Downregulated E-Cadherin Expression Indicates Worse Prognosis in Asian Patients with Colorectal Cancer: Evidence from Meta-Analysis

Xin He1*, Zhigang Chen2*, Minyue Jia3, Xiaoying Zhao1*

1 Department of Hematology, the Second Affiliated Hospital, Zhejiang University School of Medicine, Hangzhou, China, 2 Department of Oncology, the Second Affiliated Hospital, Zhejiang University School of Medicine, Hangzhou, China, 3 Department of Endocrinology, Second Affiliated Hospital, Zhejiang University School of Medicine, Hangzhou, China

Abstract

Background: Epithelial-mesenchymal transition (EMT) plays a crucial role in the progression and aggressiveness of colorectal carcinoma. E-cadherin is the best-characterized molecular marker of EMT, but its prognostic significance for patients with CRC remains inconclusive.

Methodology: Eligible studies were searched from the PubMed, Embase and Web of Science databases. Correlation between E-cadherin expression and clinicopathological features and prognosis was analyzed. Subgroup analysis was also performed according to study location, number of patients, quality score of studies and cut-off value.

Principal Findings: A total of 27 studies comprising 4244 cases met the inclusion criteria. Meta-analysis suggested that downregulated E-cadherin expression had an unfavorable impact on overall survival (OS) of CRC (n = 2730 in 14 studies; HR = 2.27, 95%CI: 1.63–3.17; Z = 4.83; P = 0.000). Subgroup analysis indicated that low E-cadherin expression was significantly associated with worse OS in Asian patients (n = 1054 in 9 studies; HR = 2.86, 95%CI: 2.13–3.7, Z = 7.11; P = 0.000) but not in European patients (n = 1552 in 4 studies; HR = 1.14, 95%CI: 0.95–1.35, Z = 1.39; P = 0.165). In addition, reduced E-cadherin expression indicated an unfavorable OS only when the cut off value of low E-cadherin expression was >50% (n = 512 in 4 studies; HR = 2.08, 95%CI 1.45–2.94, Z = 4.05; P = 0.000). Downregulated E-cadherin expression was greatly related with differentiation grade, Dukes' stages, lymphnode status and metastasis. The pooled OR was 0.36(95%CI: 0.19–0.7, Z = 3.03, P = 0.002), 0.34(95%CI: 0.21–0.55, Z = 6.61, P = 0.000), 0.49(95%CI: 0.32–0.74, Z = 3.02, P = 0.002) and 0.45(95%CI: 0.22–0.91, Z = 3.43, P = 0.001), respectively.

Conclusions: This study showed that low or absent E-cadherin expression detected by immunohistochemistry served as a valuable prognostic factor of CRC. However, downregulated E-cadherin expression seemed to be associated with worse prognosis in Asian CRC patients but not in European CRC patients. Additionally, this meta-analysis suggested that the negative threshold of E-cadherin should be >50% when we detected its expression in the immunohistochemistry stain.

Introduction

Colorectal cancer (CRC) is the third most common cause of cancer-related deaths worldwide, and its 5-year survival rate ranges from 90% for stage I patients to 10% for metastatic cases [1]. Thus, distant metastases formation is the decisive and the most lethal event during the disease course. In fact, about 25% of CRC patients present with Liver metastases at the time of diagnosis. Although Liver metastasis may be successfully treated by surgical resection, more than two thirds experience relapse [2]. Furthermore, 30–40% of cases will unfortunately develop metastases within 2 years after the resection of the primary tumour. Therefore, it is important to uncover the biological mechanisms underlying metastases of CRC and formulate strategies to intervene in this process.

Mounting evidence suggests that epithelial-mesenchymal transition (EMT) plays a crucial role in the progression and aggressiveness of colorectal carcinoma [3–6]. EMT was first recognized as an essential component of embryonic development, tissue remodeling, and wound repair [7]. Later, EMT was reported to participate in the progression and metastases of many epithelial tumors [8]. During the process of EMT, epithelial cells actively downregulate cell–cell adhesion systems, lose polarity, and acquire a mesenchymal phenotype. This phenotype enables tumor cells to infiltrate surrounding tissues, and thus license these cells to metastasize in distant sites. Several markers have been recognized...
as indicators of EMT, such as E-cadherin, vimentin, N-cadherin, and Snail [8,9]. E-cadherin is the best-characterized molecular marker of EMT and loss of E-cadherin expression is an EMT hallmark [9,10]. Therefore, E-cadherin is expected to be a useful biomarker associated with invasiveness, poor differentiation and malignant phenotype in CRC. However, the correlation between the expression of E-cadherin detected by immunohistochemistry and patient survival remains controversial, and the number of cases enrolled in numerous studies published was not large enough. Therefore, it is necessary to analyze the data of E-cadherin systematically in CRC to draw a reasonable conclusion about its prognostic significance.

In this study, we performed a meta-analysis to investigate E-cadherin expression and the prognosis of patients with CRC to determine whether low E-cadherin expression is associated with poor outcome and clinicopathologic characteristics of CRC.

