Association between LRP1 C766T polymorphism and Alzheimer’s disease susceptibility: a meta-analysis

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Low density lipoprotein receptor-related protein 1 (LRP1) C766T polymorphism (rs1799986) has been extensively investigated for Alzheimer’s disease (AD) susceptibility. However, results in different studies have been contradictory. Therefore, we conducted a meta-analysis containing 6455 AD cases and 6304 controls from 26 independent case–control studies to determine whether there was an association between the LRP1 C766T polymorphism and AD susceptibility. The combined analysis showed that there was no significant association between LRP1 C766T polymorphism and AD susceptibility (TT + CT versus CC: OR = 0.920, 95% CI = 0.817–1.037, P = 0.172). In subgroup analysis, significant decreased AD susceptibility was found among Asian population in allele model (T versus C: OR = 0.786, 95% CI = 0.635–0.974, P = 0.028) and dominant model (TT + CT versus CC: OR = 0.800, 95% CI = 0.647–0.990, P = 0.040). Moreover, T allele of LRP1 C766T was statistically associated with late onset of AD (LOAD) (T versus C: OR = 0.858, 95% CI = 0.748–0.985, P = 0.029; TT + CT versus CC: OR = 0.871, 95% CI = 0.763–0.994, P = 0.040). In conclusion, our meta-analysis suggested that LRP1 C766T polymorphism was associated with lower risk of AD in Asian, and could reduce LOAD risk especially. Considering some limitations of our meta-analysis, further large-scale studies should be done to reach a more comprehensive understanding.

Alzheimer’s disease (AD), a progressive and lethal neurodegenerative disorder, has become a global challenge for the 21st century1. It is essentially characterised by cerebral senile plaques laden with β-amyloid peptide (Aβ), dystrophic neurites in neocortical terminal fields as well as neurofibrillary tangles of hyperphosphorylated microtubule-associated protein tau. Besides, loss of neurons and white matter, congophilic angiopathy, inflammation, and oxidative damage are also important pathological features of AD. It is believed that genetic factors, lifestyle and environmental factors synergistically give rise to AD. Variants associated with AD have been detected in more than 20 genes, which are involved in metabolism, inflammation, synaptic activity and intracellular trafficking.

Low density lipoprotein receptor-related protein 1 (LRP1) has been widely studied due to its pleiotropic roles in AD pathogenesis. LRP1 is ubiquitously expressed in various tissues, especially high in liver, lung and brain. In the central nervous system, LRP1 plays an important role in controlling Aβ metabolism and maintaining brain homeostasis. There are two forms of LRP1–soluble LRP1 and cell-surface LRP1. In plasma, soluble LRP1 binds to peripheral Aβ, and consequently prevents free Aβ access to the brain. As a cell surface receptor, LRP1 can control the endocytosis of multiple ligands, mediate cell signaling transductions and regulate gene expression through its intracellular domain. For instance, the interaction between amyloid precursor protein (APP) and cell-surface LRP1 leads to increased endosomal trafficking of APP, accelerating Aβ production. Besides that, Aβ can enter multiple cell types (e.g. abluminal brain endothelial cell and hepatic cell) through cell-surface LRP1, in which the ubiquitous apolipoprotein E (APOE) and activated alpha-2-macroglobulins (A2M) are chaperones.

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and subsequently degraded by endopeptidase. Therefore, LRP1 are involved in the bulk transport, primary production, brain and systemic clearance of AD toxin Aβ, and thus plays a critical role in AD pathogenesis.

The silent C766T polymorphism in exon 3 of LRP1 gene (rs1799986) has attracted extensive attention since first reported as a risk factor for AD. However, results in different studies have been contradictory. The inconsistency is likely to relate with insufficient statistical power, racial differences or other demographic variables. Therefore, we conducted a comprehensive meta-analysis to determine whether there was an association between the LRP1 C766T polymorphism and AD susceptibility.

