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META-ANALYSIS

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Association between EXOC3L2 rs597668 Polymorphism and Alzheimer's Disease

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Keywords
Alzheimer's disease; Caucasian; East Asian; *EXOC3L2* rs597668 polymorphism; Genome-wide association studies.

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SUMMARY

Background: *EXOC3L2* gene rs597668 polymorphism was identified to be significantly associated with Alzheimer's disease (AD) in Caucasian population. However, recent studies reported consistent and inconsistent results in Caucasian and Asian populations. **Aims:** In order to assess this association, we performed a meta-analysis of rs597668 polymorphism using RevMan (v.5.1) software. **Methods:** We searched PubMed and Google scholar databases and selected 4 independent publications, which included 16 independent studies. We conducted sensitivity analysis and evaluated the publication bias. In the end, we calculated the odds ratio (OR) using fixed effect model (Mantel-Haenszel). **Results:** We observed significant association between rs597668 polymorphism and AD using allele model (P = 0.006, OR = 1.09, 95% CI 1.03–1.16) and the dominant model (P = 0.008, OR = 1.11, 95% CI 1.03–1.21). **Discussion and Conclusions:** To our knowledge, this is the first study that assesses the association between rs597668 polymorphism and AD by meta-analysis. We believe that our findings will be very useful for future genetic studies in AD.

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Introduction

Alzheimer's disease (AD) is a complex and neurodegenerative disease in the elderly [1]. It is estimated that there are 35.6 million dementia people in the world in 2010 [2]. This number will be 65.7 million in 2030 [2]. In order to identify common AD variants, large-scale genome-wide association studies (GWAS) of AD have been conducted [3–7]. Some common AD variants in new AD susceptibility genes (*CR1*, *BIN1*, *CLU*, *PICALM*, *MS4A4*/*MS4A6E*, *CD2AP*, *CD33*, *EPHA1*, *EXOC3L2* and *ABCA7*) have been reported [3–7].

A single-nucleotide polymorphism (SNP) rs597668 near *EXOC3L2* gene was reported to be significantly associated with AD in Caucasian population (P = 6.450E−09) [5]. The following studies investigated the association and reported consistent and inconsistent results in Caucasian population and Asian population. Carrasquillo et al. investigated the rs597668 polymorphism in a large dataset from the USA and Europe (3287 AD cases and 4396 controls). The result showed that rs597668 variant was not significantly associated with AD (P = 0.090) [8]. Lambert et al. analyzed rs597668 polymorphism in three contrasting European populations (Finland, Italy, and Spain) with 2816 AD cases and 2706 controls. They successfully replicated this association with P = 2.000E−03 [9].

In addition to the Caucasian population, this polymorphism was also investigated in Chinese and Japanese populations. Liu et al. investigated rs597668 polymorphism in 1205 unrelated Northern Han Chinese subjects (598 AD patients and 607 healthy controls). However, they did not report significant association between rs597668 and AD using genotype test (P = 0.653) and allele test (P = 0.603) [10]. Ohara et al. investigated rs597668 polymorphism using 3758 Japanese subjects (825 AD cases and 2933 controls). However, they still did not report any significant association (P = 0.440) [11]. Considering the inconsistent results, in this research, we investigated the association between rs597668 and AD by a meta-analysis.

Materials and Methods

Literature Search and Inclusion Criteria

We searched PubMed and Google scholar databases to select all possible studies using the key words "Alzheimer's disease", "*EXOC3L2*". The literature search was updated on 3/20/2013. We included the studies meeting the following criteria: (1) the study evaluated the association between rs597668 polymorphism and AD by a case-control design and (2) the study provided the numbers of rs597668 genotype. In the end, we selected four indepen-

dent publications, which included 16 independent studies (Figure 1).

Data Extraction

For the case-control genetics studies, the following information was extracted from each study: (1) the name of the first author, (2) the year of publication, (3) the ethnicity of the studied population, (4) the number of cases and controls, (5) the genotype numbers of rs597668 polymorphism in cases and controls, and (6) the type of AD (Table 1).

