Research Paper

Is Ep-CAM Expression a Diagnostic and Prognostic Biomarker for Colorectal Cancer? A Systematic Meta-Analysis

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ABSTRACT

Background: Cancer stem cell (CSC) epithelial cell adhesion molecule (Ep-CAM) is frequently expressed in colorectal cancer (CRC). However, the clinical significance of Ep-CAM expression in CRC is not clear. This study evaluated whether Ep-CAM provided valuable insight as a molecular biomarker for CRC diagnosis and prognosis and the potential of Ep-CAM as a novel therapeutic target in CRC.
Methods: Publications were selected online using electronic databases. The pooled odds ratios (ORs) or hazard ratios (HRs) with their 95% confidence intervals (95% CIs), and the combined sensitivity, specificity, and area under the curve (AUC) were calculated and summarized.
Results: Eleven eligible articles published in English involving 4561 cases were analyzed in this study. Ep-CAM expression was significantly higher in CRC compared with normal controls, and its overexpression was negatively linked to tumor differentiation, tumor stage, vascular invasion, depth of tumor invasion, lymph node metastasis, distant metastasis, and tumor budding in CRC patients. The loss of Ep-CAM expression positively correlated with these characteristics. Multivariate analysis of loss of Ep-CAM expression correlated with a poor prognosis in disease-free survival (DFS), disease-specific survival (DSS), and overall survival (OS). The pooled sensitivity, specificity and AUC values of Ep-CAM expression in patients with CRC vs. normal controls were 0.93, 0.90, and 0.94, respectively.
Conclusions: The present findings suggest that Ep-CAM expression may be associated with CRC carcinogenesis, while the loss of Ep-CAM expression is correlated with the progression, metastasis, and poor prognosis of CRC. Ep-CAM expression may be a useful biomarker for the clinical diagnosis of CRC.

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1. Introduction

Colorectal cancer (CRC) is the fifth most frequent malignancy and the fifth leading cause of death in all human cancers in China (Chen et al., 2016). Global cancer statistics noted approximately 1.4 million new cases were clinically diagnosed with CRC in 2012 and an estimated 693,900 deaths occurred due to CRC worldwide (Torre et al., 2015). CRC is a major public health problem because approximately 50% of CRC cases develop metastasis and exhibit a poor survival rate despite advances in early detection and treatment (Aranda et al., 2015; Ferlay et al., 2015; Van Cutsem et al., 2010). Therefore, there is a need for a potential biological marker that allows early detection and predicts clinical prognosis of CRC.

A special subpopulation of cancer cells, known as cancer stem cells (CSCs), drive cancer progression. CSCs self-renew, uncontrollably proliferate, differentiate, and form the bulk of the tumor (Majumdar et al., 2012; Chandler and Lagasse, 2010). CSCs also regulate the development, progression, and metastasis of cancer (da Silva-Diz et al., 2016, Sampetrean and Saya, 2013, Nguyen et al., 2012). Multiple studies reported evidence of CSCs in CRC (Patman, 2016; Zeuner et al., 2014). Epithelial cell adhesion molecule (Ep-CAM), also known as epithelial-specific antigen (ESA) or CD326, is a transmembrane glycoprotein cell adhesion molecule that is encoded by the Ep-CAM gene mapped to chromosomal region 4q (Balzar et al., 1999; Linnenbach et al., 1989). This cell adhesion molecule plays a key role in Ca^{2+}-independent cell-to-cell adhesion (Litvinov et al., 1994). Ep-CAM correlates with cell proliferation, migration, invasion, motility, and signal transduction (Subramanian et al., 2015; Maetzel et al., 2009). Studies suggest that Ep-CAM is associated with epithelial-to-mesenchymal transition (EMT) and enhances tumor-initiating capacity (Gupta et al., 2009; Morel et al., 2008). Ep-CAM is overexpressed in many types of cancers, such as breast cancer, ovarian cancer, and head and neck squamous cell

cancer (Moldenhauer et al., 2012). Some studies demonstrated that Ep-CAM overexpression was an unfavorable prognostic marker in breast cancer and gallbladder carcinoma (Varga et al., 2004; Gastl et al., 2000). Ep-CAM is a CSC marker, and it is frequently expressed or overexpressed in CRC (Dalerba et al., 2007; Mosolits et al., 2004). Ep-CAM is defined as a universal molecular marker for circulating tumor cell (CTC) detection, which is termed the "post-Ep-CAM era" (Nicolazzo et al., 2015; Raimondi et al., 2015). Therefore, it is important to investigate Ep-CAM further.

Some studies demonstrated inconsistent and controversial conclusions of Ep-CAM expression in CRC patients. For example, Kuhn et al. reported that Ep-CAM expression was not associated with tumor stage or grade (Kuhn et al., 2007). Gosens et al. reported that the loss of Ep-CAM expression was significantly associated with tumor grade and trended towards a correlation with tumor stage (Gosens et al., 2007). Therefore, the present study assessed whether Ep-CAM expression correlated with an increased risk of CRC vs. benign colonic lesions and normal controls. We also analyzed whether Ep-CAM overexpression or the loss of Ep-CAM expression was associated with the prognostic effect and clinicopathological features of CRC. Finally, we evaluated the use of Ep-CAM expression as a biomarker for the early diagnosis of CRC.