Results

Eligible studies. A total of 167 relevant studies were identified from initial database searching, of which 35 publications were included based on titles and abstracts (Fig. 1). Furthermore, 4 reviews, 1 duplicated publication and 3 studies with inadequate information were excluded after careful reading of the full text. Besides, manual search of references revealed 3 more articles. After primary data extracted from the 30 independent studies, 4 studies were excluded for genotype distribution of controls was not in Hardy-Weinberg equilibrium (HWE). Finally, 26 eligible studies containing 6455 AD cases and 6304 controls were included in our meta-analysis. The ethnicities of these subjects involved in the comparisons were diverse, including Caucasian (n = 16), Asian (n = 6), African (n = 1) and mixed (n = 3). Besides, LRP1 C766T genotype and allele distribution among AD cases and controls was summarized in Table 2, and the control group in all studies was in HWE.

Meta-analysis and meta-regression results. The combined analysis showed that there was no significant association between LRP1 C766T polymorphism and AD susceptibility in any genetic model (T versus C: OR = 0.905, 95% CI = 0.813–1.008, P = 0.069; TT versus CC: OR = 0.791, 95% CI = 0.622–1.005, P = 0.055; CT versus CC: OR = 0.915, 95% CI = 0.813–1.030, P = 0.139; TT + CT versus CC: OR = 0.920, 95% CI = 0.817–1.037, P = 0.172; TT versus CC + CT: OR = 0.815, 95% CI = 0.640–1.037, P = 0.095) (Table 3 and Fig. 2).

In subgroup analysis by ethnicity, T allele of LRP1 C766T was found to be associated with decreased AD susceptibility among Asian population (T versus C: OR = 0.786, 95% CI = 0.635–0.974, P = 0.028; TT + CT versus CC: OR = 0.800, 95% CI = 0.647–0.990, P = 0.040) (Fig. 3). However, we did not observe any association for all comparisons in Caucasians. When stratified by time of AD onset, we found T allele of LRP1 C766T may act as a protective factor for late onset of AD (LOAD) (T versus C: OR = 0.858, 95% CI = 0.748–0.985, P = 0.029; TT + CT versus CC: OR = 0.871, 95% CI = 0.763–0.994, P = 0.040) (Fig. 4), but no significant association was observed for early onset of AD (EOAD). Furthermore, no significant interaction was observed for APOE ε4 status (P > 0.05).
The results of univariate and multivariate meta-regression analyses showed that age, MMSE and/or APOE ɛ4 were not potential factor(s) for heterogeneity among those studies, but gender might contributed to the heterogeneity (as shown in Table 4).

Publication bias. Begg’s test and Egger’s test were performed to evaluate the publication bias of the included studies. The shape of Begg’s funnel plot appeared to be approximately symmetrical (Fig. 5). Besides, statistical significance was also not observed according to Egger’s test ($P > 0.05$, Table 3). In general, there was no publication bias in our included studies.

