The Efficacy of Bevacizumab Compared with Other Targeted Drugs for Patients with Advanced NSCLC: A Meta-Analysis from 30 Randomized Controlled Clinical Trials

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Abstract

Background: The extent of the benefit of bevacizumab combined with chemotherapy in the treatment of advanced non-small-cell lung cancer (NSCLC) is still unclear. We performed this meta-analysis to compare the efficacy of bevacizumab with other commonly used targeted drugs for different patients with advanced NSCLC.

Methods: We searched PubMed, Cochrane Library, EMBASE and abstracts from the proceedings of the American Society of Clinical Oncology (ASCO), and identified 30 randomized controlled clinical trials published within 1999 to 2011 for meta-analysis.

Results: The outcomes of treatment efficacy included response rate, PFS and OS. Comparing bevacizumab (15 mg/kg) with chemotherapy to standard chemotherapy alone, for chemotherapy-naïve patients, the pooled OR of response rate was 2.741 (95%CI: 2.046, 3.672), the pooled HR for disease progression was 0.645 (95%CI: 0.561, 0.743), and the pooled HR for death was 0.790 (95%CI: 0.674, 0.926), respectively. In addition, the adjusted HR for previously-treated patients was 0.680 (95%CI: 0.492, 0.942) comparing bevacizumab combined with chemotherapy to standard chemotherapy alone.

Conclusions: Bevacizumab accompanied by chemotherapy was found to significantly improve patients’ response rate, progression free survival (PFS), and overall survival (OS) among chemotherapy-naïve patients compared to other targeted drugs in the treatment of non-small cell lung carcinoma (NSCLC).

Introduction

Lung cancer has become the most common cancer and the leading cause of cancer death in the world [1,2]. Non-small cell lung cancer accounts for at least 85% in all lung cancer cases [3], presenting as local advanced disease in approximately 25–30% of cases and as metastatic disease in approximately 40–50% of cases [4]. Various epidemiological studies have shown that the 5-year survival rate for patients with NSCLC is extremely low, ranging from 5% to 15% [2]. For NSCLC patients with local advanced or metastatic disease, chemotherapy, radiation and supportive treatment are the principal therapies given the fact that these patients are not able to tolerate surgical operations. However, standard first-line chemotherapy has limited efficacy for NSCLC patients, with an objective response rate about 30%, median survival time 8–9 months and 1-year survival rate 30–40% [3], all of which call for a more effective and safer therapy for lung cancer.

In general, aberrant biological pathways in tumorigenesis result in the disfunction of a protein molecule or a gene fragment, mostly at the molecular level. Accordingly, recent clinical trials have focused on targeted therapies designed to interfere with specific aberrant biological pathways as a new treatment option for NSCLC [6]. Studies, including a recent meta-analysis report, have showed that the use of chemotherapy plus Bevacizumab (at a dose of 15 mg/kg, every 3 weeks) increases two year survival rate for patients diagnosed with advanced lung cancer compared to chemotherapy alone[7,8]. The main agents that have been investigated so far in NSCLC treatment are epidermal growth factor receptor (EGFR) family (tyrosine kinase) inhibitors (gefitinib...
### Table 1. Baseline characteristics of the thirty trials.

