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ORIGINAL ARTICLE

Lack of association between cytotoxic T-lymphocyte antigen-4 gene polymorphisms and lymphoid malignancy risk: evidence from a meta-analysis

Zhiming Dai[1,2] · Chuanjie Feng[3] · Wanggang Zhang[2] · Jie Liu[2] · Xingmei Cao[2] · Hui Zhang[2] · Yuhong Liu[2,4] · Meng Wang[5] · Xinghan Liu[5] · Zhijun Dai[5]

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Abstract Cytotoxic T-lymphocyte antigen-4 (CTLA-4) polymorphisms have been associated with susceptibility to lymphoid malignancies. However, results from the published single studies are inconsistent. Therefore, the present meta-analysis was conducted to get a more accurate estimation of the relationship between CTLA-4 gene polymorphisms and the lymphoid malignancy risk. We identified nine independent studies accounting for 3090 subjects up to January 30, 2016. Summary odds ratios (OR) and 95 % confidence intervals (CI) were used to evaluate the risk of lymphoid malignancies. Overall, no significant association was found between +49A/G (rs231775), −318C/T (rs5742909), and +6230A/G (rs3087243) CTLA-4 gene polymorphisms and lymphoid malignancies. Furthermore, ethnicity (Asian and Caucasian) and histopathology subgroup analyses (non-Hodgkin's lymphoma) also failed to detect an association between the studied polymorphisms and lymphoid malignancy risk. Our study shows that common CTLA-4 gene polymorphisms may not contribute to lymphoid malignancy susceptibility based on the current evidence.

Keywords Cytotoxic T-lymphocyte antigen-4 · Polymorphisms · Lymphoid malignancies · Meta-analysis

Introduction

Lymphoid malignancies, including lymphoma, lymphoid leukemia, and myeloma, are large and heterogeneous disease groups, which were classified by the World Health Organization (WHO) in 2001 [1] and updated in 2008 [2]. Lymphoid malignancies arise from the malignant transformation of normal B- and T-lymphoid cells at various stages of differentiation and comprise the seventh most common cancer worldwide among men and women [3]. However, the etiological factors contributing to most lymphoid malignancies remain poorly understood [4]. The potential molecular mechanism of lymphomagenesis is very complex. However, several studies support that immune-related genetic factors are associated with the etiology of lymphoid malignancies [5]. An increased incidence of lymphoid malignancies has been observed among individuals with autoimmune disease and congenital and acquired immunodeficiencies, suggesting that immune dysfunction might represent a risk factor for lymphoid malignancies [6].

Cytotoxic T-lymphocyte antigen-4 (CTLA-4), also known as CD152, is an immune regulatory molecule expressed exclusively in T cells. It has been known that CTLA-4 acts

Zhiming Dai and Chuanjie Feng contributed equally to this work.

✉ Wanggang Zhang
zhangwanggang2003@yahoo.com

✉ Zhijun Dai
dzj0911@126.com

1 Department of Anesthesiology, The Second Affiliated Hospital of Xi'an Jiaotong University, Xi'an 710004, Shaanxi, People's Republic of China

2 Department of Hematology, The Second Affiliated Hospital of Xi'an Jiaotong University, Xi'an 710004, Shaanxi, People's Republic of China

3 Department of Emergency, The Affiliated Hospital of Yan'an University, Yan'an 716000, Shaanxi, People's Republic of China

4 Department of Hematology and Immunology, The Affiliated Hospital of Yan'an University, Yan'an 716000, Shaanxi, People's Republic of China

5 Department of Oncology, The Second Affiliated Hospital of Xi'an Jiaotong University, Xi'an 710004, Shaanxi, People's Republic of China

as an immune checkpoint and regulates immune function through pleiotropic mechanism suppression, the activity of effector T cells, and establishment of peripheral T cell tolerance [7]. Mouse model studies have demonstrated that deletion of the CTLA-4 gene results in a lethal systemic immune activation by profound lymphoproliferation [8]. Furthermore, several studies have verified that blocking the CTLA-4 function can enhance antitumor immunity [9, 10].

The human CTLA-4 gene, located on chromosome 2q33 in an immune regulatory gene region, is composed of four exons. Several relevant polymorphisms have been described in this region, including −318C/T (rs5742909) located in the promoter region [11], +49A/G (rs231775) located in exon 1 [12], and +6230A/G (rs3087243) located in the 3′-untranslated region (3′ UTR) [13]. It has been shown that these single-nucleotide polymorphisms (SNPs) in the CTLA-4 gene might influence the host immune response by affecting gene transcription, protein expression, and interaction with the CD80/CD86 ligand [14–16].

