ORIGINAL RESEARCH

Investigation of *cyclin D1* rs9344 G>A polymorphism in colorectal cancer: a meta-analysis involving 13,642 subjects

Hao Qiu,[1,]* Chengguo Cheng,[2,]* Yafeng Wang,[3] Mingqiang Kang,[4] Weifeng Tang,[4,5] Shuchen Chen,[4] Haiyong Gu,[6] Chao Liu,[5] Yu Chen[7,8]

[1]Department of Immunology, School of Medicine, Jiangsu University, [2]Department of Pulmonary Medicine, Affiliated Hospital of Jiangsu University, Zhenjiang, [3]Department of Cardiology, The People's Hospital of Xishuangbanna Dai Autonomous Prefecture, Jinghong, [4]Department of Thoracic Surgery, Affiliated Union Hospital, Fujian Medical University, Fuzhou, [5]Department of Cardiothoracic Surgery, Affiliated People's Hospital of Jiangsu University, Zhenjiang, [6]Department of Thoracic Surgery, Shanghai Chest Hospital, Shanghai Jiaotong University, Shanghai, [7]Department of Medical Oncology, Fujian Provincial Cancer Hospital, Fujian Medical University Cancer Hospital, [8]Fujian Provincial Key Laboratory of Translational Cancer Medicine, Fuzhou, People's Republic of China

*These authors contributed equally to this work

Correspondence: Yu Chen
Department of Medical Oncology, Fujian Provincial Cancer Hospital, Fujian Medical University Cancer Hospital, 420 Fu-ma Road, Fuzhou, Fujian Province 350000, People's Republic of China
Tel +86 138 5908 9836
Email 13859089836@139.com

Abstract: The relationship between *cyclin D1* (*CCND1*) rs9344 G>A polymorphism and colorectal cancer (CRC) risk is still ambiguous. To obtain a precise estimation of the relationship, we performed an extensive meta-analysis based on the eligible studies. Crude odds ratios with their 95% confidence intervals were harnessed to determine the strength of correlation between *CCND1* rs9344 G>A polymorphism and CRC risk under the allele, the homozygote, the dominant, and the recessive genetic models, respectively (28 studies with 5,784 CRC cases and 7,858 controls). Our results indicated evidence of the association between *CCND1* rs9344 G>A polymorphism and the increased risk of CRC in four genetic models: A vs G, AA vs GG, AA+GA vs GG, and AA vs GA+GG. In a stratified analysis by cancer type of CRC, there was an increased risk of sporadic CRC found in three genetic models: A vs G, AA vs GG, and AA+GA vs GG. In a stratified analysis by ethnicity, there was an increased CRC risk found among Asians in allele comparison genetic models, as well as Caucasians in two genetic models: AA+GA vs GG and A vs T. In summary, this meta-analysis demonstrates that *CCND1* rs9344 G>A polymorphism may be a risk factor for CRC.

Keywords: polymorphism, *CCND1*, colorectal cancer, susceptibility, meta-analysis

Introduction

In 2012, colorectal cancer (CRC) is the third and second most commonly diagnosed malignancy in males and females, respectively, worldwide, with an estimated 1,360,600 new CRC cases and 693,900 CRC-related mortality occurring annually.[1] This type of malignancy involves a more frequent sporadic CRC (sCRC) and a less frequent hereditary form. The increasing CRC incidence and mortality rate have been attributed to an increasingly "Westernized lifestyle," including a decreased consumption of dietary fiber, drinking, smoking, overweight, and being physically inactive.[2] However, the etiology of CRC is very complicated. A number of altered environmental and genetic factors have been considered as risk factors for CRC.[3,4] Recently, a previous study showed that ~35% of CRC patients could be attributed to certain inherited genetic risk factors.[5] Identification of these important genetic risk factors correlated with CRC may enrich our view of this complex disease.

The *cyclin D1* (*CCND1*) gene located on chromosome 1q31-32. CCND1 is an important protein for the regulation of the G1–S phase transition of cell cycle. Overexpression or disordered regulation of the *CCND1* gene will break the balance of cell cycle and might lead to abnormalities and consequently result in cellular transformation and malignancy. Recent studies showed that *CCND1* was overexpressed in CRC,

6641

which was correlated with a poor clinical outcome and some clinicopathological characteristics.[6,7]

The human *CCND1* gene is very polymorphic (http://www.ncbi.nlm.nih.gov/SNP). The *CCND1* rs9344, a G to A polymorphism at nucleotide 870 in exon 4, increases the frequency of alternate splicing. Results of prior studies showed that the A allele of *CCND1* rs9344 G>A resulted in an increasing level of mRNA (transcript-b) encoding CCND1 protein with an altered C-terminal domain.[8,9] Results of some epidemiologic studies demonstrated that *CCND1* rs9344 G>A polymorphism might confer CRC risk.[10–18] Several meta-analyses showed that *CCND1* rs9344 G>A polymorphism might be a risk factor for CRC, especially in the subgroups of sCRC and Caucasians.[19–21] However, in these studies, as only a few case–control studies performed on the Asian populations, the power of these pooled analyses might be limited. Recently, more epidemiologic studies focusing on the relationship between *CCND1* rs9344 G>A polymorphism and CRC risk were conducted among Asians. Considering the vital role of *CCND1* rs9344 G>A polymorphism for CRC risk, an updated meta-analysis was needed to obtain a more precise assessment.

Materials and methods
Search strategy
PubMed and EMBASE online databases (updated to February 11, 2016) were searched using the corresponding keywords related to *CCND1* rs9344 G>A polymorphism and CRC: cyclin D1 or CCND1; and polymorphism, variant, or single-nucleotide polymorphism; colorectal, rectal, or colon; and cancer, carcinoma, tumor, malignancy, or neoplasm. No language restriction was applied. We also searched the bibliography of reviews, meta-analyses, and all eligible articles to retrieve the potential publications.

Inclusion and exclusion criteria
The included studies were selected according to the major criteria as follows: 1) case–control studies; 2) the association of *CCND1* rs9344 G>A polymorphism with CRC risk; 3) CRC cases diagnosed by histopathology; and 4) genotype frequencies to determine the pooled odds ratios (ORs) with their 95% confidence intervals (95% CIs). Accordingly, publications with insufficient data, reviews and meta-analyses, and comments were excluded.

Data extraction
For each included study, two authors (HQ and CC) extracted the data independently as follows: the first author's surname; year of publication; country where the study was carried out; race (included Asians, Caucasians, and Mixed); the type of CRC (included hereditary non-polyposis colorectal cancer [HNPCC] and sCRC); the source of controls (included hospital-based study [HB], population-based study, and family-based study); genotyping method; sample size (numbers of cases/controls), genotypes; and the Hardy–Weinberg equilibrium (HWE) in the controls. If these two authors could not reach a consensus, the third author (YW) was consulted to resolve the dispute by discussion.

