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Review Article

Prognostic Significance of VEGF-C Expression in Patients with Breast Cancer: A Meta-Analysis

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Abstract

Background: Vascular endothelial growth factor (VEGF)-C, as a lymphangiogenic factor, plays important roles in the progression of several malignancies. However, its clinical prognostic value in breast cancer still remains controversial. We performed a meta-analysis of available studies to assess the association between VEGF-C expression and the outcomes of breast cancer patients

Methods: We searched eligible studies in three English databases (MEDLINE, EMBASE, and Web of Science) and two Chinese databases (Wanfang and Chinese National Knowledge Infrastructure databases). Key words used in the research included "VEGF-C", "breast cancer", "immunohistochemistry", "breast neoplasma(s)", "breast carcinoma", "metastasis", and "prognosis". Fourteen studies with a total of 1, 573 breast cancer cases were finally included into the meta-analysis. The pooled odds ratios (ORs) with the corresponding 95% confidence interval (95% CIs) for lymph node metastasis, overall survival, and disease-free survival were calculated by using fixed-effects or random-effects models. Heterogeneity and publication bias were also assessed.

Results: Meta-analysis of random-effects model showed VEGF-C expression was associated with lymph node metastasis in patients with breast cancer (random-effects, OR = 2.14; 95 % CI 1.21–3.77, $P = 0.009$). VEGF-C expression was associated with poorer overall survival (fixed-effects, OR = 2.46, 95% CI: 1.46–4.14, $P < 0.001$) and disease-free survival (fixed-effects, OR = 2.10, 95% CI: 1.32–3.35, $P = 0.002$) in patients with breast cancer.

Conclusion: VEGF-C expression is positively associated with lymph node metastasis in breast cancer, and VEGF-C detection in breast cancer might be an effective and feasible means to predict outcome.

Keywords: Breast cancer, Prognosis, Meta-analysis, Vascular endothelial growth factor-C

Introduction

Breast cancer is the leading cause of cancer mortality in women worldwide and is one of the major contributors to the global health burden (1-3). Mounting clinical data suggest that the development of metastatic spread of the disease is responsible for at least 90% of the cancer-associated mortality, and the survival rate falls from 90% for localized breast cancer to 20% for metastatic breast cancer (4). Lymphatic metastasis is one of the most important pathways of breast cancer systemic metastasis, and is closely related to prognosis and therapy plans for breast cancer patients. Frequently, the initial sites of metastasis are the regional lymph nodes (5, 6), and migration of tumor cells into the lymph nodes is greatly facilitated by lymphangiogenesis, a process that generates new lymphatic vessels from pre-existing lymphatics or lymphatic endothelial progenitors (7-9). Some researchers suggest that the vascular endothelial growth factor-C (VEGF-C)/ vascular endothelial growth factor receptor 3 (VEGFR3) signaling system is the most efficient pathway in regulating lymphangiogenesis. VEGF-C, also called lymphatic vessel growth factor, belongs to the

VEGF family, and involves in tumor lymph-angiogenesis by inducing lymphatic endothelial proliferation and vessel enlargement to facilitate the shedding of tumor cells into the surrounding lymphatic vessels (10, 11). VEGF-C expression has recently been reported to be correlated with lymph node metastasis in breast (12, 13), gastric (14), colorectal (15), lung (16), prostate (17), head and neck (18), and gallbladder cancer (19). However, in breast cancer, the definite role of VEGF-C has not yet been elucidated. Some studies reported that VEGF-C expression correlated with lymph node metastasis and patients' poor survival (13, 20), while some others not (21, 22). To date, insufficient samples and some other factors have resulted in controversial results of different clinical studies.

To derive a more precise estimation of the relationship between VEGF-C expression and clinical outcomes in patients with breast cancer, we performed a meta-analysis of 14 prospective or retrospective cohort studies with a total of 1,573 breast cancer patients.

