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RESEARCH ARTICLE

Meta-analysis Reveals No Association of DNMT3B -149 C>T Gene Polymorphism With Overall Cancer Risk

Raju Kumar Mandal[1,#], Shafiul Haque[1,2,#], Mohd Wahid[1,2], Arshad Jawed[1], Naseem Akhter[2,3], Md. Ekhlaque Ahmed Khan[4], Aditya Kumar Panda[4], Mohammed Yahya Areeshi[1] and Sajad Ahmad Dar[1,*]

[1]*Research and Scientific Studies Unit, College of Nursing and Allied Health Sciences, Jazan University, Jazan-45142, Kingdom of Saudi Arabia;* [2]*Department of Biosciences, Jamia Millia Islamia (A Central University), New Delhi-110025, India;* [3]*Department of Laboratory Medicine, Faculty of Applied Medical Sciences, Albaha University, Albaha, Saudi Arabia;* [4]*Centre for Life Science, School of Natural Sciences, Central University of Jharkhand, Ranchi-835205, Jharkhand, India*

S.A. Dar

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Abstract: ***Background:*** DNA methyltransferase-3B (*DNMT3B*) plays a key role in establishment and maintenance of genomic methylation patterns. Polymorphism in promoter region -149 C>T (C46359T) of *DNMT3B* gene may alter *DNMT3B* activity which leads to increased susceptibility to cancer. Inconsistent results regarding this have been reported in a number of studies.

***Objective*:** To carry out a meta-analysis of the studies reported to assess the precise relationship between the *DNMT3B* -149 C>T polymorphism and the overall cancer risk.

***Method*:** PubMed (MEDLINE) web database was searched for the studies concerning *DNMT3B* -149 C>T polymorphism and its association with cancer risk. The pooled odds ratios (ORs) along with 95% confidence intervals (95% CIs) were calculated for all the genetic models, from the selected case-control studies, by meta-analysis.

***Results*:** Overall eighteen studies containing 5583 cancer cases and 7618 controls were analyzed. No significant risk was observed overall for T allele carrier (T vs. C: p=0.303; OR=1.032, 95% CI=0.972-1.097), homozygous (TT vs. CC: p=0.336; OR=1.063, 95% CI=0.939–1.204), heterozygous (CT vs. CC: p=0.802; OR=1.022, 95% CI=0.860-1.216), dominant (TT vs. CC+CT: p=0.298; OR=1.101, 95% CI=0.919-1.319) and recessive (TT+CT vs. CC: p=0.656; OR=1.021, 95% CI=0.931-1.121) genetic models. Subgroup analysis of Asian and Caucasian populations also did not demonstrate any cancer risk in all the genetic models studied.

***Conclusion*:** Our meta-analysis proposes that the *DNMT3B* -149 C>T polymorphism may not be an independent predisposing factor for the risk of cancer. However, larger sample size and expression studies are required to confirm the observation.

Keywords: DNA methyltransferase-3B, Polymorphism, Cancer, Meta-analysis, Asians, Caucasians.

INTRODUCTION

Cancer is recognized as one of the most common causes for death world over. Patients suffering from cancer live a poor quality of life and it impacts a serious socio-economic burden on the health care system [1]. The underlying causes of this malignant disease remain undetermined. Studies have suggested that susceptible genes, a few high penetrance, numerous moderate and some low penetrance, may play a significant role in cancer development [2]. However, these factors alone are not sufficient for progression of carcinogenesis, suggesting consideration of role of changes in epigenetic status in carcinogenesis.

Aberrant DNA methylation pattern is one of the many epigenetic changes in human cancers. The DNA methylation silences a number of tumor suppressor genes in cancer cells around the promoter regions on CpG islands and its level is lower in cancer cells than in normal cells [3]. A family of DNA methyltransferases (DNMTs), including DNMT1, DNMT3A and *DNMT3B*, mediate the DNA methylation in human cells [4, 5]. Among these three active forms, *DNMT3B,* which encodes DNA methyltransferase-3B and is located on chromosome 20q11.2, is a major mammalian DNA methyltransferase primarily responsible for de novo methylation process, thereby, playing oncogenic role in malignancies [6].

*Address correspondence to this author at the Research and Scientific Studies Unit, College of Nursing & Allied Health Sciences, Jazan University, Jazan - 45142, Kingdom of Saudi Arabia; Tel: +966500246938;
E-mail: sdar@jazanu.edu.sa

 # These authors contributed equally.

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Over expression of *DNMT3B* has been reported in carcinogenesis and it plays a crucial role in tumorigenesis [7, 8]. Single nucleotide polymorphisms (SNPs) located within the genes of DNMTs can change their expression levels which may affect the development of various cancers [9]. A SNP cytosine (C) >T (Thymine) C46359T (Gen Bank accession no. AL035071) located upstream of the transcription start site at the -149 base pair of the promoter region is reported to increase the promoter activity [10]. The association between *DNMT3B* -149 C>T polymorphism with cancer risk has been widely studied in several types of cancers but till now no consensus has been achieved because of conflicting results [10-27]. It is possible that small sample size with low power contributed to the false-positive or false-negative findings, indicating the significance of sample size as a methodological concern in the genetic association studies. Therefore, the use of meta- and pooled-analysis which combines the results from individual studies, both with statistically significant and non-significant observations, and weighs them by their precision as a function of sample size [28], is warranted. We in this study did a systematic meta-analysis by pooling all the published studies and examining the results to evaluate the overall possibility of a *DNMT3B* -149 C>T gene variation with cancer risk.