| First author | Year | Country | Ethnicity | AD Controls | Criteria for AD diagnosis | Genotyping method | Source of control | Time of AD onset | Quality score |
|--------------|------|---------|-----------|-------------|---------------------------|-------------------|-----------------|----------------|--------------|
| Yuan, Q. 50  | 2013 | China   | Asian     | 364 74.9 69.9 57% | NINCDS-ADRDA             | PCR and Direct sequencing | HB Mixed | 9              |
| Vargas, T. 50 | 2010 | Spain   | Caucasian | 746 NA 73.7 66% | NINCDS-ADRDA and DSM-IV   | TaqMan SNP Genotyping Assays | PB NA   | 12             |
| Vazquez-Higuera, J. L. 52 | 2009 | Spain   | Caucasian | 246 76.6 72.9 65% | NINCDS-ADRDA             | PCR-RFLP          | PB Mixed       | 10             |
| Chen, Y. 53  | 2009 | China   | Asian     | 67 71.9 NA 34% | NINCDS-ADRDA             | PCR-RFLP          | PB NA           | 8              |
| Bahia, V. S. 53 | 2008 | Brazil  | Mixed     | 120 75.2 71.2 68% | NINCDS-ADRDA and DSM-IV   | PCR-RFLP          | PB Mixed       | 10             |
| Rodriguez, E. 53 | 2006 | Spain   | Caucasian | 274 75.4 71.6 68% | NINCDS-ADRDA             | PCR-RFLP          | PB Mixed       | 8              |
| Forero, D. A. 53 | 2006 | Colombia | Mixed     | 106 73.3 68.8 71% | NINCDS-ADRDA             | PCR-RFLP          | NA Mixed        | 7              |
| Pritchard, A. 53 | 2005 | UK      | Caucasian | 250 NA 56.7 55% | NINCDS-ADRDA and DSM-III-R | PCR-RFLP          | PB Early        | 9              |
| Pritchard, A. 53 | 2005 | UK      | Caucasian | 183 NA 73.8 65% | NINCDS-ADRDA and DSM-III-R | PCR-RFLP          | PB Late         | 9              |
| Bia, S. 50  | 2001 | China   | Asian     | 216 NA 74.7 NA 34% | NINCDS-ADRDA and DSM-IV   | PCR-RFLP          | PB Late         | 11             |
| Panza, F. 57  | 2004 | Italy   | Caucasian | 166 69.4 NA 62% | NINCDS-ADRDA             | Roche LightCycler Genotyping | PB Mixed      | 9              |
| Zheng, W. D. 58 | 2004 | China   | Asian     | 79 72.8 >65 49%   | NINCDS-ADRDA             | PCR-RFLP          | PB Late         | 10             |
| Kolisch, H. 59 | 2003 | Germany | Caucasian | 212 73.1 NA 71% | DSM-IV                    | PCR-RFLP          | PB + HB NA      | 12             |
| Helbecque, N. 60 | 2003 | France  | Caucasian | 239 74.0 NA 65% | NINCDS-ADRDA and DSM-III-R | PCR-RFLP          | HB NA           | 10             |
| Helbecque, N. 60 | 2003 | France  | Caucasian | 56 85.0 NA 80% | NINCDS-ADRDA and DSM-III-R | PCR-RFLP          | HB NA           | 9              |
| Berry, R. T. 60 | 2003 | USA     | African   | 111 71.3 NA 78% | NINCDS-ADRDA             | PCR-RFLP          | PB NA           | 11             |
| Bi, S. 60  | 2001 | China   | Asian     | 38 70.2 NA 45% | NINCDS-ADRDA             | PCR-RFLP          | PB NA           | 8              |
| Sanchez-Guerrua, M. 61 | 2001 | Spain   | Caucasian | 305 75.5 71.8 68% | NINCDS-ADRDA             | PCR-RFLP          | PB Mixed        | 12             |
| McIlroy, S. P. 61 | 2001 | UK      | Caucasian | 219 77.5 >65 67% | NINCDS-ADRDA and DSM-IV   | PCR-SSCP          | PB Late         | 12             |
| Prince, J. A. 62 | 2001 | Sweden  | Caucasian | 204 NA 61% 171 NA 63% | NINCDS-ADRDA             | PCR-SSCP          | PB + HB NA      | 10             |
| Verpillat, P. 62 | 2001 | France  | Caucasian | 274 65.5 56% | NINCDS-ADRDA and DSM-III-R | PCR-RFLP          | PB NA           | 12             |
| Bullido, M. 63 | 2000 | Spain   | Caucasian | 199 70.4 60% | NINCDS-ADRDA             | PCR-RFLP          | PB Late         | 10             |
| Hatanaka, Y. 63 | 2000 | Japan   | Asian     | 100 NA 76.6 68% | NINCDS-ADRDA             | PCR-RFLP          | PB Late         | 8              |
| Bertram, L. 63 | 2000 | USA     | Mixed     | 276 NA 71.7 NA 194 NA | NINCDS-ADRDA             | PCR-SSCP          | PB NA           | 11             |
| Beffert, U. 64 | 1999 | Canada  | Caucasian | 225 NA 70.9 48% | NINCDS-ADRDA             | PCR-RFLP          | PB + HB NA      | 9              |
| Kamboh, M. 64 | 1998 | USA     | Caucasian | 432 75.4 68.6 62% | NINCDS-ADRDA and DSM-III-R | PCR-SSCP          | NA NA           | 9              |
| Lambert, J. C. 64 | 1998 | France  | Caucasian | 558 71.8 68.8 62% | NINCDS-ADRDA and DSM-III-R | PCR-SSCP          | NA NA           | 9              |
| Kang, D. E. 64 | 1997 | USA     | Caucasian | 157 >65 73.2 53% | NINCDS-ADRDA             | PCR-SSCP          | PB Late         | 11             |