Methodology

Literature search

We carried out a search of the PubMed, Embase and Web of Science databases using the terms: “E-cadherin”, “CDH1”, “colorectal neoplasms”, “colorectal Cancer”, “colon cancer” “rectal cancer”, “prognosis” with all possible combinations. The references of all the studies were manually searched for additional eligible studies. Review articles and bibliographies of other pertinent article were also inspected to find related articles.

Inclusion and exclusion criteria

The inclusion criteria in the meta-analysis were as follows: (1) to evaluate E-cadherin expression by immunohistochemistry in the human CRC tissues; (2) to assess the relationships between E-cadherin expression and CRC pathological features or prognosis; (3) to be published in English language; (4) to provided sufficient information to estimate hazard ratio (HR) or odds ratio (OR) and their 95% confidence intervals (CIs).

The articles were not in the scope of our analysis if they met the following criteria: (1) letters, reviews, conference abstracts, case reports; (2) articles which don’t offer enough data to calculate the HR about overall survival (OS); (3) articles published in non-English; (4) overlapping articles.

Data extraction and assessment of study quality

Two investigators (HX and JMY) reviewed each eligible study and extracted following data: the first author’s name, year of publication, country of origin, number of patients, gender of patients, tumor site, disease stage, antibody source, cut-off value, condition of adjuvant therapy and survival data. Controversial problems were arbitrated by the third investigator (ZXY). Newcastle–Ottawa quality assessment scale was used to assess the quality of each study [11].

Statistical analysis

Odds ratios (ORs) and their 95% CIs were combined to evaluate the association between E-cadherin expression and clinicopathological factors, such as differentiation grade, Dukes' stages, depth of invasion, lymphnode status and metastasis. For the pooled analysis of E-cadherin expression on survival outcome, HRs and its 95% CI were the recommended summary statistics for meta-analysis of OS. If these statistical variables were described in a literature, we pooled it directly; otherwise, they were calculated from available numerical data in the articles according to the methods described by Parmar [12]. An observed OR<1 implies unfavorable parameters for the group with decreased E-cadherin expression. An observed HR>1 implies worse survival for the group with decreased E-cadherin expression. The impact of decreased E-cadherin expression on survival or clinicopathological factors was considered to be statistically significant if the 95%CI did not overlap with 1. Heterogeneity across studies was assessed by Chi- square based Q statistical test [13]. And the I² statistic to quantify the proportion of the total variation, which is due to inter-study heterogeneity rather than sampling error and is measured from 0% to 100% [14]. A P>0.10 for the Q-test indicated a lack of heterogeneity among the studies, then the pooled ORs and HRs estimate of each study were calculated by the fixed-effects model (the Mantel-Haenszel method) [15]. Otherwise, the random-effects model (the DerSimonian and Laird method) was used [16]. Egger’s test was used to examine the possibility of publication bias. Publication bias was indicated when p value of Egger’s test <0.05. The statistical analyses were performed using STATA version 12.0 software (Stata Corporation, Collage Station, Texas, USA). All the P values were for a two-side test and considered statistically significant when p<0.05.

Results

Description of studies

A total of 549 studies were identified from a search of the above databases using the search strategy as described above (Figure 1). After scrutinizing the abstracts and full-text of these studies, a total of 27 eligible studies were ultimately chosen in this meta-analysis [17–43]. The clinical features of these 27 included studies were summarized in Table 1. These studies were published from 1996 to 2012, and total 4244 CRC patients were enrolled and investigated the relationship between E-cadherin expression and pathological features or OS. Sample sizes ranged from 37 to 1164 patients. 11 studies enrolled less than 100 patients and 4 studies included more than 200 patients. Of these 27 studies, 7 studies were conducted in Japan, 4 each in China and Greece, 2 in Turkey, 1 each in Hungary, Korea, Italy, Roumania, European, Argentina, Russia, Norway, Sweden and England. 17 studies selected the percentage of negative staining as the cut-off point, including 10 studies more than 50%, 4 studies less than 50% and 3 studies 50%.

Methodological quality of the studies

The qualities of 27 eligible studies included in our meta-analysis were assessed according to the Newcastle–Ottawa scale.
NOS assessed eight items of methodology, which were categorized into the three dimensions of selection, comparability, and outcome. For quality, scores ranged from 0 (lowest) to 9 (highest), and studies with scores of 6 or more were rated as high quality. 16 included studies obtained scores of 6 or more in methodological assessment, indicating that they were of high quality (Table 1).

**Impact of E-cadherin expression on overall survival of colorectal cancer**

The meta-analysis was performed on 14 studies assessing the association of E-cadherin expression with OS. The pooled HR was 2.27, 95%CI: 1.63–3.17; Z = 4.83; P = 0.000 (Figure 2) with heterogeneity (I² 67.3% P = 0.000). It suggested that loss of E-cadherin was significantly with the worse prognosis of CRC and low or absent E-cadherin expression was a valuable prognostic factor in CRC. Moreover, we also performed subgroup analysis by study location, number of patients, quality score and cut-off value. The results showed that the significant relation between low E-cadherin expression and OS was exhibited especially in Asian countries (HR = 2.86 95%CI 2.13–3.7, Z = 7.11; P = 0.000). Additionally, reduced E-cadherin expression indicated an unfavorable OS only when the cut off value of low E-cadherin expression.

