Original Article
Meta-analysis of the association between CCAAT/enhancer binding protein-ε polymorphism and the risk of childhood acute lymphoblastic leukemia

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Abstract: *CEBPE* rs2239633 polymorphism has been implicated in susceptibility to childhood acute lymphoblastic leukemia (ALL) risk. Several studies investigated the association of this polymorphism with ALL in different populations. However, the results were contradictory. A meta-analysis was conducted to assess the association between *CEBPE* rs2239633 polymorphism and ALL susceptibility. Databases including Pubmed, EMBASE, Chinese National Knowledge Infrastructure (CNKI) and Wangfang were searched to find relevant studies. Odds ratios (ORs) with 95% confidence intervals (CIs) were used to assess the strength of associations. A random-effects model was used. A significant association was found between *CEBPE* rs2239633 polymorphism and childhood ALL (OR = 1.19, 95% CI, 1.11-1.28). In the subgroup analyses by ethnicity, the significant association was found among Caucasians (OR = 1.19, 95% CI, 1.09-1.30) and Hispanics (OR = 1.39, 95% CI, 1.18-1.63), but not in Asians (OR = 1.05, 95% CI, 0.90-1.22). In the subgroup analysis by histology, B-cell ALL risk (OR = 1.29, 95% CI, 1.15-1.44) and B hyperdiploid ALL risk (OR = 1.84, 95% CI, 1.40-2.43) were increased. Our results suggested that *CEBPE* rs2239633 polymorphism conferred a risk factor of childhood ALL.

Keywords: Acute lymphoblastic leukemia, *CEBPE*, meta-analysis

Introduction

Acute lymphoblastic leukemia (ALL), a malignant disorder of lymphoid progenitor cells, is the most common type of leukemia among children, accounting for 75% of all childhood leukemia and 25% of all malignancy in childhood [1]. ALL is a malignant clonal proliferation of lymphoid progenitor cells, most commonly of the B-cell lineage. In the pediatric population, ALL accounts for 81% of childhood leukemias; leukemia overall accounts for one third of cancers diagnosed in children between ages 0-14 years [1]. In the United States, the majority of ALL cases occur in ages 1-4, with an incidence rate in this group of 8 per 100,000, and preponderance for males over females [2].

The CCAAT/enhancer binding proteins (*CEBPE*) are transcription factors involved in hematopoietic cell development and induction of several inflammatory mediators. Park et al. found that *CEBPE* was rapidly induced in the acute promyelocytic leukemia (APL) cell line NB4 during granulocytic differentiation after exposure to retinoic acid (RA) [3]. They also suggested that *CEBPE* was a downstream target gene responsible for RA-induced granulocytic differentiation of APL cells [3].

To identify whether the *CEBPE* polymorphism is involved in the pathogenesis of ALL in vivo, many case-control studies concerning this allelic variation and cancer risk have been broadly performed [4-14]. However, there is still uncertainty about the risk for ALL and *CEBPE* polymorphism. Therefore, we performed a meta-analysis to identify statistical evidence for an association between the *CEBPE* polymorphism and ALL risk.

