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Meta-analysis: E-cadherin immunoexpression as a potential prognosis biomarker related to gastric cancer metastasis in Asian patients

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Abstract. – OBJECTIVE: The prognostic potential of reduced E-cadherin expression is associated with an increased risk of gastric cancer. However, its role in gastric cancer remains poorly understood. This study was to quantitatively summarize available evidences for evaluating E-cadherin immunoexpression in Asian patients with gastric cancer as a prognostic indicator.

MATERIALS AND METHODS: Searches were applied to MEDLINE, EMBASE, the Cochrane Library and Chinese Biomedicine Databases until June 2012, without language restrictions. Studies were pooled and summary risk ratio (RR) or odds ratio (OR) were calculated. Potential sources of heterogeneity were sought out via subgroup and sensitivity analyses, and publication bias were also conducted.

RESULTS: Our combined results showed that reduction of E-cadherin expression in Asian patients with gastric cancer was frequently observed as compared to the counterpart normal tissue (odds ratio [OR] = 64.16, 95% confidence interval [CI] = 24.53-167.80, $p < 0.001$). All the analyses estimated favored a stronger link between the reduced E-cadherin expression and the poor 5 year overall survival (risk ratio [RR] = 1.50, 95% CI = 1.36-1.66, $p < 0.001$). When stratifying the studies by the clinical variables, the depth of invasion (OR = 2.46, 95% CI = 1.70-3.57, $p < 0.001$), lymph node spread (OR = 1.83, 95% CI = 1.49-2.26, $p < 0.001$), distant metastasis (OR = 2.04, 95% CI = 1.45-2.87, $p < 0.000$), and TNM stage (OR = 2.11, 95% CI = 1.58-2.83, $p < 0.001$) provided significant prognostic information.

CONCLUSIONS: Our findings indicate that E-cadherin appears to predict the overall survival and mark metastasis in Asian patients with gastric cancer. Importantly, E-cadherin may be implemented in the routine clinical management of gastric cancer. However, further pursuit of this possibility is warranted.

Key Words:
Gastric cancer, E-cadherin, Prognostic factor, Metastasis.

Abbreviations

RR = risk ratio; OR = odds ratio; OS = overall survival; VEGF = vascular endothelial growth factor.

Introduction

Metastases specifically occur in malignant tumors and are the most common cause of cancer-related deaths[1]. Gastric cancer is one of the most aggressive tumors and tends to be associated with lymph node metastasis, peritoneal dissemination, and hematogenous metastasis. Although recent advances in the field of oncology have resulted in the increased survival of patients with advanced disease, the prognosis of gastric cancer remains dismal. 5 year overall survival (OS) for most newly diagnosed patients with regional or distant metastatic disease is around 5 to 20%, with median overall survival being less than 1 year[2-4]. Therefore, identification and characterization of novel prognostic markers that will lead to a better understanding molecular nature of gastric cancer and help to predict recurrence or survival for these patients in early stage.

Recently, recapitulation of the developmental process of epithelial to mesenchymal transition (EMT) has been proposed as a crucial mechanism for enabling cancer cell invasion, dissemination and metastasis[5,6]. Loss of E-cadherin, one

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cell-adhersion protein, was considered as one of the best-characterized hallmarks of EMT[7]. It is responsible for calcium assisted cell-to-cell adhesion critical to the maintenance of normal tissue architecture and morphogenesis, which is known to act as one of the most important suppressors for regulating the neoplastic transformation and tumor metastasis[8,9]. Thus, E-cadherin loss could induce tumor cells to dedifferentiate and become highly metastatic, indicating that targeting Ecadherin may be of therapeutic benefit for gastric cancers.

Several meta analyses have evaluated the association between E-cadherin, immunoexpression and the prognostic potential in a few of advanced cancers, including esophageal cancer[10], ovarian cancer[11], head and neck squamous[12], non-small cell lung cancer[13], breast cancer[14]. Since then, recent studies have consecutively reported associations of *CDH1*-160C/A polymorphism with the susceptibility to gastric cancer, but with mixed or conflicting results[15-18]. Thus, the current evidences are insufficient to conclude whether E-cadherin, immunoexpression, can mark more advanced disease or whether it does serve as an independent negative predictive marker in gastric cancer. It is estimated that almost two-thirds of gastric cancer occur in Asia (China, Japan and Korea)[19]. In this investigation, we carried out a meta-analysis of data from published studies to more precisely and comprehensively estimate the prognostic influence of the reduced E-cadherin expression detected by Immunohistochemistry (IHC) on patient 5 year overall survival and to indentify various clinical factors that might confound 5 year overall survival in Asian patients with gastric cancer.

Materials and Methods

Literature Search

The meta-analysis was performed by means of a predefined written form. Databases of MEDLINE, PubMed, EMBASE and Chinese National Knowledge Infrastructure (CNKI) search for studies investigating the prognostic significance of E-cadherin in gastric cancer were performed (the upper date of June 2012), without language restrictions. Various combinations of the terms [E-cadherin and "prognosis" and ("gastric" or "stomach"), "cancer" or "carcinoma" or "tumor"] were used to screen for potentially relative studies.

