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5-HTR2A Polymorphisms rs6311 and rs6313 and Major Depressive Disorder: A Meta-Analysis

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Abstract

rs6311 and rs6313 are two Single Nucleotide Polymorphisms (SNPs) on the Serotonin Receptor 2A gene (5-HTR2A) in complete linkage disequilibrium. Numerous gene association studies have examined the relationships between one or both of these two polymorphisms and Major Depressive Disorder (MDD), with conflicting results. The present meta-analysis examined 19 case-control gene association studies, 9 of which included rs6311 (n = 3382), and 15 of which included rs6313 (n = 5590). The strength of relationship with MDD was assessed by pooled odds ratios and 95% confidence intervals for both SNPs according to four genetic models. Heterogeneity was measured by Q and I^2. Subgrouping was performed by minor allele and by ethnicity. Results were nonsignificant for all models and subgroups, suggesting that genotype alone does not play a major role in genetic susceptibility to depression. The potential for epistatic, epigenetic, and regulatory RNA interactions with these SNPs is discussed, and future areas of research are recommended.

Keywords

Genetic Predisposition to Disease, Receptors, Serotonin, Polymorphism, Single Nucleotide, Genetics, Behavioral

1. Introduction

Major Depressive Disorder (MDD) affects over 264 million people worldwide, with a lifetime prevalence rate in the United States of 12% in men and 20% in women [1] [2]. Numerous genetic features have been proposed as possible etiological factors in the development of MDD, and twin studies suggest a heritabil-

ity for MDD of about 37% [1]. Many of the genetic features studied in relation to MDD are involved in the serotonergic system, as it has been widely implicated in depression. Many aspects of the serotonergic system have demonstrable differences in function in MDD patients, especially the serotonin 2A receptors, making them likely candidates for genetic associations with depression.

Serotonin 2A receptors (5-HTR2A) are G-protein coupled receptors in the serotonin receptor family, which consists of 7 subfamilies and 14 subtypes [3] [4]. 5-HTR2A is heavily expressed in the cerebral cortex, particularly the middle layers, as well as the basolateral amygdala and anterior cingulate cortex [5]. 5-HTR2A expression has been found to be altered in relation to MDD along several metrics, perhaps suggesting an involvement in development of depression. For example, unmedicated, fully recovered MDD patients demonstrated higher 5-HTR2A binding than controls, while the binding of 5-HTR2A in the prefrontal cortex of suicide victims was also elevated [6] [7]. Central 5-HTR2A density decreased after antidepressant treatment, and 5-HTR2A downregulation following antidepressant treatment occurred in conjunction with a reduction of symptoms [8] [9]. Converging evidence points toward a role of 5-HTR2A in the development of depression and in the amelioration provided by antidepressants, and further research into genetic variants of 5-HTR2A is needed to elucidate its effects.

The human 5-HTR2A gene is located on chromosome 13q14q21 and was originally believed to consist of 3 exons and 2 introns, being 63 kilobases (kb) in total length [3]. However, recent research suggests that it may actually contain 7 exons and cover 66 kb, with a parallel antisense gene 5-HTR2A-AS1, which is transcribed into a lncRNA [10]. The 5-HTR2A promoter has 4 potential transcription initiation sites: −1157, −1137, −1127, and −496 [11]. It consists of a primary promoter followed by the first three initiation sites, which are all within 30 base pairs (bp) of each other on the same side of the double helix, followed by a silencer, a secondary promoter, and the fourth initiation site at −496 [11]. The first SNP of interest in the present study, rs6311, is located at site −1438 in the primary promoter and can be either an A or a G allele, which are found in approximately equal proportions in the population [12]. The second SNP of interest, rs6313, is located at site 102 in the middle of exon 1 and consists of a T and a C allele, also found in approximately equal proportions in the populations, both of which code for a serine in codon 34 [3]. The two SNPs, rs6311 and rs6313, are in complete linkage disequilibrium such that the A and T alleles always appear together [3].

