Using a cognitive endophenotype to identify risk genes for depression

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ABSTRACT

We theorized the cognitive vulnerability factor featured in hopelessness theory [2] to be a novel endophenotype for depression. We investigated two possible genetic contributors to individual differences in cognitive vulnerability (and, in turn, depression): the \textit{BDNF} gene and the \textit{COMT} gene. Results showed that individuals (n = 95) with the \textit{BDNF} Val\textsuperscript{66} genotype had significantly greater levels of cognitive vulnerability than individuals with a \textit{BDNF} Met\textsuperscript{66} genotype. In addition, among individuals with high levels of cognitive vulnerability, those with the Val\textsuperscript{66} genotype were significantly more likely than participants with a Met\textsuperscript{66} genotype to experience increases in depressive symptoms when faced with increased stress. The \textit{COMT} gene was not associated with cognitive vulnerability or risk for depression. Results support the use of the cognitive vulnerability factor featured in hopelessness theory as an endophenotype associated with depression as well as the role of the \textit{BDNF} gene in a cognitive subtype of depression.

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Scientists have made great strides in identifying factors that increase risk for depression. This progress is particularly evident in the identification of psychological and environmental risk factors (e.g., cognitive, interpersonal, life stress \cite{3,17}). In contrast to this work, the identification of genetic risk factors for depression has progressed much more slowly. Indeed, after nearly a decade, it is still impossible to name a specific gene or polymorphism that is unequivocally associated with risk for depression.

One reason for the slow progress in identifying “depression genes” could be due to the complexity of the phenotypes associated with depression. Complex syndromes involve multiple genetic influences, and thus, pose considerable problems for traditional genetic linkage studies, which are best suited for identifying phenotypes influenced by a few major genes. Depression is characterized by complexity in both its symptom expression (e.g., two people could meet criteria for depression and not share any of the same symptoms) and its etiology. One solution for identifying genes involved in a complex phenotype is to break it down into its more basic constituents or endophenotypes. Endophenotypes form the causal link between genes and the phenotype. They tend to be basic human processes (e.g., cognitive control) that are less complex than mental disorders, and thus, more manageable for genetic analyses.

The goal of the current study was to use the endophenotype strategy to identify genes containing specific variants that may be associated with depression. Specifically, we theorized the cognitive vulnerability factor featured in the hopelessness theory of depression \cite{3} to be an ideal endophenotype for depression. According to the hopelessness theory, cognitive vulnerability is the tendency to generate interpretations of stressful life events that have negative implications for one’s future and for one’s self-worth. Research shows that cognitive vulnerability is a trait-like risk factor that stabilizes in early adolescence, and confers risk for depression throughout the life span \cite{24}. Indeed, by early adulthood one is saddled (or blessed) with a level of cognitive vulnerability that is relatively impervious to changes in environmental conditions \cite{19}.

Importantly, the cognitive vulnerability factor featured in hopelessness theory meets the stringent criteria put forth by Kendler and Neale \cite{21} for being an endophenotype. First, research suggests that cognitive vulnerability is a cause rather than a consequence of depression. Prior studies show that it is possible to take a group of individuals who have never been depressed and predict which of them are most likely to develop a first onset of depression based solely on individual differences in their level of cognitive vulnerability \cite{2,16}. Further, experiments aimed at reducing cognitive vulnerability effectively prevent future depression \cite{25}. Second, cognitive vulnerability is state independent. High levels of cognitive vulnerability can be reliably detected in non-depressed, depressed, and formerly depressed populations \cite{15}. Third, cognitive vulnerability tends to run in families \cite{2,10}. Finally, there is strong evidence that cognitive vulnerability varies continuously in the population and tends to be normally distributed \cite{11,14}.

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Taken together, there is strong justification for using the cognitive vulnerability factor featured in hopelessness theory as an endophenotype for identifying depression genes. In this study, we investigated two possible genetic contributors to individual differences in cognitive vulnerability (and, in turn, depression): the brain-derived neurotrophic factor (BDNF) gene and the Catechol-O-Methyltransferase (COMT) gene. We chose to start with these two genes because research shows that they are associated with both cognitive functioning and possibly depression. The BDNF gene codes for a protein that is associated with neuronal survival and synaptic signaling. There is evidence that a common single-nucleotide polymorphism (SNP) in the BDNF gene (rs6265) is related to both executive functioning and mood disorders [7,20]. The polymorphism is located in exon 11 and results in an amino acid substitution from valine to methionine at codon 66 (Val<sup>66</sup>Met).

