-repeat allele (*10R) than the 9-repeat allele (*9R) (Brookes et al., 2007; Miller & Madras, 2002; Jacobsen et al., 2000; Van Dyck et al., 2005), but this has not been found in all studies (Heinz et al., 2000; Fuke et al., 2001; Mill et al., 2002; Brookes et al., 2006). These inconsistent results may emerge from several factors; i.e. from differences in subject groups including varying diagnostic and racial compositions or different methods of image analysis (see Van Dyck et al., 2005). Another explanation for these divergent findings is that 3´ UTR VNTR may not be an independent source of functional variation in the gene. Rather, another polymorphic site that modifies dopamine transporter function or itself regulates expression of the gene may be in linkage disequilibrium with the ´3 UTR polymorphism (see Jacobsen et al., 2000; Van Dyck et al., 2005; Guindalini et al., 2006).

Some studies have indicated that another DAT1 VNTR, mapped to intron 8, has functional features (Guindalini et al., 2006; Hill et al., 2009; Brookes et al., 2007), whereby the 5- repeat allele (*5R) is thought to be more active than the 6- repeat allele (*6R) (Hill et al., 2009). Accordingly, two studies reported that the *10R of 3'UTR in combination with the *6R allele of intron8 may be associated with ADHD and- it was suggested that DAT1 polymorphisms may influence the hyperactive-impulsive symptoms rather than the inattentives (Asherson et al., 2007; Brookes et al., 2006).

1.7 Association between schizophrenia and common VNTRs of DAT gene

Current research in the genetics of SCZ is driven mainly by the "common disease – common alleles" model (Chakravarti, 1999). This model is based on the assumption that 'affected' individuals are those that lie above some biological

threshold of risk, resulting from the additive and cumulative impact of multiple common small -effect- genetic variants, interacting with environmental exposures (Gottesman & Shields, 1982). Consequently, in this model the causative alleles are not necessary adverse loss-of-function mutations, but instead, are polymorphisms that modulate normal functions. Since SCZ is relatively frequent occurs worldwide, common susceptibility alleles shared across populations are plausible. The model also helps explaining the variable results of linkage studies and weak associations of various candidate genes with SCZ (see McClellan et al., 2007).

Persicio and Maccardi (1997) reported higher frequency of the 3´UTR VNTR homozygotes (genotypes *9R/*9R and *10R/*10R) and reduced frequency of heterozygotes (genotype *9R/*10R) in SCZ patients compared with controls. Similarly Saiz and co-workers reported that *10R/*10R occurred more frequently in schizophrenia patients, but only if the DRD3 genotype was taken into account (Saiz et al., 2010). Furthermore, another study discovered higher occurrence of the *10R /*10R genotype of SLC6A3 in schizoid/avoidant personality disorder (Blum et al., 1997). However, the association between 3´UTR VNTR and schizophrenia was not found in some studies (Codeiro et al., 2004; Persico & Macciardi, 1997; Yeong et al., 2004) but this could also be due to small sample sizes.

1.8 Objective and Hypothesis

As reviewed in the preceding sections, it has been shown that PPI and PPF are reduced in people suffering from SCZ (see Braff et al., 2001 & 2007; Braff, 2010; Takashi et al., 2011; Powell et al., 2016). Accordingly, a key feature of SCZ is the inability to gate out irrelevant sensory input which may result in overstimulation and aberrant information processing and as a result in SCZ patients the perception of the environment is dissimilar to the norm (see Braff et al., 2001). A common research question is if gating deficiency (low PPI) has some influence on the development of psychoses. However, the findings that PPI and PPF are heritable (Cadenhead et al., 1993 & 2000; Dawson et al., 2003 & 2004) make them promising candidates as endophenotypic markers in genetic studies of SCZ spectrum disorders (Braff &

Freedman, 2002; Braff, 2010 & 2011).

The gene of DAT1 is of great interest, as it plays a central role in dopamine neurotransmission which is considered to be disturbed in SCZ. In addition, there are findings regarding alteration of DAT in schizophrenic patients (Laasko et al., 2004), as well as findings of DAT KO mice displaying PPI deficits and some behavioural features of SCZ (Gainetdinov et al., 2006).

The primary goal of the present work was to investigate the potential functional effects of DAT genotype in healthy individuals on PPI of the ASR by identifying risk alleles for "deficient" PPI. We expected risk allele careers having lower PPI. Based on findings from Jacobsen et al. (2000), Van Dyck et al. (2005) and Guindalini et al. (2006), we considered the *10R of 3'UTR and the *6R of intron8 as risk alleles. Since the carriers of *10R are expected to have lower expression of DAT and therefore higher DA availability, we hypothesized that PPI will be lower in these individuals. Vice versa, carriers of the *9R were assumed to have more DAT binding, thereby having a faster reuptake of DA from synaptic gap, which should be associated with stronger PPI compared to the *10R carriers.

2. Methods

2.1 Experimental design

Data were collected at the Social, Cognitive and Affective Neuroscience Unit (SCAN-Unit) at the Faculty of Psychology of the University of Vienna, and at the Laboratory of Cognitive Neuroscience at the Institute of Normal and Pathological Physiology, Slovak Academy of Sciences in Bratislava, Slovakia (INPP SAS). The studies were approved by the Ethical Committee of INPP SAS.

The study had a repeated measures design, in which the dependent variables (PPI; PPF) were measured at five different prepulse lead intervals (ISIs), inducing either PPI or PPF. The independent variables were the genotypes at the 3´UTR and the intron8 loci of DAT gene (see section 2.4).