Methods

Search strategy and selection criteria

A systematic literature search of MEDLINE (1960 through February, 2013), EMBASE (1988 through February, 2013), web of Science (1960 through February, 2013) databases, and two Chinese databases (Wanfang and Chinese National Knowledge Infrastructure databases, 1960 through February, 2013) was conducted by two study investigators (D.C. and B.L.) independently for all relevant articles about the prognostic value of VEGF-C expression in breast cancer patients. Key words used in the research included "VEGF-C", "breast cancer", "immunohistochemistry", "breast neoplasma(s)", "breast carcinoma", "metastasis", and "prognosis". Studies eligible for inclusion in this meta-analysis should meet the following criteria: 1) measures VEGF-C express-ion in the cancer tissue with immunohis-tochemistry (IHC), 2) patients have pathologically confirmed breast cancer, 3) patients provides information on survival time according to VEGF-C expression; 4) patients has a follow up time exceeding 5 years. When an individual author published several articles obtained from the same patient population, only the newest or most complete article was included in the analysis. The exclusion criteria of the meta-analysis were: (a) animal studies; (b) meta-analyses, letters, reviews, meeting abstracts, or editorial comments; (c) studies with duplicate data or incomplete data.

Data abstraction

Two reviewers independently extracted the following data from each study: first author's name, year of publication, type of cohort study, country of origin, total number of cases (N), follow-up time, and numbers of the patients with positive and negative expression of VEGF-C, etc. Any disagreements were resolved by consensus.

Statistical analysis

Review manager 5.0 program (version 5.1.0; The Cochrane Collaboration, Oxford, England) and Stata (Version12.0, Stata Corporation, TX, USA) were used to perform all the statistical analysis. Two models of pooling data for dichotomous outcomes were conducted: the random-effects model and the fixed-effects model. The pooled statistical analysis was calculated using the fixed effects model, but a random-effect model was performed when the *P* value of heterogeneity test was <0.1. The odds ratio and 95% CI were calculated for each study, and the combined OR and 95% CI were calculated for the studies. OR was the proportion of the exposed population in whom disease has developed over the proportion of the unexposed population in whom disease has developed in a case-control study. By convention, OR >1 implies a worse prognosis in VEGF-C positive group. The significance of the pooled OR was determined by the Z test and a *P* value of less than 0.05 was considered significant. In order to assess the between-study heterogeneity, the I^2 statistic to quantify the proportion of the total variation due to heterogeneity was calculated. The potential publication bias was assessed by Begg's funnel plot and Egger's test (23, 24).

Results

The flow chart that displays the study design process is shown in Fig. 1. A total of 152 potentially relevant articles were reviewed, and 138 articles were excluded due to their irrelevance to the current analysis and insufficient data. Therefore, the final meta-analysis was performed on the basis of the remaining 14 studies that met the criteria set forth in the search strategy and study selection. The main features of eligible studies are summarized in Table 1. Among those 14 studies, 12 studies reported data on the lymph node metastasis, and 5 studies reported data on the VEGF-C expression on overall survival and disease-free survival. There were 1,240 cases providing the data on lymph node metastasis, among which 629 cases had metastasis and 611 cases were without metastasis, or 772 cases were with positive VEGF-C expression and 518 cases were negative VEGF-C expression.