Studies eligibility were included in the meta-analysis if they met the following criteria: (1) proven diagnosis of gastric cancer and normal gastric epithelial mucosa in humans; (2) E-cadherin evaluation by IHC methods; (3) data performed using cohorts from medical centers in Asian population, and (4) has a follow up time exceeding 5 years. References of retrieved articles were cross-searched to identify any studies missed by the computerized literature search. Authors of eligible studies were contacted for supplement of additional data relevant to meta-analysis.

Data Extraction and Methodological Assessment

As there are no generally accepted standards for measuring study quality, especially of observational studies[20], we did not weigh each study by a quality score. Data retrieved from all full publications included author, year of publication, country, antibody dilution, cut-off value for abnormal protein expression, number of readers, blinded reading, number of controls and of cases, number of association between E-cadherin expression and overall 5 year survival, and number of sex, tumor location, histo-differentiation, depth of invasion, lymph node status, distant metastasis, TNM stage, and vascular invasion of gastric cancer analyzed.

We tried carefully to avoid duplication of data, by examining each publication the names of all authors and the different medical centers involved. When an individual author published several articles obtained from the same or overlapping population, only the newest or most complete article was included in the analysis, otherwise, independent data were analyzed. Two investigators (Chen J and Li T) independently assessed study eligibility. Disagreements were resolved by consensus.

Statistical Methods

In present studies, we prospectively analyzed three categories of stratified models: the first stratified multivariate model was performed to confirm whether E-cadherin protein was abnormally expressed in gastric cancer in comparison with the normal gastric mucosa. The second outcome of meta-analysis was to measure the impact of the negative/reduced E-cadherin expression on survival by estimating the risk ratio (RR). And the third interest was to examine the prognostic value of E-cadherin expression that was corrected for the main prognostic factors including sex,

location and hiso-differentiation, depth of invasion, vascular invasion, lymph node status, distant metastasis and TNM stage.

According to clinical characteristics, T_1 and T_2 were combined, T_3 and T_4 were combined; also Stage I and Stage II were combined, Stage III and Stage IV were combined; well and moderate differentiation were combined, poor and undifferentiation were combined. When these statistical variables were described in text or tables, we obtained them directly from articles. When not given explicitly in an article, they were calculated from available numerical data in the articles using methods reported by Parmar et al[21].

All statistical analyses were conducted using STATA version 9.0, and a two-tailed *p* value of less than 0.05 was considered statistically significant. The risk ratio (RR) was used as a summary statistic for censored outcomes (5 year survival) and the odds ratio (OR) was used as the summary statistic for statistical analysis of dichotomous variables. Moreover, the in-depth analyses of study quality, the extent and sources of heterogeneity and the presence of publication bias between published studies were also tested. Heterogeneity across studies was quantified by I^2 statistical, which is generally considered significant for values ≥ 50%. In case of heterogeneity, meta-analysis was done by the random effect model after exploring the causes of heterogeneity. Otherwise, the fixed-effects model was applied. Additionally, Begg's test and the Egger's linear regression analyses were used to determine the presence of publication bias for each of the pooled study groups.

Results

Study Selection and Characteristics

After comprehensive searching, a total of 223 publications in English and 40 in Chinese were initially retrieved. 188 were excluded after title and abstract review. These included 21 reviews and 167 studies, which were either non-comparative or not relevant. The remaining 75 studies were full reviewed for detail evaluation. Of these, 8 studies performed different cohorts outside Asian (1 report originated from Iceland, 2 from Poland, 1 from Newzealand, 1 from Mexico, 1 from Italy and 2 from Greece); 8 studies overlapped with others; 13 studies assessed E-cadherin by other method rather than IHC, and 20 studies lacked extractable data. These 49 papers were excluded from the meta-analysis, leaving 26 eligible studies for the final analyses that met the criteria set forth in the search strategy and study selection as described (Figure 1A).

Among them, 18 studies were performed on the association of E-cadherin expression between gastric cancer and normal gastric mucosa[22-39]; 8