### Table 1. Characteristics of studies included for the meta-analysis.

| First author | Year | Country | Patient(M/F) | Antibody source | Definition of E-cadherin negative | HR of OS(95%CI) | Relationship with survival | Quality score |
|--------------|------|---------|--------------|----------------|----------------------------------|-----------------|--------------------------|---------------|
| Lu           | 2012 | China   | 136(84/52)   | BD             | Multiplying the intensitys core by expressions core ≤3 | NA              | NA                       | 4             |
| Andras       | 2012 | Hungary | 100(52/48)   | Transduction   | 25%                              | 1.1(0.55–2.198) | No                        | 3             |
| Chen         | 2012 | China   | 60(NA)       | Santa Cruz     | 50%                              | NA              | NA                       | 6             |
| Lampropoulos | 2012 | Greece  | 195(103/92)  | Santa Cruz     | 75%                              | NA              | NA                       | 6             |
| Ozguven      | 2011 | Turkey  | 60(38/22)    | Neomarkers     | 50%                              | NA              | NA                       | 6             |
| Kang         | 2011 | Korea   | 301(168/133) | Transduction   | Multiplying the intensities core by expressions core ≤3 | 10.753(2.95–40) | Yes                       | 4             |
| Fang         | 2010 | China   | 142(80/62)   | Dako           | 30%                              | NA              | NA                       | 6             |
| Karamitopoulos | 2010 | Greece  | 82(39/43)    | Dako           | 60%                              | 1.389(0.714–2.5) | No                        | 3             |
| Aresu        | 2010 | Italy   | 44(22/22)    | Transduction   | 50%                              | NA              | NA                       | 6             |
| Filiz        | 2009 | Turkey  | 138(83/55)   | Labvision      | Lightly/moderately staining or negative staining | 2.024(1.21–3.378) | Yes                     | 6             |
| Pap          | 2009 | Roumania | 149(87/62)   | Labvision      | Summing the intensities core and expressions core = 0 | NA              | NA                       | 6             |
| Chen         | 2008 | China   | 60(36/24)    | Beijing Zhongshan Golden Bridge | Summing the intensities core and expressions core ≤3 | 3.311(0.993–11.111) | No                      | 6             |
| Zlobec       | 2007 | Europe  | 1164(NA)     | Dako           | 5%                               | 1.09(0.89–1.319) | No                        | 2             |
| Nqan         | 2007 | Japan   | 140(79/61)   | Vector         | 75%                              | 2.849(1.362–5.952) | Yes                     | 4             |
| Shiono       | 2006 | Japan   | 86(NA)       | NA             | NA                               | 4.237(1.289–13.889) | Yes                     | 2             |
| Roca         | 2006 | Argentina | 84(47/37)   | BD             | 90%                              | 2.38(1.316–4.348) | Yes                     | 7             |
| Bravou       | 2005 | Greece  | 125(NA)      | BD             | 70%                              | NA              | NA                       | 7             |
| Delektorskaya | 2005 | Russia  | 129(NA)      | Novocastra     | 75%                              | NA              | NA                       | 3             |
| Bondi        | 2006 | Norway  | 206(197/10)  | Zymed          | 70%                              | 2.1(0.75–5.917) | No                       | 5             |
| Fernebro     | 2004 | Sweden  | 269(173/96)  | Dako           | Weak or absent staining          | NA              | NA                       | 5             |
| Garinis      | 2003 | Greece  | 37(20/17)    | Santa Cruz     | 70%                              | NA              | NA                       | 6             |
| Aoki         | 2003 | Japan   | 82(44/38)    | Transduction I | 20%                              | 3.597(1.075–12.048) | Yes                    | 8             |
| Ikekuchi     | 2000 | Japan   | 105(58/47)   | Takara         | 20%                              | 3.448(1.135–10.526) | Yes                    | 8             |
| Nanashima    | 1999 | Japan   | 44(29/15)    | Takara         | Absent staining                  | 4(1.292–12.346) | Yes                     | 7             |
| Ilyas        | 1997 | England | 68(NA)       | Dako           | 75%                              | NA              | NA                       | 5             |
| Mohri        | 1996 | Japan   | 100(59/41)   | Sigma          | 90%                              | NA              | NA                       | 7             |

NA, not available; HR, hazard ratio; OS, overall survival; 95%CI, 95% confidence interval.
doi:10.1371/journal.pone.0070858.t001
Figure 2. Forrest plot of Hazard ratio (HR) for the association of E-cadherin expression with overall survival (OS). HR > 1 implied worse survival for the group with negative/decreased E-cadherin expression and loss of E-cadherin was significantly with the worse prognosis of CRC patients.
doi:10.1371/journal.pone.0070858.g002

Table 2. Stratified analysis of pooled hazard ratios of colorectal cancer patients with reduced E-cadherin expression.