Statistical analysis
The distribution of genotypes in controls was calculated for departure from HWE by an online test (http://ihg.gsf.de/cgi-bin/hw/hwa1.pl). The crude ORs with their 95% CIs were used to determine the strength of correlation between *CCND1* rs9344 G>A polymorphism and CRC risk. Heterogeneity assumption was assessed by the chi-square-based Q-test and I^2 test. $I^2 > 50\%$ or $P < 0.10$ indicates statistical heterogeneity among studies,[22] so the pooled ORs and CIs were measured by the random-effects model (the DerSimoian and Laird method).[23] Otherwise, the fixed-effects model (the Mantel–Haenszel method) was used.[24] In order to check the ethnicity and the type of CRC effects, subgroup analyses were performed. Moreover, one-way sensitivity analysis was performed. Publication bias was tested by visual inspection of funnel plots and formally determined by Begg's adjusted rank correlation test and Egger' linear regression test.[25] All statistical calculations were conducted with STATA version 12.0 (Stata Corporation, College Station, TX, USA). All P-values were two-sided, and $P < 0.05$ was defined as statistically significant.

Results
Characteristics
A total of 198 relevant publications were retrieved. There were several subgroups in certain publications,[15,16,26] and we treated them separately. We listed the major screening process in Figure 1. Finally, there were 28 eligible studies included in the pooled analysis.[12–18,26–42] There were 9 studies conducted in Asians,[12,13,15,18,27,30,33,37] 16 studies conducted in Caucasians,[14–17,26,28,32,34–36,38–41] and 3 studies conducted in mixed populations.[29,31,42] Of these articles, 22 investigated sCRC,[12–18,26–38] and 6 investigated HNPCC.[16,26,39–42] And the detailed characteristics of the included studies[12–18,26–42] and the distribution of the *CCND1* rs9344 G>A polymorphism as well as alleles are listed in Tables 1 and 2, respectively.
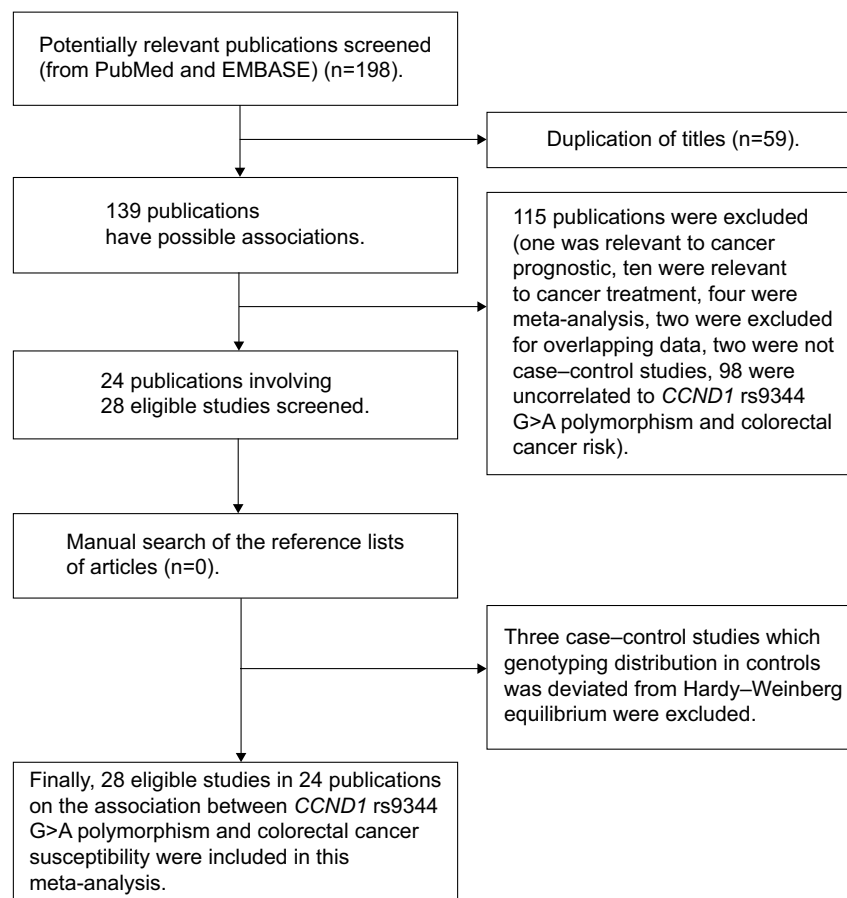
Figure 1 Flow diagram of candidate studies selection process.
Abbreviation: *CCND1*, *cyclin D1*.

Quantitative synthesis

In total, 28 eligible studies[12–18,26–42] with 5,784 CRC cases and 7,858 controls were included in our meta-analysis. Overall, the *CCND1* rs9344 G>A polymorphism was associated with the overall CRC risk in four genetic models (A vs G: OR, 1.12; 95% CI: 1.03–1.21, P=0.005; AA vs GG: OR, 1.25; 95% CI: 1.06–1.48, P=0.008; AA+GA vs GG: OR, 1.18; 95% CI: 1.05–1.33, P=0.007; AA vs GA+GG: OR, 1.13; 95% CI: 1.05–1.28, P=0.042; Table 3 and Figure 2). In a subgroup analysis by CRC type, the *CCND1* rs9344 G>A polymorphism was associated with an increased risk of sCRC in three genetic models (A vs G: OR, 1.13; 95% CI: 1.04–1.23, P=0.004; AA vs GG: OR, 1.28; 95% CI: 1.07–1.54, P=0.008; AA+GA vs GG: OR, 1.20; 95% CI: 1.06–1.36, P=0.004; Table 3 and Figure 2), but not of HNPCC. In a subgroup analysis by ethnicity, an increased CRC risk was found among Caucasians in two genetic models (A vs G: OR, 1.11; 95% CI: 1.00–1.23, P=0.049; AA+GA vs GG: OR, 1.16; 95% CI: 1.01–1.33, P=0.041; Table 3 and Figure 3), and among Asians in one genetic model (A vs G: OR, 1.17;

95% CI: 1.00–1.36, P=0.048; Table 3 and Figure 3), but not mixed populations.

Tests for publication bias, sensitivity analyses, and heterogeneity

Begg's funnel plot and Egger' linear regression test were harnessed to examine potential publication bias. As shown in Figure 4, no significant publication bias was detected in our study (Begg's test P=0.514; Egger's test P=0.259).

Influence of an individual study on the pooled ORs and CIs was also determined by omitting it in turn and repeating the meta-analysis.[43] The results indicated that no individual study significantly altered the pooled ORs and CIs (Figure 5).

As shown in Table 3, there was significant heterogeneity in all genetic models. Because ethnicity, the type of CRC, and source of controls can affect the heterogeneity, subgroup analyses were conducted. Results showed that Asians, Caucasians, population-based study, HB study, and sCRC subgroups may contribute to the major source of heterogeneity.