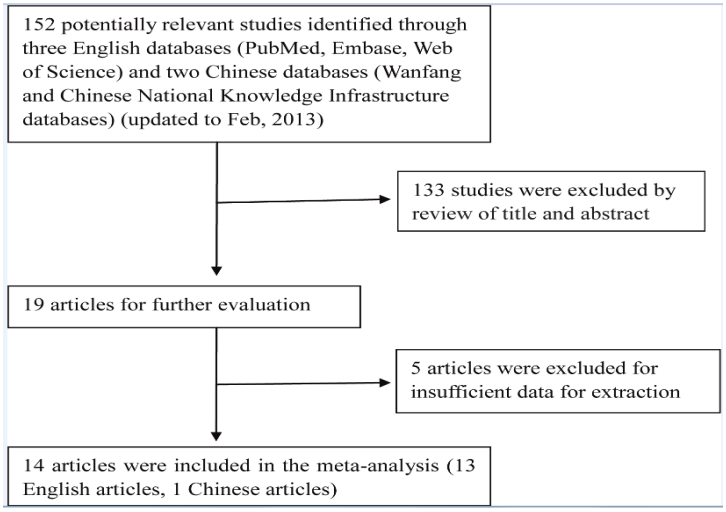

Fig. 1: Flow diagram of the literature search and trial selection process

The cumulative metastasis rate and VEGF-C expression rate of breast cancer were 50.73% (629/1240) and 62.26% (772/1240), respectively. The VEGF-C –positive cases had a cumulative metastasis rate of 56.87% (439/772) that was higher than 36.68% (190/518) in VEGF-C-negative cases. Of those 12 studies on lymph node metastasis, there was high between-study heterogeneity (I^2=75 %). Meta-analysis of random-effects model was adopted, and showed VEGF-C expression was associated with lymph node metastasis in patients with breast cancer (random-effects, OR=2.14; 95 % CI: 1.21–3.77, P<0.009) (Fig. 1). There were 5 studies with a total of 414 breast cancer cases relating the overall survival (OS) and disease-free survival (DFS). Of those 5 studies on OS and DFS, there were low between-study heterogeneities (I^2=10 % for OS; I^2=0 % for DFS). Meta-analysis of fixed-effects model showed VEGF-C expression was associated with poorer OS in patients with breast cancer (fixed-effects, OR = 2.46, 95% CI: 1.46–4.14, P < 0.001) (Fig. 2a). In addition, meta-analysis of fixed-effects model also showed VEGF-C expression was associated with poorer DFS in patients with breast cancer (fixed-effects, OR = 2.10, 95% CI: 1.32–3.35, P = 0.002) (Fig. 2b). We performed the funnel plots and Egger's test to assess the publication bias on studies related to lymph node metastasis. Funnel plot did not reflect obvious asymmetry in this meta-analysis (Figure 3). Also, no indication of publication bias was found (Begg's P value =0.373 and Egger's P value =0.192). Since there are not more than five prognostic studies about OS and DFS, publication bias of the included studies was not performed.

Table 1: Baseline characteristics of the 14 eligible studies in the meta-analysis

Studies	Country	Type of co-hort study	Patient's cohort	Age (range)	VEGF-C expression	Follow-up	Outcomes
Cai XP 2012	China	Prospective	108	48 (32-63)	84/108	NA	NA
Dai XL 2012	China	Prospective	137	50.95 (37-75)	102/154	NA	NA
Hoar FJ 2003	United Kingdom	Prospective	51	60 (30-87)	30/51	NA	NA
Huang JH 2006	China	Prospective	89	54 (37-77)	49/89	NA	NA
Kim BC 2009	Korea	Prospective	128	49 (25-79)	112/128	1-116 months	NA
Kinoshita J 2001	Japan	Prospective	98	55 (30-86)	39/98	3.9 ± 1.3 years	DFS and OS
Li XQ 2012	China	Prospective	60	43.6 (27-66)	47/60	NA	NA
Mylona E 2007	Greece	Prospective	177	56.89 (25-86)	90/177	5-135 months	NA
Okada K 2005	Japan	Prospective	56	NA	35/56	NA	NA
Yavuz S 2005	Turkey	Retrospective	217	50.05 (25-85)	180/217	NA	NA
Zhang XH 2008	China	Prospective	70	49 (30-77)	55/70	68 (28-83) months	DFS and OS
Gisterek I 2007	Poland	Retrospective	98	56 (29-86)	82/98	60 months	DFS and OS
Gu Y 2008	China	Prospective	61	57.59 (29-90)	43/61	60 months	DFS and OS
Watanabe 2005	Japan	Prospective	223	NA	38/87	91.7 months	DFS and OS