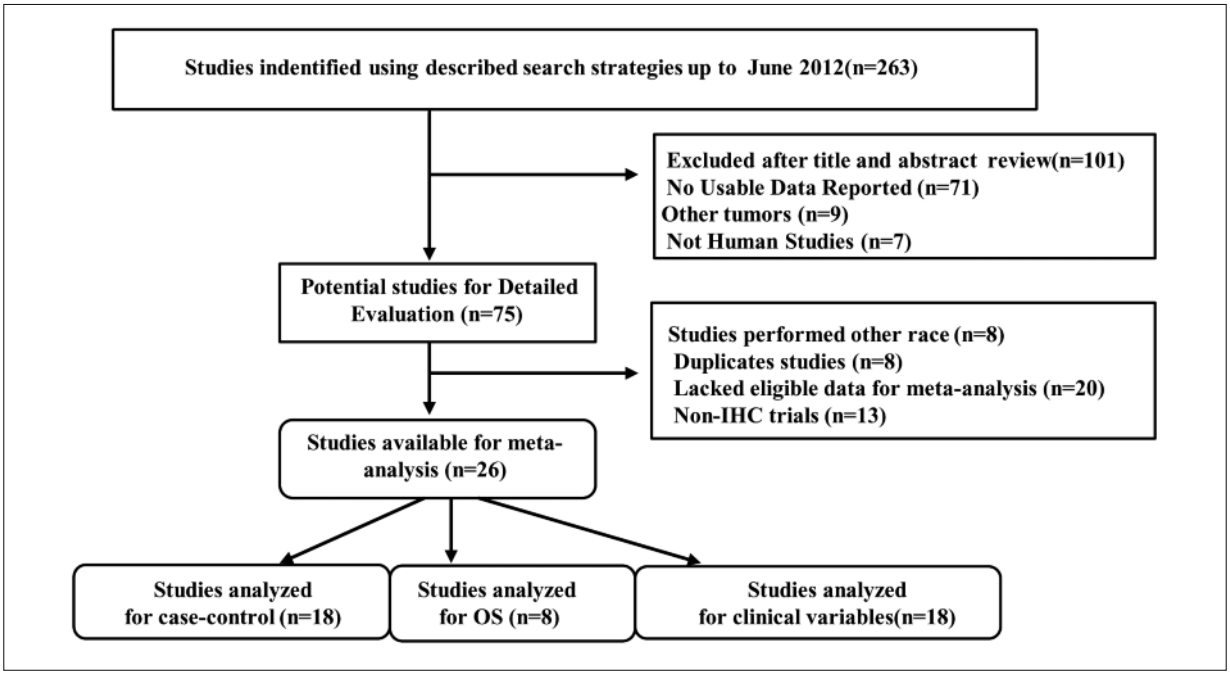

Figure 1. Methodologic flow chart of the meta-analysis.

studies predicted the impact of E-cadherin status on overall survival in patients with gastric cancer[24,27,40-45]; and 18 studies evaluated the prognostic value of E-cadherin expression and clinical factors[22-29,32-35,38-40,42,46,47]. For all the patients, measurements had been performed in the primary tumor, and all specimens had been taken before chemotherapy or radiotherapy. The main characteristics of eligible studies in our meta-analyses are reported in Table I.

Prognosis analyses were done in 26 studies encompassing 2,783 patients. Patient distribution rates according to reduced E-cadherin positive expression were as follows: 926 in gastric cancer (2081, 44.5%), 40 in normal gastric mucosa tissue (2096, 1.91%); 446 were mortal in 5 year overall prognosis (715, 62.38%), 315 were survived in prognosis (774, 40.70%); 436 were in poor differentiation (680, 64.1%), 337 in well and moderate differentiation (702, 48.0%); 517 were in positive serosa invasion (840, 61.55%), 358 in negative serosa invasion (790, 45.32%); 685 were in positive lymph node spread (1135, 60.35%), 318 in negative lymph node spread (666, 47.75%); 130 were in positive distant metastasis (204, 63.73%), 334 in negative distant metastasis (747, 44.71%); 225 were in positive venous invasion (358, 62.85%), 196 in negative venous invasion (372, 52.69%); 368 in stage III-IV (526, 69.96%), 223 in stage I-II (397, 56.17%); 211 were in female (373, 56.57%), 440 in male (794, 55.42%); 65 were in antrum (96, 67.7%), 165 in cardia and (or) corpus (249, 66.3%) (Table II).

Meta-analysis Results

Correlation of E-cadherin Expression Between Gastric Cancer and Normal Gastric Mucosa

The mean frequency of cases showing reduced or absent expression of E-cadherin was 44.5% (range, 12-69%) among 18 studies (2,081 patients and 2,096 controls). The combined OR was 64.16 (95% CI = 24.53-167.80; $p < 0.001$) (Figure 2, Table III), indicating that reduced or absent E-cadherin expression in the primary gastric cancer was an extremely significant indicator of unfavourable prognosis. However, a highly degree of heterogeneity was detected among the studies ($I^2 = 70.2\%$, $p < 0.001$) (Table III). When stratifying by ethnicity, the combined OR of China was 11.56 (95% CI = 6.42-20.82; $p < 0.001$) with low heterogeneity ($I^2 = 9.20\%$, $p = 0.357$) (Figure 2, Table III); the combined OR of Japan was 147.81 (95% CI = 50.74-430.53; $p < 0.001$) also without heterogeneity ($I^2 = 0.00\%$, $p = 0.799$) (Figure 2, Table III); Similar results were found in Korea (OR = 106.76; 95% CI = 30.33-375.78; $p < 0.001$) ($I^2 = 0.00\%$, $p = 0.628$) (Figure 2, Table III).

Correlation Bbetween E-cadherin Expression and Overall Survival in 5 Years

Meta-analysis on the prognostic value of E-cadherin expression showed that the overall survival rate at 5 years after the initial treatment was significantly lower in cases with reduced or absent expression of E-cadherin among 761 patients from 8 studies. The combined RR was 1.50 (95% CI = 1.36-1.66; $p < 0.001$), with mild between-study heterogeneity ($I^2 = 45.3\%$, $p = 0.077$) (Figure 3). When stratifying for race, results were similar among China, Japan, and Korea without significant heterogeneity (Figure 3, Table III). The potential for publication bias could be ruled out, however, the effect of bias was slight ($p = 0.033$) (Table III).