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

Study	Year	Country	Ethnicity	Type of CRC	Genotyping method	No of case/control	Source of controls
Govatati et al[12]	2014	India	Asians	sCRC	DNA sequencing	103/107	HB
Sameer et al[27]	2013	India	Asians	sCRC	PCR-RFLP	130/160	PB
Jelonek et al[17]	2010	Poland	Caucasians	sCRC	PCR-RFLP	50/153	PB
Yaylim-Eraltan et al[28]	2010	Turkey	Caucasians	sCRC	PCR-RFLP	57/117	HB
Kanaan et al[29]	2010	USA	Mixed	sCRC	PCR-HLC	75/93	HB
Liu et al[30]	2010	China	Asians	sCRC	PCR-RFLP	373/838	PB
Forones et al[31]	2008	Brazil	Mixed	sCRC	PCR-RFLP	123/120	HB
Tan et al[32]	2008	Germany	Caucasians	sCRC	PCR-RFLP	498/600	PB
Talseth et al[39]	2008	Australia/Poland	Caucasians	HNPCC	TaqMan	157/153	HB
Grunhage et al[26]	2008	Germany	Caucasians	HNPCC	PCR-RFLP	98/218	HB
Grunhage et al[26]	2008	Germany	Caucasians	sCRC	PCR-RFLP	96/218	HB
Jing et al[37]	2008	China	Asians	sCRC	TaqMan	104/205	HB
Josifovski et al[38]	2007	Macedonia	Caucasians	sCRC	PCR-RFLP	331/101	HB
Kruger et al[40]	2006	Germany	Caucasians	HNPCC	Multiplex-PCR	315/245	PB
Probst-Hensch et al[33]	2006	Singapore	Asians	sCRC	TaqMan	300/1,169	PB
Schernhammer et al[34]	2006	USA	Caucasians	sCRC	TaqMan	610/1,237	PB
Jiang et al[13]	2006	India	Asians	sCRC	PCR-RFLP	301/291	HB
Hong et al[18]	2005	Singapore	Asians	sCRC	PCR-RFLP	254/101	PB
Grieu et al[35]	2003	Australia	Caucasians	sCRC	PCR-SSCP	569/327	HB
Le Marchand et al[15]	2003	USA	Asians	sCRC	PCR-RFLP	70/83	PB
Le Marchand et al[15]	2003	USA	Asians	sCRC	PCR-RFLP	296/380	PB
Le Marchand et al[15]	2003	USA	Caucasians	sCRC	PCR-RFLP	138/161	PB
Porter et al[16]	2002	UK	Caucasians	HNPCC	PCR-RFLP	99/171	PB
Porter et al[16]	2002	UK	Caucasians	sCRC	PCR-RFLP	235/171	PB
Bala and Peltomaki[41]	2001	Finland	Caucasians	HNPCC	PCR-SSCP	146/186	FB
Kong et al[14]	2001	USA	Caucasians	sCRC	PCR-SSCP	156/152	PB
McKay et al[36]	2000	UK	Caucasians	sCRC	PCR-RFLP	100/101	PB
Kong et al[42]	2000	USA	Mixed	HNPCC	PCR-SSCP	49/37	FB

Abbreviations: FB, family-based study; HB, hospital-based study; HNPCC, hereditary nonpolyposis colorectal cancer; PB, population-based; PCR-HLC, polymerase chain reaction high-performance liquid chromatography; PCR-RFLP, polymerase chain reaction-restriction fragment length polymorphism; PCR-SSCP, polymerase chain reaction single-stranded conformation polymorphism; sCRC, sporadic colorectal cancer.

Table 2 Distribution of *CCND1* rs9344 G>A polymorphism genotypes and alleles

Study	Year	Case			Control			Case		Control		HWE
		GG	GA	AA	GG	GA	AA	A	G	A	G	
Govatati et al[12]	2014	54	39	10	71	33	3	59	147	39	175	Yes
Sameer et al[27]	2013	19	70	41	41	76	43	152	108	162	158	Yes
Jelonek et al[17]	2010	12	33	5	44	71	38	43	57	147	159	Yes
Yaylim-Eraltan et al[28]	2010	9	28	20	29	60	28	68	46	116	118	Yes
Kanaan et al[29]	2010	19	39	17	24	48	21	73	77	90	96	Yes
Liu et al[30]	2010	66	187	120	160	429	249	427	319	927	749	Yes
Forones et al[31]	2008	36	66	21	34	67	19	108	138	105	135	Yes
Tan et al[32]	2008	120	263	115	147	310	143	493	503	596	604	Yes
Talseth et al[39]	2008	34	78	45	42	80	31	168	146	142	164	Yes
Grunhage et al[26]	2008	13	50	35	48	109	61	120	76	231	205	Yes
Grunhage et al[26]	2008	24	43	29	48	109	61	101	91	231	205	Yes
Jing et al[37]	2008	11	61	32	41	113	51	125	83	215	195	Yes
Josifovski et al[38]	2007	77	153	100	25	51	25	353	307	101	101	Yes
Kruger et al[40]	2006	110	144	61	73	121	51	266	364	223	267	Yes
Probst-Hensch et al[33]	2006	56	132	112	207	548	414	356	244	1,376	962	Yes
Schernhammer et al[34]	2006	125	311	174	264	593	380	659	561	1,353	1,121	Yes
Jiang et al[13]	2006	46	130	125	56	145	90	380	222	325	257	Yes
Hong et al[18]	2005	55	128	71	12	50	39	270	238	128	74	Yes
Grieu et al[35]	2003	142	313	114	90	158	79	541	597	316	338	Yes
Le Marchand et al[15]	2003	5	35	30	18	35	30	95	45	95	71	Yes
Le Marchand et al[15]	2003	75	143	78	96	195	89	299	293	373	387	Yes
Le Marchand et al[15]	2003	29	75	34	50	85	26	143	133	137	185	Yes
Porter et al[16]	2002	30	47	22	60	81	30	91	107	141	201	Yes
Porter et al[16]	2002	55	128	52	60	81	30	232	238	141	201	Yes
Bala and Peltomaki[41]	2001	50	70	26	47	97	42	122	170	181	191	Yes
Kong et al[14]	2001	36	71	49	45	84	23	169	143	130	174	Yes
McKay et al[36]	2000	25	58	17	34	50	17	92	108	84	118	Yes
Kong et al[42]	2000	9	36	4	10	21	6	44	54	33	41	Yes

Abbreviation: HWE, Hardy–Weinberg equilibrium.

Table 3 Meta-analysis of the *CCND1* rs9344 G>A polymorphism and CRC risk

Group	No of study	A vs G			AA vs GG			AA+GA vs GG			AA vs GA+GG		
		OR (95% CI)	P-value	P-value (Q-test)	OR (95% CI)	P-value	P-value (Q-test)	OR (95% CI)	P-value	P-value (Q-test)	OR (95% CI)	P-value	P-value (Q-test)
Total	28	**1.12 (1.03–1.21)**	**0.005**	0.001	**1.25 (1.06–1.48)**	**0.008**	<0.001	**1.18 (1.05–1.33)**	**0.007**	0.013	**1.13 (1.0–1.28)**	**0.042**	0.005
Ethnicity													
Asians	9	**1.17 (1.00–1.36)**	**0.048**	0.004	1.38 (0.99–1.94)	0.059	0.002	1.26 (0.96–1.65)	0.092	0.005	1.18 (0.98–1.42)	0.074	0.092
Caucasians	16	**1.11 (1.00–1.23)**	**0.049**	0.005	1.23 (1.00–1.53)	0.055	0.090	**1.16 (1.01–1.33)**	**0.041**	0.090	1.13 (0.95–1.35)	0.167	0.005
Mixed	3	1.01 (0.79–1.30)	0.944	1.000	0.99 (0.58–1.71)	0.978	0.925	1.06 (0.71–1.58)	0.767	0.653	0.95 (0.60–1.51)	0.830	0.519
Type of CRC													
sCRC	22	**1.13 (1.04–1.23)**	**0.004**	0.002	**1.28 (1.07–1.54)**	**0.008**	0.001	**1.20 (1.06–1.36)**	**0.004**	0.045	1.14 (1.00–1.31)	0.054	0.004
HNPCC	6	1.06 (0.86–1.32)	0.578	0.035	1.13 (0.73–1.76)	0.581	0.037	1.08 (0.78–1.51)	0.630	0.051	1.10 (0.88–1.37)	0.420	0.177
Source of control													
HB	11	**1.19 (1.08–1.30)**	**<0.001**	0.161	**1.38 (1.14–1.68)**	**0.001**	0.140	**1.27 (1.08–1.48)**	**0.003**	0.484	**1.25 (1.07–1.45)**	**0.004**	0.134
PB	15	1.09 (0.99–1.21)	0.085	0.003	1.21 (0.97–1.51)	0.088	0.001	1.16 (0.99–1.37)	0.065	0.012	1.09 (0.94–1.27)	0.263	0.013
FB	2	0.80 (0.61–1.06)	0.120	0.404	0.60 (0.34–1.08)	0.089	0.778	0.77 (0.50–1.18)	0.227	0.106	0.69 (0.42–1.15)	0.157	0.516