Both rs6311 and rs6313 have been implicated in a variety of psychological disorders, often with conflicting results. One meta-analysis found that the rs6313 C allele carried increased risk of schizophrenia in studies where C was the minor allele, while a different meta-analysis found that rs6311 A, in complete linkage disequilibrium with rs6313 T, had an increased risk of schizophrenia [4] [13]. A meta-analysis by Lin *et al.* [8] found the T allele of rs6313 to be correlated with better response rates to antidepressants. Genis-Mendoza *et al.* [14] found the G

allele of rs6311 to be highly correlated with increased odds of an eating disorder. The C allele of rs6313 has been found to be associated with seasonal patterns in depression [15].

The literature regarding rs6311 and rs6313 in relation to Major Depressive Disorder is mixed. Some studies have found the rs6313 C allele [16] [17] or the rs6311 G allele [18] [19] to be correlated with increased odds of depression. On the other hand, many other studies have found no relationship between either SNP and MDD [20]-[25]. Several meta-analyses have also found no relationship with depression [13] [26] [27]. However, the most recent meta-analysis was published in 2014, and further research has been conducted since then which must be taken into account. In addition, no prior meta-analysis examining depression has subgrouped by minor allele, a detail which Sun *et al.* [4] found to be significant in the relationship of rs6313 to schizophrenia. Therefore, we conducted a meta-analysis examining rs6311 and rs6313 and their relationships with depression, subgrouping by minor allele and ethnicity, to evaluate the MDD risk associated with these two polymorphisms.

2. Methods

2.1. Search Strategy

A systematic electronic literature search was performed in the PubMed database to identify relevant articles. Three sets of keywords were used, combined with the Boolean "AND." First, the relevant gene and SNP was identified using the list: 5-HTR2A OR 5-HT2A OR "5-HT 2A" OR rs6313 OR 102T/C OR 102C/T OR T102C OR rs6311 OR 1438A/G OR 1438G/A OR A1438G. This was combined with the list: polymorphism OR genetic OR SNP OR mutation. Finally, the list "depression" OR "depressive disorder" OR "depressive disorders" was combined to select only research relating to depression. References in all selected literature were reviewed to identify additional sources missed by the database search.

2.2. Selection Criteria

All sources included in the final meta-analysis were consistent with the following inclusion criteria: 1) they involved rs6311 and/or rs6313; 2) they contained original, independent data; 3) they were case-control studies using human subjects; 4) the control groups were in Hardy-Weinberg equilibrium (HWE); 5) they were published in a peer-reviewed journal and available online in English; 6) they specified genotyping methods and diagnostic criteria; and 7) the case groups were diagnosed with Major Depressive Disorder or Unipolar Affective Disorder (UPAD). Studies were excluded according to the following exclusion criteria: 1) they used datasets overlapping with another study (in which case the larger dataset was retained); 2) they were family studies; 3) the control groups deviated from HWE; 4) cases were selected using additional criteria to an MDD or UPAD diagnosis; 5) they were meeting abstracts, case reports, editorials, or review articles; or 6) they were not available online in English. Titles and ab-

stracts were first reviewed to identify relevant studies. The full text was then examined to ensure compliance with inclusion and exclusion criteria.

2.3. Data Extraction

The following information was extracted from every participating study: first author's name, year of publication, location of research, sample ethnicity, genotyping method, diagnostic criteria, sources of case and control groups, and number of each genotype in each group. Allele frequencies, minor allele, and HWE were calculated from genotype frequencies.

2.4. Statistical Analysis

A chi-square test was used to evaluate HWE in control groups. Odds ratios and 95% confidence intervals were used to assess strength of relationship between SNPs and MDD, while the associated Z score and P value evaluated significance. Relationships were assessed using four genetic models: allelic (T vs. C and A vs. G); dominant (TT vs. TC & CC and AA vs. AG & GG); recessive (TT & TC vs. CC and AA & AG vs. GG); and homozygous (TT vs. CC and AA vs. GG). In addition, subgroup analysis was performed by minor allele (T or C and A or G) and ethnicity (Asian or Caucasian). Some ethnicities were marked "other" and were not included in either ethnic subgroup but were included in the total meta-analysis. Heterogeneity was measured with Q and I^2. A random effects model and inverse variance algorithm were used for all analyses, chosen for their robustness and ability to handle both large and small sample sizes with both high and low heterogeneity. All statistical analyses were conducted using PyMeta version 1.11 [28].