Correlation Between E-cadherin Expression and Clinical Variables

When stratifying for the clinicopathological variables by the depth of invasion of gastric cancer, statistically significance was observed. Patients with T_3 and T_4 gastric cancer had a much lower E-cadherin expression in 13 studies (1,630 patients) (OR = 2.46; 95% CI = 1.70-3.57; $p < 0.001$) than those with T_1 and T_2 gastric cancer, with moderate between-study heterogeneity ($I^2 = 56.0\%$, $p = 0.007$) (Table III). When stratifying for lymph node status of gastric cancer, statistically significant results also appeared that reduced E-cadherin expression was associated with lymph node metastasis in 16 studies (1,801 patients) (OR = 1.83; 95% CI = 1.49-2.26, $p < 0.001$), with no significant heterogeneity between studies ($I^2 = 0.0\%$, $p = 0.471$), but not among Korean in 4 studies (291 patients) (OR = 1.51; 95% CI = 0.88-2.59; $p = 0.283$) ($I^2 = 50.6\%$, $p = 0.108$) (Table III). When stratifying for the distant metastasis of gastric cancer, there was a statistical significance that E-cadherin expression was associated with distant metastasis in 8 studies (951 patients) (OR = 2.04; 95% CI = 1.45-2.87; $p < 0.001$), with no significant heterogeneity between studies ($I^2 = 0.0\%$, $p = 0.564$) (Table III). When further stratifying for TNM stage, E-cadherin expression of patients with stages III

Table I. Main characteristics of the 26 studies included in the final meta-analysis.

First author of issue (reference)	Year of publication	Language	Population	Number of patients (M/F)	Median age (years)	Treatment	Antibody dilution	Cutoff for reduced E-cadherin positivity (%)	Blinded reading	Reader(s) (n)	RR estimate	Survival analysis	Results
Cheng XX, et al	2005	English	China	–	–	–	–	–	–	–	Miss	–	–
Yuan WJ, et al	2009	English	China	107/58	56.4	Surgery	1:200	–	–	–	Miss	–	–
Zhou YN, et al	2002	English	China	123/40	54.5	Surgery	–	–	Yes	2	Reported in text	OS	Negative
Chu YQ, et la	2008	English	China	91/27	56.3	Surgery	–	> 90%	–	–	Reported in text	OS	Positive
Chen HC, et al	2003	English	China	73/11	–	Surgery	1:100	> 90%	Yes	–	Reported in text	OS	Negative
Zhong XY, et al	2008	English	China	86/32	59	Surgery	1:100	> 60%	–	–	Reported in text	OS	Positive
Zhu BH, et al	2006	Chinese	China	61/19	56	Surgery	1:200	> 90%	–	–	Reported in text	OS	Positive
Gu HP, et al	2004	Chinese	China	–	–	Surgery	–	> 70%	–	–	Reported in text	OS	Positive
Liu WT, et al	2007	Chinese	China	23/18	54	Endoscopic biopsy	1:50	–	–	–	Miss	–	–
Tetsuro Ohno, et al	2006	English	Japan	142/60	63	–	1:500	–	Yes	1	Reported in text	OS	Positive
Yutaka Shimada, et al	2004	English	Japan	183/93	63.1	Surgery	–	> 50%	Yes	2	Reported in text	OS	Positive
Andrei Cugolkov, et al	2003	English	Japan	38/18	59	Surgery	1:100	> 80%	Yes	2	Miss	–	–
Che XM, et al	1998	English	Japan	54/15	62	Surgery	–	> 90%	–	–	Miss	–	–
JiRO WATARI, et al	2004	English	Japan	–	–	–	–	> 80%	Yes	2	Miss	–	–
Yoshihide Shino, et al	1995	English	Japan	77/44	62	Surgery	–	> 90%	–	–	Reported in text	OS	Positive
Yutaka Yonemura, et al	1995	English	Japan	–	63.4	Surgery	1:200	> 60%	–	–	Reported in text	OS	Positive
Yutaka Yonemura, et al	2000	English	Japan	–	–	Surgery	1:100	> 60%	–	–	Reported in text	OS	Positive
S Lim, et al	2003	English	Japan	254/114	30	Surgery	–	> 90%	–	–	Miss	–	–
Min A Kim, et al	2009	English	Korea	396/202	54.8	Surgery	1:200	> 90%	–	–	Reported in text	OS	Positive
MEE JOO, et al	2003	English	Korea	59/40	57.6	Surgery	1:200	> 80%	–	–	Miss	–	–
Yu Jung Kim, et al	2008	English	Korea	75/34	52	Surgery	–	–	–	–	Reported in text	RFS	Negative
Hye Seung Lee, et al	2003	English	Korea	–	54.8	Surgery	1:200	–	–	–	Reported in text	OS	Positive
Young–Eun Joo, et al	2000	English	Korea	38/27	55.2	Surgery	1:50	> 90%	Yes	2	Reported in text	OS	Positive
In Mok Jung, et al	2007	English	Korea	76/35	57	Surgery	1:100	> 90%	–	–	Miss	–	–
Sang–UK Han, et al	2005	English	Korea	37/13	53.2	Endoscopic biopsy	–	–	–	–	Reported in text	OS	Positive
Masakazu Yashiro, et al	2006	English	Japan	78/34	62	Surgery	–	–	–	–	Reported in text	OS	Negative