Note: Statistically significant values are shown in bold.
Abbreviations: CRC, colorectal cancer; CI, confidence interval; FB, family-based study; HB, hospital-based study; HNPCC, hereditary nonpolyposis colorectal cancer; HWE, Hardy–Winberg equilibrium; OR, odds ratio; PB, population-based; sCRC, sporadic colorectal cancer.

Discussion

CCND1 may act as an important regulator in the evolution of malignancy by influencing cell proliferation, differentiation, and apoptosis. It has been reported that the G1–S transition of the cell cycle is controlled by sequential activation of cyclin/cyclin-dependent kinase (CDK) complexes.[44] The CCND1, a vital cell cycle regulatory protein, regulates transition of G1–S phase during cell division. High activity of CCND1 leads to premature cell passage through the G1–S transition, resulting in proliferation of unrepaired DNA damage and genetic errors, thus leading to selective advantage for abnormal cell propagation.[45] Previous studies indicated that CCND1 was overexpressed in a number of malignancies.[6,46] Owing to these important roles in carcinogenesis, polymorphisms of *CCND1* may be implicated in accelerating the development and/or progression of CRC.

Of late, numerous epidemiologic investigations focused on the relationship of the *CCND1* polymorphism with CRC risk.[12–18,26–42] The most prevalent *CCND1* gene polymorphism, rs9344 G>A, has been most widely explored. High activity of CCND1 is common in a lot of human tumors.[47,48] Several case–control studies have reported a positive signal of the *CCND1* rs9344 G>A polymorphism with the risk of CRC;[10–16] however, others have reported negative signal.[17,18] Because of conflicting results and the insufficient sample size of individual studies, the final decision was far from certain. Because meta-analysis is a powerful way for pooling the results of all included studies with a more power, it can get more robust results than an individual study.[49] Our findings showed that the presence of the *CCND1* rs9344 A allele, which elevate CCND1 activity,[8,9] might confer the susceptibility to CRC. In addition, subgroup analyses were performed regarding ethnicity and the type of CRC for this polymorphism. *CCND1* rs9344 G>A polymorphism increased the risk of CRC among Asians, Caucasians, and sCRC. Results of the current meta-analysis indicated the influence of the *CCND1* rs9344 G>A polymorphism and diversity on the type of CRC. However, our results should be interpreted with very caution. For HNPCC, only six studies with small sample sizes were included in this group, which may restrict the statistical power to obtain a final decision.[16,26,39–42] When stratified by ethnicity, the *CCND1* rs9344 G>A polymorphism was associated with CRC risk in both Asians and Caucasians. Additionally, in other genetic models, a borderline risk of CRC was also observed in these two ethnicities. Results of several previous meta-analyses showed that the *CCND1* rs9344 G>A polymorphism might be a risk factor for CRC, especially in the subgroups of sCRC and Caucasians.[19–21] Our results were very analogous to these
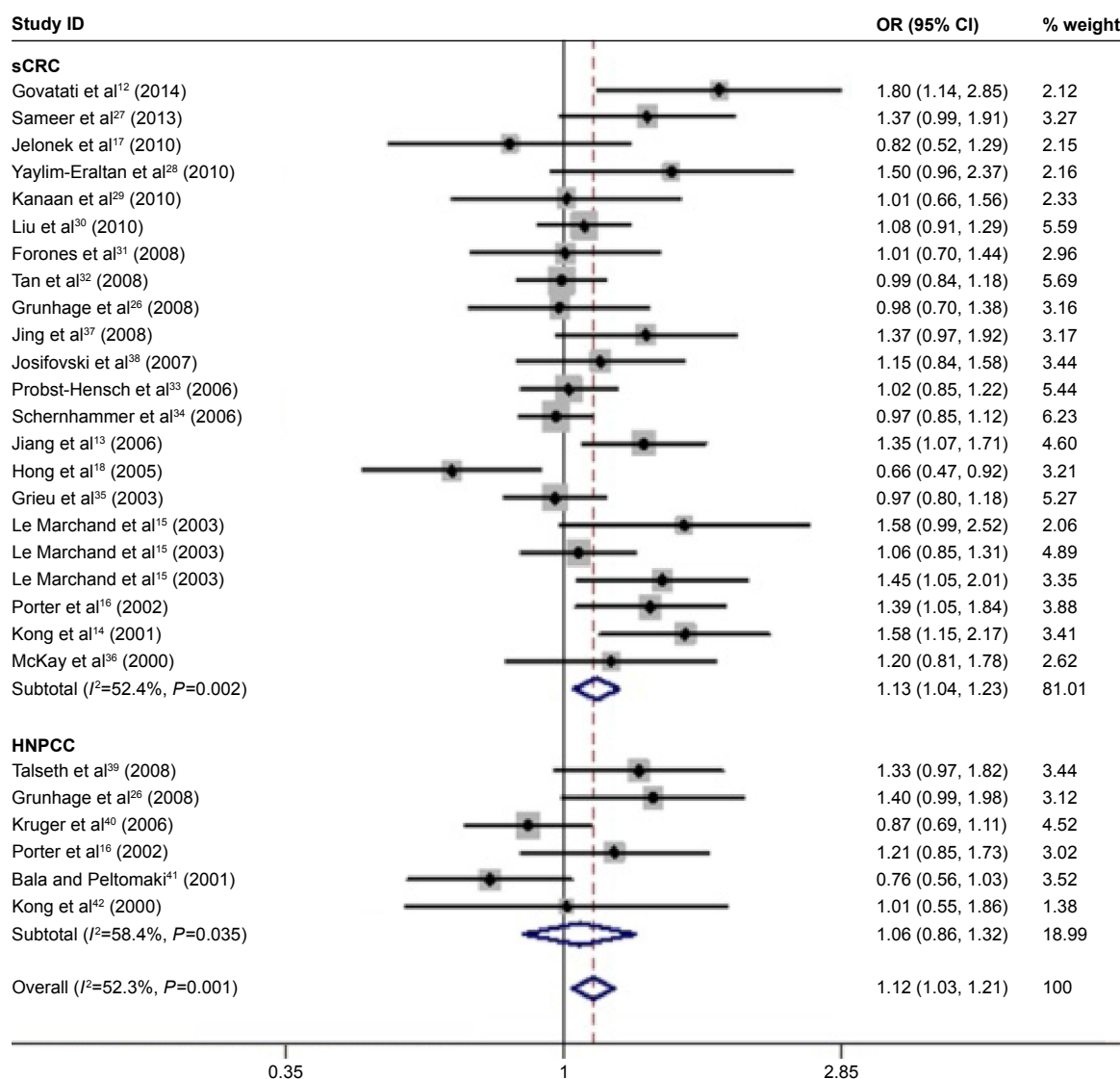
Study ID		OR (95% CI)	% weight
sCRC			
Govatati et al[12] (2014)		1.80 (1.14, 2.85)	2.12
Sameer et al[27] (2013)		1.37 (0.99, 1.91)	3.27
Jelonek et al[17] (2010)		0.82 (0.52, 1.29)	2.15
Yaylim-Eraltan et al[28] (2010)		1.50 (0.96, 2.37)	2.16
Kanaan et al[29] (2010)		1.01 (0.66, 1.56)	2.33
Liu et al[30] (2010)		1.08 (0.91, 1.29)	5.59
Forones et al[31] (2008)		1.01 (0.70, 1.44)	2.96
Tan et al[32] (2008)		0.99 (0.84, 1.18)	5.69
Grunhage et al[26] (2008)		0.98 (0.70, 1.38)	3.16
Jing et al[37] (2008)		1.37 (0.97, 1.92)	3.17
Josifovski et al[38] (2007)		1.15 (0.84, 1.58)	3.44
Probst-Hensch et al[33] (2006)		1.02 (0.85, 1.22)	5.44
Schernhammer et al[34] (2006)		0.97 (0.85, 1.12)	6.23
Jiang et al[13] (2006)		1.35 (1.07, 1.71)	4.60
Hong et al[18] (2005)		0.66 (0.47, 0.92)	3.21
Grieu et al[35] (2003)		0.97 (0.80, 1.18)	5.27
Le Marchand et al[15] (2003)		1.58 (0.99, 2.52)	2.06
Le Marchand et al[15] (2003)		1.06 (0.85, 1.31)	4.89
Le Marchand et al[15] (2003)		1.45 (1.05, 2.01)	3.35
Porter et al[16] (2002)		1.39 (1.05, 1.84)	3.88
Kong et al[14] (2001)		1.58 (1.15, 2.17)	3.41
McKay et al[36] (2000)		1.20 (0.81, 1.78)	2.62
Subtotal (*I*²=52.4%, *P*=0.002)		1.13 (1.04, 1.23)	81.01
HNPCC			
Talseth et al[39] (2008)		1.33 (0.97, 1.82)	3.44
Grunhage et al[26] (2008)		1.40 (0.99, 1.98)	3.12
Kruger et al[40] (2006)		0.87 (0.69, 1.11)	4.52
Porter et al[16] (2002)		1.21 (0.85, 1.73)	3.02
Bala and Peltomaki[41] (2001)		0.76 (0.56, 1.03)	3.52
Kong et al[42] (2000)		1.01 (0.55, 1.86)	1.38
Subtotal (*I*²=58.4%, *P*=0.035)		1.06 (0.86, 1.32)	18.99
Overall (*I*²=52.3%, *P*=0.001)		1.12 (1.03, 1.21)	100