We also investigated the association of the COMT gene with individual differences in cognitive vulnerability. COMT is not only an important determinant of cognitive functioning, but it has also been associated with depression [1,4,8]. The COMT gene has a functional and common SNP (rs4680) located in codon 158, which translates into a valine-to-methionine substitution in the peptide sequence (Val<sup>158</sup>Met). Research shows that the human Met<sup>158</sup> allele is associated with enhanced dopamine signaling and more efficient cognitive processing [26]; for exception see [23]. We hypothesized that the Val<sup>158</sup> genotype would more likely than the Met<sup>158</sup> genotype to be associated with cognitive vulnerability because of its influence on cognitive processing, its association with depressive outcomes [4,8], and its effect on reward-related behaviors [9], which are also influenced by cognitive vulnerability [13].

In summary, the search for genetic risk factors for depression has progressed slowly. One explanation for the lack of progress is the complexity of the phenotype(s) associated with depression. The goal of the current study was to reduce this complexity by using a novel endophenotype—cognitive vulnerability. This is the first study to investigate the genetics of the cognitive vulnerability factor featured in the hopelessness theory, a prominent and highly supported theory of depression. We tested two primary hypotheses: (1) specific functional polymorphisms in the BDNF and COMT genes (rs6265 and rs4680, respectively) would be associated with cognitive vulnerability to depression, and (2) these polymorphisms in the BDNF and COMT genes would interact with life stress to predict prospective changes in depressive symptoms in a cognitively vulnerable sample.

1. Method

1.1. Participants

Participants were 95 unselected undergraduates (36 women, 59 men; M age = 18.2) at a mid-sized private university in the midwestern U.S. An unselected sample was used to ensure a range of cognitive vulnerability scores. If we had selected participants based on levels of depression, then this would have truncated the range of the cognitive vulnerability variable and made it difficult to identify its genetic determinants. Specific data regarding ethnicity was not collected; however, the sample is likely representative of the diversity of the university more generally (76% Caucasian, 11% Hispanic, 8% Asian, 5% African American). Participants provided written consent to participate and were compensated monetarily. The University's Human Subjects Committee approved the study.

1.2. Measures

Acute Life Events Questionnaire (ALEQ [14]). The ALEQ was used to assess naturally occurring acute stressful life events important to college students (from school/achievement to interpersonal/romantic) that occurred over the previous 4 weeks. Scores can range from 0 to 30 with higher scores indicating the occurrence of more stressful events. The ALEQ was administered at baseline and 2 months.

Beck Depression Inventory (BDI) [6]). The BDI is a 21-item self-report inventory that assesses depressive symptoms (scores range from 0 to 63 with higher scores reflecting greater depression). The BDI has strong internal consistency, test–retest reliability, and validity [5]. The BDI was administered at baseline, 2 months, and 3 months.

Cognitive Style Questionnaire (CSQ [16]). The CSQ assesses the cognitive vulnerability factor featured in the hopelessness theory of depression. The CSQ assesses participants’ causal attributions and self-worth inferences for the 12 hypothetical negative events. Mean-item scores can range from 1 to 7, with higher scores reflecting greater cognitive vulnerability. The CSQ has high internal consistency [16,19], strong test–retest reliability over months and even years [2,16], and a high degree of construct validity [2,14,16,22]. Coefficient alpha for the CSQ in this study was .92. The CSQ was administered a baseline.

1.3. Procedure

We conducted a two-component study. The first component used a cross-sectional design and included all participants. Participants completed measures of cognitive vulnerability (CSQ), depressive symptoms (BDI), and stressful events (ALEQ); they also provided a saliva sample (using a kit produced by Oragene) for genotyping. Saliva samples were sent to Dr. Grigorenko’s lab at Yale University where DNA was extracted and genotyped using the ABI TaqMan platform. For the purposes of this project, two SNPs were genotyped: rs6265 and rs4680.