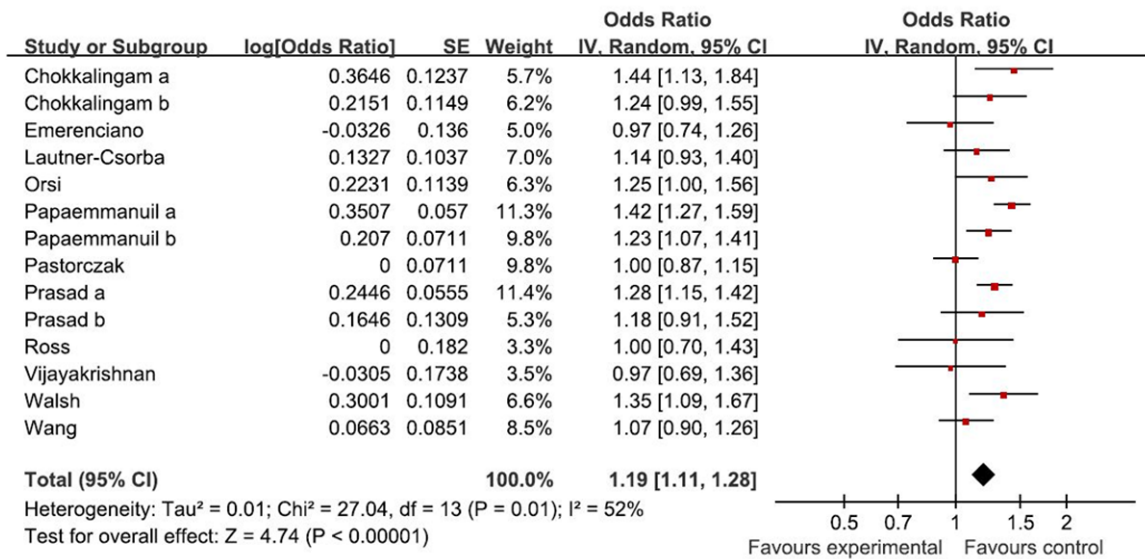## Methods

Publication search

Published studies were identified through a computerized search of Pubmed, EMBASE, Chinese National Knowledge Infrastructure

Table 1. Characteristics of the studies

First author	Gender	Race	Type of disease	No. of Case	No. of Control	Quality score
Papaemmanuil a/2009	Mixed	Caucasian	ALL	577	1438	9
Papaemmanuil b/2009	Mixed	Caucasian	ALL	404	960	8
Prasad a/2010	Mixed	Caucasian	B-cell ALL	1193	1510	9
Prasad b/2010	Mixed	Caucasian	B-cell ALL	183	352	7
Vijayakrishnan/2010	Mixed	Asian	Mixed	190	182	8
Pastorczak/2011	Mixed	Caucasian	ALL	398	731	7
Lautner-Csorba/2012	Mixed	Caucasian	ALL	543	529	8
Orsi/2012	Mixed	Caucasian	Mixed	441	1542	9
Chokkalingam a/2013	Mixed	Hispanic	Mixed	300	406	8
Chokkalingam b/2013	Mixed	Caucasian	Mixed	225	369	8
Ross/2013	Mixed	Caucasian	ALL	171	384	8
Walsh/2013	Mixed	Hispanic	B-cell ALL	297	454	7
Wang/2013	Mixed	Asian	ALL	568	673	8
Emerenciano/2014	Mixed	Caucasian	Mixed	149	483	9

ALL, acute lymphoblastic leukemia.

Study or Subgroup	log[Odds Ratio]	SE	Weight	Odds Ratio IV, Random, 95% CI
Chokkalingam a	0.3646	0.1237	5.7%	1.44 [1.13, 1.84]
Chokkalingam b	0.2151	0.1149	6.2%	1.24 [0.99, 1.55]
Emerenciano	-0.0326	0.136	5.0%	0.97 [0.74, 1.26]
Lautner-Csorba	0.1327	0.1037	7.0%	1.14 [0.93, 1.40]
Orsi	0.2231	0.1139	6.3%	1.25 [1.00, 1.56]
Papaemmanuil a	0.3507	0.057	11.3%	1.42 [1.27, 1.59]
Papaemmanuil b	0.207	0.0711	9.8%	1.23 [1.07, 1.41]
Pastorczak	0	0.0711	9.8%	1.00 [0.87, 1.15]
Prasad a	0.2446	0.0555	11.4%	1.28 [1.15, 1.42]
Prasad b	0.1646	0.1309	5.3%	1.18 [0.91, 1.52]
Ross	0	0.182	3.3%	1.00 [0.70, 1.43]
Vijayakrishnan	-0.0305	0.1738	3.5%	0.97 [0.69, 1.36]
Walsh	0.3001	0.1091	6.6%	1.35 [1.09, 1.67]
Wang	0.0663	0.0851	8.5%	1.07 [0.90, 1.26]
Total (95% CI)			100.0%	1.19 [1.11, 1.28]

Heterogeneity: Tau² = 0.01; Chi² = 27.04, df = 13 (P = 0.01); I² = 52%
Test for overall effect: Z = 4.74 (P < 0.00001)

Figure 1. Meta-analysis of the association between *CEBPE* rs2239633 polymorphism and childhood ALL.