0.35   1   2.85

Figure 2 Meta-analysis with a random–effects model in the different type for the association between *CCND1* rs9344 G>A polymorphism and CRC risk (A vs G genetic model).
Note: Weights are from random-effects analysis.
Abbreviations: CI, confidence interval; CRC, colorectal cancer; HNPCC, hereditary nonpolyposis colorectal cancer; OR, odds ratio; sCRC, sporadic colorectal cancer.

pooled analyses. In addition, we also found that the *CCND1* rs9344 G>A polymorphism might be a risk factor for CRC risk in Asians.

The *CCND1* rs9344 G allele may provide an optimal splice donor site and produce a full transcript for CCND1 (transcript a), whereas the *CCND1* rs9344 A allele results in a truncated transcript (transcript b).[47,50,51] The well-described transcript (transcript a) interacts with and activates the downstream molecules, such as G1 CDK, CDK4, and CDK6. Then, the CCND1–CDK complex phosphorylates and inhibits the retinoblastoma tumor suppressor, which is necessary for the G1–S transition.[52] However, a truncated transcript (transcript b) encodes the protein short of the point estimation by

sequential testing (PEST) region in the C-terminal domain[47] and decreases phosphorylation ability of retinoblastoma.[53] On the other hand, the transcript b has a longer half-life than transcript a, which may result in an overexpression of CCND1. Subsequently, the *CCND1* rs9344 G→A substitution could lead to facilitation of cell proliferation and increase the susceptibility of malignancy.[50] The findings of our meta-analysis were consistent with the conclusion of previous functional studies mentioned earlier. The epidemiologic investigations provided evidence suggesting that CRC carcinogenesis may be multiple steps that involve both individual's genetic and environmental factors. In the future, larger epidemiologic studies with a well-designed methodology are needed to
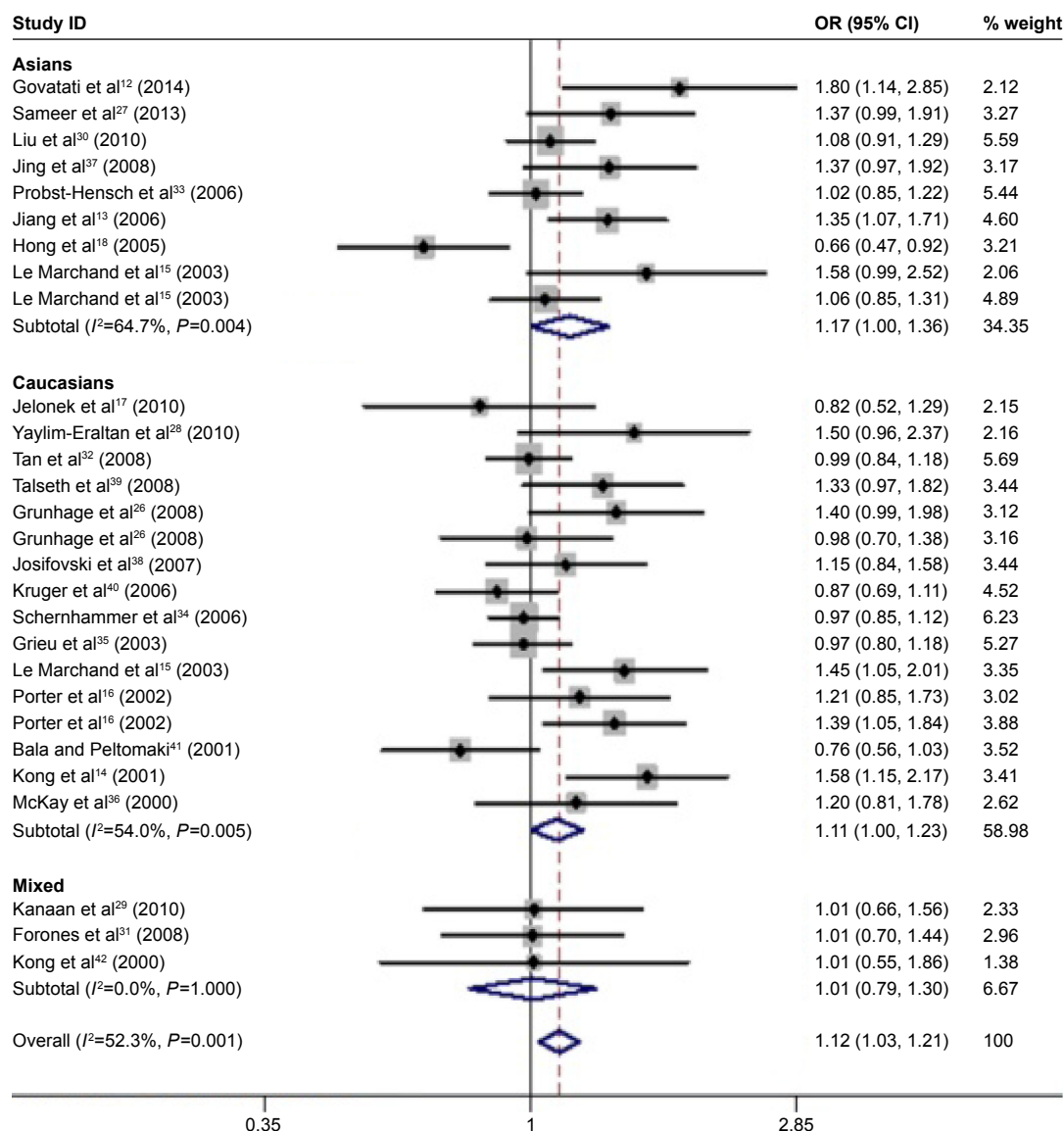
Study ID	OR (95% CI)	% weight
Asians		
Govatati et al[12] (2014)	1.80 (1.14, 2.85)	2.12
Sameer et al[27] (2013)	1.37 (0.99, 1.91)	3.27
Liu et al[30] (2010)	1.08 (0.91, 1.29)	5.59
Jing et al[37] (2008)	1.37 (0.97, 1.92)	3.17
Probst-Hensch et al[33] (2006)	1.02 (0.85, 1.22)	5.44
Jiang et al[13] (2006)	1.35 (1.07, 1.71)	4.60
Hong et al[18] (2005)	0.66 (0.47, 0.92)	3.21
