

## Association between Catechol-O-Methyltransferase Gene Variant and Bipolar Disorder

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**ABSTRACT** Emotional dysregulation is a core characteristic of many psychiatric diseases. Bipolar disorder is a brain disorder that causes unusual shifts in mood, energy level, activity level and ability to carry day to day work. Catechol-O-methyltransferase (COMT) is involved in the metabolism of dopamine and epinephrine. To a large extent it is responsible in maintaining human cognitive functioning. Polymorphisms in the COMT gene lead to various dysregulations. For the study we have considered single nucleotide polymorphism (SNP) rs 4680 (val158met), a G to A transition mutation due to which valine is substituted by methionine at codon 158, which reduces the enzyme activity by four folds. The aim of the current study is to check whether the low activity allele has any association with bipolar disorder. The study included 50 unrelated patients, diagnosed suffering from bipolar disorder. Age and sex matched control samples were taken. Blood samples were collected from all the individuals after taking informed consent. SNP was genotyped with a PCR-based restriction fragment length polymorphism analysis using the restriction enzyme NlaIII. Amongst the 50 patients, 29 were homozygous for G allele, 18 were heterozygous and 3 were homozygous for A allele. In the control population only 6 individuals were found to be heterozygous for the variation. The met allele frequency is 0.24 in patients and 0.06 in control group. Chi square value is 12.08 (df=2) and p value is 0.002. Significant association was observed between the catechol-O-methyltransferase val158met polymorphism and bipolar affective disorder.

### INTRODUCTION

Emotional dysregulation is referred to as an emotional response that is poorly modulated and that does not fall within the conventionally accepted range. This characteristic is detected in patients with bipolar disorder also. Bipolar disorder (MIM 125480), also known as manic depressive illness, is a brain disorder that causes unusual shifts in mood, energy, activity level and ability to perform. Its estimated prevalence is 0.8 to 2.6% (Kato 2007).

It has been proved that the impaired functioning of the prefrontal cortex of human brain is responsible for the disorder (McIntosh 2008). The most typical psychological term for functions carried out by the pre-frontal cortex area is executive function. Executive function relates to abilities to differentiate among conflicting thoughts, determine good and bad, better and best, same and different, future consequences of current activities, working towards a defined goal, prediction of outcomes, expectation based on actions, and social "control" (the ability to suppress urges that, if not suppressed, could lead to socially-unacceptable outcomes).

Though environment plays an important role, individual psychological variables and genetic predisposition are also responsible in develop-

ment and course of the disorder. Most of the patients seem to have gone through some psychological variables like stressful events, experience of some adversity and conflict. There are genetic dispositions involved which cumulate to give rise to the disorder. Dopamine is the neurotransmitter that maintains the neuro-chemistry of prefrontal cortex. Many genes are responsible for the proper functioning of dopamine, for example COMT.

The COMT gene located on chromosome 22q11.2 region provides instruction for making an enzyme called Catechol-o-methyltransferase. Genetic polymorphism in COMT gene which results in a valine to methionine substitution, due to a G-to-A transition was reported by Lachman et al. (1996a). Later Syvanen et al. (1997) demonstrated that this change is the basis for variation in the thermal stability and activity level of the enzyme. This enzyme helps in the breakdown of neurotransmitters like dopamine and epinephrine. COMT plays an important role in the prefrontal cortex (PFC) by maintaining levels of dopamine in the brain. It helps PFC to work and maintain its functions. Various SNPs in COMT are studied that leads to malfunctioning of PFC. For the study we have considered, functional, non-synonymous, single nucleotide polymorphism (SNP) rs 4680 (val158met), a G to

A transition mutation, due to which valine is substituted by methionine at codon 158. The methionine allele is thermo labile at 37°C and has one fourth the enzymatic activity of the valine allele (Männistö et al. 1999).

Smoller and Finn (2003) estimated a 10-fold increase in the recurrence risk (8.7%) for bipolar disorder among first-degree relatives of bipolar probands. Twin studies suggest heritability estimates from 79% to 93% for mania/bipolar disorder (Mick 2009).