(CNKI) and Wangfang databases (Last search was updated on August, 2014). The search terms were used as follows: (acute lymphoblastic leukemia or ALL) and (CCAAT/enhancer binding protein-ε or *CEBPE*). We also perused the reference lists of all retrieved articles and relevant reviews. There was no language restriction.

Inclusion and exclusion criteria

Eligible studies had to meet the following criteria: (a) only case-control and cohort studies were considered, (b) the study explored the correlation between *CEBPE* polymorphism with ALL risk. Major exclusion criteria were: (a) no control population, (b) no available genotype frequency, (c) duplication of the previous publications, the largest or most recent publication was selected.

Data extraction

Information was extracted from all eligible publications independently by two authors according to the inclusion criteria listed above. The

Table 2. Meta-analysis results and subgroup analysis of this meta-analysis

	OR (95% CI)	*P* Value	I^2 (%)	$P_{heterogeneity}$
Overall	1.19 (1.11-1.28)	< 0.00001	52	0.01
Asian	1.05 (0.90-1.22)	0.53	0	0.62
Caucasian	1.19 (1.09-1.30)	< 0.0001	56	0.02
Hispanic	1.39 (1.18-1.63)	< 0.0001	0	0.70
B-cell ALL	1.21 (1.13-1.29)	< 0.0001	0	0.56
B hyperdiploid ALL	1.84 (1.40-2.43)	< 0.0001	0	0.89
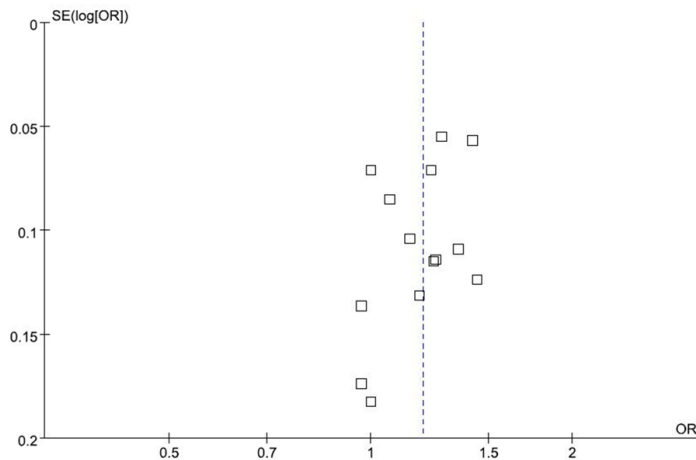
Figure 2. Funnel plot of the childhood ALL risk with *CEBPE* rs2239633 polymorphism.

following information was extracted from each study: first author, publication year, racial background, gender, number of cases and controls, polymorphisms, and odds ratios (ORs) and the corresponding 95% confidence intervals (CIs) of ALL risk.

Statistical analysis

The association between *CEBPE* polymorphism with ALL was assessed using odds ratio (OR) with 95% confidence interval (CI). Allele model was used in this meta-analysis, because most of the studies reported the results in this model. B Heterogeneity among studies was calculated using the χ^2-based Cochran's Q-statistic test (*P* < 0.10 was considered statistically significant heterogeneity), the inconsistency index I^2 statistic was also calculated to observe between-study variability that was due to heterogeneity rather than chance. This statistic, which was documented by percentage, yields result ranging from 0 to 100% (I^2 = 0-25%, no heterogeneity; I^2 = 25-50%, moderate hetero-

geneity; I^2 = 50-75%, large heterogeneity; I^2 = 75-100%, extreme heterogeneity). A random effects model using the method of DerSimonian and Laird was used in this meta-analysis. Sensitivity analyses were conducted by deleting a single study each time involved in the meta-analysis to identify the potential influence of the individual data set on the pooled ORs. Potential publication bias was assessed by Egger's weighted regression method. All statistical analyses were conducted using the RevMan software. All *P* values were two-sided.