Genetic Model

In order to ensure that interesting findings were not missed because of the different methods, we used three genetic mod-

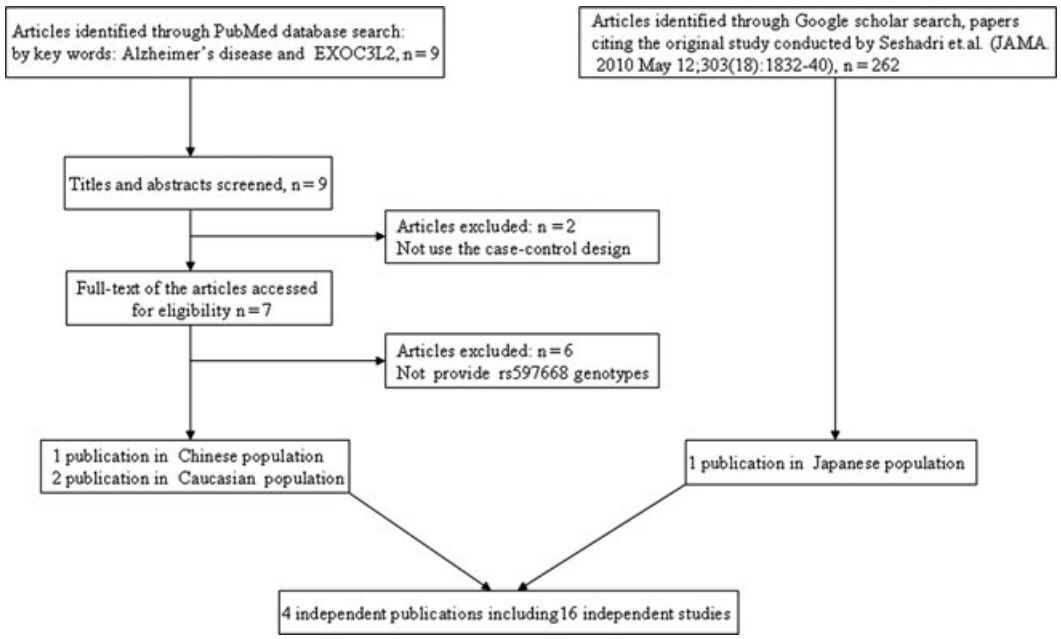

Figure 1 Flow chart of meta-analysis for exclusion or inclusion of individual articles.

Table 1 Characteristics of 16 studies selected for meta-analysis

				Case genotype #				Control genotype #		
Study	Population	Type of AD	Case #	TT	TC	CC	Control #	TT	TC	CC
Carrasquillo 2011 [8]	Autopsy	LOAD	304	205	92	7	100	73	24	3
Carrasquillo 2011 [8]	Jacksonville	LOAD	500	332	150	18	959	684	246	29
Carrasquillo 2011 [8]	Norway	LOAD	338	224	102	12	544	351	175	18
Carrasquillo 2011 [8]	Rochester	LOAD	311	215	81	15	1617	1142	436	39
Carrasquillo 2011 [8]	Southampton	LOAD	36	24	11	1	128	79	44	5
Carrasquillo 2011 [8]	Bristol	LOAD	200	141	56	3	37	23	13	1
Carrasquillo 2011 [8]	Leeds	LOAD	113	72	36	5	275	190	81	4
Carrasquillo 2011 [8]	Man/Notts	LOAD	177	111	60	6	87	57	29	1
Carrasquillo 2011 [8]	NCRAD	LOAD	698	460	209	29	209	138	64	7
Carrasquillo 2011 [8]	Oxford	LOAD	97	64	28	5	204	149	50	5
Carrasquillo 2011 [8]	Poland	LOAD	470	273	169	28	182	128	51	3
Lambert 2011 [9]	Finland	LOAD	562	265	241	56	529	302	184	43
Lambert 2011 [9]	Italy	LOAD	1541	1115	336	90	1268	980	266	22
Lambert 2011 [9]	Spain	LOAD	627	559	54	14	832	669	150	13
Liu 2012 [10]	China	LOAD	571	261	236	74	607	267	275	65
Ohara 2012 [11]	Japan	LOAD	825	265	418	142	2933	937	1449	547
All	–	–	7370	4586	2279	505	10511	6169	3537	805

LOAD, Late-onset Alzheimer's disease.

els, which included the allele model (C vs. T), the dominant model (CC + CT vs. TT), and the recessive model (CC vs. CT + TT).

Heterogeneity Test

The heterogeneity among the selected studies was evaluated using Cochran's Q test. It approximately follows a χ^2 distribution with k-1 degrees of freedom (k is the number of studies for analysis) [12]. The threshold of significant difference is 0.01. $I^2 = (Q - (k-1))/Q \times 100\%$ was also used to evaluate the heterogeneity. Low, moderate, large and extreme heterogeneity corresponded to 0–25%, 25–50%, 50–75% and 75–100%, respectively [12].