2. Materials and Methods

2.1. Literature Search

A comprehensive literature search (PubMed, EMBASE, EBSCO, Web of Science, and Cochrane Library databases) was performed to identify relevant publications on Ep-CAM expression in CRC patients prior to January 16th, 2017. The articles were identified using the following search terms and key words: 'Ep-CAM', 'epithelial cell adhesion molecule', 'EpCAM', 'CD326', 'GA733', 'CO17-1A', 'EGP', 'KS1-4', 'ESA', 'MOC31', 'BerEP4', and 'colorectal cancer' 'colorectal tumor', 'colorectal carcinoma', 'colorectal neoplasm', 'CRC' and 'expression'. The reference lists of the included articles were also screened to obtain other potential studies. This study was performed based on the preferred reporting items for systematic reviews and meta-analyses (PRISMA) statement criteria (Moher et al., 2009) (Table S1).

2.2. Selection Criteria

Eligible studies were selected based on the following inclusion criteria: 1) patients were limited to a diagnosis of CRC; 2) articles were published in English with the full text; 3) Ep-CAM was considered as a positive expression, overexpression or a loss of the expression; 4) articles provided sufficient information to evaluate the correlation of Ep-CAM expression between CRC and nonmalignant controls; 5) articles provided sufficient data to assess the relationship of Ep-CAM expression with the clinicopathological characteristics of patients with CRC; and 6) the pooled hazard ratios (HRs) and their 95% confidence intervals (CIs) were extracted to evaluate the prognostic role when the data from the original papers were available. The most complete paper containing the most information was selected when authors published several articles using duplicated data.

2.3. Data Extraction

The following data were collected from eligible studies: first author's surname, year of publication, country, ethnic population, age, detection method, clinical stage, staining patterns, cut-off values, Ep-CAM expression (overexpression or loss), expression frequency, number of cases and controls, survival data of multivariate analysis, and clinicopathological features. The clinicopathological characteristics included tumor differentiation (poor vs. well/moderate), tumor stage (3–4 vs. 1–2), vascular invasion (yes vs. no), depth of tumor invasion (pT3–4 vs. pT1–2), lymph node metastasis (yes vs. no), distant metastasis (yes

vs. no), tumor budding (yes vs. no), and tumor location (colon vs. rectum). Loss of Ep-CAM expression consisted of complete and partial loss.

2.4. Statistical Analysis

Data were analyzed using Stata software (version 12.0, Stata Corporation, College Station, TX, USA). The combined odds ratios (ORs) and their 95% confidence intervals (95% CIs) were calculated to determine the relationship of Ep-CAM expression between CRC and nonmalignant controls. The associations of Ep-CAM overexpression or loss of Ep-CAM expression with the clinicopathological features of CRC were also calculated using pooled ORs with 95% CIs. The overall hazard ratios (HRs) with 95% CIs were used to determine the impact of Ep-CAM expression on the survival of CRC patients, if possible. Between-study heterogeneity was measured using the Cochran's Q statistic (Zintzaras and Ioannidis, 2005). The random-effects model was used to increase the reliability of the results in the present analysis (heterogeneity: $P < 0.1$). A sensitivity analysis was performed to determine the influence of one study on the results and heterogeneity via omission of a single study when the pooled results with greater than two studies had substantial heterogeneity ($P < 0.1$) (Higgins et al., 2003; Lau et al., 1997). Potential publication bias was assessed using Egger's test for the results with greater than or equal to ten studies (Egger et al., 1997). The pooled sensitivity, specificity, and the summary receiver operator characteristic (SROC) curve (AUC) values were calculated and constructed according to the bivariate analysis to evaluate the performance of the diagnostic capacity of Ep-CAM expression to CRC in this study (Reitsma et al., 2005; Jones and Athanasiou, 2005).

3. Results

3.1. Characteristics of the Included Studies

Fig. 1 shows the detailed selection procedure used for the potential literature. The abovementioned inclusion criteria resulted in 11 eligible studies published in English from 2005 to 2016 (Kim et al., 2016; Chai et al., 2015; Zhou et al., 2015; Kim et al., 2014; Goossens-Beumer et al., 2014; Lugli et al., 2010; Paret et al., 2007; Kuhn et al., 2007; Gosens et al., 2007; Went et al., 2006; Karanikiotis et al., 2005) for the meta-analysis, which included 4103 CRC patients and 458 controls. Five studies of 331 patients with CRC and 458 controls analyzed the relationship of Ep-CAM expression between CRC and controls (Chai et al., 2015; Zhou et al., 2015; Paret et al., 2007; Kuhn et al., 2007; Karanikiotis et al., 2005). Six studies analyzed the correlation of Ep-CAM overexpression or loss with
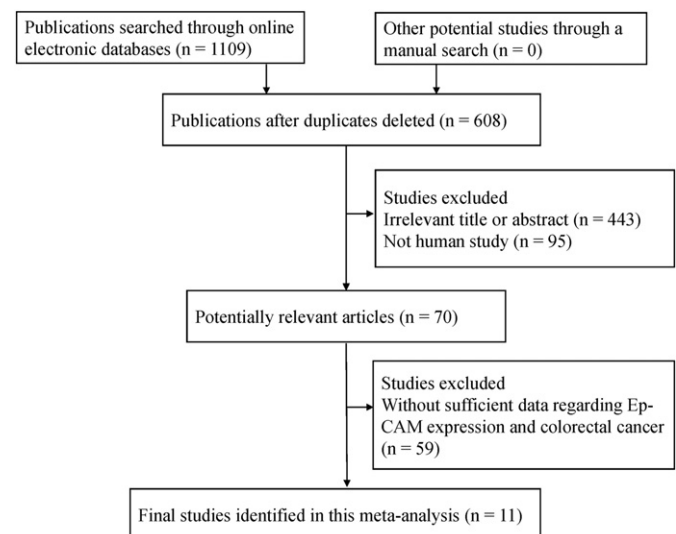

Fig. 1. Flow chart of the selection procedure.