A subset of the cross-sectional sample (n = 53; 33 women, 20 men) participated in the second component of the study, which used a 3-month longitudinal prospective design. Participants were selected for the longitudinal component if they were considered to possess elevated levels of cognitive vulnerability (scoring in the top 40% of a larger screening sample [12] on the CSQ). Participants in the longitudinal component completed a measure of depressive symptoms (BDI) at 2-months and 3-months post-baseline. They also completed a measure of stressful life events (ALEQ) at the 2-month time point. Stressful life events was assessed at the 2-month (rather than the 3-month) time point in order to ensure that their occurrence clearly preceded any changes in depressive symptoms 1 month later. Prior research indicates that this time frame is a sufficient time interval for stress to lead to the development of depressive symptoms [14,16,18,22].

2. Results

Allele frequencies for the BDNF and COMT SNPs in the sample are listed in Table 1. Both of the markers demonstrated Hardy–Weinberg Equilibrium. Given the small number of individuals homozygous for the BDNF Met<sup>66</sup> allele, we combined this group with the heterozygous group for analyses. Thus, the comparisons to follow for the BDNF gene compared participants homozygous for the Val<sup>66</sup> allele to those with the Met<sup>66</sup>/heterozygous genotypes. Means and standard deviations for the primary study variables at baseline and the 3-month follow-up are listed in Table 2.

Hypothesis 1. BDNF and COMT SNPs are associated with cognitive vulnerability.

To examine the effect of BDNF and COMT SNPs on cognitive vulnerability, we performed an ANCOVA with genotype as the independent variable and cognitive vulnerability (CSQ scores) as
the dependent variable. BDI score was used as covariate. Results revealed a significant main effect of BDNF genotype ($F(1,92) = 5.40, p = .02, \eta^2_p = .06$), but not COMT genotype ($F(2,95) = 13, p = .88, \eta^2_p < .01$). Consistent with our hypotheses, participants who were homozygous for the BDNF Val<sup>66</sup> allele had significantly higher levels of cognitive vulnerability ($M = 4.39, SE = .74$) than participants with the Met allele<sup>66</sup> (either heterozygous or homozygous; $M = 4.00, SE = .83$). Neither the BDNF nor the COMT genotype were associated with concurrent levels of depressive symptoms (both $p$s > .51). The results did not change if gender was added as a covariate.

**Hypothesis 2.** BDNF and COMT SNPs interact with life stress to predict changes in depressive symptoms in a vulnerable sample.

We used hierarchical multiple regression to analyze the data. Predictor variables were entered into the regression equation in two steps. In step one, the baseline and 2-month depression scores (BDI) were entered to create a residual change score for the same depression measure at 3-months (dependent variable). Baseline level of stress was also included in this step to control for any initial individual differences in stress levels. In step 2, the main effects of genotype (BDNF and COMT polymorphisms, respectively) and stress (at 2-months) were entered. And in step 3, the interaction of genotype and stress was entered. Individual variables within a given step were not interpreted unless the set as a whole was significant, thereby reducing Type I errors. Note that the COMT genotypes were treated categorically, using two dummy variables to represent the three possible allele combinations (homozygous for Val<sup>158</sup>, heterozygous, homozygous for Met<sup>158</sup> allele). The homozygous Val<sup>158</sup> genotype was used as the reference group.

As shown in Table 3, there was a significant BDNF genotype × Stress interaction, $b = 5.27, t = 3.16, p = .003$. Consistent with hypotheses, participants with the Val<sup>66</sup> genotype who experienced increases in stress had the greatest level of depressive symptoms at 3-months, even after controlling for baseline and 2-month BDI scores (see Fig. 1); note that results remained the same if only the BDI score at the 2-month time point was used as a covariate. The effect size of this interaction (partial correlation = .44) is considered medium to large. Indeed, as can be seen in Table 3, participants with

![Table 1](https://example.com/table1.png)