Numerous studies have shown that CTLA-4 polymorphisms play an important immunoregulatory role in lymphoid malignancies; however, the results were inconsistent and even contradictory [17–25]. Considering that a single study is unable to elucidate the overall effects, we conducted a comprehensive meta-analysis of all eligible case-control studies to evaluate the association between CTLA-4 SNPs (rs231775, rs5742909, and rs3087243) and the susceptibility to lymphoid malignancies.

Materials and methods

Publication search

We searched the PubMed, Embase, and Chinese National Knowledge Infrastructure (CNKI) databases for studies published prior to January 2016 (last search: January 30, 2016). The keywords searching were performed with and without Medical Subject Headings (MeSH) terms for "Cytotoxic T-lymphocyte antigen 4/CTLA-4/CD152," "polymorphism," and "lymphoid malignancies/lymphoid neoplasms." To identify relevant publications, reference lists of research papers were also reviewed by manual search.

Inclusion and exclusion criteria

All studies included in the meta-analysis met the following criteria: (1) evaluation of the association of CTLA-4 polymorphisms and lymphoid malignancy risk, (2) case-control design, (3) available information on genotype frequency, and (4) the controls had no malignant disease. Additionally, the following exclusion criteria were also used: (1) the study was not regarding the polymorphisms of interest; (2) it was a repeated study, a review, or an abstract; and (3) the study did not have a control group.

Data extraction

According to the inclusion and exclusion criteria listed above, two investigators independently revised all potentially relevant studies, and disagreements were resolved through discussion with a third researcher. For each study, the following information was collected: first author, year of publication, country of origin of the study, subjects' ethnicity, source of control, genotyping method, total number of cases and controls, number of different genotypes in cases and controls, and Hardy-Weinberg value of controls. All data were obtained from published articles. All patients were confirmed by histology or pathology. Histopathological subtypes of lymphoid malignancies were classified based on the WHO classification guidelines [2, 26].

Statistical analysis

We calculated the odds ratios (OR) and 95 % confidence intervals (CI) to evaluate the lymphoid malignancy risk associated with CTLA-4 polymorphisms, based on the allele frequencies in cases and controls for each eligible study. The following five different ORs were computed: (i) B vs. A (allele comparison), (ii) BB vs. AA (homozygous carriers), (iii) BB vs. AA + AB (recessive model), (iv) BB + AB vs. AA (dominant model), and (v) AB vs. AA (heterozygous carriers). Statistical heterogeneity between studies was evaluated by chi-square-based Q test and I^2 test. Higher I^2 values indicated higher levels of heterogeneity ($I^2 = 75$–100 %: extreme heterogeneity; $I^2 = 50$–75 %: large heterogeneity; $I^2 = 25$–50 %: moderate heterogeneity; $I^2 <$ 25 %: no heterogeneity). If the p value of the Q test was higher than 0.1, the pooled OR estimate of the study was calculated by the fixed-effects model. Otherwise, the random-effects model was used. Subgroup analysis was performed to assess the ethnicity-specific effects. Egger's and Begg's tests were adopted to assess publication bias. A $p < 0.05$ was considered statistically significant. Sensitivity analysis was performed to estimate the stability of the results by removing the studies, one at a time. All statistical analyses were performed with STATA 12.0 software (Stata Corp, College Station, TX).

Results

Study characteristics

As shown in Fig. 1, 102 publications were retrieved from the initial screening. After skimming the titles and