Table 1
Basic characteristics of the publications included in this study.

First author	Country	Ethnicity	Age	Method	Stage	Staining patterns	Cut off scores (positive)	Sample	Status	CRC N (E+ %)	Benign N (E+ %)	Normal N (E+ %)	Clinicopathological features	MA-survival
Karanikiotis et al. (2005)	Greece	Caucasians	NA	#	NA	NA	NA	Blood	E	27 (96.3)		20 (0)	No	NA
Went et al. (2006)	Iran	Caucasians	69.7	IHC	NA	M/C	70%	Tissue	High	1186 (97.7)			Yes	NA
Kuhn et al. (2007)	Germany	Caucasians	NA	IHC	NA	M	0%	Tissue	E	104 (94.2)	6 (100)	92 (40.2)	No	NA
Paret et al. (2007)	Germany	Caucasians	NA	IHC	NA	NA	25%	Tissue	E	100 (94)		100 (20)	No	NA
Gosens et al. (2007)	The Netherlands	Caucasians	NA	IHC	1–4	M	NA	Tissue	High	109 (64.2)			Yes	NA
Lugli et al. (2010)	Switzerland	Caucasians	NA	IHC	NA	M	100%	Tissue	High	1278 (89.6)			Yes	NA
Kim et al. (2014)	Korea	Asians	NA	IHC	1–4	M/C	90%	Tissue	High	168 (85.7)			Yes	NA
Goossens-Beumer et al. (2014)	The Netherlands	Caucasians	NA	IHC	1–4	M	89%	Tissue	High	305 (49.8)			Yes	Yes
Zhou et al. (2015)	China	Asians	NA	IHC	1–4	C	0%	Tissue	E	50 (92)	50 (6)	100 (5)	No	NA
Chai et al. (2015)	China	Asians	NA	IHC	1–3	C	15%	Tissue	E	50 (92)		90 (5.6)	No	NA
Kim et al. (2016)	Korea	Asians	NA	IHC	1–4	M/C	95%	Tissue	High	726 (93)			Yes	Yes

NA: not applicable; E: expression; MA: multivariate analysis; "*" stands for a novel chromatographic method (molecular strip) for the detection of PCR-amplified product; IHC: immunohistochemistry; M: membrane; C: cytoplasm.

the clinicopathological characteristics of 3772 patients with CRC (Kim et al., 2016; Kim et al., 2014; Goossens-Beumer et al., 2014; Lugli et al., 2010; Gosens et al., 2007; Went et al., 2006). Two studies of 1031 CRC patients reported multivariate survival analyses of Ep-CAM expression (Kim et al., 2016; Goossens-Beumer et al., 2014). Ep-CAM expression was assessed on tissues using immunohistochemistry (IHC). Tables 1 and S2 present the main characteristics of all eligible studies.

3.2. Association of Ep-CAM Expression Between CRC and Controls

The overall OR from five studies of 331 patients with CRC and 402 normal controls demonstrated that the frequency of Ep-CAM expression in CRC was notably higher than in normal controls (OR = 96.70, 95% CI = 34.85–268.29, $P < 0.001$) (Fig. 2).

Two studies of 154 CRC patients and 56 benign colonic lesions revealed no significant difference between CRC and benign colonic lesions
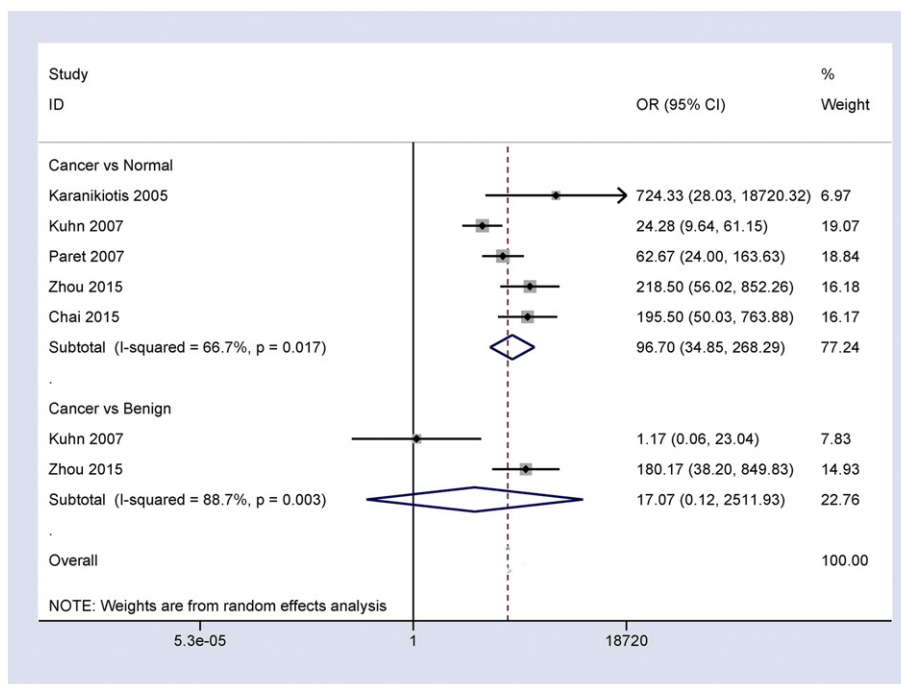

Fig. 2. Forest plot showing the relationship between Ep-CAM expression and CRC in cancer vs. controls, CRC vs. normal controls: OR = 96.70, 95% CI = 34.85–268.29, $P < 0.001$; and CRC vs. benign colonic lesions: OR = 17.07, 95% CI = 0.12–2511.93, $P = 0.265$.

in Ep-CAM expression (OR = 17.07, 95% CI = 0.12–2511.93, P = 0.265) (Fig. 2).