2.2 Study participants

For these studies, young healthy men were recruited via online announcements on job pages and in social networks. Women were not included in order to prevent increased data variability (e.g., due to hormonal status). All participants had to fulfil the initial including criteria: age between 18 and 45 years, right- handedness, absence of any auditory deficits and normal or corrected-to-normal visual acuity. Persons who responded on our announcement completed an online survey composed of the 12-item General Health Questionnaire (GHQ-12; Goldberg & Williams, 1988; Sarkova et al., 2006)), an anamnestic questionnaire including items of the M.I.N.I. International Neuropsychiatric Interview (M.I.N.I.; Sheehan et al., 1998), as well as a screening questionnaire about their use of psychoactive substances. Exclusion criteria were past or current mental or neurological illnesses, such as schizophrenia-spectrum-disorder, epilepsy or mood disorder, affecting individuals or their first-degree relatives and the abuse of psychoactive substances, like coffee, cigarette smoking, alcohol and illegal drugs, as well as long term pharmacological treatment. Furthermore, individuals who exhibited excessive spontaneous and voluntary blinks -but no visible startle responses (non-responders) were subsequently excluded from the data set. To minimize the effects of substances on PPI and startle, test subjects were directed to avoid caffeinated beverages one hour before the start of the test sessions, as well as medication intake and alcohol consumption within 24 hours before the measurement. In addition, they were instructed to arrive well-rested and not to wear contact lenses on the test day. For their study participation test subjects received a monetary remuneration in the amount of 16 Euros.

Initially, the data set contained the information of 241 test subjects. Seventy-eight subjects were excluded due to missing values. Furthermore, three subjects were excluded as carries of uncommon alleles (other than *9R or *10R at 3´UTR and *5R or *6R at intron8 locus). Further 21 subjects were excluded since they showed no PPI at ISIs of 30, 60 or 120ms. The final test sample thus consisted of 139 male participants (mean age ± SD= 24.8 ± 4.4 years).

2.3 Procedure

Upon arrival at the laboratory participants received detailed information about the study procedure and gave written informed consent. Then, participants were asked to provide an urine sample for the toxicological screenings of cotinine (i.e. nicotine consumption; Diagnostik Nord, Schwerin, Germany) and multiple drugs, such as cocaine, amphetamines, methamphetamine, MDMA, benzodiazepines, barbiturates, opiates, LSD and cannabis (DIPRO med GmbH, Wegelsdorf, Austria). Subjects showing positive results were excluded. Furthermore, subjects underwent a structured interview focused at their consumption of psychoactive substances, including caffeine, alcohol, nicotine, cannabinoids, methamphetamine, MDMA (exstasy), cocaine, amphetamines, opiates, hallucinogens, organic solvents and psychotropic drugs. For each substance, test subjects were asked to report frequency within the last six months, recency and amount of consumption. In addition, vague and distinct responses in the online questionnaire (see section 2.2) were discussed to clarify and ensure the participants' physiological and psychological health. To identify and exclude individuals with hearing impairments, participants underwent an audiometric test which presented white noise at five increasing intensities, whereby all subjects confirmed to hear noise of at least 35 dB.

Then, subjects were prepared for EMG recording (see section 2.6) and were asked to remove all metal objects and to switch their mobile phones into flight mode. To stabilize eye movements, a white linen sheet was put approximately 15 cm in front of the participants' faces and subjects were instructed to keep still with their eyes open and to avoid unnecessary blinking. Subsequently, participants underwent the PPI test-session (see section 2.5).

Finally, subjects completed the Schizotypal Personality Questionnaire (SPQ; Raine, 1991), the NEO Five Factor Inventory (NEO-FFI; Costa & McCrae, 1992 & NEO-PI-R; Ostendorf & Angleitner, 2003) and the Edinburgh Handiness Inventory (Oldfield, 1971).

Additionally, a buccal smear was taken for genetic analysis. The test session was duration of approximately two hours.

2.4 Genotyping and group assessment

DNA was extracted from buccal cells collected using swabs. Genotyping was carried out at the Institute of Molecular Biomedicine, Medical Faculty, Comenius University in Bratislava, Slovakia.

Genotype was assessed for DAT loci 3'UTR and intron8. The genotypes, more precisely both alleles of each DAT1 locus (3´UTR and intron8) were used to generate group-assignments as the independent variables. First, the subjects were assigned to groups dependent on both alleles in both gene loci. As depicted in table 1, in some subgroups there was insufficient number of subjects, thus it was not possible to conduct an analysis including both genotypes in a single analysis, i.e. a two-factorial ANOVA. Therefore, each DAT1 locus was analyzed separately. For each locus, the subjects were assigned into three groups. Group 0 consisted of homozygous individuals for the allele which was classified as non-risk- allele (*9R in 3´UTR/ *5R in intron 8). Group 1 included heterozygous individuals and group 2 homozygous individuals for the risk- allele (*10R in 3´UTR/ *6R in intron 8). Table 2 shows the group sizes of the subgroups of the allele assignments of each DAT1 locus. In both group- assignments group 0 was of small sample size compared to group 1 and 2. Therefore, groups 0 were included into groups 1. All analyses examine the effect on the dependent variable in these two groups of each polymorphism, by which high-risk individuals (group 2) were compared with low-risk individuals (group 1).

2.5 Auditory stimulation

Sound stimuli were delivered via audiometric headphones ER-2 (Etymotic Research, Groove Village, IL, USA).

The experimental session started with a 3-minute acclimation period to 55dB white noise that was present as background noise for the entire session in order to minimize the impact of eventual environmental noises. Subsequently PPI/PPF paradigm pulse stimuli (white noise, 105dB, 40ms; with instantaneous rise/fall) were either presented alone or following a prepulse (white noise, 75 dB, 20ms).There was a set of ISIs (prepulse onset to pulse onset) to elicit either PPI (30ms, 60ms and 120ms) or PPF

(2000ms and 4000ms). Overall, the auditory stimulation consisted of 69 trials in three blocks. Block 1 consisted of 5 successive PA trials. Block 2 was a combination of randomized sequences of PA and pulse to pulse stimuli with the end result of 10 PA trials and 10 trials of each ISI. Block 3 was composed of 4 PA trails. The Inter-trial intervals (ITI) were randomized within an interval ranging from 10 to 20 s. The PPI session lasted approximately 22 min.