Table 1. Characteristics of individual studies included in the meta-analysis. NINCDS: the National Institute of Neurological Disorders and Stroke; ADRDA: Alzheimer Diseases and Related Disorders Association; DSM: the Diagnostic and Statistical Manual of Mental Disorders; NA: not available; PB: population-based control; HB: hospital-based control. aNumber. bAge at survey. cAge at onset of Alzheimer’s disease. dPercentage of female.
Discussion

AD, as a continuum, bring about serious threat to human health. Considering early detection and intervention at the asymptomatic stage may offer better chance of therapeutic success, it is urgent to identify early diagnostic biomarkers. LRP1, a member of the LDL receptor family, is an endocytic receptor for more than 40 structurally diverse ligands. The findings of previous studies indicate that LRP1 and many of its ligands (eg. APOE and A2M) are co-deposited with Aβ in senile plaques in AD brains. Subsequent studies demonstrated that LRP1 modulates the clearance of Aβ via receptor-mediated pathway in central nervous system. Besides, soluble LRP1 provides an endogenous peripheral ‘sink’ activity for Aβ by preventing plasma free Aβ access to the brain. It has also been reported that LRP1 is responsible for a rapid peripheral uptake of Aβ by the liver, which plays a key role in systemic clearance of Aβ. On the other hand, endocytosis of LRP1 could modulate APP trafficking, and contribute to Aβ generation. Interestingly, LRP1 can regulate Aβ metabolism in two contrary sides.

The association between LRP1 polymorphisms and AD susceptibility also has been described extensively, especially exon 3 C766T polymorphism. Kang et al. first reported the LRP1 C766T polymorphism, and found a positive association between C allele and AD susceptibility. This finding was replicated in some following studies, but Kolsch et al. found the opposite result that carriers of a C allele were at lower risk of AD, while some failed to show any association between LRP1 C766T polymorphism and AD risk. Previously, three meta-analysis have tried to clarify the relationship between LRP1 C766T polymorphism and AD susceptibility, which one revealed a weak correlation of LRP1 CC genotype with AD, but other two separately studies showed that no positive evidence was involved in the relationship between this polymorphism and AD risk among overall and Chinese population. Since several factors could be responsible for these discrepancies, such as inadequate sample size, variability in phenotype definition and allele frequency polymorphisms in different ethnic backgrounds, we conducted a comprehensive meta-analysis with different genetic models in this study, to better clarify the association between LRP1 C766T polymorphism and AD susceptibility.

New results from our research did not show any association of LRP1 C766T polymorphism with AD susceptibility from 6455 AD cases and 6304 controls in overall population. This result is consistent with two published meta-analyses. Compared with the results from previous studies, our data from meta-analysis was relatively reliable to illustrate the association between LRP1 C766T polymorphism and AD susceptibility, because we used different genetic models with a larger number of case-controls. Due to that people in different ethnic populations may have different allele frequency, and can affect the heterogeneity, we additionally conducted subgroup analysis by ethnicity, time of AD onset and APOE ε4 status.

Table 2. LRP1 C766T genotype and allele distribution among AD cases and controls in the included studies. HWE: Hardy-Weinberg equilibrium. *P value for HWE test in controls.