3.3. Association of Ep-CAM Overexpression with Tumor Differentiation and Clinical Stage of CRC

Ep-CAM overexpression negatively correlated with tumor differentiation and clinical stage (OR = 0.25, 95% CI = 0.11–0.55, P = 0.001; OR = 0.52, 95% CI = 0.28–0.96, P = 0.036; respectively) (Fig. 3) in our analysis of the results from five studies of 3433 CRC patients and four studies with 1308 CRC patients, respectively.

3.4. Association of Ep-CAM Overexpression with Vascular Invasion and Depth of Tumor Invasion of CRC

The results from three studies of 2147 patients with CRC demonstrated a negative correlation between Ep-CAM overexpression and vascular invasion (OR = 0.44, 95% CI = 0.21–0.93, P = 0.032) (Fig. 3).

The pooled OR from five studies of 3429 patients with CRC revealed a trend towards a negative association between Ep-CAM overexpression and depth of tumor invasion (OR = 0.73, 95% CI = 0.51–1.04, P = 0.078) (Fig. 4).

3.5. Association of Ep-CAM Overexpression with Lymph Node Metastasis and Distant Metastasis of CRC

Analysis of the pooled data of five studies of lymph node metastasis with 3393 CRC patients and three studies of distant metastasis with 1348 CRC patients demonstrated that Ep-CAM overexpression negatively correlated with lymph node metastasis and distant metastasis (OR = 0.55, 95% CI = 0.42–0.71, P < 0.001; OR = 0.35, 95% CI = 0.23–0.55, P < 0.001; respectively) (Fig. 4).

3.6. Association of Ep-CAM Overexpression with Tumor Budding and Tumor Location of CRC

The results of a study of 726 CRC patients revealed a negative relationship between Ep-CAM overexpression and tumor budding (OR = 0.18, 95% CI = 0.06–0.50, P = 0.001) (Fig. 4).

The pooled OR from two studies of 1031 CRC patients revealed that Ep-CAM overexpression was similar in colon and rectal cancer (OR = 0.86, 95% CI = 0.56–1.31, P = 0.486) (Fig. 4).

3.7. Association of the Loss of Ep-CAM Expression with the Clinicopathological Features of CRC

Ep-CAM overexpression negatively correlated with the clinicopathological characteristics of CRC patients, including tumor differentiation, tumor stage, vascular invasion, depth of tumor invasion, lymph node metastasis, distant metastasis, and tumor budding. Therefore, we further evaluated whether the loss of Ep-CAM expression positively correlated with these clinicopathological features.

Figs. 5 and 6 show that the loss of Ep-CAM expression positively associated with tumor differentiation, tumor stage, vascular invasion, depth of tumor invasion, lymph node metastasis, distant metastasis, and tumor budding in CRC patients (OR = 4.05, 95% CI = 1.80–9.10, P = 0.001; OR = 1.92, 95% CI = 1.04–3.54, P = 0.036; OR = 2.18, 95% CI = 1.11–4.27, P = 0.024; OR = 1.38, 95% CI = 0.96–1.97, P = 0.078; OR = 1.82, 95% CI = 1.40–2.37, P < 0.001; OR = 2.82, 95% CI = 1.81–4.39, P < 0.001; OR = 4.33, 95% CI = 1.70–11.05, P = 0.001, respectively). No correlation was found between the loss of Ep-CAM expression and tumor location (OR = 1.16, 95% CI = 0.76–1.77, P = 0.486).

3.8. Sensitivity Analysis

Heterogeneity was observed in the comparison of CRC and normal controls, in tumor differentiation, clinical stage, and vascular invasion
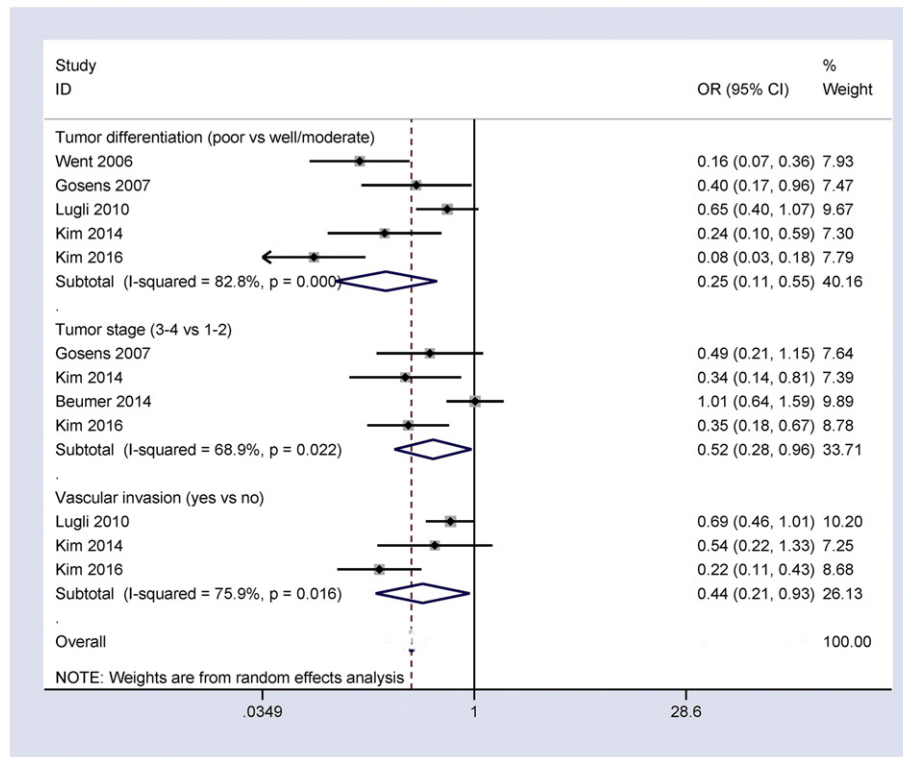