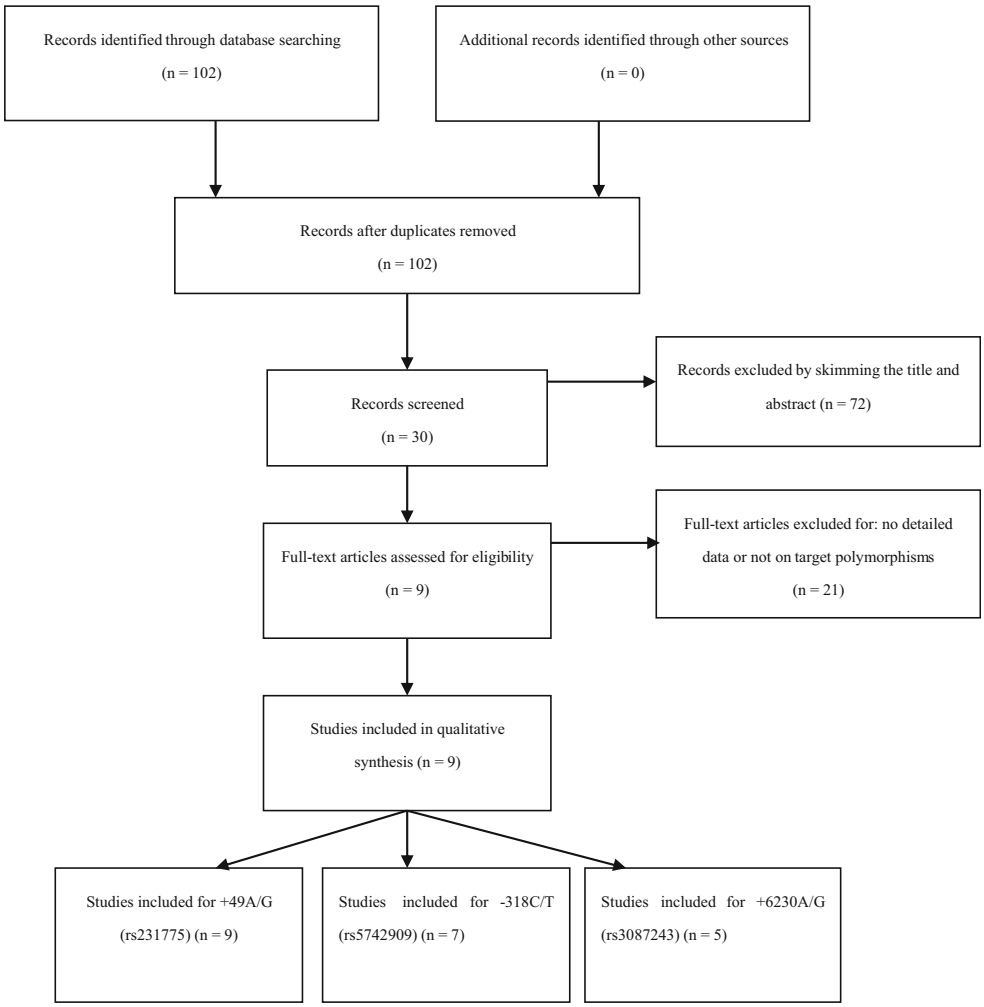


Fig. 1 Studies identified with criteria for inclusion and exclusion

reviewing the abstracts, 72 studies were excluded by their irrelevance to the current analysis. Furthermore, 21 studies were excluded because of lack of detailed data or lack of data regarding the target polymorphisms. Thus, nine case-control studies involving 3090 subjects were selected for the meta-analysis. The main characteristics of the included studies are summarized in Table 1. Eligible studies presented data for several different lymphoid malignancies including chronic lymphocytic leukemia (CLL), mucosa-associated lymphoid tissue (MALT) lymphoma, non-Hodgkin's lymphoma (NHL), and multiple myeloma. There were seven studies based on Caucasian and two on Chinese population. Additionally, six studies were hospital-based, and three were population-based case-control studies. Detailed data on genotype distribution in each eligible study and minor allele frequency (MAF) as well as Hardy-Weinberg equilibrium (HWE) of controls are shown in Table 2. Among the nine case-control studies, in one study regarding CTLA-4 rs231775 polymorphism, a significant departure from HWE was observed [24].

Meta-analysis results

Nine studies assessed the association between CTLA-4 rs231775 polymorphism and lymphoid malignancy susceptibility. As shown in Fig. 2, the heterogeneity of GG vs. AA + AG was assessed for all case-control studies. The I^2 value was 0 %, indicating no heterogeneity. Meta-analysis results were as follows: $\chi^2 = 7.66$, degrees of freedom = 8, and $p = 0.41$ in a fixed-effects model. We observed a lack of association between the rs231775 polymorphism and lymphoid malignancy risk under a recessive model. Further studies confirmed that neither the subgroup analyses by histologic type nor those by ethnicity revealed a noteworthy association. All comparison results are listed in Table 3.

Seven studies evaluated the association between rs5742909 polymorphism and lymphoid malignancy risk including 1037 cases and 1332 healthy controls. As shown in Table 3, the meta-analysis results indicate that the rs5742909 polymorphism is not associated with lymphoid malignancy predisposition in the overall population, neither

Table 1 Characteristics of the studies included in the meta-analysis

First author	Year	Country	Cancer type	Ethnicity	Study design	Genotyping method	Source of control	Total sample size (case/control)	SNP no.
Pavkovic	2003	Macedonia	CLL	Caucasian	CC	PCR-RFLP	HB	130/100	1
Monne	2004	Italy	NHL	Caucasian	CC	PCR-RFLP	HB	44/76	1, 2
Piras	2005	Italy	NHL	Caucasian	CC	PCR-RFLP	HB	100/128	1
Cheng	2006	China	MALT	Chinese	CC	PCR-RFLP	HB	62/250	1, 2, 3
Suwalska	2008	Poland	CLL	Caucasian	CC	SNaPshot	HB	170/224	1 2
Bonzheim	2008	Germany	T-NHL	Caucasian	CC	PCR-RFLP	PB	94/173	1, 2, 3
Karabon	2012	Poland	MM	Caucasian	CC	PCR-RFLP	PB	200/380	1, 2, 3
Liu	2013	China	T-NHL	Chinese	CC	PCR-LDR	HB	300/291	1, 2, 3
Khorshied	2014	Egyptian	B-NHL	Caucasian	CC	PCR-RFLP	PB	181/200	1, 2, 3