| First Author | No. of centers | Jadad Score | EGFR mutation | CT-naive | Asian origin | Group | n | Median age | Female (%) | ECOG > 2 (%) | Stage > IV (%) |
|--------------|----------------|-------------|---------------|----------|--------------|-------|---|------------|------------|--------------|---------------|
| Reck M. (2010) | 150 | 1 | 2 | 1 | No | Yes | No | GP+bev | 351 | 59 | 37.6 | 0 | 84 |
| Sandler A. (2006) | NR | 1 | 0 | 1 | No | Yes | No | PCp+bev | 417 | NR | 50 | 0 | 88 |
| Johnson DH. (2004) | 12 | 2 | 1 | 1 | No | No | No | PCp+bev | 35 | NR | 54.28 | 11.43 | 80 |
| Nishio M. (2009) | NR | 1 | 0 | 1 | No | No | Yes | PCp+bev | 121 | NR | NR | NR | NR |
| Herbst RS. (2007) | 51 | 1 | 0 | 1 | No | Yes | No | D/M+bev | 40 | 63.5 | 42.5 | 0 | NR |
| Herbst RS. (2011) | 177 | 2 | 2 | 1 | No | Yes | No | erl+bev | 319 | 64.8 | 46 | 7 | NR |
| Lynch T.J. (2010) | 96 | 1 | 0 | 1 | No | Yes | No | TC+cet | 325 | 64 | 43 | 0 | 93 |
| Pirker R. (2009) | 155 | 2 | 0 | 1 | Yes | Yes | No | NP+cet | 557 | 59 | 31 | 17 | 94 |
| Rosell R. (2008) | 16 | 1 | 0 | 0 | Yes | Yes | No | NP+cet | 43 | 58 | 23.3 | 93 | 93 |
| Butts CA. (2007) | 32 | 1 | 0 | 1 | No | Yes | No | GP+cet | 65 | 66 | 61.5 | 1.5 | 846 |
| Cappuzzo F. (2010) | 110 | 2 | 1 | 1 | No | Yes | No | erl | 438 | 60 | 27 | 0 | 74 |
| Gatzemeier U. (2007) | 164 | 1 | 1 | 1 | No | Yes | No | GP+erl | 580 | 61 | 22 | <1 | 65 |
| Mok T. (2009) | 19 | 2 | 1 | 1 | No | Yes | Yes | GP+erl | 76 | 57.5 | 29 | 0 | 83 |
| Herbst RS. (2005) | multi | 1 | 1 | 1 | No | No | No | PCp+erl | 539 | 63 | 40.3 | 0 | 844 |
| Lilenbaum R. (2008) | 14 | 1 | 0 | 1 | No | Yes | No | erl | 52 | NR | 56 | NR | NR |
| Shepherd FA. (2005) | 82 | 2 | 1 | 1 | No | Yes | No | erl | 488 | 62 | 35.5 | 25.8 | NR |
| Mitsudomi T. (2010) | 36 | 2 | 0 | 1 | Yes | Yes | Yes | gef | 86 | 64.0 | 68.6 | 0 | 884 |
| First Author          | No. of centers | Jadad Score | EGFR mutation | CT-naive | Asian origin | Group               | n   | Median age | Female (%) | ECOG> 2 (%) | Stage > IV (%) |
|----------------------|----------------|-------------|---------------|----------|--------------|---------------------|-----|------------|------------|-------------|----------------|
| **EGFR mutation**    | **random**     | **blind**   | **dropouts**  |          |              |                     |     |            |            |             |                |
|                      |                |             |               |          |              |                     |     |            |            |             |                |
| Herbst RS. (2004)    | multi          | 1           | 1             | 0        | No           | Pcp+gef             | 345 | 61         | 42.3       | 10.4        | 97.4           |
| Giaccone G. (2004)   | 155            | 1           | 1             | 0        | No           | GP+gef              | 365 | 59         | 23.3       | 9.6         | 98.1           |
| Maemondo M. (2010)   | 43             | 1           | 0             | 1        | Yes          | gef                 | 114 | 63.9       | 63.2       | 0.9         | 86.8           |
| Crino L. (2008)      | 41             | 1           | 0             | 1        | No           | gef                 | 97  | 74         | 22.7       | 23.7*        | NR             |
| Goss G. (2009)       | 37             | 1           | 2             | 1        | No           | gef                 | 100 | 74         | 39.0       | 100         | NR             |
| Takeda K. (2010)     | 39             | 2           | 0             | 1        | No           | gef                 | 300 | 62         | 36*        | 0           | 81.7           |
| Gaafar RM. (2011)    | 24             | 2           | 1             | 1        | No           | gef                 | 86  | 61         | 22         | 7           | 100            |
| More`re JF. (2010)   | 29             | 1           | 0             | 1        | No           | gef                 | 43  | 70         | 12*        | 100         | 100            |
| Kim ES. (2008)       | 149            | 2           | 0             | 1        | No           | gef                 | 733 | 61         | 36.4       | 11.7        | 77.9           |
| Thatcher N. (2005)   | 210            | 2           | 2             | 1        | No           | gef                 | 1129| 62         | 33         | 0           | 81             |
| Cufer T. (2006)      | 25             | 2           | 0             | 1        | No           | gef                 | 68  | 63         | 31         | 36.8*       | NR             |
| Maruyama R. (2008)   | 50             | 1           | 0             | 1        | No           | gef                 | 245 | NR         | 38.4       | 4.5         | 80.8           |
| Lee D.H. (2010)      | 6              | 1           | 0             | 1        | No           | gef                 | 82  | 57         | 32.9*      | 7.3         | 86.6           |