Fig. 3. Forest plot showing the relationship of Ep-CAM overexpression with some clinicopathological features of CRC, including tumor differentiation: OR = 0.25, 95% CI = 0.11–0.55, P = 0.001; clinical stage: OR = 0.52, 95% CI = 0.28–0.96, P = 0.036; and vascular invasion: OR = 0.44, 95% CI = 0.21–0.93, P = 0.032.
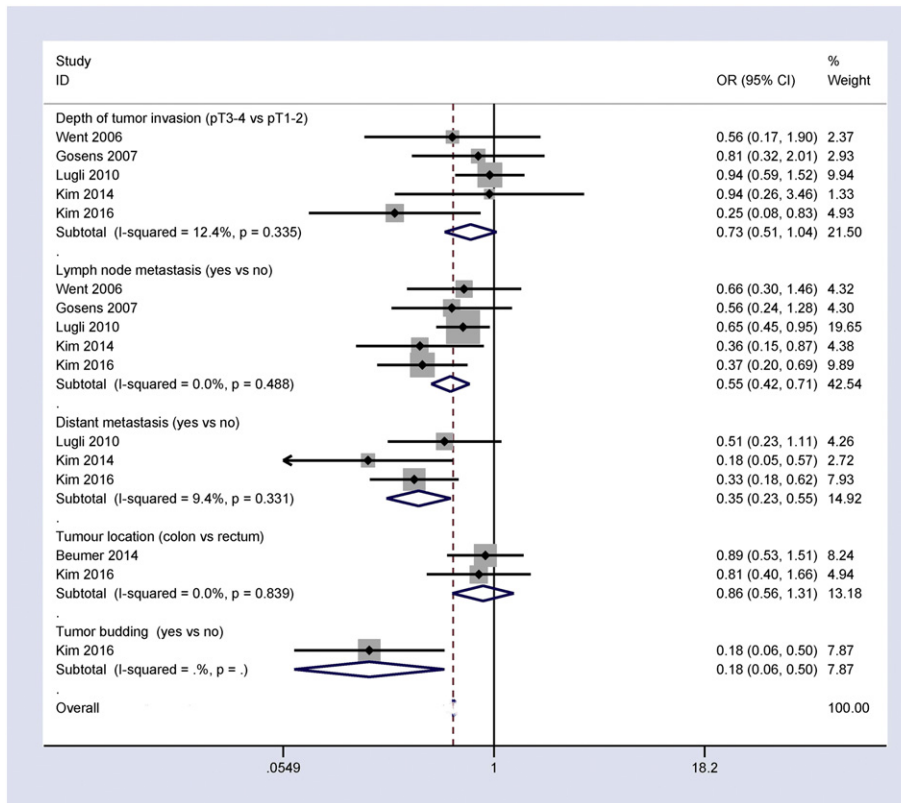
Fig. 4. Forest plot showing the association of Ep-CAM overexpression with other clinicopathological features of CRC, including depth of tumor invasion: OR = 0.73, 95% CI = 0.51–1.04, P = 0.078; lymph node metastasis: OR = 0.55, 95% CI = 0.42–0.71, P < 0.001; distant metastasis: OR = 0.35, 95% CI = 0.23–0.55, P < 0.001; tumor budding: OR = 0.18, 95% CI = 0.06–0.50, P = 0.001; and tumor location: OR = 0.86, 95% CI = 0.56–1.31, P = 0.486.
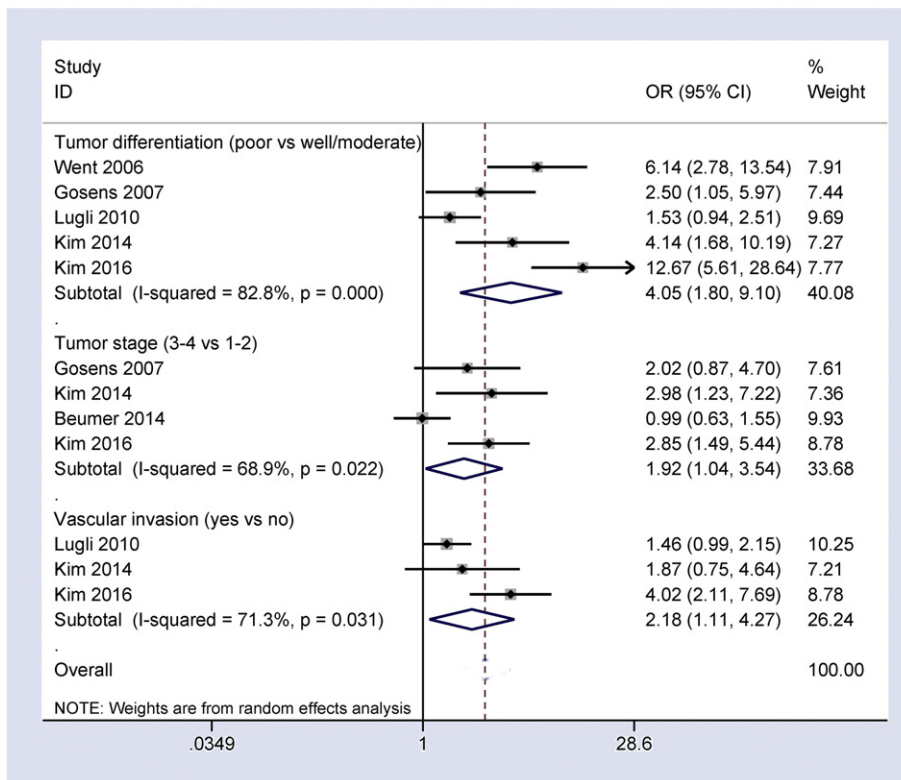