RR: risk ratio; OS: overall survival; RFS: relapse-free survival; Positive: inverse relationship between reduced E-cadherin expression and survival; Negative: no relationship. 'Reader' are readers of the histologic slides, 'blinded reading' means that readers of the slides without knowledge of the clinical outcome, and '–' corresponds to missing data

Table II. Main characteristics of E-cadherin expression on prognostic factors.

Stratification of gastric cancer	Type	Total (n)	Distribution of E-cadherin			
			Reduced		Preserved	
			n	(%)	n	(%)
Case-control	Carcinomas	2081	926	44.50	1155	55.50
	Non-neoplastic mucosa	2096	40	1.91	2056	98.01
Overall 5 year survival	Mortal	715	446	62.38	269	37.62
	Survival	774	315	40.70	459	59.30
The depth of invasion	Positive	840	517	61.55	323	38.45
	Negative	790	358	45.32	432	54.68
Lymph node status	Positive	1135	685	60.35	450	39.65
	Negative	666	318	47.75	348	52.25
Distant metastasis	Positive	204	130	63.73	74	36.27
	Negative	747	334	44.71	413	55.29
TNM stage	III-IV	526	368	69.96	158	30.04
	I-II	397	223	56.17	174	43.83
Histological differentiation	Pooly	680	436	64.12	242	35.59
	Well/moderate	702	337	48.01	365	51.99
Vascular invasion	Positive	358	225	62.85	133	37.15
	Negative	372	196	52.69	176	47.31
Sex	Male	794	440	55.42	354	44.58
	Female	373	211	56.57	162	43.43
Location	Antrum	96	65	67.70	31	32.29
	Cardia and (or) Corpus	249	165	66.30	84	33.73

Case: gastric cancer; Control: normal gastric mucosa; T: the depth of invasion; N: lymph node status; M: distant metastasis; Positive: patients have E-cadherin reduced expression; Negative: patients have no E-cadherin reduced expression; n: number of patients.

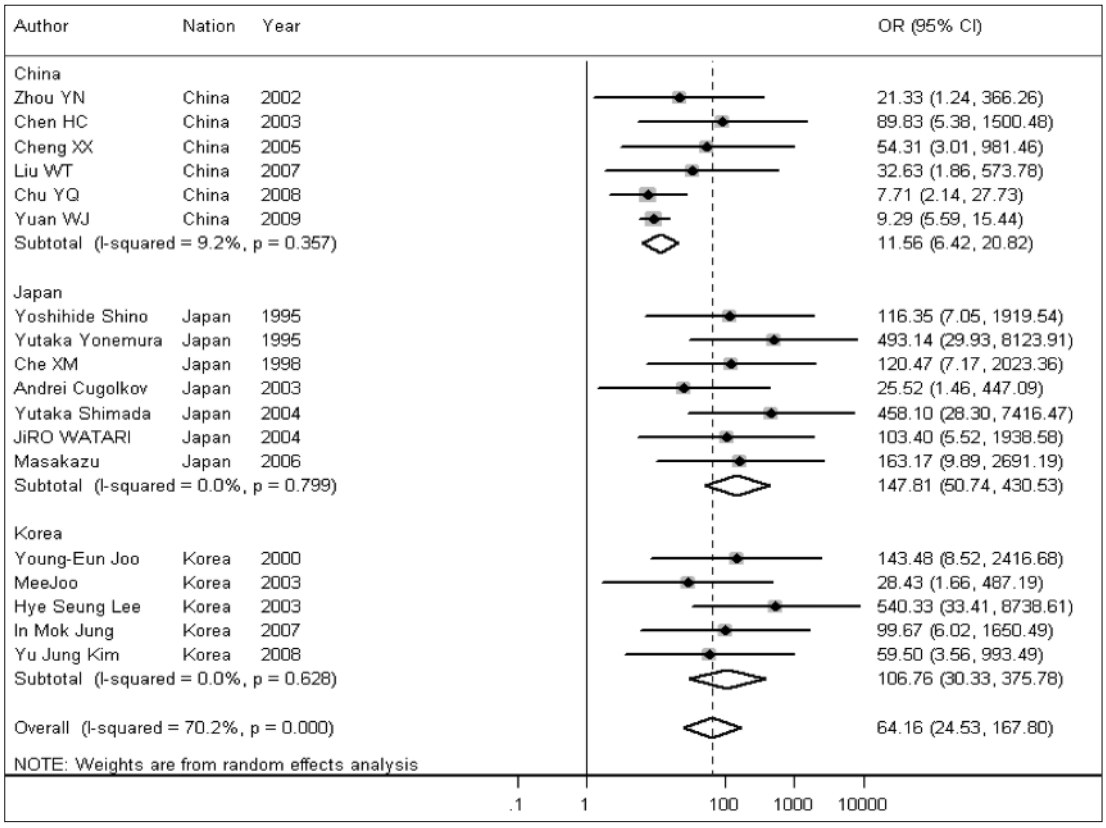

Figure 2. Meta-analysis on the relation between E-cadherin expression in gastric cancer and normal gastric mucosa.