MATERIALS AND METHODS

Identification and Eligibility of Relevant Studies

We performed a systematic literature search through PubMed (Medline), EMBASE web databases covering all research articles published till June, 2015 using the following key words alone or in combination: ''*DNMT3B* gene AND (variant OR polymorphism OR mutation) AND Cancer or Carcinoma or malignancy". The studies showing potential relevance were examined for genetic association by scrutinizing their titles and abstracts. The studies matching with the above mentioned eligible criteria were retrieved and included in this meta-analysis.

Inclusion and Exclusion Criteria

The eligible studies had to meet the following criteria in order to minimize heterogeneity and facilitate the proper elucidation of results: (i) evaluation of the *DNMT3B* -149 C46359T (C>T) and risk of cancer, (ii) case-control study, (iii) recruited pathologically confirmed cancer cases and cancer free controls, (iv) availability of subject's genotype frequency, and (v) in English language. In case a study of same case series was published in more than one article, the study containing largest number of subjects was included. The main exclusion criteria were: (i) data overlapping, and (ii) studies including cases only and review articles.

Data Extraction and Quality Assessment

Two independent investigators assessed the methodological quality, extracted and abstracted the data for each retrieved and eligible study using a standard protocol. A data-collection form was used to ensure the accuracy of the data following the inclusion criteria listed above. Any disagreement on the collected data from the retrieved studies was discussed fully to reach a consensus. The following were the main characters abstracted from the included studies: first author's name, publication year, country of origin, source and number of cases and controls, type of study, and genotype frequencies.

Statistical Analysis

The pooled odds ratios (ORs) and their corresponding 95% confidence intervals (CIs) were calculated for each study to evaluate the relation between the *DNMT3B* -149 C>T polymorphism and the risk of cancer. The association was examined using allelic, recessive and dominant genetic models. The chi-square based Q-test was used to examine the heterogeneity assumptions [29], where *p*-value less than 0.05 indicated lack of heterogeneity among the studies. When the heterogeneity among studies was not significant, pooled ORs were calculated by the fixed-effects model [30]; otherwise, random-effects model was used [31]. To quantify inter-study variability (ranged between 0% and 100%, where a value of 0% indicates no observed heterogeneity and larger values indicate an increasing degree of heterogeneity), I^2 statistics was employed [32]. In the control group, the Hardy–Weinberg equilibrium (HWE) was measured via the chi-square test to find the departure of DNMT3B -149 polymorphism frequencies from the expected frequencies. To test the publication bias, funnel plot asymmetry was estimated by the Egger's linear regression test, where t test was used to determine the significance of the intercept and *p*-value <0.05 was considered to be representing statistically significant publication bias [33]. The comprehensive meta-analysis (CMA) version 2 software (Biostat Inc., USA) was used to perform all the statistical analyses for this study. The comparison of various meta-analysis programs is available on the web through http://meta-analysis.com/pages/comparisons.html.

RESULTS

Characteristics of Published Studies

Through literature search from the PubMed (Medline) and the EMBASE database, a total of 63 articles were included initially. These articles were examined by reading their titles, abstracts, and the full texts, and their suitability for meta-analysis was also checked. For other potentially relevant articles to be included in the study, the reference list of these retrieved articles was also screened. Further, survival studies on the *DNMT3B* polymorphism patients, and those indicating therapeutic response were excluded. After following the stringent criteria in article search, only case-control or cohort design studies, with frequency of all the three genotypes available, were included. Careful screening and application of the above mentioned stringent inclusion and exclusion criteria resulted in 18 eligible original published studies to be included in the study (Table **1**). The detailed flowchart for this selection process is shown (Fig. **1**). The genotype distribution for all the subjects, HWE (*p*-values) for the controls, and cancer susceptibility is depicted (Table **2**).

Publication Bias

To evaluate the publication bias among the included studies, the Begg's funnel plot and Egger's test were performed. No evidence of publication bias for all the comparison

Table 1. Main characteristics of *DNMT3B* -149 C>T based studies included in the meta-analysis.