3. Results

3.1. Search Results

The initial PubMed database search returned 432 results. 36 studies were selected on the basis of title and abstract, and 20 of these studies were eliminated upon examination of full text. 4 studies were eliminated because they measured depressive symptoms rather than diagnoses, 3 because the control groups were not in HWE, 2 because they contained overlapping datasets, 2 because they did not distinguish between MDD and other mood disorders, 2 because they provided allele frequencies but not genotype frequencies, 2 because they did not use controls, 2 because cases were not only depressed but also suicidal, 1 because it did not contain original data, 1 because it was not relevant to the SNPs at hand, and 1 because the full text was not available in English. Sixteen studies were chosen on the basis of full text and another 3 were identified through literature tracing, for a total number of 19 studies.

3.2. Study Characteristics

A total of 15 studies were identified for rs6313 (n = 5590) and 9 studies for

rs6311 (n = 3382). For rs6313, T was the minor allele in 6 studies (n = 1732) and C was the minor allele in 8 studies (n = 3657). Five studies were performed with Caucasian subjects (n = 1590) and 6 studies were performed with Asian subjects (n = 3194). For rs6311, A was the minor allele in 6 studies (n = 3073) and G was the minor allele in 3 studies (n = 759). Three studies were performed with Caucasian subjects (n = 1190) and 4 studies were performed with Asian subjects (n = 2237). Detailed ethnicity, as well as genotyping method, diagnostic criteria, and genotype frequencies for each study can be found in **Tables 1-4**.

Table 1. Characteristics of the studies included in the meta-analysis for rs6311.

Study	Country/Area	Ethnicity	Genotyping Method	Diagnostic Criteria	Case (n)	Control (n)
Bonnier *et al.*, 2002 [29]	France	Caucasian	PCR	DSM-IV	65	142
Terayama *et al.*, 2003 [23]	Japan	Asian	PCR-Msp I	DSM-IV	39	112
Choi *et al.*, 2004 [18]	Korea	Asian	PCR-Msp I	DSM-IV	189	148
Illi *et al.*, 2009 [21]	Finland	Caucasian	Taqman Assay	DSM-IV	86	395
Noskova *et al.*, 2009 [19]	Russia	Caucasian	PCR-Msp I	ICD-10	174	328
Tencomnao *et al.*, 2010 [30]	Thailand	Asian	PCR-Msp I	DSM-IV	180	183
Yusup *et al.*, 2013 [31]	China-Uighur (China)	Other	PCR-Msp I	CCMD-3	195	50
Zhang *et al.*, 2016 [25]	China	Asian	MALDI-TOF	DSM-IV	558	842
Basu *et al.*, 2019 [20]	India	Other	SEQUENOME	DSM-IV	80	80

Table 2. Characteristics of the studies included in the meta-analysis for rs6313.

Study	Country/Area	Ethnicity	Genotyping Method	Diagnostic Criteria	Case (n)	Control (n)
Zhang *et al.*, 1997 [32]	Japan	Asian	PCR-SSCP	DSM-IV	51	150
Frisch *et al.*, 1999 [33]	Israel	Caucasian	PCR-Msp I	DSM-IV	99	172
Tsai *et al.*, 1999 [34]	Taiwan (China)	Asian	PCR-HpaII	DSM-IV	79	96
Bondy *et al.*, 2000 [35]	Germany	Caucasian	PCR-Msp I	DSM-IV	84	125
Arias *et al.*, 2001 [15]	Spain	Caucasian	PCR-Msp I	DSM-IV	159	164
Minov *et al.*, 2001 [36]	Germany	Caucasian	PCR-Msp I	DSM-IV	172	121
Correa *et al.*, 2002 [37]	Brazil	Other	PCR-Hpa II	DSM-IV	78	52
Oswald *et al.*, 2003 [38]	Europe	Caucasian	PCR-Msp I	DSM-IIIR	142	142
Terayama *et al.*, 2003 [23]	Japan	Asian	PCR-Msp I	DSM-IV	39	112
Illi *et al.*, 2009 [21]	Finland	Caucasian	Taqman Assay	DSM-IV	86	395
Kishi *et al.*, 2009 [22]	Japan	Asian	Taqman Assay	DSM-IV	325	802
Yusup *et al.*, 2013 [31]	China-Uighur (China)	Other	PCR-Msp I	CCMD-3	195	50
Guo *et al.*, 2014 [39]	China	Asian	PCR-Msp I	DSM-IV	72	70
Zhang *et al.*, 2016 [25]	China	Asian	MALDI-TOF	DSM-IV	558	842
Basu *et al.*, 2019 [20]	India	Other	SEQUENOME	DSM-IV	80	80