| First author | AD | Control | HWE |
|--------------|----|---------|-----|
|              | CC | CT      | TT  | T  | C  | T  | CC | CT | TT | T  | C  | T  | P*  |
| Yuan, Q.     | 304| 54      | 6   | 662| 66 | 232| 52 | 7  | 516| 66 | 0.058 |
| Vargas, T.   | 559| 172     | 15  | 1290| 202| 442| 138| 18 | 1022| 174| 0.079 |
| Vazquez-Higuera, J. L. | 193| 51      | 2   | 437| 55 | 198| 35 | 4  | 431| 43 | 0.107 |
| Chen, Y.     | 59 | 8       | 0   | 126| 8  | 56 | 19 | 2  | 131| 23 | 0.800 |
| Bahia, V. S. | 87 | 28      | 5   | 202| 38 | 86 | 30 | 4  | 202| 38 | 0.497 |
| Rodriguez, E. | 211| NA      | NA  | NA | NA | 233| NA | NA | NA | NA | 0.576 |
| Forero, D.A. | 84 | 22      | 0   | 190| 22 | 78 | 18 | 1  | 174| 20 | 0.972 |
| Pritchard, A. | 337| 115     | 14  | 789| 143| 334| 132| 11 | 800| 154| 0.629 |
| Bin, L.      | 189| 26      | 1   | 404| 28 | 179| 21 | 0  | 379| 21 | 0.433 |
| Panza F.     | 115| 49      | 2   | 279| 53 | 160| 63 | 2  | 383| 67 | 0.116 |
| Zheng, W. D. | 72 | 6       | 1   | 150| 8  | 139| 16 | 1  | 294| 18 | 0.478 |
| Kolosch, H.  | 145| 59      | 8   | 349| 75 | 250| 84 | 3  | 584| 90 | 0.156 |
| Helbecque, N. | 216| 70      | 9   | 562| 88 | 290| 108| 14 | 688| 136| 0.321 |
| Perry, R. T. | 97 | 14      | 0   | 208| 14 | 74 | 4  | 0  | 152| 4  | 0.816 |
| Bi, S.       | 31 | 6       | 1   | 68 | 8  | 24 | 13 | 3  | 61 | 19 | 0.516 |
| Sanchez-Guerra, M. | 237| 65      | 3   | 539| 71 | 249| 51 | 4  | 549| 59 | 0.457 |
| McIlroy, S. P. | 193| 24      | 2   | 410| 28 | 198| 37 | 2  | 433| 41 | 0.852 |
| Prince, J. A. | 155| 47      | 2   | 357| 51 | 124| 41 | 6  | 289| 53 | 0.269 |
| Verpillar, P. | 198| 71      | 5   | 467| 81 | 214| 66 | 10 | 494| 86 | 0.092 |
| Bullido, M. I. | 151| 47     | 1   | 349| 49 | 173| 66 | 4  | 412| 74 | 0.417 |
| Hatanaka, Y. | 83 | 17      | 0   | 183| 17 | 200| 45 | 1  | 445| 47 | 0.358 |
| Bertram, L.  | 186| 82      | 8   | 454| 98 | 135| 55 | 4  | 325| 63 | 0.556 |
| Befert, U.   | 158| 58      | 9   | 374| 76 | 125| 57 | 5  | 307| 67 | 0.619 |
| Kamboh, M. I. | 310| 111     | 11  | 731| 133| 71 | 29 | 6  | 171| 41 | 0.205 |
| Lambert, J. -C. | 428| 119     | 11  | 975| 141| 407| 168| 21 | 982| 210| 0.480 |
| Kang, D. E.  | 127| 26      | 4   | 280| 34 | 65 | 34 | 3  | 164| 40 | 0.563 |
The outcomes by subgroups revealed that T allele of LRP1 C766T could reduce the risk of AD in allele model (T vs. C) and dominant model (TT + CT vs. CC) among Asian population, no significant role was found in Caucasian group. In terms of onset age, the results from subgroup analysis showed that T allele of LRP1 C766T could act as a protective factor for late onset of AD, but no significant association with early onset of AD. This is also consistent with previous report.