Meta-Analysis

The fixed effect model (Mantel-Haenszel) or random-effect model (DerSimonian-Laird) was used to calculate the odds ratio (OR). If there is no significant heterogeneity among the studies included ($P > 0.01$ and $I^2 < 50\%$), odds ratio (OR) is calculated by the fixed effect model (Mantel-Haenszel). Otherwise, the OR is calculated by random-effect model (DerSimonian-Laird). The significance level of OR was determined using the *Z*-test. We used the RevMan (v.5.1) software to calculate all the tests above (http://ims.cochrane.org/revman).

Sensitivity Analysis

We performed a sensitivity analysis to illustrate the influence of specific studies. In sensitivity analysis, relative influence of each study was assessed by omitting one study at a time.

Publication Bias Analysis

The results of meta-analysis may be influenced by potential publication bias [13]. The likely presence or absence of bias should be routinely examined [13]. Here, we evaluated the potential publication bias using funnel plots. If there is no bias, the plot will be a symmetrical inverted funnel. Otherwise, it will be an asymmetrical inverted funnel [12].

Results

The Results from Meta-Analysis

We observed significant heterogeneity among these 16 studies using the allele ($P < 1.000E{-}05$ and $I^2 = 71\%$), the recessive ($P = 7.00E{-}04$ and $I^2 = 61\%$), and the dominant ($P < 1.00E{-}05$ and $I^2 = 71\%$) models. We then calculated the overall OR using random-effect model. Significant result was identified using recessive model ($P = 6.000E{-}03$, OR = 1.48, 95% CI 1.12–1.95). However, we did not identify significant association between rs597668 polymorphism and AD using allele model ($P = 0.100$, OR = 1.11, 95% CI 0.98–1.26) and dominant model ($P = 0.280$, OR = 1.08, 95% CI 0.94–1.25).

The Results from Sensitivity Analysis

To verify the findings from meta-analysis, we performed a sensitivity analysis and the relative influence of each study was assessed by omitting one study at a time. We found that the study from Lambert et al. (Italy and Spain) [9] played an important role in causing the different association between rs597668 polymorphism and AD (Table 2).

Table 2 Sensitivity analysis with each study omitted in meta-analysis

	C vs. T			CC vs. CT + TT			CC vs. CT + TT		
Study omitted	OR	95% CI	*P*	OR	95% CI	*P*	OR	95% CI	*P*
None	1.11	0.98–1.26	0.10	1.48	1.12–1.95	0.006	1.08	0.94–1.25	0.28
Carrasquillo 2011 Autopsy	1.11	0.97–1.26	0.13	1.51	1.14–2.01	0.004	1.07	0.92–1.25	0.36
Carrasquillo 2011 Jacksonville	1.10	0.98–1.26	0.16	1.51	1.12–2.05	0.007	1.07	0.91–1.25	0.41
Carrasquillo 2011 Norway	1.12	0.98–1.28	0.09	1.52	1.13–2.04	0.006	1.10	0.94–1.28	0.24
Carrasquillo 2011 Rochester	1.11	0.97–1.26	0.14	1.44	1.07–1.92	0.01	1.08	0.93–1.27	0.32
Carrasquillo 2011 Southampton	1.12	0.99–1.27	0.08	1.50	1.13–1.99	0.005	1.09	0.94–1.26	0.25
Carrasquillo 2011 Bristol	1.12	0.99–1.27	0.07	1.50	1.13–1.99	0.005	1.10	0.95–1.27	0.22
Carrasquillo 2011 Leeds	1.10	0.97–1.25	0.15	1.44	1.09–1.91	0.01	1.07	0.92–1.25	0.36
Carrasquillo 2011 Man/Notts	1.11	0.97–1.26	0.12	1.46	1.10–1.94	0.008	1.08	0.93–1.26	0.31
Carrasquillo 2011 NCRAD	1.12	0.98–1.27	0.11	1.50	1.12–2.01	0.007	1.09	0.93–1.27	0.28
Carrasquillo 2011 Oxford	1.10	0.97–1.25	0.15	1.46	1.10–1.94	0.009	1.07	0.92–1.24	0.37
Carrasquillo 2011 Poland	1.08	0.96–1.22	0.22	1.44	1.08–1.87	0.01	1.05	0.91–1.21	0.50
Lambert 2011 Finland	1.09	0.96–1.24	0.19	1.52	1.11–2.07	0.009	1.05	0.91–1.20	0.49
Lambert 2011 Italy	1.08	0.96–1.22	0.21	1.13	0.99–1.30	**0.08**	1.06	0.91–1.24	0.44
Lambert 2011 Spain	1.16	1.05–1.29	**0.006**	1.49	1.11–2.00	0.009	1.15	1.07–1.23	**3.00E−04**
Liu 2012 China	1.12	0.98–1.28	0.11	1.52	1.11–2.10	0.01	1.10	0.94–1.28	0.24
Ohara 2012 Japan	1.12	0.98–1.29	0.09	1.62	1.36–1.93	5.96E−08	1.09	0.93–1.28	0.29

The bold indicates the significance level is 0.05.