SNP no. 1: +49A/G (rs231775), 2: −318C/T (rs5742909), 3: +6230A/G (rs3087243); *CC* case-control, *SNP* single-nucleotide polymorphism, *NHL* non-Hodgkin's lymphoma, *MALT* mucosa-associated lymphoid tissue lymphoma, *CLL* chronic lymphocytic Leukemia, *MM* multiple myeloma, *B-/T-NHL* B-/T cells non-Hodgkin's lymphoma, *HB* hospital-based, *PB* population-based

Table 2 CTLA-4 polymorphisms genotype distribution and allele frequency in cases and controls

First author	Genotype (*N*)								Genotype (*N*)				MAF	HWE
	Case				Control				Case		Control			
	Total	AA	AB	BB	total	AA	AB	BB	A	B	A	B		
+49 A/G (rs231775)														
Pavkovic 2003	130	61	53	16	100	51	39	10	175	85	141	59	0.33	Yes
Monne 2004	44	36	7	1	76	38	32	6	79	9	108	44	0.51	Yes
Piras 2005	100	74	23	3	128	74	43	11	171	29	191	65	0.15	Yes
Cheng 2006	62	2	26	34	250	29	102	119	30	94	160	340	0.76	Yes
Suwalska 2008	170	56	84	30	224	71	106	47	196	144	248	200	0.42	Yes
Bonzheim 2008	94	37	38	19	173	78	65	30	112	76	221	125	0.40	No
Karabon 2012	199	48	103	48	368	124	169	75	199	199	319	417	0.50	Yes
Liu 2013	300	34	118	148	291	26	127	138	186	414	179	403	0.69	Yes
Khorshied 2013	181	72	88	21	200	106	76	18	232	130	288	112	0.36	Yes
−318C/T (rs5742909)														
Monne 2004	44	33	9	2	76	62	6	8	75	13	130	22	0.15	Yes
Cheng 2006	62	59	3	0	250	209	40	1	121	3	458	42	0.02	Yes
Suwalska 2008	170	121	42	7	333	267	62	4	284	56	596	70	0.16	Yes
Bonzheim 2008	94	78	13	3	173	140	29	4	169	16	309	37	0.10	Yes
Karabon 2012	195	155	40	0	367	297	68	2	350	40	662	72	0.10	Yes
Liu 2013	291	222	64	5	300	222	73	5	508	74	517	83	0.13	Yes
Khorshied 2014	181	145	26	10	200	166	21	13	316	46	353	47	0.13	Yes
+6230A/G (rs3087243)														
Cheng 2006	62	39	20	3	250	154	79	17	98	26	387	113	0.21	Yes
Bonzheim 2008	94	29	41	24	173	43	95	35	99	89	181	165	0.47	Yes
Karabon 2012	193	81	88	24	374	128	180	66	250	136	436	312	0.35	Yes
Liu 2013	291	197	84	10	300	208	82	10	478	104	498	102	0.18	Yes
Khorshied 2014	181	36	94	51	200	44	96	60	166	196	184	216	0.54	Yes