Table 3. Distribution of rs6311 genotype between cases and controls.

Study	Case			Control			HWE P value
	AA	AG	GG	AA	AG	GG	
Bonnier *et al.*, 2002 [29]	10	34	21	20	64	58	0.7291
Terayama *et al.*, 2003 [23]	9	16	14	29	61	22	0.3221
Choi *et al.*, 2004 [18]	24	117	48	39	82	27	0.1602
Illi *et al.*, 2009 [21]	11	41	34	45	178	172	0.9181
Noskova *et al.*, 2009 [19]	17	75	82	49	154	125	0.8891
Tencomnao *et al.*, 2010 [30]	110	69	1	116	64	3	0.0775
Yusup *et al.*, 2013 [31]	63	112	20	18	21	11	0.3112
Zhang *et al.*, 2016 [25]	109	254	183	166	423	251	0.6108
Basu *et al.*, 2019 [20]	15	36	29	12	38	30	0.9953

Table 4. Distribution of rs6313 genotype between cases and controls.

Study	Case			Control			HWE P value
	TT	TC	CC	TT	TC	CC	
Zhang *et al.*, 1997 [32]	12	18	21	45	69	36	0.3477
Frisch *et al.*, 1999 [33]	34	45	20	46	77	49	0.1710
Tsai *et al.*, 1999 [34]	26	44	9	36	50	10	0.2239
Bondy *et al.*, 2000 [35]	12	49	23	27	58	40	0.4892
Arias *et al.*, 2001 [15]	35	81	43	30	81	53	0.9222
Minov *et al.*, 2001 [36]	31	90	51	26	56	39	0.4844
Correa *et al.*, 2002 [37]	20	43	15	15	27	10	0.7284
Oswald *et al.*, 2003 [38]	38	70	34	31	71	40	0.9617
Terayama *et al.*, 2003 [23]	8	22	7	33	62	17	0.1682
Illi *et al.*, 2009 [21]	11	41	34	45	181	169	0.7408
Kishi *et al.*, 2009 [22]	87	154	84	220	386	196	0.3007
Yusup *et al.*, 2013 [31]	64	113	18	19	20	11	0.2057
Guo *et al.*, 2014 [39]	18	28	26	19	29	22	0.1552
Zhang *et al.*, 2016 [25]	194	254	110	251	420	171	0.8456
Basu *et al.*, 2019 [20]	31	32	17	27	41	12	0.5771

3.3. Meta-Analysis Results

Using odds ratios and confidence intervals, no significant relationships were found between MDD and either SNP in any of the four genetic models. The odds ratios and confidence intervals for each model of rs6311 were: allelic model (OR = 1.09, CI: 0.95 - 1.26, P = 0.230); dominant model (OR = 1.17, CI: 0.93 - 1.47, P = 0.174); recessive model (OR = 1.06, CI: 0.81 - 1.39, P = 0.653); and

homozygous model (OR = 1.15, CI: 0.80 - 1.67, P = 0.457) (Table 5). The odds ratios and confidence intervals for each model of rs6313 were: allelic model (OR = 0.96, CI: 0.89 - 1.04, P = 0.343); dominant model (OR = 0.95, CI: 0.83 - 1.07, P = 0.389); recessive model (OR = 0.95, CI: 0.81 - 1.11, P = 0.505); and homozygous model (OR = 0.94, CI: 0.79 - 1.10, P = 0.423) (Table 6, Figure 1). No significant results were found for any subgroup model. There was a trend toward increased risk in the Asian subgroup of rs6311 for the allelic and recessive model: allelic model (OR = 1.20, CI: 0.99 - 1.47, P = 0.064); recessive model (OR: 1.36, CI: 0.97 - 1.88, P = 0.075) (Table 5, Figure 2).