Fig. 5. Forest plot indicating the correlation of loss of Ep-CAM expression with some clinicopathological features of CRC, including tumor differentiation: OR = 4.05, 95% CI = 1.80–9.10, P = 0.001; clinical stage: OR = 1.92, 95% CI = 1.04–3.54, P = 0.036; and vascular invasion: OR = 2.18, 95% CI = 1.11–4.27, P = 0.024.
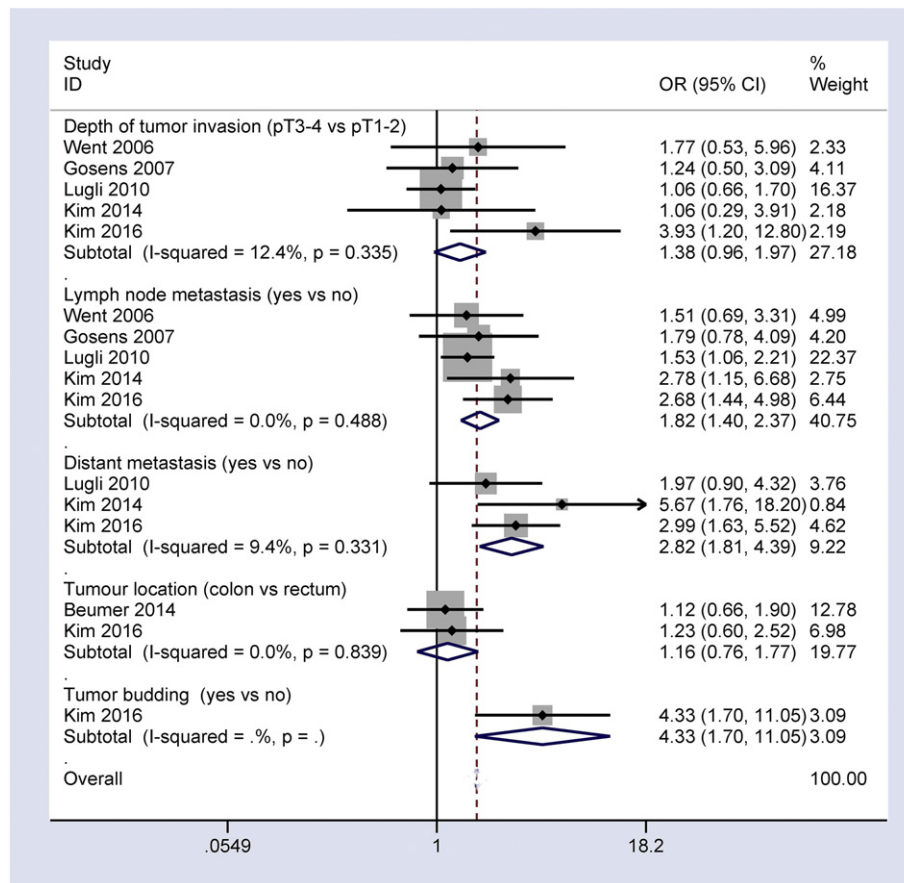
Fig. 6. Forest plot indicating the correlation of loss of Ep-CAM expression with other clinicopathological features of CRC, including depth of tumor invasion: OR = 1.38, 95% CI = 0.96–1.97, $P = 0.078$; lymph node metastasis: OR = 1.82, 95% CI = 1.40–2.37, $P < 0.001$; distant metastasis: OR = 2.82, 95% CI = 1.81–4.39, $P < 0.001$; tumor budding: OR = OR = 4.33, 95% CI = 1.70–11.05, $P = 0.001$; and tumor location: OR = 1.16, 95% CI = 0.76–1.77, $P = 0.486$.

(all $Ps < 0.1$). We removed the study (Kuhn et al., 2007) and recalculated the overall OR (OR = 137.14, 95% CI = 59.32–317.08, $P < 0.001$) in CRC vs. normal controls, which dramatically reduced the heterogeneity ($P = 0.243$). We successively removed two studies (Lugli et al., 2010) and (Kim et al., 2016) from our correlation analyses between the loss of Ep-CAM expression and tumor differentiation, and the pooled OR was 4.09 (95% CI = 2.42–6.92, $P < 0.001$), with no heterogeneity ($P = 0.319$). The combined OR between loss of Ep-CAM expression and tumor stage was 2.62 (95% CI: 1.68–4.08, $P < 0.001$) based on the omission of the study (Goossens-Beumer et al., 2014), with no evidence of heterogeneity ($P = 0.776$). One study(Kim et al., 2016) was removed from the correlation analyses of the loss of Ep-CAM expression with vascular invasion, and the pooled OR did not change substantially (OR = 1.52, 95% CI = 1.06–2.17, $P = 0.023$), but heterogeneity was significantly reduced ($P = 0.624$). The above analyses demonstrated that our results were stable and credible.