Le Marchand et al[15] (2003)	1.58 (0.99, 2.52)	2.06
Le Marchand et al[15] (2003)	1.06 (0.85, 1.31)	4.89
Subtotal (I^2=64.7%, P=0.004)	1.17 (1.00, 1.36)	34.35
Caucasians		
Jelonek et al[17] (2010)	0.82 (0.52, 1.29)	2.15
Yaylim-Eraltan et al[28] (2010)	1.50 (0.96, 2.37)	2.16
Tan et al[32] (2008)	0.99 (0.84, 1.18)	5.69
Talseth et al[39] (2008)	1.33 (0.97, 1.82)	3.44
Grunhage et al[26] (2008)	1.40 (0.99, 1.98)	3.12
Grunhage et al[26] (2008)	0.98 (0.70, 1.38)	3.16
Josifovski et al[38] (2007)	1.15 (0.84, 1.58)	3.44
Kruger et al[40] (2006)	0.87 (0.69, 1.11)	4.52
Schernhammer et al[34] (2006)	0.97 (0.85, 1.12)	6.23
Grieu et al[35] (2003)	0.97 (0.80, 1.18)	5.27
Le Marchand et al[15] (2003)	1.45 (1.05, 2.01)	3.35
Porter et al[16] (2002)	1.21 (0.85, 1.73)	3.02
Porter et al[16] (2002)	1.39 (1.05, 1.84)	3.88
Bala and Peltomaki[41] (2001)	0.76 (0.56, 1.03)	3.52
Kong et al[14] (2001)	1.58 (1.15, 2.17)	3.41
McKay et al[36] (2000)	1.20 (0.81, 1.78)	2.62
Subtotal (I^2=54.0%, P=0.005)	1.11 (1.00, 1.23)	58.98
Mixed		
Kanaan et al[29] (2010)	1.01 (0.66, 1.56)	2.33
Forones et al[31] (2008)	1.01 (0.70, 1.44)	2.96
Kong et al[42] (2000)	1.01 (0.55, 1.86)	1.38
Subtotal (I^2=0.0%, P=1.000)	1.01 (0.79, 1.30)	6.67
Overall (I^2=52.3%, P=0.001)	1.12 (1.03, 1.21)	100

0.35   1   2.85

Figure 3 Meta-analysis with a random–effects model in different races for the association between the *CCND1* rs9344 G>A polymorphism and CRC risk (A vs G genetic model).
Note: Weights are from random-effects analysis.
Abbreviations: CI, confidence interval; CRC, colorectal cancer; OR, odds ratio.
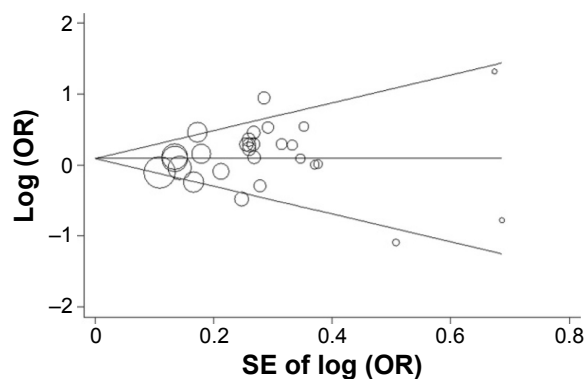
Figure 4 Begg's funnel plot of meta-analysis of the relationship between the *CCND1* rs9344 G>A polymorphism and CRC risk (AA vs GA+GG genetic model).
Abbreviations: CRC, colorectal cancer; OR, odds ratio; SE, standard error.

confirm or refute these associations. Results of our pooled analysis may prompt further clinic investigation of diagnosis and prevention strategies.

There were some merits in this meta-analysis. First, the current meta-analysis was the most extensively study which explored the relationship of the *CCND1* rs9344 G>A polymorphism with CRC susceptibility. Second, our results first confirmed that the *CCND1* rs9344 G>A polymorphism was associated with CRC susceptibility among Asians.