After both studies were excluded, we did not identify significant heterogeneity among the remaining 14 studies for allele model ($P = 0.030$ and $I^2 = 47\%$), the recessive model ($P = 0.170$ and $I^2 = 26\%$), and the dominant model ($P = 0.050$ and $I^2 = 41\%$). We calculated the OR using fixed effect model. The robust association between rs597668 polymorphism and AD was detected using allele model ($P = 0.006$, OR = 1.09, 95% CI 1.03–1.16) and the dominant model ($P = 0.008$, OR = 1.11, 95% CI 1.03–1.21; Figure 2).

The Result from Publication Bias Analysis

We evaluated the potential publication bias among the 14 studies using funnel plots. Interestingly, all the three plots were symmetrical inverted funnels for allele model, the recessive model, and the dominant model, which indicated no publication bias (Figure 3).

Discussion

Previous studies investigated the association between rs597668 and AD in Caucasian and East Asian populations and reported consistent and inconsistent results. In this research, we performed a meta-analysis. We first evaluated the genetic heterogeneity among the selected studies and found significant heterogeneity. After the sensitivity analysis, we found that the study from Lambert et al. (Italy and Spain) contributed to the large heterogeneity. After removing both studies, no heterogeneity was observed. We calculated the OR using fixed effect model and found that rs597668 polymorphism was significantly

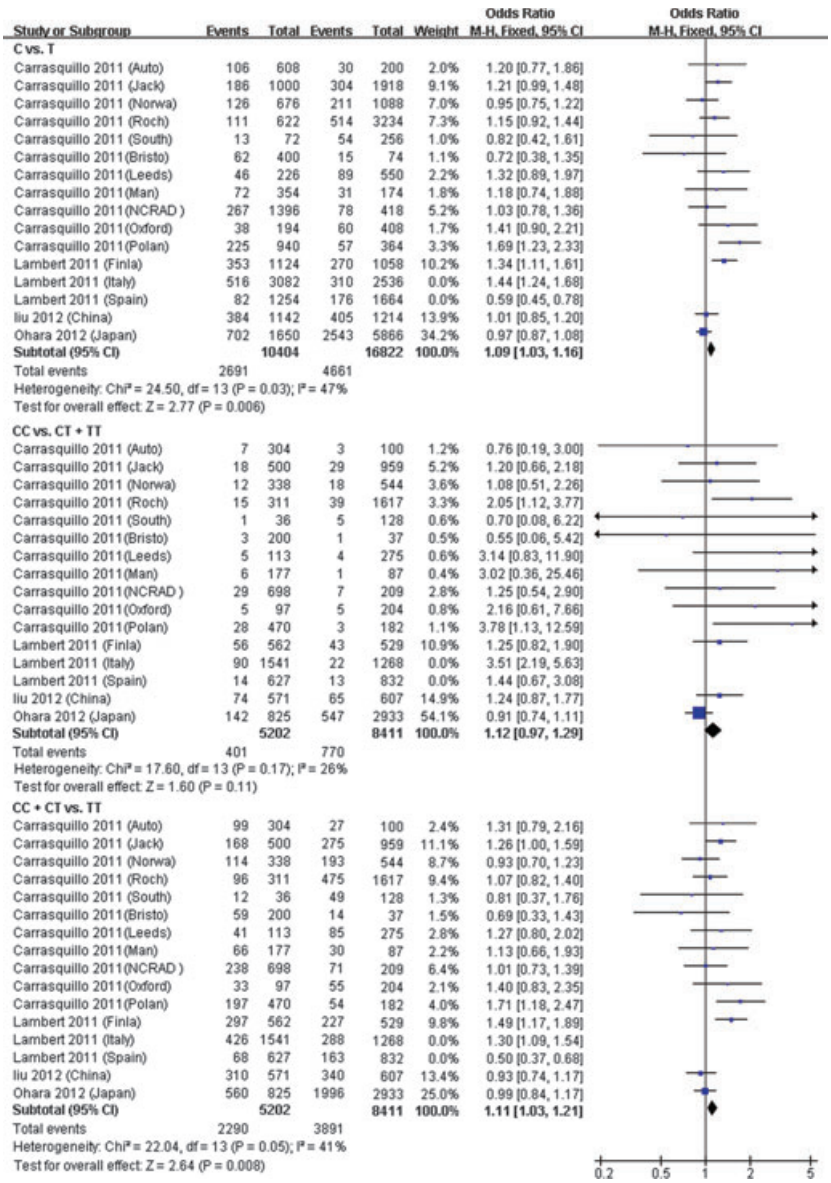

Figure 2 Forest plot for the meta-analysis of rs597668 polymorphism in AD.