First Author and Year	Cancer	Country	Ethnicity	Control	Cases	Source
Eftekhar *et al.*, 2014[a]	Breast	Iran	Caucasian	138	100	Tissue
Succi *et al.*, 2014[b]	Head and Neck	Brazil	Caucasian	488	237	Blood
Lao *et al.*, 2013[c]	Hepatocellular	China	Asian	216	108	Blood
Mostowska *et al.*, 2013[d]	Ovarian	Poland	Caucasian	180	159	Blood
Bao *et al.*, 2011[e]	Colorectal	China	Asian	533	544	Blood
Hu *et al.*, 2010[f]	Gastric	China	Asian	262	259	Blood
Karpinski *et al.*, 2010[g]	Colorectal	Poland	Caucasian	140	186	Tissue
de Vogel *et al.*, 2009[h]	Colorectal	Netherland	Caucasian	1,810	703	Mouth swab
Ezzikouri *et al.*, 2009[i]	Hepatocellular	Morocco	Mixed	222	96	Blood
Iacopetta *et al.*, 2009[j]	Colorectal	Australia	Caucasian	949	828	Buccal scrape
Liu *et al.*, 2008[k]	Squamous Cell	USA	Caucasian	843	832	Blood
Chang *et al.*, 2008[l]	Nasopharyngeal	Taiwan	Asian	250	259	Tissue
Fan *et al.*, 2008[m]	Colorectal	China	Asian	308	137	Blood
Wu and Lin, 2007[n]	Hepatocellular	China	Asian	140	100	Blood
Wang *et al.*, 2005[o]	Gastric	China	Asian	294	212	Blood
Aung *et al.*, 2005[p]	Gastric	Japan	Asian	247	152	Blood
Montgomery *et al.*, 2004[q]	Breast	UK	Caucasian	258	352	Blood
Shen *et al.*, 2002[r]	Lung	USA	Caucasian	340	319	Blood

[a] Reference 11, [b] Reference 12, [c] Reference 13, [d] Reference 14, [e] Reference 15, [f] Reference 16, [g] Reference 17, [h] Reference 18, [i] Reference 19, [j] Reference 20, [k] Reference 21, [l] Reference 22, [m] Reference 23, [n] Reference 24, [o] Reference 25, [p] Reference 26, [q] Reference 27, [r] Reference 10.

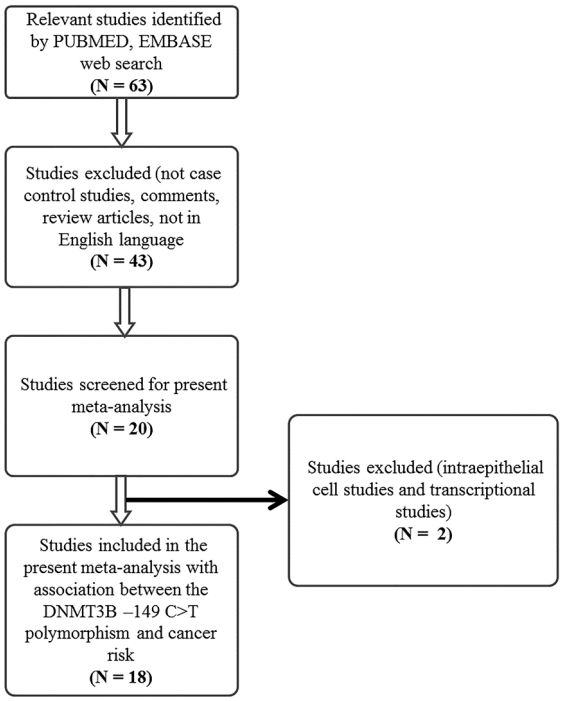

Fig. (1). Flow chart depicting the procedure of identification and selection of studies for the meta-analysis.

Table 2. Genotypic distribution of *DNMT3B*- 149 C>T gene polymorphism based studies included in the meta-analysis.

Authors and Year	Controls				Cancer Cases				HWE[b]
	Genotype			Minor Allele	Genotype			Minor Allele	
	CC	CT	TT	MAF[a]	CC	CT	TT	MAF	*p*-value
Eftekhar *et al.*, 2014[d]	27	93	18	0.46	27	47	26	0.49	<0.001
Succi *et al.*, 2014[e]	111	261	116	0.5	57	118	62	0.51	0.12
Lao *et al.*, 2013[f]	0	6	210	0.98	0	1	107	0.99	0.83
Mostowska *et al.*, 2013[g]	51	91	38	0.46	46	86	27	0.44	0.82
Bao *et al.*, 2011[h]	0	12	521	0.98	0	6	538	0.99	0.79
Karpinski *et al.*, 2010[i]	45	67	28	0.43	56	91	39	0.45	0.73
Hu *et al.*, 2010[j]	0	3	259	0.99	0	2	257	0.99	0.92
Ezzikouri *et al.*, 2009[k]	37	63	27	0.46	18	34	6	0.39	0.98
de Vogel *et al.*, 2009[l]	597	895	318	0.42	240	348	115	0.41	0.57
Iacopetta *et al.*, 2009[m]	274	463	212	0.46	247	414	167	0.45	0.53
Liu *et al.*, 2008[n]	266	433	144	0.42	259	384	189	0.45	0.15
Chang *et al.*, 2008[o]	0	0	250	1	0	0	259	1	ND[c]
Fan *et al.*, 2008[p]	0	4	304	0.99	0	2	135	0.99	0.98
Wu and Lin, 2007[q]	0	1	139	0.99	0	3	97	0.98	0.96
Wang *et al.*, 2005[r]	0	15	279	0.97	0	7	205	0.98	0.65
Aung *et al.*, 2005[s]	0	0	247	1	0	0	152	1	ND
Montgomery *et al.*, 2004[t]	120	173	59	0.41	82	116	60	0.45	0.8
Shen *et al.*, 2002[u]	119	142	79	0.44	71	181	67	0.49	0.004