| Stratified analysis       | Number of studies | Number of patients | Pooled HR(95% CI) | P value | I²(%) | P value |
|---------------------------|-------------------|--------------------|-------------------|---------|-------|---------|
| Study location            |                   |                    |                   |         |       |         |
| Asia                      | 9                 | 1054               | 2.86(2.13–3.7)    | 0       | 0     | 0.507   |
| Europe                    | 4                 | 1552               | 1.14(0.95–1.35)   | 0.165   | 0     | 0.587   |
| Number of patients        |                   |                    |                   |         |       |         |
| >100                      | 8                 | 2192               | 2.08(1.33–3.23)   | 0.001   | 73.7  | 0       |
| ≤100                      | 6                 | 538                | 2.38(1.67–3.33)   | 0.007   | 73.1  | 0.001   |
| Cut off value             |                   |                    |                   |         |       |         |
| ≤50%                      | 4                 | 1451               | 1.56(0.89–2.7)    | 0.116   | 59.7  | 0.059   |
| >50%                      | 4                 | 512                | 2.08(1.45–2.94)   | 0       | 0     | 0.477   |
| Quality score             |                   |                    |                   |         |       |         |
| ≤5                        | 7                 | 2079               | 1.27(1.2–3.33)    | 0.007   | 73.1  | 0.001   |
| >5                        | 7                 | 651                | 2.5(1.85–3.33)    | 0       | 0     | 0.889   |

doi:10.1371/journal.pone.0070858.t002
Correlation of E-cadherin expression with clinicopathological parameters

Thirteen studies evaluated the correlation of E-cadherin expression with differentiation grade. The pooled OR was 0.36 (95% CI: 0.19–0.7, Z = 3.03, P = 0.002) with heterogeneity (I² 64.9%, P = 0.000) (Table 3). The authors suggested that low E-cadherin expression and poor patient outcome was associated with decreased differentiation grade (HR = 2.31, 95% CI: 1.73–3.09) (Table 4). Although heterogeneity was detected (I² = 72.4%, P = 0.000), the significant relationship was not changed in a sensitivity analysis removing each study. Subgroup analysis revealed that low E-cadherin expression was only significantly associated with poor prognosis in Asian countries, while not in European countries. Recently, E-cadherin was also reported to be related with gastric cancer or non-small cell lung cancer among Asians but not Europeans [47,48]. These observations concurred with our finding. Moreover, Egger’s test indicated that there was no evidence of significant publication bias after assessing the funnel plot (Figure S1–S5) for the studies included in our meta-analysis.

Discussion

E-cadherin is a well-described cosuppressor that was important in cell adhesion. Decreased production of E-cadherin, one of the central events underlying EMT, has been linked to increased invasiveness in several cancers [9,44–46]. However, there is no consensus on the association between reduced E-cadherin expression detected by IHC and poor survival in patients with CRC at present. Meta-analysis is a systematical approach applied widely to the evaluation of prognostic indicators in different trials. Thus, we performed a quantitative meta-analysis to determine the association between E-cadherin expression and the survival and clinicopathological features of CRC.

To explore the connection with the CRC survival, our analysis combined the outcomes of 14 studies comprising 2730 CRC patients, indicating that the relationship between reduced E-cadherin expression and worse prognosis of CRC was obviously (HR = 2.27, 95% CI: 1.63–3.17; Z = 4.83; P = 0.000). In addition, the significant relationship was not changed in a sensitivity analysis removing each study. Subgroup analysis revealed that low E-cadherin expression was only significantly associated with poor prognosis in Asian countries, while not in European countries. Recently, E-cadherin was also reported to be related with gastric cancer or non-small cell lung cancer among Asians but not Europeans [47,48]. These observations concurred with our finding and suggested that E-cadherin expression could be racial different as a prognostic factor. In addition, we found that the cut off value of low E-cadherin expression also altered the prognostic significance. The prognostic value of low E-cadherin expression existed when the threshold was >50% rather than ≤50%. Moreover, significant correlations were also observed between E-cadherin expression and clinicopathological features including differentiation grade, Dukes’ stages, lymphnode status and metastasis.

In this meta-analysis, we had dealt with highly significant heterogeneity among the 27 studies. Although we used random effects models to analyze the data, heterogeneity was still a potential problem to affect meta-analysis results. Meanwhile, we only chose studies with methods of immunohistochemistry to reduce heterogeneity as soon as possible, but source and dilutions of primary antibodies, evaluation standards, clinicopathological parameters, study location, number of patients, sex and age of patients and quality score were quite different, which contributed to the heterogeneity inevitably. When the analysis on OS was performed without consideration of other factors, obvious heterogeneity was detected (I² = 67.3%, P = 0.000). Thus, we performed stratified analysis according to study location, quality score, number of patients and cut-off value to identify the source of the great heterogeneity, and found that when the analysis was carried out on the basis of study location, heterogeneity