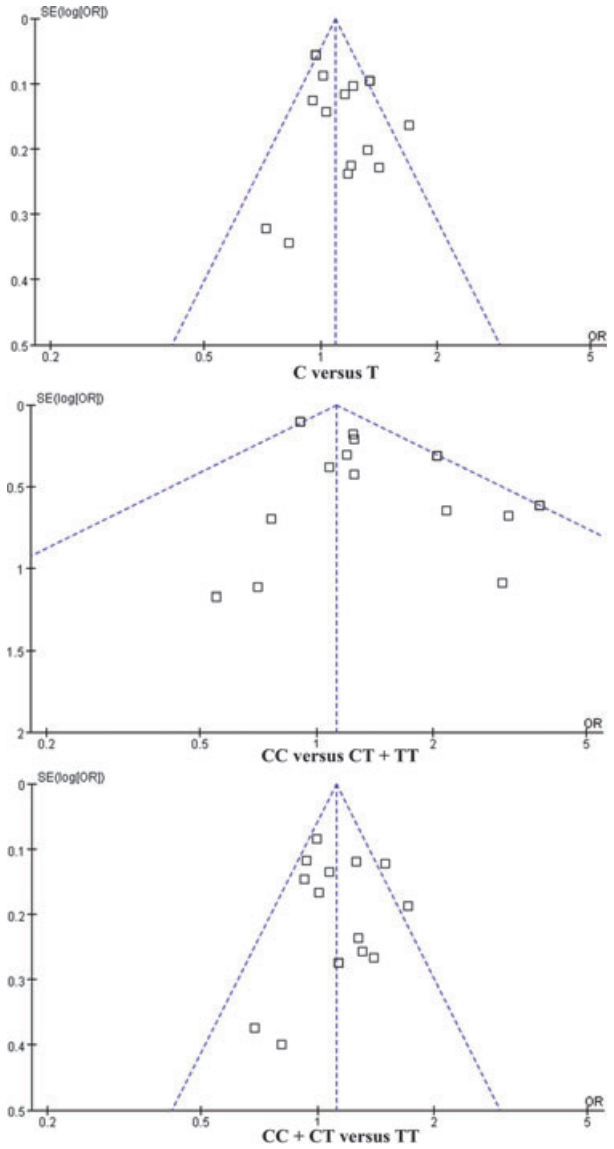

Figure 3 The publication bias analysis of rs597668 polymorphism in AD.

associated with AD. Using the funnel plots, we identified no publication bias.

Recent study investigated the mechanisms of rs597668 polymorphism in AD pathogenesis [14]. Schmidt et al. analyzed rs597668 polymorphism of 40 AD patients by a longitudinal study. They identified that *EXOC3L2* rs597668 polymorphism was significantly associated with more aggressive disease courses [14]. The C allele was associated with the risk of faster decline [14]. For rs597668 polymorphism, in addition to being a risk factor for disease development, it also might act as prognostic disease marker [14]. Considering the findings from our study and previous studies, rs597668 polymorphism may be a new therapeutic target to treat or prevent AD.

In our research, considering only two studies in Asia population and the results were consistent, we did not separately perform a meta-analysis in Caucasian and East Asian populations. The reason can be seen from Li et al.'s study [15]. In order to collect all possible studies with various types of study design from all over the world, we searched PubMed and Google scholar databases. In the end, we selected 17 independent studies, among which 16 studies evaluated the association between rs597668 polymorphism and AD by a case-control design. Considering that only one study analyzed rs597668 polymorphism of 40 AD patients by a longitudinal study [14], we only performed a meta-analysis of rs597668 polymorphism using the results from case-control design.

Before our submission (3/22/2013), we accessed the AlzGene (www.alzgene.org) [16] and PubMed (www.ncbi.nlm.nih.gov/pubmed) databases. We found no study investigating the association between rs597668 polymorphism and AD by a meta-analysis method. To our knowledge, this is the first study that assesses the association between rs597668 polymorphism and AD by meta-analysis. We believe that our findings will be very useful for future genetic studies in AD.

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Conflict of Interest

The authors declare no conflict of interest.

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