Limitations

There were some limitations of our study. First, in some included studies, controls were selected from family member
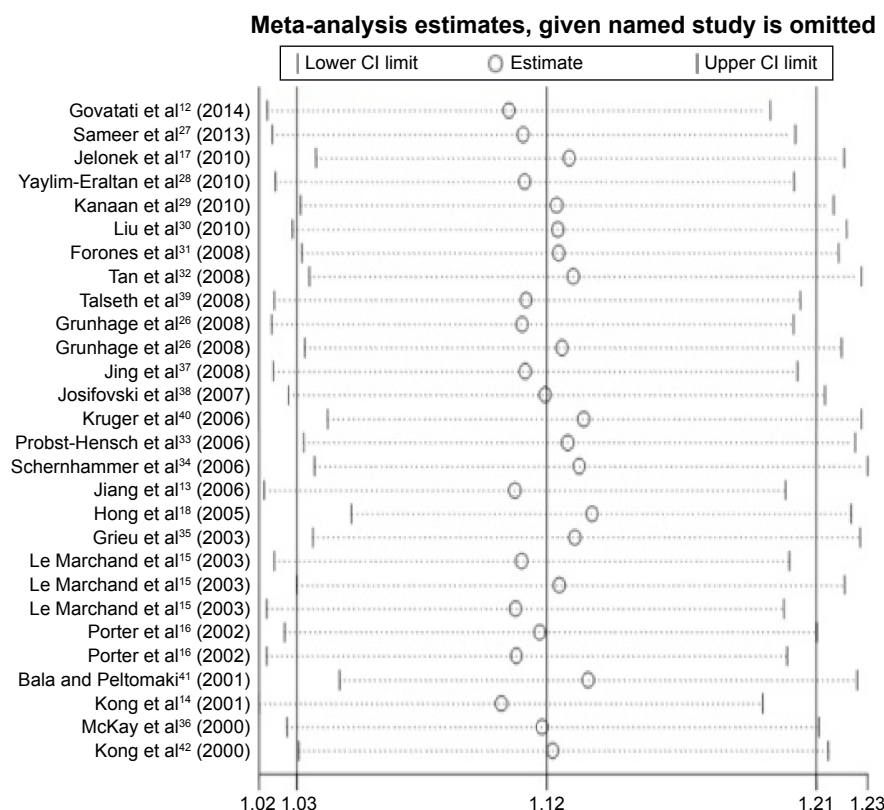
Meta-analysis estimates, given named study is omitted

Figure 5 Sensitivity analysis of the influence of A vs G genetic model in overall CRC meta-analysis (random–effects estimates).
Abbreviations: CI, confidence interval; CRC, colorectal cancer.

and non-cancer hospital patients, which might result in misclassification bias. Second, large heterogeneity was observed in our meta-analysis, which means our findings should be interpreted with caution. Finally, our findings were based on unadjusted ORs and CIs, while a more precise measurement should be adjusted by multiple risk factors, such as family history, smoking status, drinking, diabetes, body mass index, etc.

Conclusion

In summary, this meta-analysis suggests that the *CCND1* rs9344 G>A polymorphism is correlated with increased risk of CRC. Moreover, these relationships were different across different cancer types of CRC, suggesting that large sample and well-designed epidemiologic studies are warranted to confirm or refute our findings.

Acknowledgments

Disclosure

The authors report no conflicts of interest in this work.

References

1. Torre LA, Bray F, Siegel RL, Ferlay J, Lortet-Tieulent J, Jemal A. Global cancer statistics, 2012. *CA Cancer J Clin*. 2015;65(2):87–108.
2. Center MM, Jemal A, Smith RA, Ward E. Worldwide variations in colorectal cancer. *CA Cancer J Clin*. 2009;59(6):366–378.
3. Raskov H, Pommergaard HC, Burcharth J, Rosenberg J. Colorectal carcinogenesis – update and perspectives. *World J Gastroenterol*. 2014; 20(48):18151–18164.
4. Weinberg DS, Myers RE, Keenan E, et al. Genetic and environmental risk assessment and colorectal cancer screening in an average-risk population: a randomized trial. *Ann Intern Med*. 2014;161(8): 537–545.
5. Markowitz SD, Bertagnolli MM. Molecular origins of cancer: Molecular basis of colorectal cancer. *N Engl J Med*. 2009;361(25):2449–2460.
6. Huang MY, Chen HC, Yang IP, et al. Tumorigenesis and tumor progression related gene expression profiles in colorectal cancer. *Cancer Biomark*. 2013;13(4):269–279.
7. Li Y, Wei J, Xu C, Zhao Z, You T. Prognostic significance of cyclin D1 expression in colorectal cancer: a meta-analysis of observational studies. *PLoS One*. 2014;9(4):e94508.
8. Sobti RC, Kaur P, Kaur S, et al. Effects of cyclin D1 (CCND1) polymorphism on susceptibility to lung cancer in a North Indian population. *Cancer Genet Cytogenet*. 2006;170(2):108–114.

9. Fan YZ, Fu JY, Zhao ZM, Chen CQ. Inhibitory effect of norcantharidin on the growth of human gallbladder carcinoma GBC-SD cells in vitro. *Hepatobiliary Pancreat Dis Int*. 2007;6(1):72–80.

10. Zahary MN, Ahmad Aizat AA, Kaur G, Yeong Yeh L, Mazuwin M, Ankathil R. Polymorphisms of cell cycle regulator genes G870A and C215G: association with colorectal cancer susceptibility risk in a Malaysian population. *Oncol Lett*. 2015;10(5):3216–3222.

11. Huang CY, Tsai CW, Hsu CM, Chang WS, Shui HA, Bau DT. The significant association of CCND1 genotypes with colorectal cancer in Taiwan. *Tumour Biol*. 2015;36(8):6533–6540.

12. Govatati S, Singamsetty GK, Nallabelli N, et al. Contribution of cyclin D1 (CCND1) and E-cadherin (CDH1) alterations to colorectal cancer susceptibility: a case-control study. *Tumour Biol*. 2014;35(12): 12059–12067.

13. Jiang J, Wang J, Suzuki S, et al. Elevated risk of colorectal cancer associated with the AA genotype of the cyclin D1 A870G polymorphism in an Indian population. *J Cancer Res Clin Oncol*. 2006;132(3): 193–199.

14. Kong S, Wei Q, Amos CI, et al. Cyclin D1 polymorphism and increased risk of colorectal cancer at young age. *J Natl Cancer Inst*. 2001;93(14): 1106–1108.

15. Le Marchand L, Seifried A, Lum-Jones A, Donlon T, Wilkens LR. Association of the cyclin D1 A870G polymorphism with advanced colorectal cancer. *JAMA*. 2003;290(21):2843–2848.

16. Porter TR, Richards FM, Houlston RS, et al. Contribution of cyclin d1 (CCND1) and E-cadherin (CDH1) polymorphisms to familial and sporadic colorectal cancer. *Oncogene*. 2002;21(12):1928–1933.

17. Jelonek K, Gdowicz-Klosok A, Pietrowska M, et al. Association between single-nucleotide polymorphisms of selected genes involved in the response to DNA damage and risk of colon, head and neck, and breast cancers in a polish population. *J Appl Genet*. 2010;51(3): 343–352.

18. Hong Y, Eu KW, Seow-Choen F, Fook-Chong S, Cheah PY. GG genotype of cyclin D1 G870A polymorphism is associated with increased risk and advanced colorectal cancer in patients in Singapore. *Eur J Cancer*. 2005;41(7):1037–1044.

19. Yang Y, Wang F, Shi C, Zou Y, Qin H, Ma Y. Cyclin D1 G870A polymorphism contributes to colorectal cancer susceptibility: evidence from a systematic review of 22 case-control studies. *PLoS One*. 2012;7(5):e36813.

20. Yang J, Zhang G, Chen J. CCND1 G870A polymorphism is associated with increased risk of colorectal cancer, especially for sporadic colorectal cancer and in Caucasians: a meta-analysis. *Clin Res Hepatol Gastroenterol*. 2012;36(2):169–177.

21. Chen B, Cao L, Yang P, Zhou Y, Wu XT. Cyclin D1 (CCND1) G870A gene polymorphism is an ethnicity-dependent risk factor for digestive tract cancers: a meta-analysis comprising 20,271 subjects. *Cancer Epidemiol*. 2012;36(2):106–115.