A represents the major allele, *B* represents the minor allele

MAF minor allele frequencies, *HWE* Hardy-Weinberg equilibrium

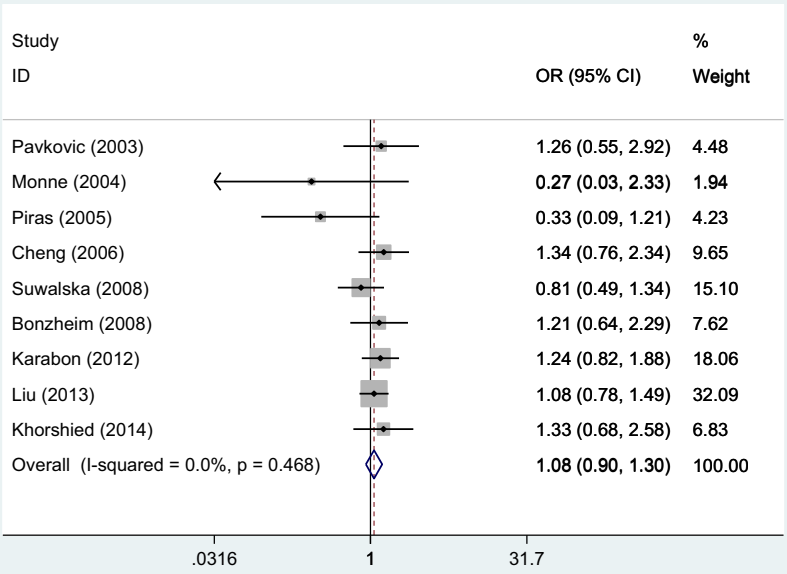


Fig. 2 Forest plots of CTLA-4 rs231775 polymorphism and NHL risk based on recessive model (GG vs. AA + AG). The *squares* and *horizontal lines* correspond to the study-specific OR and 95 % CI, respectively. The area of the squares reflects the weight (inverse of the variance). The *diamond* represents the summary OR and 95 % CI. *OR* odd ratio, *CI* confidence interval

in the NHL subgroup nor in Caucasian or Chinese populations. However, trends were observed in a heterozygous comparison model in Caucasians (CT vs. CC: 95 % CI = 1.00–1.86, $P = 0.05$).

Five studies including those on three Caucasian and two Chinese populations evaluated the association between CTLA-4 rs3087243 polymorphism and risk of lymphoid malignancies. A fixed-effects model was selected for all genetic comparisons showing no significant differences in terms of heterogeneity. The results of quantitative synthesis evidenced the lack of a significant relationship in the overall population and subgroup analyses by histological type or ethnicity (Table 3).

Heterogeneity assessment and sensitivity analysis

Because of significant heterogeneity in the literature regarding the CTLA-4 rs231775 variant (G vs. A: $p < 0.001$; GG vs. AA: $p = 0.03$; GG + AG vs. AA: $p < 0.001$; AG vs. AA: $p < 0.001$), sensitivity analysis was conducted to identify the source, by sequentially omitting one study at a time. The results indicate that the studies by Monne et al. [18] and Piras et al. [19] contributed to the major heterogeneity. The exclusion of these two studies significantly decreased the heterogeneity (G vs. A: $p = 0.21$; GG vs. AA: $p = 0.18$; GG + AG vs. AA: $p = 0.09$; AG vs. AA: $p = 0.12$). It was noted that the initial lack of association was quantitatively altered, when the two studies were excluded in the homozygous model (OR = 1.27, 95 % CI = 0.99–1.62). Sensitivity analyses were also conducted for the remaining two variants, and the pooled results were not statistically influenced when each study was sequentially omitted.

Publication bias

Begg's funnel plot and Egger's test were performed to evaluate the publication bias for rs231775, rs5742909, and rs3087243 polymorphisms. As shown in Fig. 3, the funnel plots do not evidence any obvious asymmetry (Begg: $p = 0.10$, $p = 0.88$, $p = 0.62$, respectively), and the results of Egger's test suggest no evidence of publication bias (Egger: $p = 0.36$, $p = 0.82$, $p = 0.84$, respectively).

Discussion

Lymphoid tumors differ from other tumors in that the malignancy originates from the immune system itself. The vast majority of lymphoid neoplasms arise from mature B and T lymphocytes. Therefore, the role of immune pathways in lymphomagenesis is very complex. As an archetypal immune regulatory checkpoint, CTLA-4 plays an important role in T cell anergy and in T and B cell suppression responses [7]. Therefore, an abnormal expression of CTLA-4 may have an effect on the pathogenesis of lymphoid malignancies. Furthermore, SNPs in the CTLA-4 gene have been extensively considered to modify promoter activity and regulate the expression levels of CTLA-4 [27, 28].

In the present study, we summarized the association between three CTLA-4 gene SNPs (rs231775, rs5742909, and rs3087243) and the risk of lymphoid malignancies including lymphoma, multiple myeloma, and CLL. To our knowledge, this is the first meta-analysis focusing on CTLA-4 polymorphisms and lymphoid malignancy susceptibility. The results of

Table 3 Meta-analysis of the association CTLA-4 polymorphisms and lymphoid malignancy risk