2.6 EMG recording

The magnitude of startle- induced eye blinks was measured by EMG. In preparation of the EMG recording, the skin beneath the electrodes was disinfected with ethyl alcohol and scarified with a sterile needle. Then electrodes were attached using electrode adaptors and double-sided adhesive collars and were filled with EMG conductive gel (Electro-Cap International Inc., USA).

To record EMG, two reusable sintered Ag/AgCl ring electrodes (EASYCAP, Herrsching, Germany) were fixed beneath each eye over the left and right orbicularis oculi muscle. One electrode was placed below the lower eye-lid in unison with the pupil in forward stare while the second one was placed approximately two centimeters laterally to the first one. The ground electrode was attached to the center of the forehead.

Electrode impedances were measured and kept below 3 kΩ. The EMG signal was recorded using NeXus-10 (Mind Media B.V.) with a sampling rate of 2048 Hz.

2.7 EMG processing and analysis

EMG waveforms were band-pass filtered in the range 28-800 Hz. Epochs from -100 to 400 ms with respect to startle stimulus onset were created, visually inspected and the epochs containing artifacts were removed. The magnitude of the startle response was measured as the peak value of rectified EMG signal within a time window 21-150 ms following stimulus onset. Prepulse inhibition (or facilitation) was

calculated according to the formula (1-PP/PA)*100%, where PP and PA denote mean startle amplitude in prepulse-pulse and pulse alone trials respectively. Values for the left and right eye were averaged.

2.8 Statistical analysis

For all statistical analyses Statistical Package for the Social Sciences (SPSS, IBM, USA), version 23.0, was used. Descriptive Statistics and boxplots were used to explore startle reflex data and identify outlying values.

Individuals who displayed negative values of relative startle modulation at ISIs of 30ms, 60ms or 120ms, which indicates facilitation of startle instead of anticipated inhibition, were excluded from the analysis (see section 2.2). In order to reduce the effect of outliers in the startle parameters regarding ISIs of 2000ms and 4000ms, the PPF- values were winsorized within groups before statistical analyses. Observations lower/higher than the 25th/75th percentile minus/plus 1.5 times the interquartile range of the group were replaced with the minimum/maximum value within the respective interval (replaced values: 6,47%).

The effects of genotype on PPI/PPF were analyzed for each ISI separately using independent samples t-test. In case of significant deviation from normality Mann-Whitney U-test was used. For each polymorphism, the two genotype groups were compared using t-tests with PPI-values within ISI of 30ms and PPF- values within ISI of 2000ms. Mann-Whitney U-tests were used for PPI-values within ISIs of 60 and 120ms, as well as for PPF-values within ISI4000.

To identify covariates, Spearman correlation coefficients were calculated between PPI/PPF values and the scores of NEO-FFI subscales, the SPQ subscales and total score. Analysis of covariance (ANCOVA) was used to control for the effects of these covariates on PPI/PPF.

For all statistical tests, the α-level for statistical significance was set at .05.

3. Results

Table 3 contains descriptive statistics of PPI and PPF at different ISIs and relevant trait- variables. According to the literature (Braff et al., 2001), PPI varies with the ISI and is weaker for ISIs of 30ms than ISIs of 60 and 120ms.

Statistical analyses of the effect of each polymorphism (3´UTR- and intron8) on PPI at different ISIs (30ms, 60ms, 120ms) was performed with unpaired t-tests and Mann-Whitney U-tests and revealed no significant results (Table 4). Table 5 shows the results of the statistical analyses of PPF at different ISIs (2000ms, 4000ms). Again, no significant effects of genotype were revealed.

To identify covariates, Spearman correlation coefficients were calculated between PPI/PPF values and the scores of NEO-FFI subscales, the SPQ subscales and total score. There were no significant correlations between PPI/PPF values and the SPQ total score. However, the SPQ domain “unusual perceptual experiences” correlated significantly with PPI at ISI of 30ms, $r_s(136) = 0.18$, $p < 0.05$. Based on literature suggesting a link between PPI and illusionary perceptions (Kumari et al., 2008a; Kumar et al., 2009), the effects of the DAT genotype on PPI at ISI of 30ms were analyzed using one- way – ANCOVA with “unusual perceptual experiences” as a covariate and revealed no significant results (table 6).

Regarding the questionnaire NEO-FFI there was a significant negative correlation between PPI at ISI of 30ms and neuroticism, $r_s(134) = 0.25$, $p < 0.05$. Based on significant correlation between PPI30 and neuroticism and on authors suggesting neuroticism as a mediator between schizotypy and PPI (Corr et al., 2002), further analyses were conducted. Therefore, statistical analyses of experimental effect of each polymorphism on PPI30 were conducted using a one way- ANCOVA with neuroticism as a covariate (Table 7). There was a significant effect of 3´UTR genotype on PPI30 after controlling for neuroticism, $F(1, 118) = 5.41$, $p < 0.05$, a significant effect of neuroticism on PPI30, $F(1,118) = 13.34$, $p < 0.05$, and a significant interaction, $F(1,118) = 6.08$, $p < 0.05$, between 3´UTR genotype and neuroticism. There was no significant effect of intron8 genotype on PPI30 after controlling for neuroticism, $F(1, 131) = 0.39$, $p > 0.05$. The interaction between the covariate neuroticism and the 3´UTR genotype groups was significant, indicating inhomogeneity of regression slopes.

This raises concern about the main effect of genotype revealed by the ANCOVA, as the effects of the polymorphism at 3´UTR are dependent on neuroticism. Therefore, the sample was split by median of neuroticism in two groups: subjects were assigned to either low-neuroticism group (N= 61) or high-neuroticism group (N= 78). As depicted in figure 2, PPI at ISI of 30ms was reduced in the high- neuroticism group compared to the low- neuroticism group. The analysis of effect of 3´UTR genotype on PPI30 was rerun separately in these two groups using unpaired t-tests (Table 8). Risk allele carriers who scored high in neuroticism had lowest PPI, but the group comparisons yielded no statistically significant effects.