3.9. Prognostic Impact of Ep-CAM Expression in CRC Patients Using Multivariate Analysis

Only two studies reported a multivariate survival analysis of Ep-CAM expression in CRC patients (Kim et al., 2016; Goossens-Beumer et al., 2014). The loss of Ep-CAM expression correlated with a poor prognosis in disease-specific survival (DSS) and a trend towards a poor prognosis between loss of Ep-CAM expression and a 5-year overall survival (OS) in 305 CRC patients (Goossens-Beumer et al., 2014). Multivariate analysis of 725 CRC patients demonstrated that the loss of Ep-CAM expression was an unfavorable prognostic factor in disease-free survival (DFS) (HR = 1.57, 95% CI = 1.04–2.39, $P = 0.03$) (Kim et al., 2016).

3.10. Diagnostic Capacity of Ep-CAM Expression in CRC vs. Normal Controls

We also evaluated the diagnostic value of Ep-CAM expression as a potential biomarker in the discrimination of CRC patients and normal controls. The pooled sensitivity, specificity and AUC values of Ep-CAM expression were 0.93 (95% CI = 0.90–0.96), 0.90 (95% CI = 0.74–0.97), and 0.94 (95% CI = 0.92–0.96) (Fig. 7), respectively, which suggests that Ep-CAM expression exhibited good diagnostic performance in CRC.

4. Discussion

Cancer stem cells (CSCs) are a special subpopulation of cells within a tumor. Ep-CAM overexpression is a CSC marker of tumor cells that closely correlates with tumor progression, including colorectal cancer cell lines (Liao et al., 2015). Ep-CAM overexpression is a poor prognostic factor in some cancers, such as breast and gallbladder carcinomas (Schmidt et al., 2010; Varga et al., 2004), but its overexpression is a favorable prognostic factor in ovarian cancer (Battista et al., 2014). The present study found that Ep-CAM was commonly expressed in CRC, and its expression was significantly higher in CRC than in normal controls (Chai et al., 2015; Zhou et al., 2015; Paret et al., 2007; Kuhn et al., 2007; Karanikiotis et al., 2005). Ep-CAM was slightly less frequently expressed in CRC than in benign lesions (Kuhn et al., 2007), but its expression level was notably higher in CRC than in benign lesions (Zhou et al., 2015). The current results revealed that the frequency of Ep-CAM expression was significantly more common in CRC compared with normal controls, which suggests that Ep-CAM expression is associated with the carcinogenesis of CRC. Ep-CAM expression stimulates cell
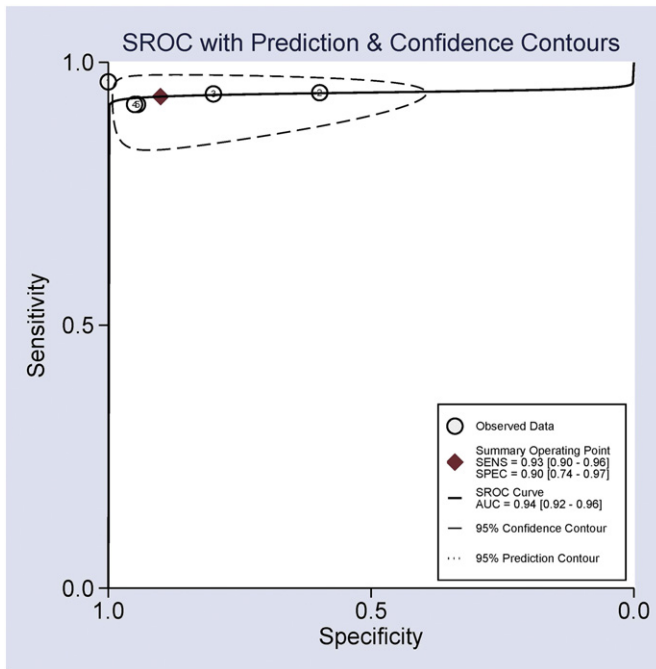
Fig. 7. Summary receiver operating characteristics (SROC) estimation of Ep-CAM expression in patients with CRC vs. normal controls, sensitivity = 0.93 (95% CI = 0.90–0.96), specificity = 0.90 (95% CI = 0.74–0.97), and AUC = 0.94 (95% CI = 0.92–0.96).

differentiation and cell proliferation via up-regulation of the proto-oncogene c-myc, which causes carcinogenic effects (Munz et al., 2004). However, no correlation in Ep-CAM expression was found between CRC and benign colonic lesions because the sample size was small. No association between Ep-CAM expression and DNA mismatch repair protein expression was reported in 218 microsatellite instability (MSI-high) CRCs (Kim et al., 2016).

Ep-CAM overexpression exhibited a high frequency in the present study (88.68%). We analyzed the relationship of Ep-CAM overexpression with the clinicopathological characteristics of CRC patients, including tumor differentiation, tumor stage, vascular invasion, depth of tumor invasion, lymph node metastasis, distant metastasis, and tumor budding. Our findings demonstrated that Ep-CAM overexpression negatively correlated with these clinicopathological characteristics, which indicates that Ep-CAM overexpression was a favorable factor in CRC progression, metastasis, and relapse.