Polymorphisms	Comparison	Population	Association				Heterogeneity	
			OR	95 % CI	*p* value	I^2 (%)	*p* value	Model
+49A/G (rs231775)	G vs. A	All	1.00	0.80–1.27	0.97	75	0.000	R
		HWE	0.98	0.75–1.27	0.85	76	0.000	R
		NHL	0.90	0.63–1.33	0.64	82	0.000	R
		Caucasian	0.94	0.70–1.27	0.70	79	0.000	R
		Chinese	1.09	0.88–1.35	0.44	57	0.13	F
	GG vs. AA	All	1.11	0.76–1.64	0.58	53	0.03	R
		HWE	1.08	0.69–1.68	0.75	58	0.02	R
		NHL	1.01	0.54–1.89	0.97	62	0.02	R
		Caucasian	1.09	0.71–1.67	0.71	52	0.05	R
		Chinese	1.59	0.33–7.79	0.57	76	0.04	R
	GG vs. AA + AG	All	1.08	0.90–1.30	0.41	0	0.47	F
		HWE	1.07	0.88–1.29	0.49	7	0.37	F
		NHL	1.09	0.86–1.37	0.48	14	0.33	F
		Caucasian	1.04	0.81–1.32	0.76	15	0.31	F
		Chinese	1.14	0.86–1.51	0.36	0	0.52	F
	GG + AG vs. AA	All	1.00	0.69–1.45	0.99	77	0.000	R
		HWE	0.96	0.63–1.47	0.86	79	0.000	R
		NHL	0.88	0.48–1.63	0.69	83	0.000	R
		Caucasian	0.96	0.63–1.45	0.84	80	0.000	R
		Chinese	1.51	0.30–7.54	0.62	77	0.04	R
	AG vs. AA	All	1.00	0.70–1.44	0.98	72	0.000	R
		HWE	0.97	0.65–1.47	0.90	75	0.000	R
		NHL	0.88	0.49–1.60	0.69	80	0.000	R
		Caucasian	0.99	0.66–1.46	0.94	74	0.001	R
		Chinese	1.40	0.28–7.01	0.68	76	0.04	R
−318C/T (rs5742909)	T vs. C	All	1.04	0.80–1.36	0.76	49	0.07	R
		NHL	0.90	0.72–1.13	0.37	17	0.31	F
		Caucasian	1.20	0.97–1.47	0.09	10	0.35	F
		Chinese	0.57	0.18–1.83	0.34	74	0.05	R
	TT vs. CC	All	1.10	0.67–1.82	0.70	0	0.45	F
		NHL	0.88	0.49–1.57	0.67	0	0.92	F
		Caucasian	1.12	0.64–1.96	0.69	30	0.22	F
		Chinese	1.02	0.32–3.29	0.97	0	0.93	F
	TT vs. CC + CT	All	1.06	0.64–1.74	0.83	0	0.44	F
		NHL	0.85	0.48–1.51	0.59	0	0.84	F
		Caucasian	1.06	0.61–1.84	0.85	32	0.21	F
		Chinese	1.06	0.33–3.43	0.92	0	0.89	F
	TT + CT vs. CC	All	1.07	0.80–1.42	0.66	47	0.08	R
		NHL	0.91	0.71–1.17	0.46	38	0.17	F
		Caucasian	1.25	0.99–1.58	0.06	0	0.53	F
		Chinese	0.55	0.17–1.80	0.32	73	0.05	R
	CT vs. CC	All	1.09	0.79–1.52	0.60	53	0.05	R
		NHL	0.98	0.59–1.62	0.93	60	0.04	R
		Caucasian	1.28	1.00–1.65	0.05	10	0.35	F
		Chinese	0.56	0.18–1.75	0.32	71	0.06	R
+6230A/G (rs3087243)	G vs. A	All	0.92	0.80–1.06	0.27	0	0.47	F
		NHL	1.01	0.85–1.19	0.95	0	0.96	F

Table 3 (continued)

Polymorphisms	Comparison	Population	Association				Heterogeneity	
			OR	95 % CI	*p* value	I^2 (%)	*p* value	Model
		Caucasian	0.89	0.75–1.05	0.16	20	0.29	F
		Chinese	1.02	0.79–1.31	0.90	0	0.59	F
	GG vs. AA	All	0.83	0.61–1.13	0.23	0	0.56	F
		NHL	1.00	0.68–1.45	0.98	0	0.95	F
		Caucasian	0.81	0.58–1.61	0.23	25	0.27	F
		Chinese	0.91	0.44–1.88	0.80	0	0.60	F
	GG vs. AA + AG	All	0.90	0.69–1.17	0.42	0	0.48	F
		NHL	1.02	0.74–1.40	0.91	0	0.70	F
		Caucasian	0.90	0.67–1.19	0.45	38	0.20	F
		Chinese	0.90	0.44–1.84	0.77	0	0.62	F
	GG + AG vs. AA	All	0.91	0.75–1.10	0.33	0	0.42	F
		NHL	1.00	0.79–1.26	0.99	0	0.66	F
		Caucasian	0.82	0.63–1.06	0.13	13	0.32	F
		Chinese	1.04	0.77–1.40	0.79	0	0.70	F
	AG vs. AA	All	0.93	0.76–1.14	0.47	1	0.40	F
		NHL	1.00	0.78–1.28	1.00	0	0.43	F
		Caucasian	0.84	0.64–1.10	0.20	27	0.25	F
		Chinese	1.06	0.78–1.44	0.72	0	0.83	F