Results

Characteristics of eligible studies

After a comprehensive literature search applying our inclusion criteria, 11 relevant studies which comprised 5639 cases and 10036 controls were identified in the final analysis. Three studies reported two case-control studies. Therefore, a total of 14 studies were included in this meta-analysis. There were only two studies with Hispanics or Asians, while the rest of the studies used Caucasians. All the studies investigated the association between *CEBPE* rs2239633 polymorphism and childhood ALL. The main study characteristics are summarized in **Table 1**.

Results of meta-analysis

A significant association was found between *CEBPE* rs2239633 polymorphism and childhood ALL (OR = 1.19; 95% CI, 1.11-1.28; **Figure 1**). Results of this meta-analysis are showed in **Table 2**. In the subgroup analyses by ethnicity, the significant association was found among Caucasians (OR = 1.19, 95% CI, 1.09-1.30) and Hispanics (OR = 1.39, 95% CI, 1.18-1.63), but not in Asians (OR = 1.05, 95% CI, 0.90-1.22). In the subgroup analysis by histology, B-cell ALL risk (OR = 1.29, 95% CI, 1.15-1.44) and B hyperdiploid ALL risk (OR = 1.84, 95% CI, 1.40-2.43) were increased. We conducted one-way sensitivity analysis to evaluate the stability of the meta-analysis. The statistical significance of the results was not altered when any single

study was omitted (data not shown). Egger's test did not indicate significant publication bias (P = 0.473, **Figure 2**).

Discussion

This current meta-analysis of 11 studies including 5639 cases and 10036 controls systematically evaluated the association between *CEBPE* rs2239633 polymorphism and childhood ALL risk. The results indicated that *CEBPE* rs2239633 polymorphism was a conspicuous high risk factor for developing childhood ALL. In the subgroup analysis by ethnicity, no significant association was found in Asians. However, cancer risk was increased in Caucasians and Hispanics who carried rs2239633 polymorphism, suggesting a possible influence among environmental exposures and different genetic backgrounds. After stratification by histology, this association remained significant in B-cell ALL risk and B hyperdiploid ALL risk. This result indicated that *CEBPE* rs2239633 polymorphism might paly a same role in the etiology of different ALLs.

C/EBPε is expressed only in myeloid lineages, with some low but detectable expression in the lymphoblastic cell line MOLT4 and the ovaries [15]. The lack of expression of secondary and tertiary granule protein mRNAs in C/EBPε-deficient neutrophils may reflect the direct loss of the transcriptional activation of C/EBPε. Blotting of RNA from C/EBPε-deficient bone marrow demonstrates specific loss of lactoferrin and gelatinase B, with normal to elevated expression of primary granule proteins neutrophil elastase, myeloperoxidase, and proteinase-3 [16].

The present meta-analysis should be interpreted with caution, several limitations merit consideration. First, due to lacking of the original data of the eligible studies, we could not perform other subgroup analyses based on gender, life style, and so on. Second, the numbers of published studies were not sufficient for a comprehensive analysis, particularly for Africans. However, our meta-analysis also had some merits. First, we investigated the association between *CEBPE* rs2239633 polymorphism and ALL risk. Second, there was low heterogeneity in this meta-analysis.

This meta-analysis suggested that the *CEBPE* rs2239633 polymorphism may be associated with ALL development. Further studies with a larger sample size are needed to further assess the presence of an association.

Disclosure of conflict of interest

None.

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