Studies of adult subjects with bipolar disorder strongly suggest a genetic component in etiology of the disorder. The COMT gene has been extensively studied as a candidate gene for a variety of psychiatric disorders including schizophrenia, bipolar disorder, and other psychiatric conditions because of its known function in dopamine metabolism. Egan et al. (2001) observed that the functional polymorphism of the enzyme (Val 108/158Met) leads to difference in cognitive functioning of the prefrontal cortex. Several studies were conducted to analyze COMT val/met polymorphism and risk of schizophrenia but very few studies were attempted on bipolar disorder. Therefore, the objective of the present study is to find an association between COMT val158met polymorphism and bipolar disorder.

#### METHODOLOGY

The study includes 50 unrelated patients from the Government Hospital for Mental Care, Visakhapatnam, diagnosed suffering from bipolar disorder, according to the Diagnostic and Statistical Manual of Mental Disorders (DSM-IV). The exclusion criteria excluded subjects diagnosed with at least one of the following disorders: mood disorder due to a general medical condition, substance-induced mood disorder, major depressive disorder, dysthymic disorder, bipolar II disorder, cyclothymic disorder, psychotic disorders, secondary mania induced by hyperthyroidism, and neurologic disorder. The patients were from Visakhapatnam district of Andhra Pradesh, India.

Equal number of age and sex matched psychiatrically healthy individuals were considered as controls. Blood samples were collected from all the individuals after taking informed consent. DNA was isolated by Lahiri and Nurnberger (1991) method. COMT Val158Met

genotypes were determined by restriction fragment length polymorphism, using the restriction enzyme NlaIII. A 116-base-pair polymerase chain reaction (PCR) product was generated using forward primer 5' TCATCACCATCGAGATCAACC 3' and reverse primer 5' CCCTTTTCCAGGTCTGACA 3'. Thermal cycling was carried out as follows: initial denaturation at 94°C for 5 minutes followed by 35 cycles with denaturation at 94°C for 30 sec, annealing at 60°C for 30 sec and extension at 72°C for 30 sec. The final extension was carried out at 72°C for 5 min.

The val and met alleles were discriminated by digesting the PCR product with 2.5U of NlaIII at 37°C for overnight, followed by 3% agarose gel electrophoresis. The val/val homozygotes (116 base pairs), met/met homozygotes (68 and 48 base pairs), and val/met heterozygotes (116, 68 and 48 base pairs) were visualized by ethidium bromide staining.

#### RESULTS AND DISCUSSION

Amongst the 50 patients, 29 were homozygous for val allele, 18 were heterozygous and 3 were homozygous for met allele. In the control population, only 6 individuals were found to be heterozygous for the variation. The met allele frequency is 0.24 in patients and 0.06 in control group. Significant association was observed between SNP rs 4680 and bipolar disorder, with met allele being over represented in cases versus controls ( $\chi^2 = 12.08$ ;  $.001 > p > .01$ ).

Studies done with COMT showed varying results across the globe. Lachman et al. (1996b) reported an association between met allele and development of bipolar disorder in patients with velocardiofacial syndrome. Goghari and Sponheim (2008) reported that met homozygosity was associated with greater positive symptomatology in bipolar disorder.

Some studies done on Chinese patients report an association between low activity allele and patients (Li et al. 1997), while some studies done with the Caucasoid population (Gutiérrez et al. 1997; Burdick et al. 2007; Shifman et al. 2004) observed no association. The present study agrees with the former in reporting significant association.

Gupta et al. (2009) did association study between COMT and schizophrenia in heterogeneous South Indian population. Association study of MLC1 and SYGR11 gene is done with

bipolar disorder in the south Indian population (Verma et al. 2005).

Szegedi et al. (2005) investigated whether functionally relevant Val<sup>108/158</sup>Met gene variant is associated with differential antidepressant response to mirtazapine and paroxetine in patients with major depression. The authors find that polymorphisms within the COMT gene seem to influence the time course of response and clinical efficacy of mirtazapine but not paroxetine, thus determining the frequency of the SNP in a population can help in better medication. Thus research of this kind helps in pharmacogenomics.

### CONCLUSION

A significant association was found between COMT val158met polymorphism and bipolar disorder in Coastal Andhra Pradesh. Therefore, we suggest that met allele, which has low activity of COMT is one of the risk allele for bipolar affective disorder.

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