**Table 3.** HRs (95% CI) of sensitivity analysis for the meta-analysis.

| Study omitted       | Estimated HR | low value of 95%CI | High value of 95%CI |
|---------------------|--------------|--------------------|---------------------|
| Andras (2012)       | 2.4472163    | 1.7096629          | 3.5029523           |
| Kang (2011)         | 2.071373     | 1.520251           | 2.822878            |
| Karamitopoulou (2010)| 2.4139702   | 1.6753392          | 3.4782522           |
| Filiz (2009)        | 2.3332052    | 1.6147903          | 3.3712401           |
| Chen (2008)         | 2.2308643    | 1.5846187          | 3.1406643           |
| Zlobec (2007)       | 2.3679984    | 1.813562           | 3.0919354           |
| Nqan (2007)         | 2.296138     | 1.5749115          | 3.1564808           |
| Shirori (2006)      | 2.2920065    | 1.602922           | 3.2771797           |
| Shiono (2006)       | 2.1931612    | 1.5658236          | 3.0718379           |
| Roca (2006)         | 2.276603     | 1.5928479          | 3.253871            |
| Bondi (2006)        | 2.2962635    | 1.6179354          | 3.2589839           |
| Aoki (2003)         | 2.2190771    | 1.5786371          | 3.119338            |
| Ikeguchi (2000)     | 2.2186153    | 1.5769041          | 3.1214669           |
| Nanashima (1999)    | 2.1959348    | 1.5661801          | 3.0789113           |
| Combined            | 2.2725907    | 1.6285183          | 3.1713911           |

**Table 4.** Combined HRs (95% CI) and high value of 95%CI of sensitivity analysis for the meta-analysis.

| Study omitted       | Estimated HR | low value of 95%CI | High value of 95%CI |
|---------------------|--------------|--------------------|---------------------|
| Andras (2012)       | 2.4472163    | 1.7096629          | 3.5029523           |
| Kang (2011)         | 2.071373     | 1.520251           | 2.822878            |
| Karamitopoulou (2010)| 2.4139702   | 1.6753392          | 3.4782522           |
| Filiz (2009)        | 2.3332052    | 1.6147903          | 3.3712401           |
| Chen (2008)         | 2.2308643    | 1.5846187          | 3.1406643           |
| Zlobec (2007)       | 2.3679984    | 1.813562           | 3.0919354           |
| Nqan (2007)         | 2.296138     | 1.5749115          | 3.1564808           |
| Shirori (2006)      | 2.2920065    | 1.602922           | 3.2771797           |
| Shiono (2006)       | 2.1931612    | 1.5658236          | 3.0718379           |
| Roca (2006)         | 2.276603     | 1.5928479          | 3.253871            |
| Bondi (2006)        | 2.2962635    | 1.6179354          | 3.2589839           |
| Aoki (2003)         | 2.2190771    | 1.5786371          | 3.119338            |
| Ikeguchi (2000)     | 2.2186153    | 1.5769041          | 3.1214669           |
| Nanashima (1999)    | 2.1959348    | 1.5661801          | 3.0789113           |
| Combined            | 2.2725907    | 1.6285183          | 3.1713911           |

doi:10.1371/journal.pone.0070858.t003

E-Cadherin in Colorectal Cancer
### A

**Study** | **OR (95% CI)** | **Weight%**
--- | --- | ---
Chen (2012) | 0.29 (0.10, 0.86) | 9.15
Ozguven (2011) | 0.29 (0.09, 0.90) | 8.82
Fang (2010) | 0.34 (0.12, 0.97) | 9.34
Filiz (2009) | 0.41 (0.09, 1.90) | 7.30
Pap (2009) | 0.04 (0.01, 0.14) | 8.50
Chen (2008) | 0.07 (0.00, 1.27) | 3.61
Shioiri (2006) | 0.76 (0.19, 3.01) | 7.91
Roca (2006) | 1.04 (0.33, 3.30) | 8.82
Bravou (2005) | 0.26 (0.11, 0.62) | 10.06
Garinis (2003) | 5.78 (1.26, 26.53) | 7.32
Namashima (1999) | 0.15 (0.02, 1.18) | 5.58
Ilyas (1997) | 1.36 (0.27, 6.85) | 6.97
Mohri (1996) | 0.11 (0.02, 0.61) | 6.63

**NOTE:** Weights are from random effects analysis

### B

**Study** | **OR (95% CI)** | **Weight%**
--- | --- | ---
Lu (2012) | 0.38 (0.17, 0.82) | 18.57
Fang (2010) | 0.04 (0.01, 0.17) | 8.28
Pap (2009) | 0.60 (0.20, 1.76) | 12.92
Chen (2008) | 0.35 (0.10, 1.16) | 11.01
Shioiri (2006) | 0.38 (0.19, 0.76) | 20.53
Ikeguchi (2000) | 0.39 (0.15, 1.07) | 14.24
Mohri (1996) | 0.44 (0.17, 1.19) | 14.44

**Overall (I−squared = 41.1%, p = 0.117)**

0.34 (0.21, 0.55) 100.00

**NOTE:** Weights are from random effects analysis

### C

**Study** | **OR (95% CI)** | **Weight%**
--- | --- | ---
Lu (2012) | 0.23 (0.10, 0.52) | 8.71
Lampropoulos (2012) | 0.56 (0.31, 1.00) | 10.62
Fang (2010) | 0.02 (0.00, 0.15) | 3.26
Filiz (2009) | 1.21 (0.50, 2.89) | 8.46
Pap (2009) | 0.61 (0.27, 2.39) | 7.08
Chen (2008) | 0.31 (0.09, 1.06) | 6.30
Nqan (2007) | 1.06 (0.48, 2.33) | 9.07
Shioiri (2006) | 0.45 (0.23, 0.89) | 9.84
Roca (2006) | 0.92 (0.38, 2.11) | 8.46
Bravou (2005) | 0.31 (0.13, 0.75) | 8.41
Garinis (2003) | 0.08 (0.01, 0.72) | 2.75
Ikeguchi (2000) | 0.54 (0.11, 2.59) | 4.66
Namashima (1999) | 1.81 (0.38, 8.64) | 4.68
Mohri (1996) | 0.38 (0.14, 1.01) | 7.71