It’s recognized that APOE ε4 is an important pathogenic factor for the development of AD. Several studies have revealed a possible protective effect of TT genotypes in carriers of APOE ε4 alleles. However, APOE ε4 status did not show that the influence of the association between LRP1 C766T polymorphism and AD susceptibility in our study. Moreover, our meta-regression analysis also showed that APOE ε4 status, age, and MMSE were not responsible for heterogeneity.

| Population | Comparison | Sample size | Association | Heterogeneity | Publication bias |
|------------|------------|-------------|-------------|---------------|-----------------|
| Overall    | T vs. C    | 6181        | 0.905 (0.813, 1.008) | Random        | 0.031 43.0 0.849 |
|            | TT vs. CC  | 6074        | 0.791 (0.622, 1.005) | Fixed         | 0.623 0 0.971  |
|            | CT vs. CC  | 6181        | 0.915 (0.813, 1.030) | Random        | 0.031 37.5 0.758 |
|            | TT + CT vs. CC | 6455       | 0.920 (0.817, 1.037) | Random        | 0.008 44.7 0.829 |
|            | TT vs. CC + CT | 6074       | 0.815 (0.640, 1.037) | Fixed         | 0.683 0 0.972  |
| Caucasian  | T vs. C    | 4704        | 0.905 (0.801, 1.022) | Random        | 0.019 48.4 0.959 |
|            | TT vs. CC  | 4704        | 0.777 (0.595, 1.013) | Fixed         | 0.329 11.1 0.901 |
|            | CT vs. CC  | 4704        | 0.916 (0.795, 1.055) | Random        | 0.021 47.7 0.950 |
|            | TT + CT vs. CC | 4978       | 0.926 (0.806, 1.065) | Random        | 0.008 52.3 0.861 |
|            | TT vs. CC + CT | 4704       | 0.799 (0.612, 1.043) | Fixed         | 0.353 8.9 0.941  |
| Asian      | T vs. C    | 864         | 0.786 (0.635, 0.974) | Fixed         | 0.156 37.5 0.460 |
|            | TT vs. CC  | 864         | 0.642 (0.297, 1.386) | Fixed         | 0.764 0 0.786  |
|            | CT vs. CC  | 864         | 0.810 (0.648, 1.011) | Fixed         | 0.351 10.1 0.279 |
|            | TT + CT vs. CC | 864       | 0.800 (0.647, 0.990) | Fixed         | 0.232 27.0 0.388 |
|            | TT vs. CC + CT | 864       | 0.687 (0.315, 1.498) | Fixed         | 0.825 0 0.732  |
| EOAD       | T vs. C    | 355         | 0.966 (0.743, 1.257) | Fixed         | 0.332 9.3 0.977 |
|            | TT vs. CC  | 321         | 1.506 (0.477, 4.750) | Fixed         | 0.719 0 NA  |
|            | CT vs. CC  | 355         | 0.906 (0.699, 1.174) | Fixed         | 0.435 0 0.922  |
|            | TT + CT vs. CC | 355       | 0.933 (0.727, 1.198) | Fixed         | 0.363 1.2 0.947 |
|            | TT vs. CC + CT | 321       | 1.536 (0.484, 4.873) | Fixed         | 0.769 0 NA  |
| LOAD       | T vs. C    | 1524        | 0.858 (0.748, 0.985) | Fixed         | 0.423 1.7 0.346 |
|            | TT vs. CC  | 1524        | 0.678 (0.374, 1.229) | Fixed         | 0.889 0 0.994  |
|            | CT vs. CC  | 1524        | 0.880 (0.767, 1.009) | Fixed         | 0.176 29.2 0.702 |
|            | TT + CT vs. CC | 1524      | 0.871 (0.763, 0.994) | Fixed         | 0.255 20.4 0.520 |
|            | TT vs. CC + CT | 1524      | 0.714 (0.394, 1.294) | Fixed         | 0.875 0 0.861  |
| APOE ε4+   | T vs. C    | 924         | 0.706 (0.436, 1.145) | Random        | 0.051 54.6 0.446 |
|            | TT vs. CC  | 815         | 0.743 (0.320, 1.723) | Fixed         | 0.532 0 0.378  |
|            | CT vs. CC  | 924         | 0.716 (0.407, 1.257) | Fixed         | 0.048 55.2 0.683 |
|            | TT + CT vs. CC | 1073      | 0.790 (0.475, 1.333) | Random        | 0.030 57.1 0.683 |
|            | TT vs. CC + CT | 815       | 0.770 (0.331, 1.791) | Fixed         | 0.528 0 0.369  |
| APOE ε4−   | T vs. C    | 819         | 1.054 (0.894, 1.242) | Fixed         | 0.591 0 0.546  |
|            | TT vs. CC  | 819         | 0.883 (0.475, 1.641) | Fixed         | 0.924 0 0.776  |
|            | CT vs. CC  | 819         | 1.095 (0.926, 1.295) | Fixed         | 0.491 0 0.360  |
|            | TT + CT vs. CC | 944       | 1.120 (0.967, 1.298) | Fixed         | 0.403 2.90 0.386 |
|            | TT vs. CC + CT | 819       | 0.876 (0.470, 1.632) | Fixed         | 0.924 0 0.665  |