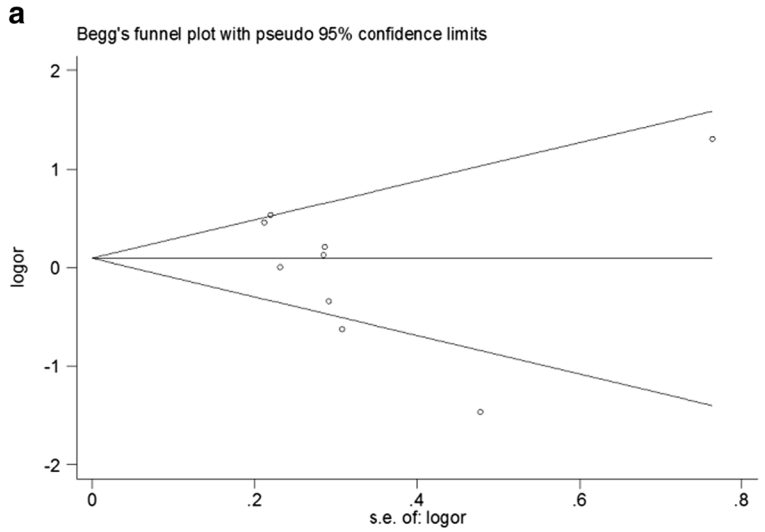
F fixed-effects model, *R* random-effects model, *OR* odd ratio, *CI* confidence interval, *HWE* Hardy-Weinberg equilibrium, *NHL* non-Hodgkin's lymphoma

our study suggest that the studied SNPs were not associated with an increased risk of lymphoid malignancies.

The rs231775 polymorphism of the CTLA-4 gene was investigated in several case-control studies on lymphoid malignancy risk; however, the results are controversial. Reports by Khorshied et al. [23] and Piras et al. [19] showed that rs231775 A alleles were associated with an increased risk of non-Hodgkin's lymphoma; however, the sample sizes in the two studies were relatively small and included different histological types of lymphomas. Contrary results were presented by Liu et al. [22] and Bonzheim et al. [24], who showed that rs231775 variant was not associated with the risk of T cell non-Hodgkin's lymphoma. Pavkovic et al. [17] found that rs231775 G allele was associated with increased risk of CLL. However, no significant association between this polymorphism and the risk of CLL development was observed by Suwalska et al. [21]. Our meta-analysis suggested that the rs231775 variant might not contribute to the risk of lymphoid malignancies in the overall population, in NHL population, or in Caucasian or Chinese populations.

The CTLA-4 rs5742909 polymorphism has been widely studied in several different cancers. However, reports on lymphoid malignancy patients remain scarce, and the association between this locus and lymphoid malignancy risk is inconclusive. A study carried out by Cheng et al. [20] examined the role of this SNP in MALT lymphoma development in Chinese population and evidenced that the rs5742909 CT genotype was associated with a lower risk of developing MALT lymphoma. Conversely, the lack of association between this variant and lymphoid malignancy susceptibility has been reported in Chinese populations by Liu et al. [22], in Egyptian populations by Khorshied et al. [23], and in Europe Caucasians by Suwalska et al. [21] and Bonzheim et al. [24]. The results of the present meta-analysis suggest that the rs5742909 polymorphism is not associated with lymphoid malignancy risk, although trends were observed in Caucasians based on the heterozygous model (CT vs. CC: 95 % CI = 1.00–1.86). Further multicenter clinical studies on the CTLA-4 rs5742909 polymorphism association with lymphoid malignancy risk in Caucasians are necessary.