[a] Minor allele frequency, [b] Hardy Weinberg equilibrium, [c] Not determined, [d] Reference 11, [e] Reference 12, [f] Reference 13, [g] Reference 14, [h] Reference 15, [i] Reference 17, [j] Reference 16, [k] Reference 19, [l] Reference 18, [m] Reference 20, [n] Reference 21, [o] Reference 22, [p] Reference 23, [q] Reference 24, [r] Reference 25, [s] Reference 26, [t] Reference 27, [u] Reference 10.

models was observed by the shape of the funnel plots and the results of Egger's test (Table **3**).

Test of Heterogeneity

The heterogeneity among the included studies was tested by Q-test and I^2 statistics. We observed heterogeneity in two genotype models, heterozygous (CT vs. CC) and recessive (TT vs CC+CT), in overall analysis. These were included for the analysis and thus random effect model was applied to calculate their pooled ORs and 95% CI (Table **3**).

Meta-analysis of *DNMT3B* -149 (C>T) Polymorphism and Cancer Susceptibility

The eighteen included studies, accumulating to a total of 7618 controls and 5583 cancer cases, were pooled together and used to assess the overall association between the *DNMT3B* -149 C>T polymorphism and cancer risk. Overall, none of the genetic models - allele (T vs. C: p = 0.303; OR= 1.032, 95% CI = 0.972-1.097), homozygous (TT vs. CC: p=0.336; OR= 1.063, 95% CI = 0.939–1.204), heterozygous (CT vs. CC: p=0.802; OR= 1.022, 95% CI = 0.860-1.216), dominant (TT vs. CC+ CT: p= 0.298; OR= 1.101, 95% CI = 0.919-1.319) and recessive (TT+CT vs. CC: p= 0.656; OR= 1.021, 95% CI = 0.931-1.121) showed any risk of developing overall cancer (Fig. **2**).

Sensitivity Analysis

For sensitivity analysis, one study at a time was excluded from the analysis to assess its influence on the pooled OR. No individual study affected the pooled OR significantly indicating the relative stability of this meta-analysis.

Subgroup Analysis by Ethnicity

We stratified the included studies into two subgroups (Asian and Caucasian) by participant's ethnicity. We did not observe any heterogeneity in all the five genetic models in Asian subgroup, hence fixed effect model was applied. Also, no publication bias existed in this subgroup (Table **3**). We observed no significant cancer risk with all the genetic models - allele (T vs. C: p=0.324; OR=1.148, 95% CI=0.873 - 1.510), homozygous (TT vs. CC: p=0.724; OR=1.119, 95% CI=0.600 - 2.089), heterozygous (CT vs. CC: p=0.733; OR=1.091, 95% CI=0.660 - 1.806), recessive (TT+CT vs. CC: p=0.694; OR=1.100, 95% CI=0.685 - 1.765) and dominant (TT vs. CC+CT: p=0.272; OR=1.249, 95% CI=0.840 - 1.857) as shown (Fig. **3**).

Table 3. Statistics to test publication bias and heterogeneity in this meta-analysis.

Comparisons	Egger's Regression Analysis			Heterogeneity Analysis			Model Used for Meta-analysis
	Intercept	95% Confidence Interval	*p*-value	Q value	$P_{heterogeneity}$	I^2 (%)	
Overall population							
T vs C	0.35	-0.60 to 1.30	0.44	17.53	0.28	14.46	Fixed
TT vs CC	0.02	-2.42 to 2.47	0.98	14.22	0.11	36.73	Fixed
CT vs CC	0.23	-2.79 to 3.27	0.86	22.28	0.008	59.61	Random
TT+CT vs CC	0.22	-2.32 to 2.76	0.84	15.75	0.72	42.87	Fixed
TT vs CC+CT	0.25	-1.11 to 1.62	0.69	29.53	0.014	49.2	Random
Asian population							
T vs C	0.22	-1.66 to 2.12	0.75	4.77	0.44	<0.001	Fixed
TT vs CC	-	-	-	<0.001	1	<0.001	Fixed
CT vs CC	-	-	-	<0.001	1	<0.001	Fixed
TT+CT vs CC	-	-	-	<0.001	1	<0.001	Fixed
TT vs CC+CT	-0.07	-2.61 to 2.46	0.93	4.51	0.47	<0.001	Fixed
Caucasian population							
T vs C	0.005	-2.07 to 2.08	0.99	10.31	0.24	22.47	Fixed
TT vs CC	-0.47	-3.57 to 2.62	0.72	12.18	0.09	42.54	Fixed
CT vs CC	0.01	-4.34 to 4.37	0.99	21.99	0.003	68.17	Random
TT+CT vs CC	-0.17	-3.72 to 3.37	0.90	14.89	0.037	53.01	Random
TT vs CC+CT	-0.07	-3.17 to 3.01	0.95	22.54	0.004	64.52	Random

Note: (-) = 95%CI could not be calculated due to absence of genotype(s) in Asian population studies.