**Overall (I−squared = 59.0%, p = 0.003)**

0.49 (0.32, 0.74) 100.00

**NOTE:** Weights are from random effects analysis

### D

**Study** | **OR (95% CI)** | **Weight%**
--- | --- | ---
Chen (2012) | 2.93 (1.02, 8.45) | 12.05
Filiz (2009) | 1.76 (0.56, 5.54) | 11.50
Pap (2009) | 0.17 (0.04, 0.66) | 10.13
Shioiri (2006) | 0.47 (0.21, 1.06) | 13.54
Delektorskaya (2005) | 0.14 (0.06, 0.32) | 13.62
Fernebro (2004) | 0.56 (0.31, 1.02) | 14.80
Namashima (1999) | 0.22 (0.07, 0.65) | 11.81
Mohri (1996) | 0.25 (0.09, 0.66) | 12.56

**Overall (I−squared = 77.6%, p = 0.000)**

0.45 (0.22, 0.91) 100.00

**NOTE:** Weights are from random effects analysis
Table 4. E-cadherin expression and clinicopathological features of colorectal cancer.

| Clinicopathological features       | Number of studies | Number of patients | Pooled OR (95%CI) | P value | I²(%) | P value |
|-----------------------------------|-------------------|--------------------|-------------------|---------|-------|---------|
| Differentiation grade (G1/G2 vs. G3/G4) | 13                | 1205               | 0.36(0.19–0.7)    | 0.002   | 67.4  | 0       |
| Dukes’ stages (A/B vs. C/D)       | 7                 | 725                | 0.34(0.21–0.55)   | 0       | 41.1  | 0.117   |
| Lymphnode status (No vs. Yes)     | 13                | 1593               | 0.49(0.32–0.74)   | 0.001   | 59    | 0.003   |
| Metastasis (No vs. Yes)           | 8                 | 1027               | 0.45(0.22–0.91)   | 0.025   | 77.6  | 0.000   |
| AJCC stage (I/II vs. III/IV)      | 7                 | 831                | 0.74 (0.54–1.0)   | 0.051   | 4     | 0.396   |
| Depth of invasion (No vs. Yes)    | 5                 | 656                | 0.47(0.19–1.16)   | 0.1     | 49.3  | 0.096   |

doi:10.1371/journal.pone.0070858.t004

disappeared. Therefore, the heterogeneity in this study might be explained by the patient ethnicity.

Meanwhile, there were some limitations in this meta-analysis. First, the study included in our meta-analysis was restricted only to articles published in English, which probably brought about additional bias. Second, the credibility of HRs calculated from data or extracted from survival curves might be less than that of direct analysis of variance.

In summary, we showed that low or absent E-cadherin expression was significantly connected with metastasis and worse prognosis of CRC in Asian patients in this study. Furthermore, a cut off value of more than 50 percent was recommended when the negative definition of E-cadherin was determined according to the negative percentage of tumor cells in the immunohistochemical staining. However, large, well-designed prospective studies are required to further confirm our results.

Supporting Information

Figure S1 Egger’s publication bias plot showed no publication bias for studies regarding E-cadherin expression and differentiation grade in the meta-analysis. (TIF)

Figure S2 Egger’s publication bias plot showed no publication bias for studies regarding E-cadherin expression and Dukes’ stages in the meta-analysis. (TIF)

Figure S3 Egger’s publication bias plot showed the presence of publication bias for studies regarding E-cadherin expression and lymphnode status in the meta-analysis. (TIF)

Figure S4 Egger’s publication bias plot showed no publication bias for studies regarding E-cadherin expression and metastasis in the meta-analysis. (TIF)

Table S1 PRISMA checklist. (DOC)

Author Contributions

Conceived and designed the experiments: XH. Performed the experiments: MJ. Contributed reagents/materials/analysis tools: XH. Wrote the paper: XH XZ.