Table 3. Meta-analysis of LRP1 C766T polymorphism and AD susceptibility. OR: odds ratio; CI: Confidence interval; EOAD: early onset of AD; LOAD: late onset of AD. *Number of comparisons.

The outcomes by subgroups revealed that T allele of LRP1 C766T could reduce the risk of AD in allele model (T versus C) and dominant model (TT + CT versus CC) among Asian population, no significant role was found in Caucasian group. In terms of onset age, the results from subgroup analysis showed that T allele of LRP1 C766T could act as a protective factor for late onset of AD, but no significant association with early onset of AD. This is also consistent with previous report.

It’s recognized that APOE ε4 is an important pathogenic factor for the development of AD. Several studies have revealed a possible protective effect of TT genotypes in carriers of APOE ε4 alleles. However, APOE ε4 status did not show that the influence of the association between LRP1 C766T polymorphism and AD susceptibility in our study. Moreover, our meta-regression analysis also showed that APOE ε4 status, age, and MMSE were not responsible for heterogeneity.
LRP1 C766T polymorphism is a silent mutation, which does not change the amino acid sequence or splice site. Therefore, it is unlikely to alter the biological function by a direct causal effect with the polymorphism. Some studies consider that the LRP1 C766T polymorphism might be responsible for susceptibility to AD by interacting with other genes, such as APOE, MAPT, and MAPK8IP1. In addition, some speculate that LRP1 C766T may be in linkage disequilibrium with a deleterious mutation in the LRP1 gene, or with other biologically relevant mutations on neighboring genes, which affect LRP1 expression. Besides, several studies have a hypothesis that the LRP1 C766T polymorphism might alter the secondary structure of the LRP mRNA to affect the translation and stability of the protein. To date, the conclusion with LRP1 C766T polymorphism with AD susceptibility is conflicting, further genetic analyses of this locus are needed to illuminate the potential mechanism and the functional interactions with AD.

Some limitations of our meta-analysis should be acknowledged. The sample size in some subgroup analysis was small, which may increase the risk of false negatives or false positives. Besides, we did not perform subgroup analysis based on other factors participated in the progression of AD, such as educational background, due to a lack of sufficient information. Larger and broader independent investigations are required to better understand the role of LRP1 C766T polymorphism in AD pathogenesis.

In conclusion, our meta-analysis suggested that LRP1 C766T polymorphism was associated with lower risk of AD in Asian, and could reduce LOAD risk especially. Furthermore, large-scale studies should be performed to reach more understanding of this association.

Materials and Methods

Search strategy. We searched electronic databases PubMed, Embase and CNKI (up to August 2016) using the following keywords: ("Alzheimer's disease" or "Alzheimer disease" or "AD") and ("low density lipoprotein receptor-related protein 1" or "LDL receptor-related protein 1" or "LRP1") and ("polymorphism" or "SNP"
or "variant" or "genotype") without language restriction. The bibliographies of the retrieved studies were also screened to identify relevant publications.

**Inclusion and exclusion criteria.** The eligible studies had to meet all the following criteria: (1) a case-control study to evaluate the association between LRP1 C766T polymorphism and risk of AD; (2) useful data including sample size, allele or genotype distribution were given; (3) genotype distribution of controls followed the HWE. Accordingly, the exclusion criteria were as follows: (1) reviews, meta-analysis or editorial articles; (2) studies were provided with inadequate information; (3) for the studies with overlapping data, only the most relevant articles with the largest dataset were included in the final analysis.