4. Discussion

This study examined the effects of common 3´UTR and intron 8 polymorphisms of DAT gene on PPI and PPF at several ISIs. Findings are consistent with previous research that PPI varies with the ISI and is weaker for ISIs of 30ms than ISIs of 60 and 120ms. In contrast to our expectations, some participants, who as a result had been excluded from the analysis, displayed an augmentation of startle reflex at ISIs of 30, 60 and 120ms where the literature states an inhibition of startle (Braff et al., 2001). It could indicate that these subjects were of high arousal while testing, but it is still unclear how such an effect could be explained.

As mentioned in the introductory sections, SCZ patients, their unaffected relatives (Cadenhead et al., 2000; Kumari et al., 2005; Takahashi et al., 2010) and patients with schizotypal personality disorder (Cadenhead et al., 1993 & 2000) display PPI deficits. In addition, other studies suggest a negative relationship between schizotypal traits and PPI in the general population (Kumari et al., 2008; Corr et al., 2013), providing evidence for a psychopathology-continuum. In our sample we found no significant relationship between SPQ-total and PPI- or PPF-values at different ISIs, but a significant negative correlation between PPI30 and the subdomain unusual perceptual experiences of SPQ. Accordingly, the occurrence of illusionary perceptions is a common symptom in SCZ and previous studies demonstrated that low PPI is related to having no control over their manifestation.

Also in concordance with previous research there was a relationship between PPI and trait neuroticism of NEO-FFI (Corr et al., 2002). The connection between PPI and emotion processing is theorised to arise from PPI deficits exhibiting an inability to gate intrusive internal stimuli such as ruminative thoughts of guilt, failure or anxiety-provoking thoughts (Dawson et al., 2000). In our sample, there was a significant effect of 3´UTR polymorphism on PPI30, controlling for covariate neuroticism. The result could not be considered due to a significant interaction between 3´UTR genotype and neuroticism. After splitting the data set in a low-neuroticism and high-neuroticism group, the analysis was rerun separately in both groups using unpaired t-tests which revealed no significant results. Further analysis using ANCOVA with neuroticism as a covariate indicated that the subjects were not sufficiently assigned to neuroticism-groups. Due to the small sample size an assignment of three neuroticism- groups would have been questionable.

The lack of participants was also responsible for the small sample sizes of the subgroups of allelic group-assignment consisting of both 3´UTR and intron 8 genotypes. Therefore, it was not possible to conduct an analysis which combined both factors.
Moreover, the study was planned as repeated measures design, but as the residuals of PPI- and PPF- values within the groups of 3´UTR- and intron8- group-assignment were not normally distributed, it was not possible to operate repeated measures analyses, and therefore potential significant results could not have been attained.

5. Conclusion

The present study found no considerable influence of 3´UTR and/or intron8 polymorphisms of DAT1 on PPI or PPF. Indeed, results showed a significant influence of the polymorphism in the 3´UTR genotype on PPI30 after controlling for neuroticism, but also a significant interaction between 3´UTR and neuroticism. Analysis of the genotype effect separately for participants with high and low neuroticism yielded no statistically significant results. Thus, the mutual relationship between 3´UTR genotype, neuroticism and PPI remains unspecified and its exploration requires further analyses on lager samples.

References

Asherson, P., Brookes, K., Franke, B., Chen, W., Gill, M., Ebstein, R. P., Buitelaar, J., Banaschewski, T., Sonuga-Barke, E., Eisenberg, J., Manor, I., Miranda, A., Oades, R. D., Roeyers, H., Rothenberger, A., Sergeant, J., Steinhausen, H. C., Faraone, S. V. (2007). Brief Report: Confirmation That a Specific Haplotype of the Dopamine Transporter Gene Is Associated With Combined-Type ADHD. *American Journal of Psychiatry*, 164(4), 674-7.

Banaschewski, T., Becker, K., Scherag, S., Franke, B., Coghill, D. (2010). Molecular genetics of attention-deficit/hyperactivity disorder: an overview. *European Child & Adolescent Psychiatry*, 19(3), 237–257.

Bitsios, P., Giakoumaki, S. G. (2005). Relationship of prepulse inhibition of the startle reflex to attentional and executive mechanisms in man. *International Journal of Psychophysiology*, 55, 229–241.

Blum, K., Braverman, E. R., Wu, S. et al. (1997). Association of polymorphisms of dopamine D2 receptor (DRD2), and dopamine transporter (DAT1) genes with schizoid/avoidant behaviors (SAB). *Molecular Psychiatry*, 2, 239-246.

Blumenthal, T. D., Schicatano, E. J., Chapman, J. G., Norris, C. M., Ergenzinger, E. R. (1996) Prepulse effects on magnitude estimation of startle-eliciting stimuli and startle responses. *Perception & Psychophysics*, 58, 73–80.

Blumenthal, T. D., Cuthbert, B. N., Filion, D. L., Hackley, S., Lipp, O. V., & van Boxtel, A. (2005). Committee report: Guidelines for human startle eyeblink electromyographic studies. *Psychophysiology*, *42*(1), 1–15.

Braff, D. L., & Geyer, M. A. (1990). Sensorimotor gating and schizophrenia: Human and animal model studies. *Archives of General Psychiatry*, *47*(2), 181–188.

Braff, D. L., Geyer, M. A. & Swerdlow, N. R. (2001). Human studies of prepulse inhibition of startle: Normal subjects, patient groups, and pharmacological studies. *Psychopharmacology*, *156*(2-3), 234–258.

Braff, D. L., Freedman, R. (2002). Endophenotypes in studies of the genetics of schizophrenia. *Neuropsychopharmacology*, 69, 703–716.

Braff, D. L. & Geyer, M. A. (2005). The use of neurophysiological endophenotypes to understand the genetic basis of schizophrenia. *Dialogues in Clinical Neurosciences*, 7(2), 125–135.