Based on heterogeneity, random effect model was applied in three genetic models in Caucasian population - CT vs. CC; TT+CT vs. CC; and TT vs. CC+CT. However, publication bias did not exist in this subgroup also (Table **3**). We found no significant association with cancer risk under all genetic models - allele (T vs. C: p=0.163; OR=1.052, 95% CI=0.980 - 1.130), homozygous (TT vs. CC: p=0.137; OR=1.117, 95% CI=0.965 - 1.291), heterozygous (CT vs. CC: p=0.842; OR=1.024, 95% CI=0.813 - 1.290), Recessive (TT+CT vs. CC; p=0.594; OR=1.050, 95% CI=0.877 - 1.257), and dominant (TT vs. CC+CT; p=0.428; OR=1.102, 95% CI=0.867 - 1.402) as shown (Fig. **4**). Sensitivity analysis was also performed for both the ethnicities and the pooled OR was not affected significantly by any of the individual study.

DISCUSSION

DNA methylation plays an important role in the pathogenesis of malignancies by altering the expression of genes involved in cell proliferation and differentiation [34]. The DNMTs are believed to act cooperatively and maintain DNA methylation patterns, and their altered expression in tumors may partly explain aberrant methylation phenomenon in cancerous tissues or cells [35]. A number of studies have suggested the aberrant role of DNA methylation in carcinogenesis [36]. Studies have shown that the *DNMT3B* -149 C>T polymorphism may change the enzyme methylation activity and thereby influence the cancer susceptibility. This has resulted in increasing number of case-control studies in the literature performed to explore the possible association between *DNMT3B* -149 C>T polymorphism and modulations of cancer risk in different populations around the world. But, inconsistency in their results has been found prevalent which incited us to assess their overall contribution in understanding the role of this polymorphism in genetic susceptibility to cancer. Also, the inability to reproduce the results of several of these genetic variation studies has been reported, suggesting a large number of "false positive" reports [37]. Therefore, we performed the meta-analysis, in order to improve the statistical power and reliability in conclusion, of eighteen studies of *DNMT3B* -149 C>T polymorphism and overall cancer susceptibility. A meta-analysis is an emerging and powerful tool for analyzing cumulative data from different research studies with small sample sizes and low statistical power [38].

The overall pooled results of this meta-analysis revealed no increased or decreased influence of *DNMT3B* -149 C>T polymorphism on overall cancer risk in all the genetic models. When we stratified the selected studies by the ethnicity-Asian and Caucasian populations, again we failed to detect