22. Higgins JP, Thompson SG, Deeks JJ, Altman DG. Measuring inconsistency in meta-analyses. *BMJ*. 2003;327(7414):557–560.

23. DerSimonian R, Laird N. Meta-analysis in clinical trials. *Control Clin Trials*. 1986;7(3):177–188.

24. Mantel N, Haenszel W. Statistical aspects of the analysis of data from retrospective studies of disease. *J Natl Cancer Inst*. 1959;22(4): 719–748.

25. Egger M, Davey Smith G, Schneider M, Minder C. Bias in meta-analysis detected by a simple, graphical test. *BMJ*. 1997;315(7109): 629–634.

26. Grunhage F, Jungck M, Lamberti C, et al. Association of familial colorectal cancer with variants in the E-cadherin (CDH1) and cyclin D1 (CCND1) genes. *Int J Colorectal Dis*. 2008;23(2):147–154.

27. Sameer AS, Parray FQ, Dar MA, et al. Cyclin D1 G870A polymorphism and risk of colorectal cancer: a case control study. *Mol Med Rep*. 2013;7(3):811–815.

28. Yaylim-Eraltan I, Arikan S, Yildiz Y, et al. The influence of cyclin D1 A870G polymorphism on colorectal cancer risk and prognosis in a Turkish population. *Anticancer Res*. 2010;30(7):2875–2880.

29. Kanaan Z, Eichenberger MR, Young M, et al. An alternative cyclin-D1 splice site is not linked to inflammatory bowel disease-associated neoplasia. *Int J Biol Markers*. 2010;25(1):27–31.

30. Liu B, Zhang Y, Jin M, et al. Association of selected polymorphisms of CCND1, p21, and caspase8 with colorectal cancer risk. *Mol Carcinog*. 2010;49(1):75–84.

31. Forones NM, de Lima JM, de Souza LG, da Silva ID. Cyclin D1 A870G polymorphism in Brazilian colorectal cancer patients. *J Gastrointest Cancer*. 2008;39(1–4):118–123.

32. Tan XL, Nieters A, Kropp S, Hoffmeister M, Brenner H, Chang-Claude J. The association of cyclin D1 G870A and E-cadherin C-160A polymorphisms with the risk of colorectal cancer in a case control study and meta-analysis. *Int J Cancer*. 2008;122(11):2573–2580.

33. Probst-Hensch NM, Sun CL, Van Den Berg D, Ceschi M, Koh WP, Yu MC. The effect of the cyclin D1 (CCND1) A870G polymorphism on colorectal cancer risk is modified by glutathione-S-transferase polymorphisms and isothiocyanate intake in the Singapore Chinese Health Study. *Carcinogenesis*. 2006;27(12):2475–2482.

34. Schernhammer ES, Tranah GJ, Giovannucci E, et al. Cyclin D1 A870G polymorphism and the risk of colorectal cancer and adenoma. *Br J Cancer*. 2006;94(6):928–934.

35. Grieu F, Malaney S, Ward R, Joseph D, Iacopetta B. Lack of association between CCND1 G870A polymorphism and the risk of breast and colorectal cancers. *Anticancer Res*. 2003;23(5b):4257–4259.

36. McKay JA, Douglas JJ, Ross VG, et al. Cyclin D1 protein expression and gene polymorphism in colorectal cancer. Aberdeen Colorectal Initiative. *Int J Cancer*. 2000;88(1):77–81.

37. Jing JC XW, Jin H, Ji BN. Relationship between CCND1 gene A870G polymorphism and genetic susceptibility to colorectal cancer. *World Chin J Digestol*. 2008;16(22):4.

38. Josifovski T, Matevska N, Hiljadnikova-Bajro M, et al. Cyclin D1 G870A variant is associated with increased risk of microsatellite instability-positive colorectal cancer in young male patients. *Balkan J Med Med Genetics*. 2007;10(2):8.

39. Talseth BA, Ashton KA, Meldrum C, et al. Aurora-A and Cyclin D1 polymorphisms and the age of onset of colorectal cancer in hereditary nonpolyposis colorectal cancer. *Int J Cancer*. 2008;122(6):1273–1277.

40. Kruger S, Engel C, Bier A, et al. Absence of association between cyclin D1 (CCND1) G870A polymorphism and age of onset in hereditary nonpolyposis colorectal cancer. *Cancer Lett*. 2006;236(2): 191–197.

41. Bala S, Peltomaki P. CYCLIN D1 as a genetic modifier in hereditary nonpolyposis colorectal cancer. *Cancer Res*. 2001;61(16):6042–6045.

42. Kong S, Amos CI, Luthra R, Lynch PM, Levin B, Frazier ML. Effects of cyclin D1 polymorphism on age of onset of hereditary nonpolyposis colorectal cancer. *Cancer Res*. 2000;60(2):249–252.

43. Azzam A, Mathews CA. Meta-analysis of the association between the catecholamine-O-methyl-transferase gene and obsessive-compulsive disorder. *Am J Med Genet B Neuropsychiatr Genet*. 2003;123B(1): 64–69.

44. Ekholm SV, Reed SI. Regulation of G(1) cyclin-dependent kinases in the mammalian cell cycle. *Curr Opin Cell Biol*. 2000;12(6):676–684.

45. Hall M, Peters G. Genetic alterations of cyclins, cyclin-dependent kinases, and Cdk inhibitors in human cancer. *Adv Cancer Res*. 1996;68: 67–108.

46. Quintayo MA, Munro AF, Thomas J, et al. GSK3beta and cyclin D1 expression predicts outcome in early breast cancer patients. *Breast Cancer Res Treat*. 2012;136(1):161–168.

47. Knudsen KE, Diehl JA, Haiman CA, Knudsen ES. Cyclin D1: polymorphism, aberrant splicing and cancer risk. *Oncogene*. 2006;25(11): 1620–1628.

48. Pabalan N, Bapat B, Sung L, Jarjanazi H, Francisco-Pabalan O, Ozcelik H. Cyclin D1 Pro241Pro (CCND1-G870A) polymorphism is associated with increased cancer risk in human populations: a meta-analysis. *Cancer Epidemiol Biomarkers Prev*. 2008;17(10):2773–2781.

49. Munafo MR, Flint J. Meta-analysis of genetic association studies. *Trends Genet*. 2004;20(9):439–444.

50. Betticher DC, Thatcher N, Altermatt HJ, Hoban P, Ryder WD, Heighway J. Alternate splicing produces a novel cyclin D1 transcript. *Oncogene*. 1995;11(5):1005–1011.

51. Solomon DA, Wang Y, Fox SR, et al. Cyclin D1 splice variants. Differential effects on localization, RB phosphorylation, and cellular transformation. *J Biol Chem*. 2003;278(32):30339–30347.

52. Adams PD. Regulation of the retinoblastoma tumor suppressor protein by cyclin/cdks. *Biochim Biophys Acta*. 2001;1471(3):M123–M133.

53. Leveque C, Marsaud V, Renoir JM, Sola B. Alternative cyclin D1 forms a and b have different biological functions in the cell cycle of B lymphocytes. *Exp Cell Res*. 2007;313(12):2719–2729.