NA, non applicable. OS, overall survival, DFS, disease-free survival

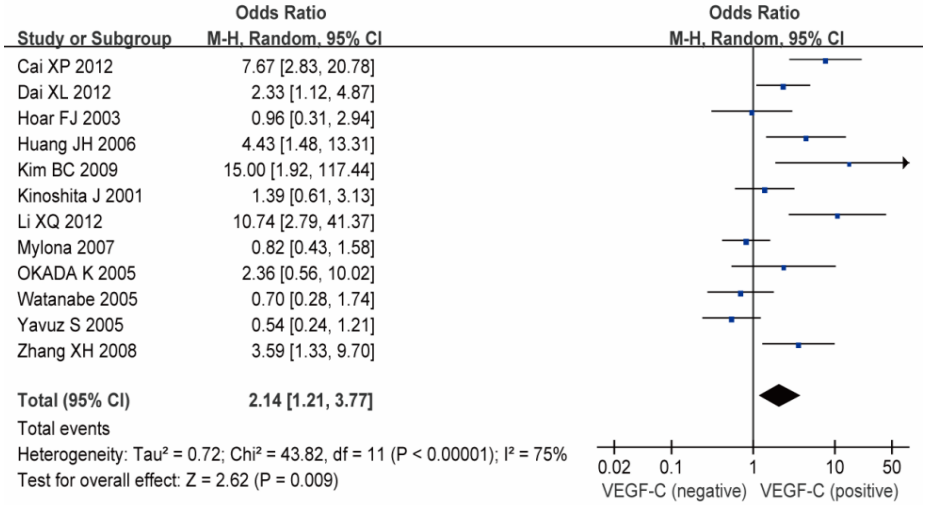

Fig. 2: Forrest plot (random-effects model) of odds ratios (ORs) for the association of VEGF-C expression with lymph node metastasis in patients with breast cancer

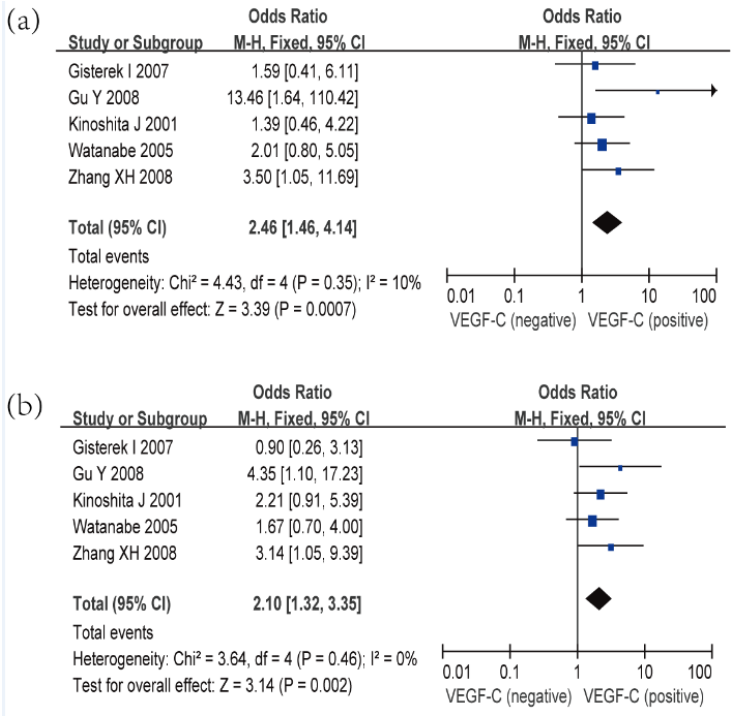

Fig. 3: Forest plot (fixed-effects model) of odds ratios (ORs) for 5 contributing studies assessing the effect of VEGF-C expression on survival in patients with breast cancer. (a) overall survival; (b) disease-free survival

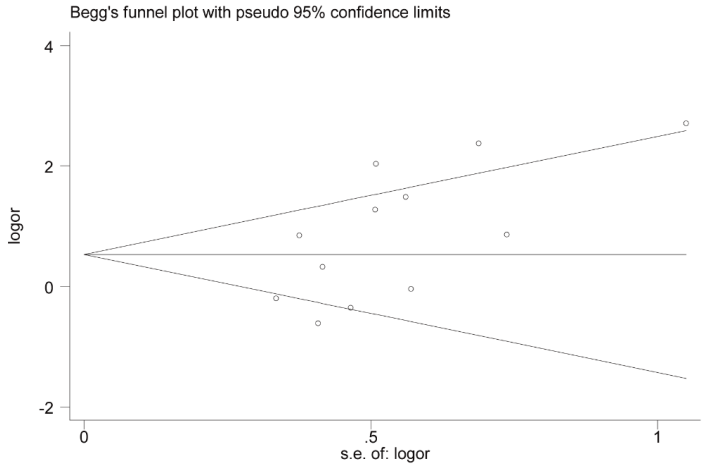

Fig. 4: Begg's funnel plot for publication bias test with pseudo 95% confidence limits for the studies related to lymph node metastasis