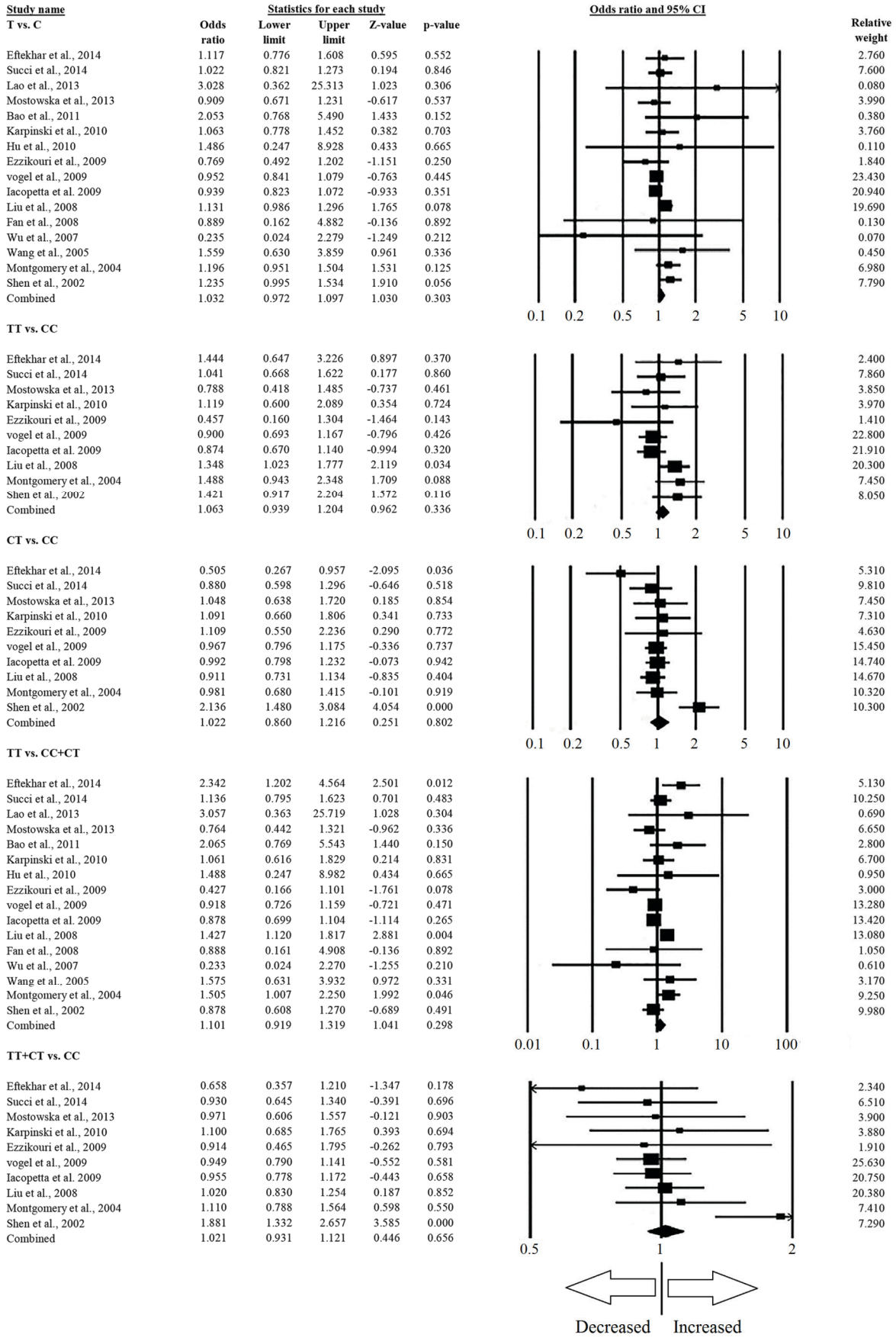

Fig. (2). Forest plot with odds ratio (OR) on overall cancer risk associated with *DNMT3B* -149 C>T gene polymorphism. The squares and horizontal lines correspond to the study specific OR and 95% confidence interval (95% CI).