The literature retrieval and inclusion were carried out in duplication by two independent reviewers.
Data extraction. Two reviewers independently extracted the following information: first author, year of publication, country, ethnicity, total number of cases and controls, mean age of cases and controls, proportion of female in cases and controls, AD diagnosis criteria, genotyping method, source of controls, time of AD onset, genotype or/and allele distribution in cases and controls. If conflicting results produced, two reviewers would review the publications again and reached a consensus by discussion.

Quality assessment. Two reviewers independently assessed the quality of each included studies in the meta-analysis according to the criteria of quality assessment (as referred in the Reference of 54, 55), and the disagreements were judged by the third reviewer to ensure a consistent outcome. Quality scores of studies ranged from 0 (the lowest) to 15 (the highest). Studies with quality scores among 10 to 15 were grouped into high quality studies and other studies scored between 0 and 9 were categorized into low quality studies.

Statistical analysis. HWE in controls was tested by a chi-square test. Summary odds ratio (OR) with confidence interval (95% CI) for genotypes and alleles were used to evaluate the strength of association between LRP1 C766T polymorphism and AD susceptibility. The significance of the pooled OR was measured using the Z-test. Four genetic models were performed in our meta-analysis: allele model (T versus C), codominant model (homozygote comparison (TT versus CC) and heterozygote comparison (CT versus CC)), dominant model (TT + CT versus CC), and recessive model (TT versus CC). The heterogeneity was also quantified with I² statistics. If no significant heterogeneity was found between the studies, the pooled OR was calculated by using the fixed effects model (the Mantel-Haenszel method) 56. Otherwise, the random effects model (the DerSimonian and Laird method) was applied 57. Both of univariate and multivariate meta-regression analyses were also carried out to explore potential sources of heterogeneity among studies. The log of the ORs from involved studies was using as dependent variables, and age, gender, Mini-Mental State Exam (MMSE) and/or APOE ε4 status as covariates. Publication bias was tested by Begg's test and Egger's test 58, 59. We also performed subgroup analysis according to ethnicity, time of AD onset and APOE ε4 status, respectively. Statistical analyses were conducted with Stata Version 11.0 (College Station, TX, USA), and a two-sided P < 0.05 was considered statistically significant.

| Heterogeneity factors | Coefficient | 95% CI       | SE  | P    |
|-----------------------|-------------|--------------|-----|------|
| Age                   |             |              |     |      |
| Univariate            | 0.008       | [−0.027, 0.043] | 0.017 | 0.644 |
| Multivariate          | −0.018      | [−0.051, 0.015] | 0.015 | 0.251 |
| Gender                |             |              |     |      |
| Univariate            | 1.864       | [0.383, 3.345] | 0.712 | 0.016 |
| Multivariate          | 2.193       | [0.233, 4.152] | 0.907 | 0.031 |
| MMSE                  |             |              |     |      |
| Univariate            | −0.081      | [−0.344, 0.182] | 0.127 | 0.532 |
| Multivariate          | 0.004       | [−0.268, 0.277] | 0.126 | 0.975 |
| APOE ε4 status        |             |              |     |      |
| Univariate            | −0.048      | [−0.440, 0.343] | 0.186 | 0.798 |
| Multivariate          | 0.190       | [−0.252, 0.632] | 0.204 | 0.37  |

Table 4. The potential sources of heterogeneity between LRP1 polymorphism and AD risk were evaluated by both of univariate and multivariate meta-regression analyses. SE = standard error; 95%CI = 95% confidence interval.

Figure 5. Funnel plot of association between LRP1 C766T polymorphism (TT + CT vs. CC) and AD susceptibility.
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Author Contributions

Y.W. and S.L. contributed equally to this work, and they designed the study and wrote the main manuscript. J.W. and J.Z. collected the information of included articles. J.W. and Y.H. analyzed the data. H.L. and H.T. prepared figures and tables. B.K. and B.W. checked and revised the results. S.S. reviewed and approved the manuscript.

Additional Information

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