Table III. Meta-analysis of E-cadherin expression and gastric cancer.

Stratification of gastric cancer	Nation	Number of studies	Total patients	Model	OR (RR) (95% CI)	*p*-value	I^2 for heterogeneity	*p*-value for bias
Gastric cancer-normal gastric mucosa	China	6	616	Fixed	11.56 (6.42, 20.82)	< .001	9.20%	0.000
	Japan	7	782	Fixed	147.81 (50.74, 430.53)	< .001	0.00%	
	Korea	5	683	Fixed	106.76 (30.33, 375.78)	< .001	0.00%	
	All	18	2.081	Random	64.16 (24.53, 167.80)	< .001	70.2	
Overall 5 year survival	China	4	188	Fixed	1.83 (1.41, 2.36)	< .001	14.20%	
	Japan	3	333	Fixed	1.61 (1.38, 1.89)	< .001	29.00%	
	Korea	1	240	–	1.30 (1.12, 1.51)	< .001	–	
	Asia	8	761	Fixed	1.50 (1.36, 1.66)	< .001	45.3	0.033
The depth of invasion	China	6	679	Random	2.85 (1.46, 5.60)	< .001	68.00%	0.114
	Japan	5	836	Fixed	2.01 (1.22, 3.30)	0.006	49.70%	
	Korea	2	115	Fixed	3.12 (1.09, 8.90)	0.033	38.00%	
	Asia	13	1.630	Random	2.46 (1.70, 3.57)	< .001	56.0	
Lymph node status	China	6	607	Fixed	1.96 (1.38, 2.79)	0.002	14.1%	0.304
	Japan	6	903	Fixed	1.85 (1.37, 2.50)	< .001	0.00%	
	Korea	4	291	Fixed	1.51 (0.88, 2.59)	0.283	50.60%	
	Asia	16	1.801	Fixed	1.83 (1.49, 2.26)	< .001	0.0%	
Distant metastasis	China	2	202	Fixed	2.86 (1.25, 6.53)	0.013	0.0%	0.338
	Japan	4	634	Fixed	1.74 (1.16, 2.60)	0.024	6.70%	
	Korea	2	115	Fixed	3.28 (1.08, 9.95)	0.039	0.00%	
	Asia	8	951	Fixed	2.04 (1.45, 2.87)	< .001	0.0%	
TNM stage	China	3	319	Fixed	2.78 (1.69, 4.57)	< .001	0.00%	0.407
	Japan	1	202	–	2.98 (1.56, 5.67)	< .001	–	
	Korea	5	402	Fixed	1.47 (0.94, 2.28)	0.088	37.70%	
	Asia	9	923	Fixed	2.11 (1.58, 2.83)	< .001	36.9	
Histological differentiation	China	5	605	Fixed	2.11 (1.44, 3.09)	< .001	0.00%	0.055
	Japan	4	712	Fixed	1.58 (1.16, 2.16)	0.004	0.00%	
	Korea	1	65	–	8.71 (2.77, 27.37)	< .001	–	
	Asia	10	1382	Fixed	1.98 (1.48, 2.65)	< .001	29.6	
Vascular invasion	China	2	196	Fixed	1.62 (0.88, 2.97)	0.122	0.0%	0.676
	Japan	3	435	Fixed	1.12 (0.56, 2.22)	0.757	63.5%	
	Korea	1	99	–	1.45 (0.49, 4.26)	0.499	–	
	Asia	6	730	Fixed	1.31 (0.90, 1.90)	0.155	21.8%	
Location	China	–		–	–	–	–	0.543
	Japan	2	271	Fixed	0.95 (0.52, 1.71)	0.856	38.60%	
	Korea	1	74	–	1.31 (0.47, 3.66)	0.602		
	Asia	3	345	random	1.03 (0.62, 1.72)	0.914	0	
Sex	China	3	367	Random	0.84 (0.51, 1.38)	0.494	0.00%	0.604
	Japan	3	547	Fixed	1.02 (0.70, 1.49)	0.900	47.20%	
	Korea	3	253	Fixed	1.04 (0.61, 1.78)	0.879	7.50%	
	Asia	9	1167	Random	0.97 (0.75, 1.27)	0.846	0	

OR: odd ratio; RR: risk ratio; CI: confidence interval. '–' corresponds to missing data and do not be analyzed in meta-analysis.

and IV gastric cancer was much lower than those with stage I and II gastric cancer in 9 studies (923 patients) (OR = 2.11; 95% CI = 1.58-2.83; $p < 0.001$), with low between-study heterogeneity ($I^2 = 36.9\%$, $p = 0.123$) (Table III).