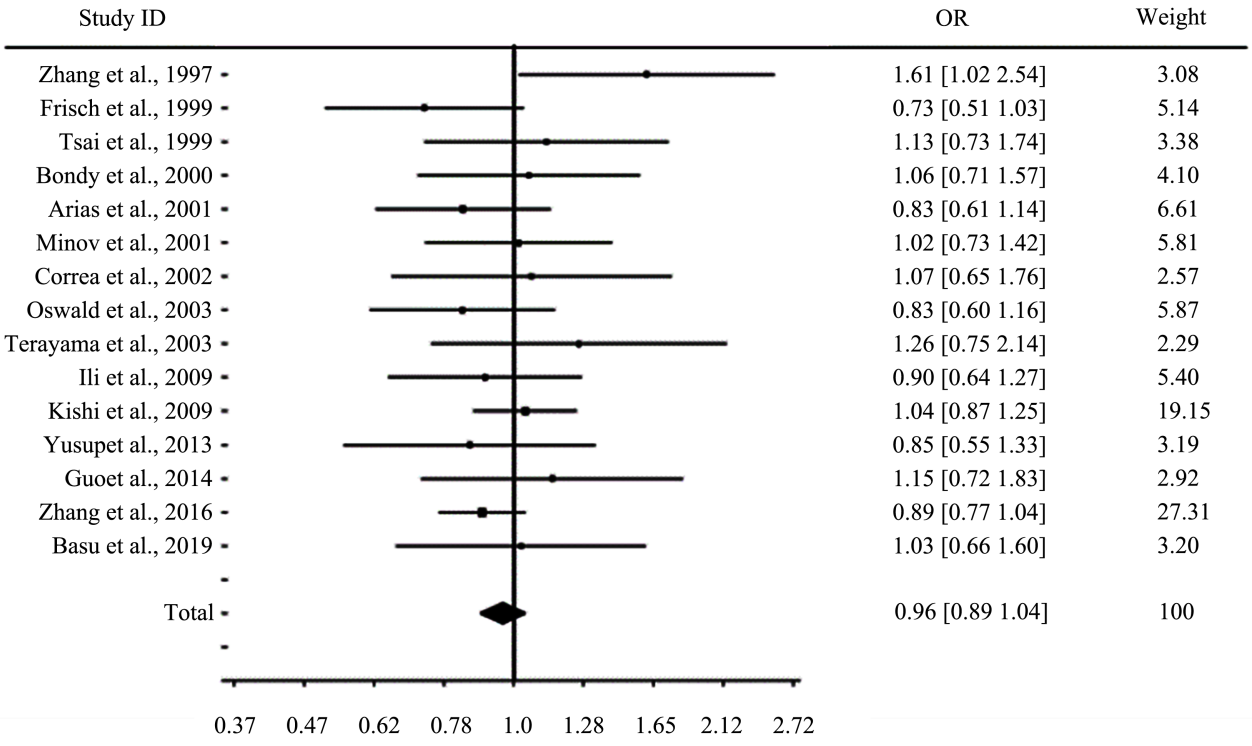

Figure 1. Forest plot of rs6313 allelic model.