Braff, D. L., Freedman, R., Schork, N. J., Gottesman, II. (2007). Deconstructing schizophrenia: an overview of the use of endophenotypes in order to understand a complex disorder. *Schizophrenia Bulletin*, 33, 21–32.

Braff, D. L. (2010). Prepulse inhibition of the startle reflex: A window on the brain in schizophrenia. In N. R. Swerdlow (Ed.), *Behavioral neurobiology of schizophrenia and its treatment* (pp. 349–371). Berlin Heidelberg: Springer-Verlag.

Braff, D. L. (2011). Gating in schizophrenia: from genes to cognition (to real world function?). *Biological Psychiatry*, 69, 395–396.

Brookes, K. J., Mill, J., Guindalini, C., Curran, S., Xu, X., Knight, J., Chen, C., Huang, Y, Sethna, V., Taylor, E., Chen, W., Breen, G., Asherson, P. (2006). A Common Haplotype of the Dopamine Transporter Gene Associated With Attention-Deficit/Hyperactivity Disorder and Interacting With Maternal Use of Alcohol During Pregnancy. *Archive of Genetics and Psychiatry*, 63, 1, 74-81.

Brookes, K. J., Neale, B. M., Sugedon, K., Khan, N., Asherson, P., D´Souza, U. M. (2007). Relationship Between VNTR Polymorphisms of the Human Dopamine Transporter Gene and Expression in Post-Mortem Midbrain Tissue. *American*

Journal of Medical Genetics, 144(B), 1070-1078.

Cadenhead, K. S., Geyer, M. A., & Braff, L. D. (1993). Impaired startle prepulse inhibition and habituation in patients with schizotypal personality disorder. *American Journal of Psychiatry*, *150*(12), 1862–1867.

Cadenhead, K. S., Swerdlow, N. R., Shafer, K. M., Diaz, M., & Braff, D. L. (2000). Modulation of the startle response and startle laterality in relatives of schizophrenic patients and in subjects with schizotypal personality disorder: Evidence of inhibitory deficits. *American Journal of Psychiatry*, *157*(10), 1660–1668.

Campanella, S., Guerit, J. M. (2009). How clinical neurophysiology may contribute to the understanding of a psychiatric disease such as schizophrenia. *Clinical Neurophysiology*, 39(1), 31-39.

Cannon, T. D. & Keller, M. C. (2006). Endophenotypes in the genetic analyses of mental disorders. *Annual Review of Clinical Psychology*, 2, 267-90.

Carpenter, A. C., Saborido, T. P., Stanwood, G. D. (2012). Development of hyperactivity and anxiety responses in dopamine transporter- deficient mice. *Developmental Neurosciences*, 34, 250–257.

Chakravati, A. (1999). Population genetics – making sense out of sequence. *Natural Genetics*, 21(1), 56-60.

Cordeiro, Q., Talkowski, M., Wood, J. , Ikenaga, E., Vallada, H. (2004). Lack of association between VNTR polymorphism of dopamine transporter gene (SLC6A3) and schizophrenia in a Brazilian sample. *Arquivos de Neuro-Psiquiatria*, 62, 973–976.

Corr, P.J., Tynan, A., Kumari, V. (2002). Personality correlates of prepulse inhibition of

the startle reflex at three lead intervals. *Journal of Psychophysiology*, 16, 82–91.

Costa, P. T., & McCrae, R. R. (1992). *NEO PI-R:* Revised NEO Personality Inventory and NEO Five-Factor Inventory (NEO-FFI). *Psychological Assessment Resources.*

Dawson, M.E., Hazlett, E.A., Filion, D.L., Nuechterlein, K.H., Schell, A.M. (1993). Attention and schizophrenia: Impaired modulation of the startle reflex. *Journal of Abnormal Psychology*, 102, 633–641.

Dawson, M.E., Schell, A.M, Hazlett, E.A., Nuechterlein, K.H., Filion, D.L. (2000). On the clinical and cognitive meaning of impaired sensorimotor gating in schizophrenia. *Psychiatry Research*, 96, 187–197.

De Koning, M. B., Bloemen, O. J., Van Duin, E. D., Booij, J., Abel, K. M., De Haan, L., Linszen, D. H., Van Amelsvoort, T. A. (2014). Pre-pulse inhibition and striatal dopamine in subjects at an ultra-high risk for psychosis. *Journal of Psychopharmacology*, 28(6), 553-60.

Ettinger, U., Kumari, V., Crawford, T. J., Flak, V., Sharma, T., Davis, R. E., Corr, P. J. (2005). Saccadic eye movements, schizotypy, and the role of neuroticism. *Biological Psychology*, 68, 61–78.

Franklin, J. C., Bowker, K. B., Blumenthal, T. D. (2009). Anxiety and prepulse inhibition of acoustic startle in a normative sample: The importance of signal-to-noise ratio. *Personality and Individual Differences*, 46(3), 369–373.

Fuke, S., Suo, S., Takahashi, N., Koike, H., Sasagawa, N., Ishiura, S. (2001). The VNTR polymorphism of the human dopamine transporter (DAT1) gene affects gene expression. *Pharmacogenomics Journal*, 1, 152–156.

Gadow, K. D., Roohi, J., DeVincent, C. J., Hatchwell, E. (2008). Association of ADHD, tics, and anxiety with dopamine transporter (*DAT1*) genotype in autism spectrum

disorder. *Journal of Child Psychology and Psychiatry*, 49(12), 1331–1338.

Gainetdinov, R. R., Mohn, A. R., Caron, M. C. (2001). Genetic animal models: focus on schizophrenia. *Trends in Neurosciences*, 24, 527-533.

Geyer, M. A. (2006). The family of sensorimotor gating disorders: comorbidities or diagnostic overlaps? *Neurotoxity Research*, 10(3-4), 211-20.