We further investigated whether the loss of Ep-CAM expression was an unfavorable factor in relation to these features. The loss of Ep-CAM expression was significantly associated with tumor differentiation (Kim et al., 2016; Kim et al., 2014; Gosens et al., 2007; Went et al., 2006), and another study demonstrated a positive trend towards an association between the loss of Ep-CAM expression and tumor differentiation (Lugli et al., 2010). An association between the loss of Ep-CAM expression and tumor stage was found in two studies (Kim et al., 2016; Kim et al., 2014), but the other studies demonstrated no association (Goossens-Beumer et al., 2014; Gosens et al., 2007). Loss of Ep-CAM expression correlated with vascular invasion (Kim et al., 2016; Lugli et al., 2010), but there was no correlation with vascular invasion in a small population (168 patients) (Kim et al., 2014). No significant relationship was observed between loss of Ep-CAM expression and depth of tumor invasion (Kim et al., 2014; Lugli et al., 2010; Gosens et al., 2007; Went et al., 2006), but the remaining one study reported a significant correlation (Kim et al., 2016). Three studies demonstrated a correlation between loss of Ep-CAM expression and lymph node metastasis (Kim et al., 2016; Kim et al., 2014; Lugli et al., 2010), but two studies failed to demonstrate any correlation (Gosens et al., 2007; Went et al., 2006). Two studies demonstrated that the loss of Ep-CAM expression

significantly correlated with distant metastasis (Kim et al., 2016; Kim et al., 2014), and another study demonstrated a trend towards the relationship between loss of Ep-CAM expression and distant metastasis (Lugli et al., 2010). A study of 726 CRC patients reported a close correlation between loss of Ep-CAM expression and tumor budding (Kim et al., 2016). Therefore, we integrated the eligible studies into a large population, and the results suggested a close association of the loss of Ep-CAM expression with tumor differentiation, tumor stage, vascular invasion, lymph node metastasis, distant metastasis, and tumor budding. We also found that loss of Ep-CAM expression exhibited a positive trend with depth of tumor invasion (OR = 1.38, 95% CI = 0.96–1.97, P = 0.078). These analyses indicate that loss of Ep-CAM expression plays a crucial role in CRC progression, metastasis, and relapse.

Loss of Ep-CAM expression with multivariate analysis was a poor prognostic factor in disease-specific survival (DSS) (Goossens-Beumer et al., 2014), 5-year overall survival (OS) (Goossens-Beumer et al., 2014), and disease-free survival (DFS) (Kim et al., 2016), which suggests that loss of Ep-CAM expression may be a potential prognostic biomarker.

Some studies demonstrated the use of CSCs as a marker for the detection and diagnosis of cancer (Takaishi et al., 2008; Yang et al., 2008; Nagrath et al., 2007). We evaluated whether Ep-CAM expression was a useful diagnostic biomarker for CRC. A comparison of CRC and normal controls demonstrated combined sensitivity, specificity and AUC values of Ep-CAM expression of 0.93, 0.90, and 0.94, respectively, which was very good (all values ≥0.9). These results suggest that Ep-CAM expression exhibits good diagnostic capacity as a promising molecular biomarker for CRC diagnosis. The importance of Ep-CAM was validated in an Ep-CAM-based assay, the CellSearch® System, which is the only Food and Drug Administration (FDA)-approved assay for the enrichment and detection of circulating tumor cells (CTCs) (Cohen et al., 2008). We also found that the detection of Ep-CAM expression in the blood exhibited a sensitivity of 96.3% and specificity of 100% (Karanikiotis et al., 2005), which indicates that the detection rate of Ep-CAM-expressing circulating tumor cells may be higher in blood samples and serve as a potential noninvasive marker. Additional large-scale and well-designed prospective studies are essential to further confirm the diagnostic effect of Ep-CAM expression in clinical applications, especially the use of blood samples.

There was evidence of heterogeneity in CRC vs. normal controls in tumor differentiation, clinical stage, and vascular invasion (all P < 0.1). We removed one study (Kuhn et al., 2007) in cancer vs. normal controls, two studies (Lugli et al., 2010) and (Kim et al., 2016) for tumor differentiation, the study (Goossens-Beumer et al., 2014) for tumor stage, and the study (Kim et al., 2016) for vascular invasion. The pooled OR remained significant with no evidence of heterogeneity, which indicated the stability and reliability of our results. The reasons for the heterogeneity were not clear, perhaps because of the use of inappropriate or different conditions in immunohistochemistry (IHC) methods, which may have led to the observed bias.

5. Limitations

This meta-analysis has some limitations. First, our study primarily included only Asian and Caucasian populations, and data on other populations, such as African populations, were lacking. Second, the analysis included only available articles published in English. Publications in other languages were excluded due to incomprehensible information, which may result in selection bias. Third, only two studies with small populations were examined to assess the correlation between CRC and benign lesions. Finally, only one study involving blood samples was analyzed in the present meta-analysis, and more studies with large sample sizes are essential to further validate the diagnostic value of Ep-CAM expression.

6. Conclusions

Our findings suggest that Ep-CAM expression is notably higher in CRC patients than in normal controls. Loss of Ep-CAM expression correlated with tumor differentiation, tumor stage, vascular invasion, depth of tumor invasion, lymph node metastasis, distant metastasis, and tumor budding in CRC. Loss of Ep-CAM expression may be a poor prognostic factor of CRC in DFS, DSS and OS. The use of Ep-CAM expression may be a potential biomarker for the diagnosis of CRC patients in the clinical setting. However, more clinical prospective studies using large populations with CRC are necessary to further confirm the diagnostic and prognostic effects.

Supplementary data to this article can be found online at http://dx.doi.org/10.1016/j.ebiom.2017.05.025.

Authors' Contributions

Susu Han, Fenggang Hou and Wen Li contributed to the conception and design of this research. Susu Han, Shaoqi Zong and Qi Shi contributed to the drafting of the article and final approval of the submitted version. Susu Han, Shaoqi Zong, Qi Shi, Hongjia Li, Shanshan Liu, Wei Yang, Wen Li, and Fenggang Hou contributed to data analyses and interpretation and completion of the figures and tables. All authors read and approved the final manuscript.

Competing Financial Interests

All authors declare they have no competing financial interests.

Ethical Review from Patients

Our study was not primary research involving human samples, but rather a secondary analysis of human subject data published in the public domain.

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