**Table 1** Allele frequencies for BDNF (rs6265) and COMT (rs4680) polymorphisms.

<table>
<thead>
<tr>
<th>Genotype</th>
<th>rs6265</th>
<th>rs4680</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cross-sectional component</td>
<td></td>
<td></td>
</tr>
<tr>
<td>GG (Val)</td>
<td>67</td>
<td>67</td>
</tr>
<tr>
<td>GA</td>
<td>28</td>
<td>28</td>
</tr>
<tr>
<td>AA (Met)</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>Longitudinal component</td>
<td></td>
<td></td>
</tr>
<tr>
<td>GG (Val)</td>
<td>68</td>
<td>68</td>
</tr>
<tr>
<td>GA</td>
<td>28</td>
<td>28</td>
</tr>
<tr>
<td>AA (Met)</td>
<td>4</td>
<td>4</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Predictor</th>
<th>$b$</th>
<th>$p$</th>
<th>$t$</th>
<th>$R^2$ change</th>
</tr>
</thead>
<tbody>
<tr>
<td>Step 1</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>BDI at baseline</td>
<td>.39</td>
<td>.25</td>
<td>1.75</td>
<td></td>
</tr>
<tr>
<td>BDI at 2-months</td>
<td>.53</td>
<td>.34</td>
<td>2.40$^*$</td>
<td></td>
</tr>
<tr>
<td>ALEQ baseline</td>
<td>.01</td>
<td>.00</td>
<td>.00</td>
<td></td>
</tr>
<tr>
<td>Step 2</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ALEQ at 2-months</td>
<td>.73</td>
<td>.19</td>
<td>1.23</td>
<td></td>
</tr>
<tr>
<td>BDNF</td>
<td>1.36</td>
<td>-10</td>
<td>-6.4</td>
<td></td>
</tr>
<tr>
<td>Step 3</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>BDNF × ALEQ at 2-months</td>
<td>.52</td>
<td>.44</td>
<td>3.16$^*$</td>
<td></td>
</tr>
</tbody>
</table>

Note. BDI = Beck Depression Inventory; ALEQ = Acute Life Events Questionnaire. BDNF = genotypes at the BDNF Val<sup>66</sup>Met polymorphism. Note that results remain the same if baseline BDI at baseline is removed from analyses. $^*$ $p < .05$.

Val<sup>66</sup> genotype and high levels of stress exhibited double the level of depressive symptoms as the rest of the sample. The COMT genotype did not interact with stress to predict depression (both product term $p$s > .35). Also the COMT and BDNF genotypes did not interact with each other to predict depression. The results did not change if gender was added as a covariate.

### 3. Discussion

This study was the first to investigate genes associated with the cognitive vulnerability factor featured in the hopelessness theory, a novel endophenotype for depression. Consistent with hypotheses, results showed that the BDNF Val<sup>66</sup>Met polymorphism was associated with individual differences in cognitive vulnerability levels, even after controlling for current depressive symptoms. Specifically, individuals with the Val<sup>66</sup> genotype had significantly greater levels of cognitive vulnerability than individuals who had the Met<sup>66</sup> allele (either heterozygous or homozygous). Not only was the Val<sup>66</sup> genotype associated with the cognitive vulnerability endophenotype, but it also conferred risk for future depressive symptoms. Results showed that cognitively vulnerable participants with the Val<sup>66</sup> genotype were significantly more likely than vulnerable participants with the Met<sup>66</sup> allele (either heterozygous or homozygous) to experience increases in depressive symptoms when faced with increases in life stress. In contrast to the positive results for the BDNF gene, the COMT Val<sup>158</sup>Met polymorphism was not associated with cognitive vulnerability or risk for future depressive symptoms.