Gottesman, I. I. & Shields, J. (1982). Schizophrenia: The Epigenetic Puzzle. *Cambridge University Press.*

Graham, F. K. (1975). The more or less startling effects of weak prestimulation. *Psychophysiology*, *12*(3), 238–248.

Guindalini, C., Howard, M., Haddley, K., Laranjeira, R., Collier, D., Ammar, N., Craig, I., O'Gara, C., Bubb, V. J., Greenwood, T., et al. (2006). A dopamine transporter gene functional variant associated with cocaine abuse in a Brazilian sample. *Proceedings of the National Academy of Science*, 103(12), 4552–455.

Heinz, A., Goldman, D., Jones, D. W., Palmour, R., Hommer, D., Gorey, J.G., Lee, K. S., Linnoila, M., Weinberger, D. R. (2000). Genotype influences in vivo dopamine transporter availability in human striatum. *Neuropsychopharmacology Journal*, 22, 133–139.

Jaber, M., Jones, S., Giros, B., Caron, M. G. (1997). The dopamine transporter: a crucial component regulating dopamine transmission. *Movement Disorders*, 12, 629-633.

Jacobsen, L. K., Staley, J. K., Zoghbi, S. S., Seibyl, J. P., Kosten, T. R., Innis, R. B., Gelernter, J. (2000). Prediction of Dopamine Transporter Binding Availability by Genotype: A Preliminary Report. *American Journal of Psychiatry*, 157, 10, 1700-1703.

Kapur, S. (2003). Psychosis as a State of Aberrant Salience: A Framework Linking Biology, Phenomenology, and Pharmacology in Schizophrenia. *American Journal of Psychiatry*, 160 (1), 13-23.

Kessler, R. C.; Adler, L.; Ames, M., Demler, O., Faraone, S., Hiripi, E., Howes, M. J., Jin, R., Secnik, K., Spencer, T., Ustun, T. B., Walters E. E. (2005). "The World Health Organization Adult ADHD Self-Report Scale (ASRS): a short screening scale for use in the general population". *Psychological Medicine,* 35(2), 245–56.

Kohl, S., Heekeren, K., Klosterkötter, J., & Kuhn, J. (2013). Prepulse inhibition in psychiatric disorders – Apart from schizophrenia. *Journal of Psychiatric Research*, *47*(4), 445–452.

Kumari, V., Antonova, E., and Geyer, M. A. (2008). Prepulse inhibition and “psychosis-proneness” in healthy individuals: an fMRI study. *European Psychiatry,* 23, 274–280.

Kumari, V., Peters, E. R., Fannon, D., Premkumar, P., Aasen, I., Cooke, M. A., Anilkumar, A. P., Kuipers, E. (2008a). Uncontrollable voices and their relationship to gating deficits in schizophrenia. *Schizophrenia Research*, 101(1-3), 185–194.

Kumar, S., Soren, S., Chaudhury, S. (2009). Hallucinations: Etiology and clinical implications. *Industrial Psychiatry Journal,* 18(2), 119-126.

Laakso, A., Vilkman, H., Alakare, B., Haaparanta, M., Bergman, J., Solin, O. *et al* (2000). Striatal dopamine transporter binding in neuroleptic-naive patients with schizophrenia studied with positron emission tomography. *American Journal of Psychiatry*, 157, 269–271.

Light, G. A. & Braff, D. L. (1999). Human and animal studies of schizophrenia-related gating deficits. *Current Psychiatry Reports*, *1*(1), 31–40.

Maher, B.A. (1974). Delusional thinking and perceptual disorder. *Journal of Individual Psychology*, 30, 98–113.

McClellan, J. M., Susser, E., King M. C. (2007). Schizophrenia: a common disease caused by multiple rare alleles. *The British Journal of Psychiatry*, 190(3), 194-199.

Mill, J., Asherson, P., Browes, C., D'Souza, U., Craig, I. (2002). Expression of the dopamine transporter gene is regulated by the 3' UTR VNTR: Evidence from brain and lymphocytes using quantitative RT-PCR. *American Journal of Medical Genetics*, 114, 8, 975-9.

Miller, G. M., Madras, B. K. (2002). Polymorphisms in the 3'-untranslated region of human and monkey dopamine transporter genes affect reporter gene expression. *Molecular Psychiatry*, 7, 44–5.

Oldfield, R. C. (1971). The assessment and analysis of handedness: the Edinburgh inventory. *Neuropsychologia*, 9(1), 97-113.

Ostendorf, F. & Angleitner, A. (2003). NEO-Persönlichkeitsinventar nach Costa und McCrae, Revidierte Fassung (NEO-PI-R). Manual. Göttingen: Hogrefe.

Patton, J. H., Stanford, M. S., Barratt, E. S. (1995). Factor structure of the Barratt Impulsiveness scale. *Journal of Clinical Psychology*, 51, 768–764.

Persicio, A. M., Macciardi, F. (1997). Genotypic association between dopamine transporter gene polymorphisms and schizophrenia. *American Journal of Medical Genetics*, 74, 53-57.

Pogorelov, V. M., Rodriguiz, R. M., Insco, M. L., Caron, M. G., Wetsel, W. C. (2005). Novelty Seeking and Stereotypic Activation of Behavior in Mice with Disruption of the *Dat1* Gene. *Neuropsychopharmacology*, 30, 1818–1831.

Powell, S. B., Weber, M., Geyer, M. A. (2012). Genetic Models of Sensorimotor Gating: Relevance to Neuropsychiatric Disorders. *Current Topics in Behavioral Neurosciences*, 12, 251–318.

Powell, S. B., Swerdlow, N. R. (2016). Social Isolation Rearing and Sensorimotor Gating in Rat Models of Relevance to Schizophrenia: What We Know, and What We Do Not Know. Handbook of *Behavioral Neuroscience*, 23(9), 125-138.

Raine, A. (1991). The SPQ: A scale for the assessment of Schizotypal Personality based on *DSM-III-R* criteria. *Schizophrenia Bulletin*, 17(4), 555-564.