The CTLA-4 rs3087243 polymorphism has been poorly explored in lymphoid malignancy case-control studies. A study involving 200 multiple myeloma patients and 380 controls showed that the rs3087243 G allele was associated with an increased risk of developing multiple myelomas in the Polish population [25]. Other studies by Khorshied et al. [23], Liu et al. [22], and Bonzheim et al. [24] had not observed a correlation between the rs3087243 polymorphism and lymphoid malignancy risk. The results of our meta-analysis support the negative findings of the latter studies, as no significant correlation was determined between CTLA-4 rs3087243 polymorphism and the risk of lymphoid malignancies.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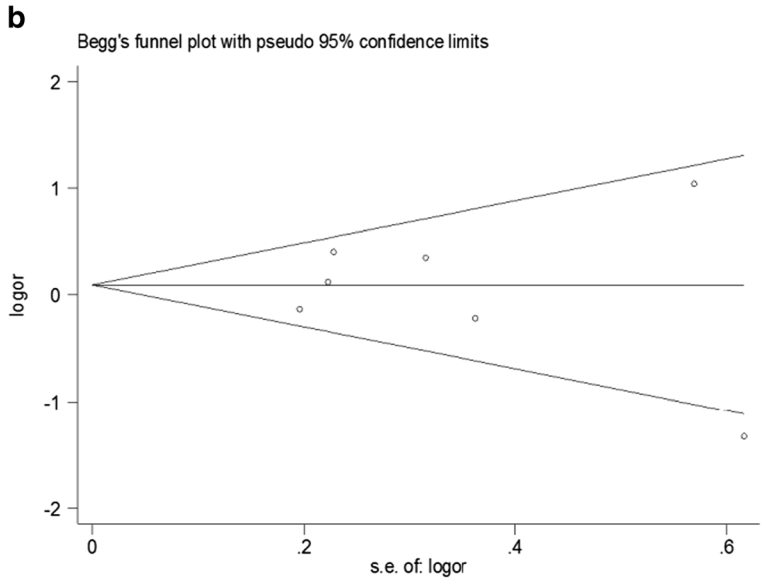
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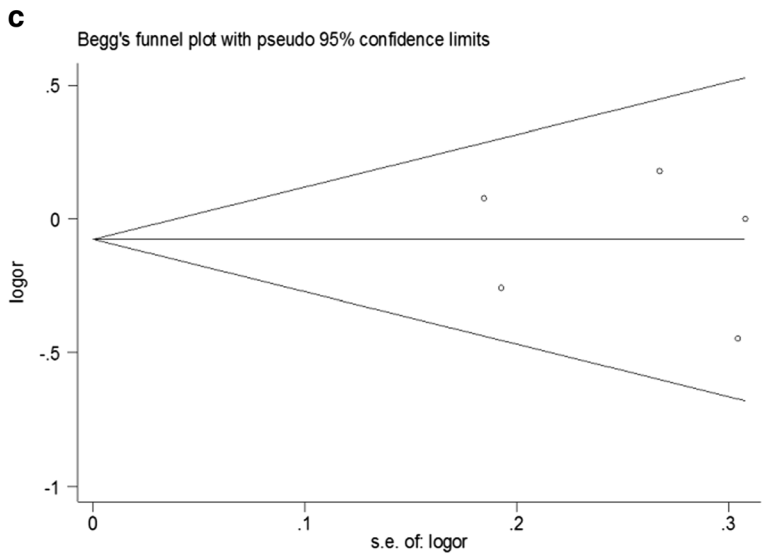
s.e. of: logor


c
Begg's funnel plot with pseudo 95% confidence limits
logor
.5
0
-.5
-1
0
.1
.2
.3
s.e. of: logor

◀ **Fig. 3** Funnel plots of CTLA-4 polymorphisms and lymphoid malignancy risk for the publication bias test in heterozygous model. Each *point* represents an independent study for the indicated association. **a** rs231775. **b** rs5742909. **c** rs3087243

Meta-analysis can overcome a few issues caused by a single study, such as small sample size, selection bias, and low test power; therefore, it is considered a powerful tool for integrating conflicted results from different studies [29]. However, it is inevitable to note limitations in our meta-analysis. First, significant heterogeneity was observed among some comparisons, especially for CTLA-4 rs231775. The heterogeneity may come from the classification of histopathological differences of lymphoid malignancies, the source of eligible studies in different countries, and the differences in gene detection methods. Although we analyzed NHL as a subgroup, it was not possible to conduct a further subgroup analysis based on a single tumor type, because of the lack of detailed data on pathological classification of malignant lymphomas in most of the eligible studies. Second, owing to the absence of detailed original data for conducting appropriate haplotype analysis, we could not assess potential effects of combined genotypes of these three SNPs. Third, tumorigenesis in humans is a multistep process; other factors such as gene-gene, gene-microbe, and gene-environment interactions may also contribute to the development of lymphoid malignancies. Finally yet importantly, we have not found published studies on the association between CTLA-4 gene polymorphisms and Hodgkin's lymphoma risk.

In conclusion, our study represents, for the first time, a comprehensive meta-analysis of the role of CTLA-4 polymorphisms on lymphoid malignancy risk. The results of the present study indicate that the three CTLA-4 gene SNPs studied were not significantly associated with lymphoid malignancy susceptibility. More large-scale epidemiological studies are necessary to confirm our findings and clarify the real genetic effect.

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Compliance with ethical standards

Competing interests The authors declare that they have no competing interests.

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