Discussion

There are no doubts that the members of VEGF family are important cancer players, thus the field of VEGF research is very promising in the area of cancer research. VEGF-C has been reported to be a lymphatic-specific growth factor, which is the first ligand to be identified for VEGFR-3 (25). Since the expression of VEGFR-3 is predominantly restricted to the lymphatic endothelium in adults. The major function of VEGF-C appears to be the regulation of lymphatic vessel growth (26).

To date, many studies have reported that the overexpression of VEGF-C significantly correlated with lymph node metastasis and lymphangiogenesis in primary tumours, such as thyroid (27), prostate (28), gastric (29), colorectal (30), ovarian (31), and breast cancers (32). However, other reports could not confirm such correlations, or opposite relationships were found (33). At present, there is no consensus on the association between increased VEGF-C expression detected by IHC and poor survival in patients with breast cancer. More accurate evaluation of the impact of VEGF-C expression on patient survival is needed. The findings from our study suggest that VEGF-C expression is associated with the prognosis of patients with breast cancer, and patients with higher VEGF-C expression have poorer survival.

In breast cancer, metastasis occurs primarily through the lymphatic system, and the extent of lymph node involvement is a key prognostic factor for the disease (7, 34). There has been accumulating evidence showing that VEGF-C is the central regulator of lymphangiogenesis (35). As a result of the sample size limitation for the individual studies, broad agreement on the association between VEGF-C expression and lymph node metastasis in breast cancer has not yet been reached. In the present study, our analyses, combining 12 independent studies that included 1,240 cases with breast cancer, revealed that VEGF-C expression is positively associated with lymph node metastasis. These findings might be important for prognosis and treatment of breast cancer, in addition to improve the understanding of biology.

On the other hand, there is significant effect of VEGF-C expression on OS and DFS in the complete patient sample set, including 5 independent studies. Our study suggested that VEGF-C expression was associated with poorer DFS in patients with breast cancer. In addition, VEGF-C expression was also associated with poorer OS in patients with breast cancer. The findings from the present study suggest that VEGF-C expression is associated with the prognosis of breast cancer patients, and patients with higher VEGF-C expression have poorer survival.

Our results should be interpreted cautiously since some limitations exist in this present meta-analysis. First, only published studies were included in the meta-analysis. Therefore, the publication bias may have occurred, even though the use of a statistical test did not show it. Second, the number of included studies was relatively small with only about 1,240 cases. Moreover, other clinical factors such as age and different chemotherapies in each study might lead to bias. Determining whether or not these factors influence the results of this meta-analysis would need further investigation. Third, the effect from our meta-analysis could be overestimated because there are two retrospective cohort studies which had high risk of reporting biases. Therefore, adequately prospective studies with large sample size are required to further assess the precise prognostic effect of VEGF-C expression in breast cancer. Finally, the studies included in this meta-analysis were from different populations, it is possible that demographic factors can confound our results.

Conclusion

Despite the limitations listed above, our study shows a significant correlation between VEGF-C expression and lymph node metastasis in breast cancer patients. Moreover, VEGF-C expression might be a potential prognostic factor for survival in patients with breast cancer, if detected by immunochemistry. In order to become a useful prognostic factor at the level of individual patient and in the context of targeted therapy, these results need to be confirmed by an adequately designed prospective study, and larger clinical trials with widely accepted assessment methods are necessary to define the precise prognostic significance for VEGF-C in breast cancer patients.

Ethical considerations

Ethical issues (Including plagiarism, Informed Consent, misconduct, data fabrication and/or falsification, double publication and/or submission, redundancy, etc) have been completely observed by the authors.

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