Saiz, P. A., Garcia-Ortilla, M. P., Arango, C., Morales, B., Arias, B., Corcoran, P., Fernandez, J. M., Alvarez, V., Coto, E., Bascaran, M. T., Bousono, M., Fananas, L., Bobes, J. (2010). Genetic polymorphisms in the dopamine-2 receptor (DRD2), dopamine-3 receptor (DRD3), and dopamine transporter (SLC6A3) genes in schizophrenia: Data from an association study. *Progress in Neuro-Psychopharmacology and Biological Psychiatry,* 34(1), 26-31.

Swerdlow, N. R., Fillion, D., Geyer, M. A., Braff, D. L (1995). "Normal" personality correlates of sensorimotor, cognitive, and visuospatial gating. *Biological Psychiatry*, 37, 286-299.

Swerdlow, N. R., Geyer, M. A. (1998). Using an animal model of deficient sensorimotor gating to study the pathophysiology and new treatments of schizophrenia. *Schizophrenia Bulletin*, 24(2), 285-301.

Swerdlow, N. R., Weber, M., Qu, Y., Light, G. A., Braff, D. L. (2008). Realistic expectations of prepulse inhibition in translational models for schizophrenia research. *Psychopharmacology*, 199(3), 331-338.

Takahashi, H., Iwase, M., Canuet, L., Yasuda, Y., Ohi, K., Fukumoto, M., Ilke, N., Nakahachi, T., Ikezawa, K., Azechi, M., Kurimoto, R., Ishii, R., Yoshida, T., Kazui,

H., Hashimoto, R., Takeda, M. (2010). Relationship between prepulse inhibition of acoustic startle response and schizotypy in healthy Japanese subjects. *Psychophysiology*, *47*(5), 831–837.

Takashi, H., Hashimoto, R., Iwase, M., Ishii, R., Kamio, Y., Takeda, M. (2011). Prepulse Inhibition of Startle Response: Recent Advances in Human Studies of Psychiatric Disease. *Clinical Psychopharmacology and Neuroscience*, 9(3), 102–110.

Van Dyck, C. H., Malison, R. T., Jacobsen, L. K., Seibyl, J. P., Staley, J. K., Laruelle, M., Baldwin, R. M., Innis, R. B., Gelernter, J. (2005). Increased Dopamine Transporter Availability Associated with the 9-Repeat Allele of the SLC6A3 Gene. *Journal of Nuclear Medicine,* 5, 745-751.

Vanderbergh, D. J., Persico, A. M., Hawkins, A. L., Griffin, C. A. , Li, X., Jabs, E. W. (1992). Human Dopamine Transporter Gene (DAT1) Maps to Chromosome 5pl 5.3 and Displays a VNTR. *Genomics*, 14(4), 1104-6.

Wong, P., Chang, C. C. R., Marx, C. E., Caron, M. G., Wetsel, W. C., Zhang, X. (2012). Pregnenolone Rescues Schizophrenia-Like Behavior in Dopamine Transporter Knockout Mice. *Trends in Neurosciences*, 24(9), 527-33.

Wynn, J. K., Dawson, M. E., Schell, A. M., McGee, M., Salveson, D., Green, M. F. (2003). Prepulse facilitation deficits in a passive task in schizophrenia patients and their siblings. *Schizophrenia Research*, 60(1), p. 264.

Wynn, J. K., Dawson, M. E, Schell, A. M., McGee, M., Salveson, D., Green, M. F. (2004). Prepulse facilitation and prepulse inhibition in schizophrenia patients and their unaffected siblings. Biological Psychiatry, 55(5), 518-523.

Wynn, J. K., Sergi, M. J., Dawson, M. E., Schell, A. M., Green, M. F. (2005). Sensorimotor gating, orienting and social perception in schizophrenia. *Schizophrenia Research*, 73(2), 319-325.

Yang, B., Chan, R. C. K., Jing, J., Li, T., Sham, P., Chen, R. Y. L. (2007). A Meta-Analysis of association studies between the 10-repeat allele of a VNTR polymorphism in the 3´UTR of dopamine transporter gene and Attention Deficit Hyperactivity Disorder. *American Journal of Medical Genetics*, 144(B), 541-550.

Yeong, S. H., Joo, E. J., Ahn, Y. M., Kim, Y. S. (2004). Association study of dopamine transporter gene and schizophrenia in Korean population using multiple single nucleotide polymorphism markers. *Progress in Neuropsychopharmacology and Biological Psychiatry*, 28, 6, 975-983.

Figures

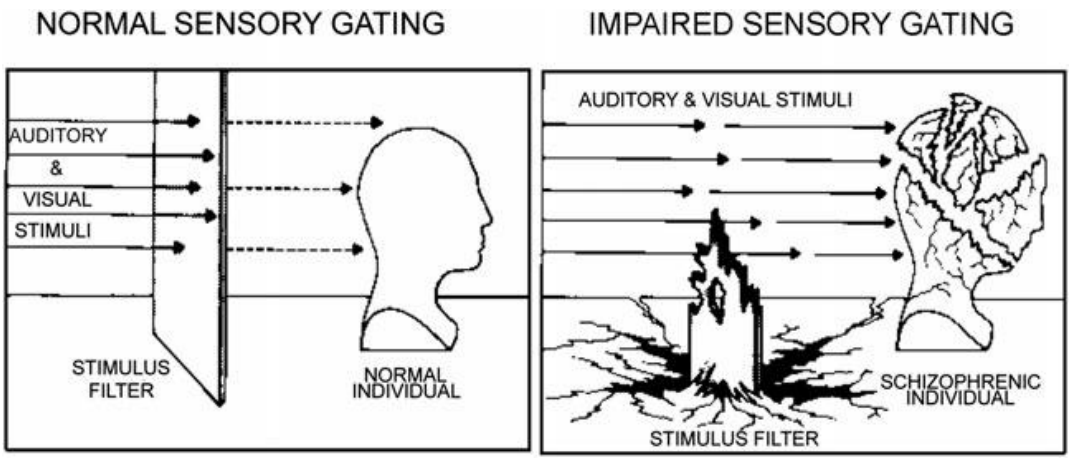

Figure 1: Illustration of impaired sensory gating: How a loss of "gating" of environmental stimuli is associated with cognitive fragmentation (Braff, 2010)

