Background: Neuroticism, as defined and measured by the NEO Personality Inventory (Neuroticism Extraversion and Openness Personality Inventory), is a core personality trait reflecting an individual’s emotional reactivity. High neuroticism is thought to be an important vulnerability factor for various psychiatric disorders in the general population, including substance abuse, depression, anxiety, and psychosis. Recent findings support the hypothesis that genetic factors underlying the neuroticism trait could increase the susceptibility to psychiatric disorders. The current study aimed to replicate genetic associations with high neuroticism previously reported in the literature. Methods: We genotyped four polymorphisms: CNR1 (rs7786029), GABRA2 (rs9291283), GABRA6 (rs3219151) and MAMDC1 (rs7151262) in 215 healthy Caucasian subjects, who completed a short version of the NEO-PI. NEO neuroticism scores of the three genotype groups were compared using ANCOVA, with age as a covariate.

Results: All four genetic polymorphisms were found to be significantly associated with NEO neuroticism scores (p < 0.0025), but not with any other NEO personality domain.

Conclusion: Our results corroborate other studies proposing a role for the GABAergic and cannabinoid systems in the modulation of affective states and stress responses, as measured through neuroticism scores. It is important to replicate the genetic findings of neuroticism, in order to gain a better understanding of this personality domain that has been reported as an important risk factor for mood and anxiety disorders and substance addiction.

Keywords: NEO; Neuroticism; Genetics

Introduction

Neuroticism is a core personality trait that reflects an individual’s emotional reactivity, with an estimated heritability of 40 to 60% [1-3]. Individuals who score high on measures of neuroticism tend to be more sensitive to negative mood states and to have excessive responses to environmental stresses. Maladaptive efforts to regulate affect and cope with negative emotional life events can result in the development of an addiction or a psychiatric disorder [4-7]. In fact, high levels of neuroticism have been extensively linked to psychiatric disorders, such as depression [8-10], anxiety [7,11] and substance use disorders [12,13]. Neuroticism, as assessed by the NEO Personality Inventory (Neuroticism Extraversion and Openness Personality Inventory), is therefore considered an important vulnerability factor for psychopathology [14,15], and a strong positive correlation exists between NEO neuroticism scores and the risk of relapse in several psychiatric conditions, including psychosis, depression and substance abuse [12,16,17]. Furthermore, neuroticism scores and negative life events were found to be the two strongest predictors of onset of psychotic and depressive symptoms during remission and relapse from both alcoholism and opioid addiction [12,17,18]. Taken together, these results suggest that neuroticism may be an underlying vulnerability factor for mental illness and addiction.

Recent findings support the hypothesis that genetic factors underlying individual differences in neuroticism could increase the susceptibility to both addiction and mental illness [19,20]. Twin studies have suggested a heritability coefficient in the range of 0.4-0.6 for this personality trait, which has spurred attempts to map the genetic basis of neuroticism. The current study aimed to verify if previously reported associations of genetic variants with NEO neuroticism [3,21-26] scores would be replicated in a sample of healthy subjects of European Caucasian descent.

Subjects and Methods

Our sample of 215 unrelated healthy European Caucasian subjects (35% male, age 48.9±16.1) was collected as part of the “Leisure, Lifestyle, and Lifecycle” Project in the province of Alberta, Canada [27]. Presence of a lifetime psychiatric disorders was an exclusion criteria and was assessed through the Composite International Diagnostic Interview (CIDI) [28]. Personality traits were assessed using the NEO-FFI, which is a 60-item short version of the NEO-PI-R (Revised NEO Personality Inventory). This personality assessment measures...
the following five personality dimensions: Neuroticism, Extraversion, Openness to Experience, Agreeableness and Conscientiousness [14].

DNA extraction was performed on blood samples using a standard high-salt method. We genotyped four polymorphisms previously associated with neuroticism in the literature: rs7766029 in the CNR1 [24], rs9291283 in GABRA2 [26,29], rs3219151 in GABRA6 [21,25] and rs7151262 in MAMDC1 [3,23,30]. Genotyping was performed using TaqMan allele-specific assays (ABI Prism 7000/7500, Applied Biosystems, Foster City, CA). According to published guidelines, we have included four negative controls in each 96-well genotyping plate (Biosystems, Foster City, CA). Our sample had over 80% statistical power to detect association of the GABRA2 polymorphism (lowest allele frequency among the evaluated polymorphisms) with a minimum \( r^2 \) of 2.5% [32]. All four variants were found to be significantly associated with neuroticism scores in our sample \( (p < 0.0025) \). We have not observed significant association of these polymorphisms with any of the other four NEO personality dimensions analyzed. The common allelic variants of CNR1 rs7766029 and GABRA2 rs9291283 were associated with higher neuroticism scores than the rare alleles; while the common allelic of GABRA6 rs3219151 and MAMDC1 rs7151262 were associated with lower neuroticism scores (Table 1).