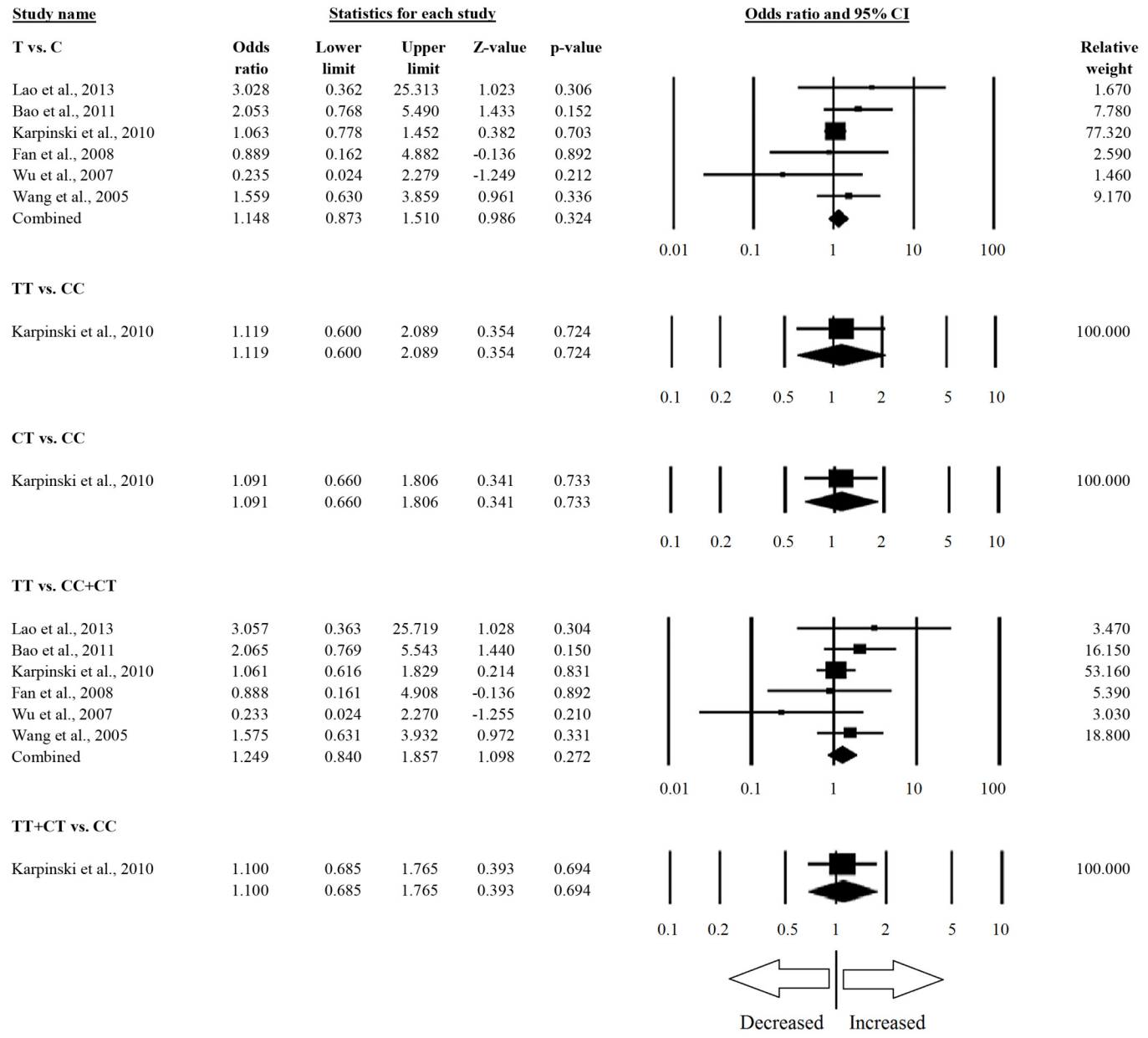

Fig. (3). Forest plot with odds ratio (OR) for the association between cancer risk and *DNMT3B* -149 C>T gene polymorphism in Asian population (subgroup analysis). The squares and horizontal lines correspond to the study specific OR and 95% confidence interval (95% CI).

significant risk of this polymorphism on cancer risk. These findings clearly indicate that the *DNMT3B* -149 C>T polymorphism may not be a potential susceptibility factor to cancer and its development in both Asian and Caucasian populations. However, the precise biological mechanism of this relationship remains unclear. In our opinion, the possible explanation may be that the *DNMT3B* -149 C>T polymorphism is not involved directly in cancer susceptibility but may be interacting in conjunction with other causative germ line polymorphisms found in linkage disequilibrium (LD). The susceptibility of cancer is multifactorial involving diverse genetic factors and pathways along with various conferred multiple loci, each with a small effect on cancer risk [39]. Hence, it is rationally inadequate to predict the cancer risk as a consequence of single genetic variation.

There were some limitations in the current meta-analysis which are acknowledged here - first, only english language studies were included; second, studies indexed by PubMed and EMBASE were included (this may have resulted in missing out on articles published in languages other than english and those indexed in other databases); third, our results were based on single-factor estimates without any adjustment for age, gender and other risk factors (e.g. smoking, drinking status etc.) because of the lack of original data. Though, there were several strengths in the current meta-analysis - first, we did not find any publication bias which indicates the statistical robustness of our results; second, our data extraction strategy was very stringent which was based on computer assisted and manual searches in order to make a trustworthy conclusion.

Study name	Odds ratio	Lower limit	Upper limit	Z-value	p-value	Relative weight
T vs. C						
Eftekhar et al., 2014	1.117	0.776	1.608	0.595	0.552	3.850
Succi et al., 2014	1.022	0.821	1.273	0.194	0.846	10.610
Mostowska et al., 2013	0.909	0.671	1.231	-0.617	0.537	5.560
Hu et al., 2010	1.486	0.247	8.928	0.433	0.665	0.160
Ezzikouri et al., 2009	0.769	0.492	1.202	-1.151	0.250	2056.000
Iacopetta et al. 2009	0.939	0.823	1.072	-0.933	0.351	29.200
Liu et al., 2008	1.131	0.986	1.296	1.765	0.078	27.460
Montgomery et al., 2004	1.196	0.951	1.504	1.531	0.125	9.730
Shen et al., 2002	1.235	0.995	1.534	1.910	0.056	10.870
Combined	1.052	0.980	1.130	1.396	0.163	
TT vs. CC						
Eftekhar et al., 2014	1.444	0.647	3.226	0.897	0.370	3.280
Succi et al., 2014	1.041	0.668	1.622	0.177	0.860	10.730
Mostowska et al., 2013	0.788	0.418	1.485	-0.737	0.461	5.260
Ezzikouri et al., 2009	0.457	0.160	1.304	-1.464	0.143	1.920
Iacopetta et al. 2009	0.874	0.670	1.140	-0.994	0.320	29.920
Liu et al., 2008	1.348	1.023	1.777	2.119	0.034	27.730
Montgomery et al., 2004	1.488	0.943	2.348	1.709	0.088	10.720
Shen et al., 2002	1.421	0.917	2.204	1.572	0.116	11.000
Combined	1.117	0.965	1.291	1.486	1.137	
CT vs. CC						
Eftekhar et al., 2014	0.505	0.267	0.957	-2.095	0.036	7.970
Succi et al., 2014	0.880	0.598	1.296	-0.646	0.518	12.980
Mostowska et al., 2013	1.048	0.638	1.720	0.185	0.854	10.510
Ezzikouri et al., 2009	1.109	0.550	2.236	0.290	0.772	7.080
Iacopetta et al. 2009	0.992	0.798	1.232	-0.073	0.942	17.280
Liu et al., 2008	0.911	0.731	1.134	-0.835	0.404	17.230
Montgomery et al., 2004	0.981	0.680	1.415	-0.101	0.919	13.480
Shen et al., 2002	2.136	1.480	3.084	4.054	0.000	13.460
Combined	1.024	0.813	1.290	0.199	0.842	
TT vs. CC+CT						
Eftekhar et al., 2014	2.342	1.202	4.564	2.501	0.012	7.960
Succi et al., 2014	1.136	0.795	1.623	0.701	0.483	14.160
Mostowska et al., 2013	0.764	0.442	1.321	-0.962	0.336	9.960
Hu et al., 2010	1.488	0.247	8.982	0.434	0.665	1.650
Ezzikouri et al., 2009	0.427	0.166	1.101	-1.761	0.078	4.920
Iacopetta et al. 2009	0.878	0.699	1.104	-1.114	0.265	17.370
Liu et al., 2008	1.427	1.120	1.817	2.881	0.004	17.040
Montgomery et al., 2004	1.505	1.007	2.250	1.992	0.046	13.070
Shen et al., 2002	0.878	0.608	1.270	-0.689	0.491	13.870
Combined	1.102	0.867	1.402	0.793	0.428	
TT+CT vs. CC						
Eftekhar et al., 2014	0.658	0.357	1.210	-1.347	0.178	6.530
Succi et al., 2014	0.930	0.645	1.340	-0.391	0.696	12.600
Mostowska et al., 2013	0.971	0.606	1.557	-0.121	0.903	9.350
Ezzikouri et al., 2009	0.914	0.465	1.795	-0.262	0.793	5.580
Iacopetta et al. 2009	0.955	0.778	1.172	-0.443	0.658	19.620
Liu et al., 2008	1.020	0.830	1.254	0.187	0.852	19.530
Montgomery et al., 2004	1.110	0.788	1.564	0.598	0.550	13.450
Shen et al., 2002	1.881	1.332	2.657	3.585	0.000	13.340
Combined	1.050	0.877	1.257	0.533	0.594	