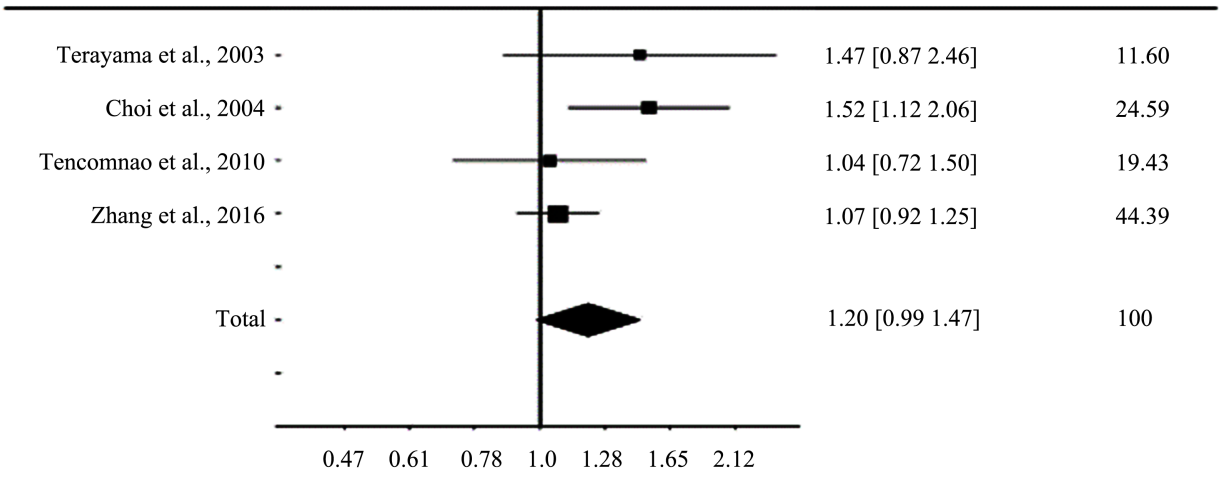

Figure 2. Forest plot of rs6311 Asian subgroup allelic model.

Table 5. Meta-analysis results for rs6311.

Genetic Model	Subgroup	OR	95% CI	P value	Heterogeneity		
					Q	P value	I^2
Allelic		1.09	[0.95, 1.26]	0.230	13.58	0.094	41.08
	A minor allele	1.10	[0.92, 1.32]	0.309	10.95	0.053	54.34
	G minor allele	1.06	[0.80, 1.40]	0.684	2.49	0.289	19.84
	Caucasian	1.02	[0.73, 1.43]	0.901	5.78	0.056	65.38
	Asian	1.20	[0.99, 1.47]	0.064	5.11	0.171	41.32
Dominant		1.17	[0.93, 1.47]	0.174	11.42	0.183	29.92
	A minor allele	1.19	[0.84, 1.71]	0.331	11.38	0.045	56.06
	G minor allele	1.13	[0.81, 1.57]	0.467	0.03	0.984	0
	Caucasian	1.16	[0.76, 1.76]	0.492	2.23	0.332	10.13
	Asian	1.28	[0.86, 1.90]	0.221	8.27	0.041	63.71
Recessive		1.06	[0.81, 1.39]	0.653	17.38	0.027	53.97
	A minor allele	1.13	[0.91, 1.39]	0.267	7.15	0.211	30.07
	G minor allele	0.78	[0.19, 3.12]	0.721	9.62	0.008	79.2
	Caucasian	0.99	[0.63, 1.56]	0.968	5.38	0.068	62.8
	Asian	1.36	[0.97, 1.88]	0.075	4.21	0.244	28.79
Homozygous		1.15	[0.80, 1.67]	0.457	17.10	0.029	53.22
	A minor allele	1.24	[0.83, 1.85]	0.298	11.59	0.041	56.85
	G minor allele	0.85	[0.28, 2.52]	0.764	4.66	0.098	57.06
	Caucasian	1.10	[0.58, 2.07]	0.780	4.31	0.121	53.57
	Asian	1.56	[0.81, 3.01]	0.187	8.15	0.043	63.19

Table 6. Meta-analysis results for rs6313.