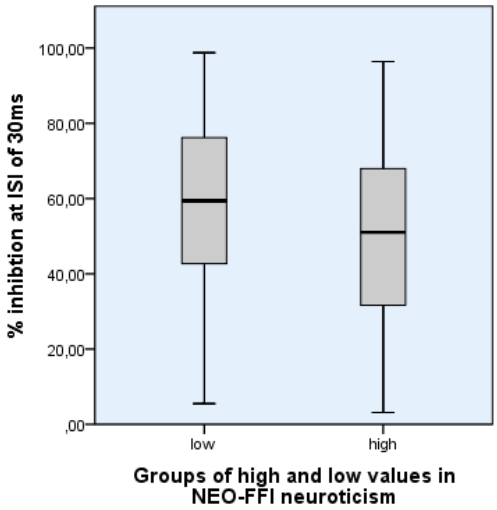

Figure 2: PPI values at ISI of 30ms in Neuroticism- low and Neuroticism- high- group

Tables

Table 1: Number of subjects for each 3'UTR/Int8 DAT allelic combination

		Int8		
		5/5	**5/6**	**6/6**
3´UTR	**9/9**	6	9	1
	9/10	3	34	16
	10/10	1	3	51

N= 124

Table 2: Number of subjects in DAT genotype groups for 3'UTR and Int8 locus

	3´UTR	**Int8**
0: homozygote non-risk	15	10
1: heterozygote	55	53
2: homozygote risk	55	75

3´UTR: N= 125; Intron8: N= 138

Table 3: Descriptive Statistics of PPI/PPF and trait-variables

	Mean	**SD**	**Min**	**Max**	**N**
PPI30	54.42	22.32	3.14	58.75	139
PPI60	74.86	15.41	30.56	98.95	139
PPI120	73.16	15.72	28.64	97.28	139
PPI2000	5.55	28.27	-82.28	53.77	139
PPI4000	-5.03	32.69	-116.37	52.76	139
Neo-FFI- Neuroticism	17.75	8.25	2	42	136
SPQ- Total score	18.53	11.50	0	53	138
SPQ- Unusual exp.	1.33	1.74	0	7	138
Age	24.8	4.42	19	40	139

Table 4: Statistical analysis of the effects of the DAT genotype on PPI at different ISIs

		Test	Test-statistic	Sign.*	Mean (1)	Mean (2)
3´UTR	**PPI30**	t-test	t= - 0.52	0.59	53.41	55.56
	PPI60	M-W test	z= - 0.34	0.73	62.01	64.25
	PPI120	M-W test	z= - 0.04	0.97	62.89	63.15
Int8	**PPI30**	t-test	t= 0.62	0.53	55.70	53.31
	PPI60	M-W test	z= - 0.65	0.52	67.10	71.52
	PPI120	M-W test	z= - 0.57	0.57	71.62	67.72

** two- tailed significance; (1) = Low-risk group, (2) = High-risk group; N=125 (n1=70, n2=55) for 3UTR polymorphism; N= 138 (n1=63, n2=75) for intron 8 polymorphism*

Table 5: Statistical analysis of the effects of the DAT genotype on PPF at different ISIs

		Test	Test-statistic	Sign.*	Mean (1)	Mean (2)
3´UTR	**PPI2000**	t-test	t= 0.24	0.81	7.05	5.86
	PPI4000	M-W test	z= - 0.21	0.72	61.99	64.29
Int8	**PPI2000**	t-test	t= 0.95	0.34	8.25	3.83
	PPI4000	M-W test	z= - 0.35	0.68	67.98	70.77

** two- tailed significance; (1) = Low-risk group, (2) = High-risk group; N=125 (n1=70, n2=55) for 3UTR polymorphism; N= 138 (n1=63, n2=75) for intron 8 polymorphism*

Table 6: Statistical analysis of the effects of the DAT genotype on PPI at ISI of 30ms using one- way – ANCOVA with "unusual perceptual experiences" (SPQ) as a covariate

		F	Sign.*
3´UTR	**3´UTR**	0.41	0.22
	UE	4.40	0.40
	3´UTR x UE	0.32	0.57
Int8	**Int8**	0.04	0.84
	UE	2.22	0.14
	Int8 x UE	0.44	0.51

** two- tailed significance; N=125(n1=70, n2=55) for 3UTR polymorphism; N= 138(n1=63, n2=75) for intron8 polymorphism; UE= unusual perceptual experiences*

Table 7: Statistical analysis of the effects of the DAT genotype on PPI at ISI of 30ms using one- way – ANCOVA with neuroticism (NEO-FFI) as a covariate

		F	Sign.*
3´UTR	**3´UTR**	5.41	0.02
	N	13.34	0.00
	3´UTR x N	6.08	0.02
Int8	**Int8**	0.39	0.53
	N	8.57	0.00
	Int8 x N	0.84	0.36

** two- tailed significance; N=125(n1=70, n2=55) for 3UTR polymorphism; N= 138(n1=63, n2=75) for intron8 polymorphism*

Table 8: Statistical analysis of effect of 3´UTR polymorphism on PPI at ISI of 30ms in low/high- neuroticism groups using unpaired t-tests

	Test-statistic	Sign.*	df	Mean (1)	SD (1)	Mean (2)	SD (2)
Low neuroticism	- 1.34	0.19	56	53.76	17.50	61.10	23.14
High neuroticism	0.89	0.38	65	53.21	22.57	47.85	25.26

**two- tailed significance; (1) = Low-risk group, N=58(n1= 26, n2= 32); (2) = High-risk group, N=67(n1= 44, n2= 23)*

Curriculum Vitae

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