We also observed trends toward a correlation of decreased E-cadherin expression with poor histological differentiation on the grounds that the pooled ORs (95% CI; *p* value) were 1.98 (1.48-2.65; $p < 0.001$), but not for vascular invasion 1.31 (0.90-1.90; $p = 0.155$), location 1.03 (0.62-1.72; $p = 0.914$) and sex 0.97 (0.75-1.27; $p = 0.846$) among the whole Asians (Table III). Most notably, no publication bias for above subgroup analyses was detected by the Begg's and Egger's test (Table III).

Discussion

In Asia, especially China, gastric cancer constitutes the peak lethal malignancy[19]. Moreover, most mortality of cancer patients is largely caused by metastases rather than their primary

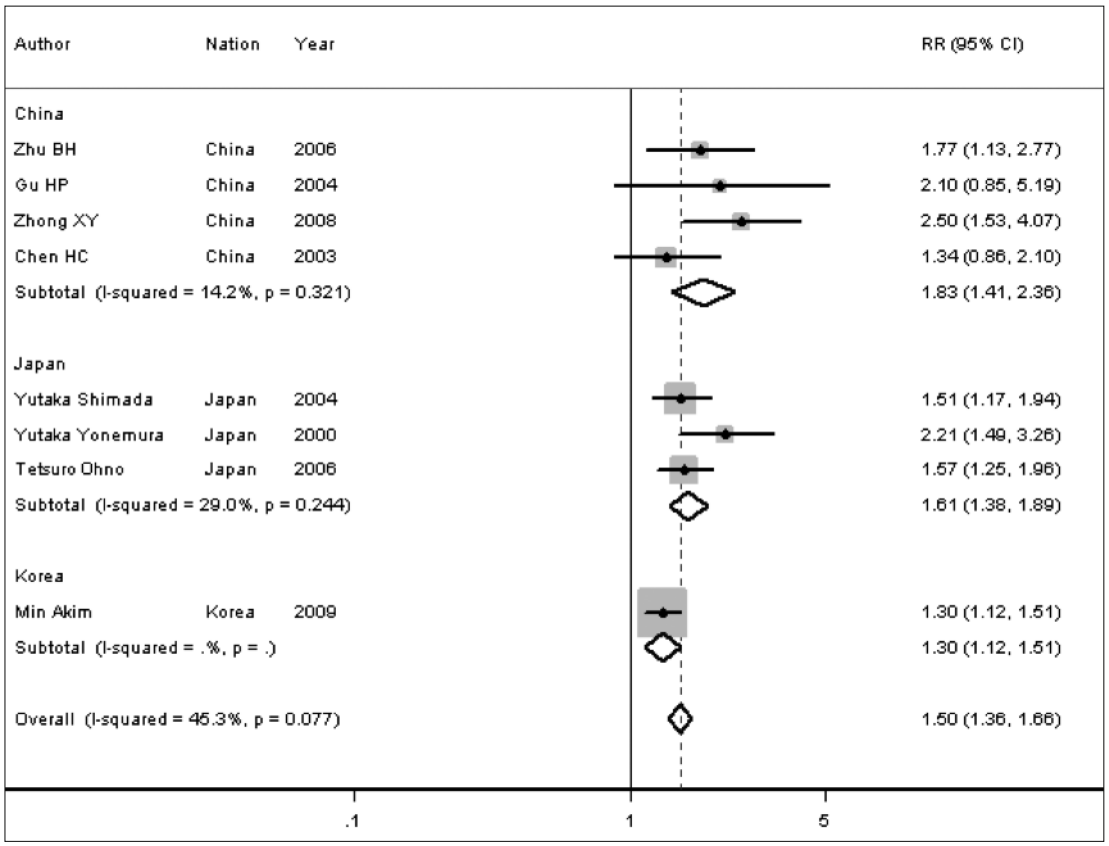

Figure 3. Meta-analysis on the relation between E-cadherin expression and 5-year overall survival (OS).

tumors at the time of diagnosis. The present analysis is the first general overview of associations between the well characterized metastatic protein, E-cadherin, and the prognosis value for gastric cancer patients based on 26 up-to-date case-control studies from cohorts of medical centers in Asia. Our analyses, combining 18 independent studies that included 2,081 patients, revealed that E-cadherin loss in patients with gastric cancer was frequently observed as compared to the counterpart normal tissue. However, heterogeneity of between-study has been shown to be significant (I^2 = 70.2%). When stratified by ethnicity, studies included in subgroup analyses displayed better homogeneity with an increased risk among China (I^2 = 9.20%), Japan (I^2 = 0.00%), and Korea (I^2 = 0.00%). Our findings, thereby, that differences in ethnic background of the study populations among the studies might be responsible for the high heterogeneity. Similar results were reported on the association between the E-cadherin expression and prognosis risk in head and neck squamous cell carcinoma and NSCLC by Zhao et al[12] and Wu et al[13]. However, assumption that needs to be further investigated in future well-designed high quality studies among different ethnicities.