References

1. Siegel R, Naishadham D, Jemal A (2012) Cancer statistics, 2012. CA: A Cancer Journal for Clinicians 62: 10–29.
2. Geoghegan JG, Scheele J (1999) Treatment of colorectal liver metastases. British Journal of Surgery 86: 136–169.
3. Barker N, Clevers H (2000) Tumor environment: a potent driving force in colorectal cancer? Trends Mol Med 7: 535–537.
4. Hur K, Toiyama Y, Takahashi M, Balaguer F, Nagasaka T, et al. (2012) MicroRNA-200c modulates epithelial-to-mesenchymal transition (EMT) in human colorectal cancer metastasis. Gut.
5. Bates RC (2005) Colorectal cancer progression: integrin alphavbeta6 and the epithelial-mesenchymal transition (EMT). Cell Cycle 4: 1350–1352.
6. Brabitz T, Huhbek F, Spaderna S, Schmalhofer O, Hiendlmeyer E, et al. (2005) Invasion and metastasis in colorectal cancer: epithelial-mesenchymal transition, mesenchymal-epithelial transition, stem cells and beta-catenin. Cells Tissues Organs 179: 56–65.
7. Thiery JP (2003) Epithelial-mesenchymal transitions in development and pathologies. Current Opinion in Cell Biology 15: 740–746.
8. Arias AM (2004) Epithelial-mesenchymal interactions in cancer and development. Cell 105: 425–431.
9. Thiery JP (2002) Epithelial-mesenchymal transitions in tumour progression. Nat Rev Cancer 2: 442–454.
10. Ye J, Wu D, Shen J, Wu P, Ni C, et al. (2012) Enrichment of colorectal cancer stem cells through epithelial-mesenchymal transition via CDH1 knockdown. Mol Med Report 6: 507–512.
11. Stang A (2010) Critical evaluation of the Newcastle-Ottawa scale for the assessment of the quality of nonrandomized studies in meta-analyses. European Journal of Epidemiology 25: 603–605.

12. Parmar MK, Torri V, Stewart L (1998) Extracting summary statistics to perform meta-analyses of the published literature for survival endpoints. Statistics in Medicine 17: 2193–2213.

13. Handoll HH (2006) Systematic reviews on rehabilitation interventions. Archives of Physical Medicine and Rehabilitation 87: 875.

14. Ioannidis JP, Patsopoulos NA, Evangelou E (2007) Uncertainty in heterogeneity estimates in meta-analyses. BMJ 335: 964–966.

15. Mantel N, Haenszel W (1959) Statistical aspects of the analysis of data from retrospective studies of disease. Journal of the National Cancer Institute 22: 719–748.

16. Higgins JP, Thompson SG, Deeks JJ, Altman DG (2003) Measuring inconsistency in meta-analyses. BMJ 327: 565–568.

17. Lu MH, Huang CC, Pan MR, Chen HH, Hung WC (2012) Prospero homeobox 1 promotes epithelial-mesenchymal transition in colon cancer cells by inhibiting E-cadherin via miR-9. Clinical Cancer Research 18: 6416–6425.

18. Andras C, Toth L, Molnar C, Tanyi M, Colli Z, et al. (2012) Correlations between clinicopathological parameters and molecular signatures of primary tumors for patients with stage T3N0 colorectal adenocarcinomas: a single center retrospective study on 100 cases. Hepato-Gastroenterology 59: 1051–1059.

19. Chen X, Wang Y, Xia H, Wang Q, Jiang X, et al. (2012) Loss of E-cadherin promotes the growth, invasion and drug resistance of colorectal cancer cells and is associated with liver metastasis. Molecular Biology Reports 39: 6707–6714.

20. Lampropoulos P, Zisi-Sermpezoglou A, Rizos S, Kostakis A, Nikitas N, et al. (2012) Prognostic significance of transforming growth factor beta (TGF-beta) signaling axis molecules and E-cadherin in colorectal cancer. Tumour Biology 33: 1005–1014.

21. Oguzen BY, Karacetin D, Kabukcuoglu F, Taskin T, Yener S (2011) Immunohistochemical study of E-cadherin and beta-catenin expression in colorectal carcinomas. Polish Journal of Pathology 62: 19–24.

22. Kang H, Min BS, Lee KY, Kim NK, Kim SN, et al. (2011) Loss of E-cadherin and MUC2 correlations evidenced with poor survival in patients with stages II and III colorectal carcinoma. Annals of Surgical Oncology 18: 711–719.

23. Fang QX, Li LZ, Yang B, Zhao ZS, Wu Y, et al. (2010) L1, beta-catenin, and E-cadherin expression in patients with colorectal cancer: correlation with clinicopathological features and its prognostic significance. Journal of Surgical Oncology 102: 433–442.

24. Karanmitopoulou E, Zieber I, Ksormarianos A, Patsouris ES, Peros G, et al. (2010) Expression of p16 in lymph node metastases of adjuvantly treated stage III colorectal cancer patients identifies poor prognostic subgroups: a retrospective analysis of biomarkers in matched primary tumor and lymph node metastases. Cancer 116: 4474–4486.

25. Areu L, Perelli P, Zanetti R, Callari D, Biglatti B, et al. (2010) E-cadherin and beta-catenin expression in canine colorectal adenocarcinoma. Research in Veterinary Science 89: 409–414.

26. Filiz AI, Sendil Z, Sucuolu I, Kurt Y, Demirbas S, et al. (2010) The survival effect of E-cadherin and catenins in colorectal carcinomas. Colorectal Dis 12: 1223–1230.

27. Pap Z, Paviu Z, Denes I, Kovalsky I, Jung J (2009) An immunohistochemical study of colon adenomas and carcinomas: E-cadherin, Syndecan-1, Ets-1. Pathology Oncology Research 15: 579–587.

28. Chen S, Liu J, Li G, Mo F, Xu X, et al. (2006) Altered distribution of beta-catenin and peritumoral lymphocytic infiltration in tumour budding in colorectal cancer. Journal of Pathology 212: 260–268.