**Results**

We obtained 100% genotyping accuracy and all polymorphisms were in Hardy-Weinberg equilibrium \((p > 0.05)\). Our sample had over 80% statistical power to detect association of the GABRA2 polymorphism (lowest allele frequency among the evaluated polymorphisms) with a minimum \( r^2 \) of 2.5% [32]. All four variants were found to be significantly associated with neuroticism scores in our sample \( (p < 0.0025) \). We have not observed significant association of these polymorphisms with any of the other four NEO personality dimensions analyzed. The common allelic variants of CNR1 rs7766029 and GABRA2 rs9291283 were associated with higher neuroticism scores than the rare alleles; while the common allelic of GABRA6 rs3219151 and MAMDC1 rs7151262 were associated with lower neuroticism scores (Table 1).

**Discussion**

We replicated previously reported associations of polymorphisms in GABRA2 and 6, CNR1, and MAMDC1 with the personality trait of neuroticism. Taken together with previous results, the current study supports the roles of the endocannabinoid and GABAergic neurotransmitter systems in the regulation of emotionality and responsiveness to stress. Limitations to this study would include a moderate sample size, but our power calculations showed we would have 80% power to detect medium genetic effects [33]. Also, the functional role of the polymorphisms investigated here is yet to be determined.

The first genome-wide association study (GWAS) for neuroticism highlighted the significance of a common polymorphism in the mam domain-containing glycosylphosphatidylinositol anchor 2 gene (MAMDC1) [3], which was recently confirmed in a second GWAS [23]. Although no functional studies of rs7151262 have been published, the MAMDC1 gene is predominantly expressed in areas associated with memory and emotional regulation (hippocampus and amygdala), and is thought to be involved in regulating neuronal migration and axonal guidance [34].

Cannabinoids are known to have both anxiolytic and anxiogenic properties, implicating the endocannabinoid system in mood and stress response regulation. It has been hypothesized that temporal changes in the endocannabinoid system during development can affect a person’s emotional stability and stress responsiveness later in life [35-37]. For example, cannabis use during adolescence has been recently highlighted as a significant risk factor for the development of psychosis symptoms in adulthood [37]. Moreover, adolescent exposure to cannabinoids is associated with an earlier onset of schizophrenia [35]. Not surprisingly, genetic variants in the cannabinoid receptor

![Table 1: Association analysis between CNR1, GABRA2, GABRA6 and MAMDC1 polymorphisms with the five personality domains assessed by the NEO-FFI scale, using age as covariate (ANCOVA).](image-url)

<table>
<thead>
<tr>
<th>Genetic Variant</th>
<th>Personality Domain</th>
<th>Genotype</th>
<th>mean</th>
<th>n</th>
<th>SD</th>
<th>mean</th>
<th>n</th>
<th>SD</th>
<th>mean</th>
<th>n</th>
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<th>F</th>
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<td>86</td>
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</table>

*The significance level was set to \( p=0.0025 \) after a Bonferroni correction for multiple testing (0.05/ 20 tests)
I gene (CNR1) have been reported to be significantly associated with high neuroticism scores [24].

Similarly, the inhibitory neurotransmitter gamma-aminobutyric acid (GABA) has long been implicated in modulating anxiety, and may therefore partially underlie the personality dimension of neuroticism [38,39]. Post-mortem studies have found that the prefrontal cortex of patients who suffered from depression or an anxiety disorder contained less GABAergic neurons [38]. Furthermore, prior genetic studies have shown associations of neuroticism with polymorphisms located in the GABAergic system genes. Common polymorphisms within the glutamic acid decarboxylase 1 (GAD1) gene, which encodes the enzyme responsible for GABA synthesis, have been significantly associated with high neuroticism scores [11]. Moreover, variants in the GABA receptor subunit alpha (GABRA) have been previously found to be associated with neuroticism and other emotional reactivity-associated traits (e.g. attenuated stress response and harm avoidance), and could confer a genetic susceptibility to major depression and anxiety disorders [25,26,29,40].

The current study investigated the association of four previously reported polymorphisms with neuroticism and these associations were replicated. Neuroticism is thought to have a moderate genetic component and may be important in the acquisition, development, and maintenance of psychiatric disorders, including substance addiction. Thus, together with previous research, our results may help clarify the genetic underpinnings of neuroticism and its role as a vulnerability factor for both mood and anxiety disorders and substance addiction.

References