![Table 2](https://example.com/table2.png)

**Table 2** Means and Standard deviations for measures at baseline and 3-month follow-up as a function of genotype.

<table>
<thead>
<tr>
<th></th>
<th>BDNF Val M (SD)</th>
<th>BDNF Met M (SD)</th>
<th>COMT Val M (SD)</th>
<th>COMT Het M (SD)</th>
<th>COMT Met M (SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline</td>
<td>4.39 (.74$^a$)</td>
<td>4.00 (.83)</td>
<td>4.30 (.74)</td>
<td>4.23 (.77)</td>
<td>4.30 (.90)</td>
</tr>
<tr>
<td>BDI</td>
<td>7.38 (7.12)</td>
<td>7.26 (5.00)</td>
<td>6.78 (6.45)</td>
<td>6.82 (5.27)</td>
<td>8.09 (8.70)</td>
</tr>
<tr>
<td>3-month</td>
<td>9.09 (9.42)</td>
<td>6.62 (7.01)</td>
<td>6.20 (6.27)</td>
<td>9.52 (9.39)</td>
<td>7.25 (9.03)</td>
</tr>
</tbody>
</table>

Note. $^a$ Participants homozygous for the BDNF Val<sup>66</sup> allele had significantly greater levels of cognitive vulnerability than participants with the Met allele<sup>66</sup>.

![Fig. 1](https://example.com/fig1.png)

**Fig. 1.** Depressive symptoms at 3-months as a function of BDNF genotype and stress in a sample of individuals with elevated levels of cognitive vulnerability ($n=53$). Participants were considered to have elevated levels of cognitive vulnerability if they scored in the top 40% of a larger screening sample on the CSQ (i.e., had scores > 4.2). Higher scores on the y-axis indicate greater levels of depressive symptoms.
The current results have a number of important implications. First, the findings from the cross-sectional component provide further support for using an endophenotype strategy for identifying genes for depression. It was only by examining an intermediate risk factor (cognitive vulnerability) that we were able to identify the BDNF gene as a possible target gene for depression. There was no association found between genotype and concurrent levels of depressive symptoms.

The results of the longitudinal component underscore the importance of conceptualizing genes as risk factors rather than markers of depression. Genes for complex phenotypes such as depression are not going to be deterministic. Rather, they most likely require specific environmental conditions (e.g., stress) and/or the presence of other risk genes to influence the phenotype. Thus, studies need to examine the moderating effects rather than main effects of genes on depression. This means that the cross-sectional designs typically used in this area of work might not be optimal for detecting an association between genes and depression because they only provide a single snapshot of time and do not allow adequate examination of moderating factors. In contrast, a longitudinal design allows time for risk factors and moderators to exert their influence on the development of depression.

Finally, these findings provide additional support for the role of BDNF in depression as well as help to explain prior inconsistencies. Our study suggests that BDNF Val66Met polymorphism might confer specific risk for a cognitive subtype of depression. Indeed, both our study and Hilt et al. [20] found that the Val66 allele was associated with a cognitive vulnerability factor for depression (Hilt and colleagues examined rumination). Thus, one explanation for the inconsistent findings in the literature for BDNF is that prior studies have used samples that may or may not have comprised individuals with high levels of cognitive vulnerability.

It is important to acknowledge limitations of the current study. For example, the study used a college sample so it is possible that the results may not generalize to community samples. Also, the effect of ethnicity on the study outcomes remains unclear as this demographic information was not collected. However, it is important to note that the results of studies using college samples often do generalize to community and clinical samples, particularly when basic processes are being studied. Another potential limitation of the current study is that it examined depressive symptoms, but not clinical diagnoses. Thus, we cannot make conclusions about clinically significant forms of depression. However, given research suggesting that depressive symptoms and depressive syndromes lie on a continuum, we expect that future research will provide evidence that our pattern of results also extends to depressive disorders. Finally, our longitudinal study design did not allow us to test whether or not cognitive vulnerability mediates the association between the variation in the BDNF gene and future depressive symptoms. We selected participants with high levels of cognitive vulnerability, which created a restricted range incapable of predicting future depression. Thus, we can only conclude that among individuals with elevated levels of cognitive vulnerability, the BDNF gene combines with stress to confer risk for future depressive symptoms (that said, it is important to note that this effect was in the medium to large range).

In conclusion, this study adds to the small, but important body of research using the endophenotype strategy to identify risk genes for depression. Results support the use of the cognitive vulnerability factor in hopelessness theory as depressive endophenotype. By using this cognitive vulnerability factor, we found that variability in the BDNF gene, but not the COMT gene, increased risk for future depressive symptoms in those experiencing high levels of stress. We look forward to future work that uses this novel endophenotype to reveal additional genes implicated in depression.

References