29. Zlobec I, Lugli A, Baker K, Roth S, Minoo P, et al. (2007) Role of APAF-1, E-cadherin and peritumoral lymphocytic infiltration in tumour budding in colorectal cancer. Journal of Pathology 212: 260–268.

30. Ngan CY, Yamamoto H, Seshimo I, Enumi K, Terayama M, et al. (2007) A multivariate analysis of adhesion molecules expression in assessment of colorectal cancer. Journal of Surgical Oncology 95: 652–662.

31. Shioso M, Shida T, Koda K, Oda K, Seike K, et al. (2006) Slug expression is an independent prognostic parameter for poor survival in colorectal carcinoma patients. British Journal of Cancer 94: 1016–1022.

32. Shioso S, Ishii G, Nagai K, Murata Y, Tsuchi K, et al. (2006) Immunohistochemical prognostic factors in resected colorectal lung metastases using tissue microarray analysis. European Journal of Surgical Oncology 32: 308–309.

33. Roca F, Mauro LV, Morandi A, Bonadecio F, Vaccaro C, et al. (2006) Prognostic value of E-cadherin, beta-catenin, MMP7, 7and TIMP1 and 2 in patients with colorectal cancer. Journal of Surgical Oncology 95: 151–160.

34. Bravou V, Klonizomos G, Papakota E, Taraviras S, Varaklas J (2006) IL8 overexpression in human colon cancer progression correlates with activation of beta-catenin, down-regulation of E-cadherin and activation of the Akt-FKHR pathway. Journal of Pathology 208: 91–99.

35. Delekoeskaya VV, Perevoshchikov AG, Golovkov DA, Kushlinski NE (2005) Expression of E-cadherin, beta-catenin, and CD4-Hv6 cell adhesion molecules in primary tumors and metastases of colorectal adenocarcinoma. Bulletin of Experimental Biology and Medicine 139: 706–710.

36. Bondi J, Buhlohn G, Nesland JM, Bakka A, Buhlohn IR (2006) An increase in the number of adhesion proteins with altered expression is associated with an increased risk of cancer death for colon carcinoma patients. International Journal of Colorectal Disease 21: 231–237.

37. Fernebro E, Bendahl PO, Dicke M, Persson A, Ferno M, et al. (2004) Immunohistochemical patterns in rectal cancer: application of tissue microarray with prognostic correlations. International Journal of Cancer 111: 921–928.

38. Gairinis GA, Spanakiz NE, Menounos PG, Manolis EN, Peros G (2003) Transcriptional impairment of beta-catenin/E-cadherin complex is not associated with beta-catenin mutations in colorectal carcinomas. British Journal of Cancer 88: 206–209.

39. Aoki S, Shimamura T, Shibata T, Nakashima Y, Moriya Y, et al. (2003) Prognostic significance of dysadherin expression in advanced colorectal carcinoma. British Journal of Cancer 88: 726–732.

40. Ikoguchi M, Taniguchi T, Makino M, Kabaza N (2000) Reduced E-cadherin expression and enlargement of cancer nuclei strongly correlate with hematogenic metastasis in colorectal adenocarcinoma. Scandinavian Journal of Gastroenterology 35: 839–846.

41. Nanashima A, Yamauchi H, Sawai T, Yasutake T, Tsujii T, et al. (1999) Expression of adhesion molecules in hepatic metastases of colorectal carcinoma. Scandinavian Journal of Gastroenterology 34: 839–846.

42. Nanashima A, Yamauchi H, Sawai T, Yasutake T, Tsujii T, et al. (1999) Expression of adhesion molecules in hepatic metastases of colorectal carcinoma. Scandinavian Journal of Gastroenterology 34: 839–846.

43. Nanashima A, Yamauchi H, Sawai T, Yasutake T, Tsujii T, et al. (1999) Expression of adhesion molecules in hepatic metastases of colorectal carcinoma. Scandinavian Journal of Gastroenterology 34: 839–846.

44. Kroepil F, Fluegen G, Totikov Z, Baldus SE, Vay C, et al. (2012) Down-regulation of CDH1 is associated with expression of SNAI1 in colorectal cancer tissue. Surgery Today 27: 606–612.

45. Byas M, Tomlinson IP, Hanby A, Talbot RC, Bodmer WF (1997) Allele loss, replication errors and loss of expression of E-cadherin in colorectal cancers. Gut 44: 654–659.

46. Mohri Y (1997) Prognostic significance of dysadherin expression in human colorectal cancer tissue. Surgery Today 27: 606–612.

47. Nagatani Kh, Merchant NB (2012) Sex-mediated regulation of E-cadherin and EMT in pancreatic cancer. Frontiers in Bioscience 17: 2059–2069.

48. Cheng JC, Auerpere N, Leung PC (2012) EGF-induced EM and invasiveness in serous borderline ovarian tumor cells: a possible step in the transition to low-grade serous carcinoma cells? Plos One 7: e34071.

49. Xing X, Tang YB, Yuan G, Wang Y, Wang J, et al. (2012) The prognostic value of E-cadherin in gastric cancer: A meta-analysis. International Journal of Cancer.