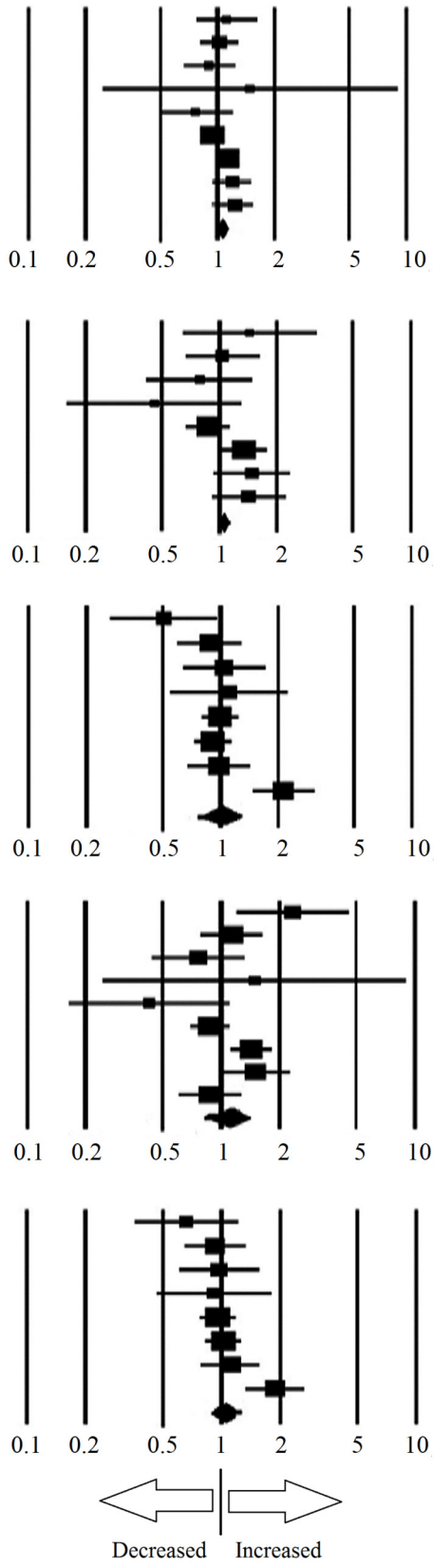

Fig. (4). Forest plot with odds ratio (OR) for the association between cancer risk and *DNMT3B* -149 C>T gene polymorphism in Caucasian population (subgroup analysis). The squares and horizontal lines correspond to the study specific OR and 95% confidence interval (95% CI).

CONCLUSION

The meta-analysis indicated that *DNMT3B* -149 C>T gene polymorphism is not associated with cancer risk overall or in subgroup ethnicities - Asian and Caucasian populations. This limits the utility of this polymorphism as a predictor or screening marker of cancer risk in asymptomatic individuals. The heterogeneity in cancer poses a great challenge to researchers focusing on cancer pathogenesis and therapy. To further validate this negative association, large scale and well-designed studies in diverse populations incorporating the role of environmental factors are needed.

LIST OF ABBREVIATIONS

DNMT3B	=	DNA Methyltransferase-3B
SNP	=	Single Nucleotide Polymorphism
T	=	Thymine
C	=	Cytosine
95%CI	=	95% Confidence Interval
ORs	=	Odds Ratio
HWE	=	Hardy Weinberg Equilibrium
LD	=	Linkage Disequilibrium

CONFLICT OF INTEREST

The author(s) confirm that this article content has no conflict of interest.

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