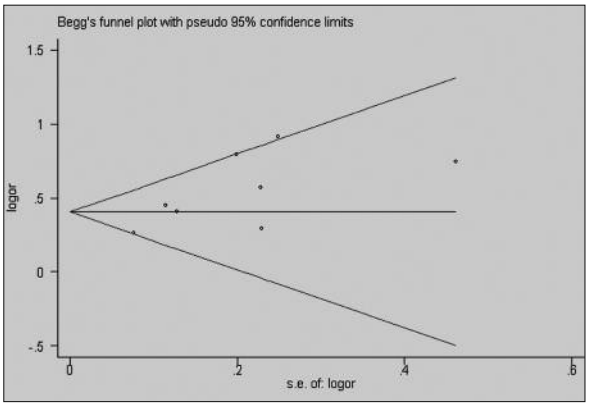

Figure 4. Begg's Funnel Plot analysis to detect publication bias for 5 year overall survival (OS).

Although recent several meta-analysis studies have reported the role of *CHD1*-160C > A polymorphism in gastric cancer risk[15-18], the results are inconclusive, partially due to small samples,

the possible small effect of the polymorphism on gastric cancer and the quality control of genotyping documented in some published studies. Additionally, those findings had no direct evidences on the relation of the status of E-cadherin protein expression with the overall survival rate at 5 years in gastric cancer. In the present study, we evaluated studies not only analyzing the direct correlation of E-cadherin protein expression status with survival, but also stratifying with clinical profiles that might confound that in gastric carcinomas. The pooled statistical data showed that reduced E-cadherin expression does decrease the overall 5 year survival of gastric cancer patients among Asians, with a meta-risk for OS (RR = 1.50), which was similar with the other three meta-analyses thus far published on head and neck squamous (HR = 1.96)[12], non-small cell lung cancer (HR = 1.49)[13], breast cancer (HR = 1.55)[14]. The reduced survival was strikingly correlated with dedifferentiation (OR = 1.98), advanced tumor invasion (OR = 2.46), lymph node spread (OR = 1.83), distant metastasis (OR = 2.04) and higher TNM stage (OR = 2.11). As a rule of the thumb, a prognostic factor with RR (OR) > 2 is considered as useful practical value[48]. Therefore, our current results confirmed that E-cadherin represents one of the most important prognosis biomarkers related to tumor metastases. The findings supported the that aggressive E-cadherin-lost gastric tumors have, like more typical gastric cancer that is prone to metastasis, EMT features, which contributed to the functional analyses and drug-targeted therapy in the prevention and treatment of gastric cancer. Furthermore, the combined results also found that reduced E-cadherin expression has no association with vascular invasion. Since carcinogenesis is a multiple-step process, any single molecule can not independently predict the survival of the patients completely. Based on our previous observations, the vascular endothelial growth factor (VEGF) appears to be a significant prognostic factor for hematogenous metastasis of gastric cancer (RR = 2.45, 95% CI = 2.11-2.83, p = 0.000)[49]. Therefore, combinations of both E-cadherin and VEGF, two prognostic biomarkers, should have increased prognostic power over individual markers themselves and be comprehensive and necessary.

We have also noticed the publicated meta analyses of E-cadherin mainly involved in gene polymorphism of *CHD1*-160C > A. However, it was the first time to investigate the relationship between E-cadherin and overall survival on gastric cancer at protein level. IHC is a cost-effective and well-documented method for the characterization of gene expression at protein level. In solid tumor pathology, almost all established diagnostic and prognostic markers are currently assessed by using this method. In this study, we found that cut-offs points for decreased E-cadherin expression in tissues (80-90%) positively stained cells were selected in most studies in terms of antibody dilution ranging from 1:100 to 1:200. Our results, therefore, make a strong case for international consensus on staining and scoring protocols. However, this meta-analysis was done at a study level, the results should be interpreted with caution.

We tried to conduct a thorough review of the existing publications, however, our meta analysis has the following limitations. First, the studies included in the analysis were mostly done at various clinical cancer centres and major institutions and, therefore, the patients included might not reflect the corresponding natural patient populations. Second, only published studies were included in the present meta analysis and it is unavoidable that some data could still be missing. Missing information may reflect ''negative'' or more conservative association of E-cadherin with overall survival, which could reduce the significance of E-cadherin expression as a predictor of mortality. However, we laid more emphasis on assessing biases across studies and pinpointing the potential sources of heterogeneity *via* subgroup and sensitivity analyses. We comprehensively assessed the publication biases by performing funnel plots qualitatively, and estimating by Begg's and Egger's tests quantitatively. In view of this, we convince that the results of our meta-analysis, in essence, are sound and reliable.

Conclusions

Our meta analysis of the association between loss of E-cadherin expression and overall survival in Asian patients with gastric cancer suggests that reduced levels of E-cadherin protein, compared to normal gastric tissue, appears to predict the 5 year overall survival risk and mark tumor metastasis. Importantly, E-cadherin may be converted from candidate to the routine clinical practice for clinicians to predict the outcome of single patient with gastric carcinoma. Therefore, further pursuit of this possibility is warranted.

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Conflict of Interest

The Authors declare that there are no conflicts of interest.

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