Genetic Model	Subgroup	OR	95% CI	P value	Heterogeneity		
					Q	P value	I^2 (%)
Allelic		0.96	[0.89, 1.04]	0.343	13.64	0.478	0
	C minor allele	0.95	[0.86, 1.05]	0.328	6.10	0.529	0
	T minor allele	0.93	[0.81, 1.08]	0.355	2.40	0.792	0
	Caucasian	0.91	[0.79, 1.06]	0.248	1.57	0.813	0
	Asian	1.06	[0.91, 1.24]	0.443	7.86	0.171	36.41
Dominant		0.95	[0.83, 1.07]	0.389	12.08	0.600	0
	C minor allele	0.92	[0.79, 1.06]	0.260	6.62	0.471	0
	T minor allele	0.99	[0.76, 1.27]	0.920	4.13	0.532	0
	Caucasian	0.97	[0.74, 1.28]	0.832	4.01	0.405	0.23
	Asian	0.97	[0.81, 1.16]	0.723	5.52	0.358	9.48
Recessive		0.95	[0.81, 1.11]	0.505	17.79	0.218	21.32
	C minor allele	0.94	[0.74, 1.18]	0.584	9.58	0.216	26.9
	T minor allele	0.86	[0.69, 1.07]	0.186	1.34	0.917	0
	Caucasian	0.83	[0.66, 1.04]	0.113	0.21	0.981	0
	Asian	1.11	[0.91, 1.34]	0.293	5.36	0.376	6.68
Homozygous		0.94	[0.79, 1.10]	0.423	14.23	0.434	1.59
	C minor allele	0.91	[0.74, 1.11]	0.348	7.19	0.410	2.58
	T minor allele	0.90	[0.67, 1.21]	0.478	2.88	0.718	0
	Caucasian	0.86	[0.63, 1.18]	0.349	2.25	0.690	0
	Asian	1.08	[0.83, 1.40]	0.564	6.12	0.295	18.34

4. Discussion

In the current study, we performed a systematic analysis of the relationship between rs6311, rs6313, and susceptibility to MDD based on 19 studies containing 2827 cases and 4174 controls. No significant associations were found in any of four genetic models. In addition, the effect of ethnicity was controlled for by dividing into subgroups based on ethnicity, with a subgroup for studies with Asian subjects and a subgroup for studies with Caucasian subjects. No significant association was found in either subgroup for any of the four models. Finally, effect of allele frequency was assessed by dividing into subgroups based on minor allele. No significant association was found for either minor allele in any of the four genetic models. There was a trend toward increased risk in the Asian subgroup of rs6311 in the recessive and allelic model, but it was not significant. Therefore, this study suggests the absence of a relationship between either rs6311 or rs6313 and MDD. These findings are consistent with those of meta-analyses conducted by Gu *et al.* [13], Jin *et al.* [26], and Tan *et al.* [27].

While neither rs6313 nor rs6311 appear to have a significant association with MDD, it is possible that confounding factors such as epistasis, epigenetic regulation, or RNA regulation mask a true effect. For example, the SNP −783A/G exists in significant linkage disequilibrium with rs6311 and displays some epistatic effects [12]. The −783A/G minor allele, G, has a prevalence rate of 9% and nearly always occurs with the G allele of rs6311 [12]. A study of promoter effectiveness by Myers *et al.* [12] found that, when expressed with the A allele of −783, there was no discernible difference in promoter strength between the A and G alleles of rs6311. However, the rs6311G/−783G haplotype had significantly reduced promoter effectiveness. It is possible that the rs6311G/−783G haplotype also corresponds with increased odds of MDD, but since no meta-analysis has included −783G, this effect has not been noticed. To the best of our knowledge, no study has ever investigated the relationship of −783A/G to depression, much less the relationships of rs6311/−783A/G haplotypes to depression. Future studies investigating the haplotype relationships of rs6311 and −783A/G to MDD could provide needed clarity to these interactions.

Epigenetic regulation may also account for a real but nuanced relationship of rs6311 and rs6313 with MDD. Both SNPs create a CpG site that is differentially methylated in individuals with bipolar disorder or schizophrenia. Specifically, site −1439 was found to be hypermethylated and site 102 was found to be hypomethylated in both individuals with schizophrenia and individuals with bipolar disorder [40]. Genotype plays a significant role in methylation, as individuals with the rs6311 GG genotype had increased methylation at −1439, and individuals with the rs6313 TT genotype were obviously unmethylated at 102 [41]. In addition to −1439, there are two other common methylation sites in the promoter region, −1421 and −1522. Methylation at all three sites is highly correlated with each other, so that individuals with the rs6311 GG genotype displayed increased methylation at all three sites in the promoter region [41]. 5-HTR2A is

also differentially expressed in some psychiatric disorders, as individuals with schizophrenia and individuals with bipolar disorder who had been antipsychotic free for at least a month displayed reduced levels of gene expression [40]. Gene expression was associated with genotype, with conflicting results. Abdolmaleky *et al.* [40] found increased expression in individuals with the rs6313 CC genotype compared to the rs6313 TT genotype. However, Polesskaya and Sokolov [42] found reduced expression of both mRNA and protein in individuals with the CC genotype compared to the TT genotype, and reduced expression of the C allele compared to the T allele in heterozygous individuals. Differential methylation at −1439 and 102 and differential expression of 5-HTR2A in individuals with MDD could indicate a real risk of MDD, but the mechanisms are not yet clear, and future studies are needed to investigate not only SNPs but also epigenetic regulation and gene expression of 5-HTR2A in relation to MDD.

An effect of rs6311 and rs6313 on RNA expression and regulation has also been proposed. The rs6311G allele has been associated with increased expression of an isoform transcript of 5-HTR2A containing an extended 5' UTR, the effects of which are unknown [10]. The rs6311A and rs6313T alleles have also been associated with increased expression of 5-HTR2A-AS1, particularly exon 14 [10]. This parallel antisense lncRNA spans 474kb and consists of 18 exons, which overlap with 3 5-HTR2A exons [10]. This suggests that 5-HTR2A-AS1 is involved in post-transcriptional processing and regulation of 5-HTR2A, although such a role has not yet been confirmed. In addition, total mRNA expression of 5-HTR2A was lower in individuals with the rs6313 TT allele than individuals with the rs6313 CC allele, and lower in rs6311 heterozygotes than either rs6311 homozygote genotype [10]. While the mechanisms of RNA transcription, processing, and regulation of 5-HTR2A have not yet been fully described, it is possible that rs6313 and rs6311 play a role in these processes, and further research is needed to elucidate it.

Heterogeneity was low for rs6313, but reached significance in several models and subgroups of rs6311, with multiple possible explanations. Allele frequency has been shown to vary by ethnicity [13], and our studies included numerous different ethnicities, with heterogeneity differing across ethnic subgroups. In addition, control subjects were selected in several different ways across studies. Some studies sampled individuals from the general population, some from a database, some from individuals attending a hospital for a regular health screening, and some from staff and students at a hospital. Many did not state the source of controls. Some studies matched controls to cases by gender, age, and/or ethnicity, but many did not. Most studies screened controls to exclude any with mental health disorders, but some did not. All of these variations could introduce heterogeneity to the study. Diagnostic criteria also differed across studies, with most diagnosing MDD according to the DSM-IV definition, but some using the DSM-IIIR, ICD-10, or CCMD-3 criteria. Behavioral differences such as suicidality and seasonal patterns may also have been obscured by a blanket diagnosis,

and have been suggested to be differentially associated with rs6311 and rs6313 [15] [42] [43]. In addition, subjects were not separated by sex in most studies, so differences in genotype effect across sex could not be analyzed.

This meta-analysis has several limitations. First, heterogeneity may influence the reliability of results. Second, key features of depression such as suicidality, seasonality, and treatment resistance were not available in most studies and therefore could not be classified into subgroups, possibly obscuring relationships between these aspects and rs6311 or rs6313. Third, publication bias is possible, as unpublished studies and studies not published in English were excluded from consideration. Fourth, some subgroups suffered from small size, especially the G minor allele subgroup for rs6311.

5. Conclusion

Depression is known to have underlying genetic factors, but specific causal factors are not known. Understanding these factors would allow us to identify individuals at risk for depression and potentially tailor pharmacological approaches to the individual. This study used statistical techniques to pool results from numerous studies and evaluate two potential causal factors. This meta-analysis failed to find a significant association between rs6311 or rs6313 and MDD in four genetic models. No association was found in subgroups by ethnicity or by minor allele. Therefore, rs6311 and rs6313 may not be risk factors for MDD. Future studies should investigate rs6311/−783A/G haplotypes, methylation at sites −1439 and 102, and RNA expression and regulation of −5HTR2A in association with MDD.

Conflicts of Interest

The authors declare no conflicts of interest associated with the publication of this paper.

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