NR: not reported.
*unbalanced between groups.
CT: chemotherapy; bev: bevacizumab; erl: erlotinib; cet: cetuximab; gef: gefitinib.
GP: Cisplatin-Gemcitabine; Pcp: Paclitaxel-carboplatin; TC: Taxane-carboplatin; NP: cisplatin-vinorelbine; D/M:docetaxel/pemetrexed.
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and erlotinib), monoclonal antibodies targeting EGFR (cetuximab), and anti-VEGF monoclonal antibody (bevacizumab).

In different clinical trials, the hazard ratios for PFS and OS of bevacizumab use ranged from 0.55 to 0.85 and from 0.71 to 1.03, respectively [9–14]. In terms of gefitinib use, the ranges of hazard ratios for PFS and OS were from 0.30 to 1.09 and from 0.77 to 1.64, respectively [15–17], which overlapped those of bevacizumab. Similarly, controversial and inefficient results have been reported for other targeted drugs in studies with small sample size and/or different inclusion and exclusion criteria.

In this study we performed an updated meta-analysis to systematically study the efficacy of bevacizumab combined with chemotherapy for advanced NSCLC patients. Our meta-analysis is different from the previous ones in that we target to provide information for future research in comparisons between bevacizumab and other targeted drugs. Information used in the study was obtained from reported and unreported randomized controlled clinical trial studies, and targeted drugs included gefitinib, erlotinib and cetuximab. Our meta-analysis has a higher power in testing efficacy compared to previously reported individual clinical trials, and will help make evidence-based clinical decisions for the treatment of NSCLC.

Materials and Methods

1. Searching method

An electronic search of the PubMed database, the Cochrane Library, and the EMBASE was performed, with the keywords ((non-small-cell lung cancer) OR nsclc) AND (target* therapy). The published language was limited to English and the years were limited from 1999 to 2011. MeSH terms searching was performed in PubMed. The American Society of Clinical Oncology (ASCO) Annual Meeting abstracts were also searched from 2000 to 2011. At the same time, the reference of related systematic reviews and clinical trials were screened.

2. Inclusion Criteria

The relevant clinical trials were manually selected carefully based on the following criteria: (1) randomized controlled trial (RCT); (2) patients with confirmed stage IIIB, stage IV or recurrent NSCLC based on historical or cytological evidence; (3) placebo-controlled or other types of superiority trial as well as non-inferiority trial; (4) Information collected including response rate, hazard ratio for progression free survival and overall survival, along with their 95% CIs or relevant data.

When searched references referred to same studies, the most recently published papers were chosen.

3. Efficacy indicators

Objective response rate (ORR) is defined as the proportion of complete response (CR) plus partial response (PR) among evaluable patients. Progression free survival (PFS) is defined as the duration of time from random assignment to documented disease progression or death, whichever occurs first. Overall survival (OS) is defined as the time from random assignment to death, irrespective of the cause of death. For patients with no event observed, the time to censor refers to the time to last follow-up. The treatment efficacy of targeted drug compared to alternative drugs was measured by odds ratio for response rate (OR ORR), and
hazard ratio for progression free survival and overall survival (HRPFS or HR OS).

4. Quality assessment
The methodological quality of trials was evaluated using the Jadad scale [a 5-point scale assessing randomization (0–2 points), double-blinding (0–2 points), and follow-up (0–1 points)] [18]. The Jadad scale has a total range from 0 to 5, and clinical trials are defined as ‘good’ when the scale is 3–5 [18]. Two reviewers independently assessed trial quality, and disagreements were resolved by consensus.

5. Data extraction
Two investigators searched the publications independently using standardized data-abstraction forms. When the two investigators discovered different results, an independent expert in oncology made the final decision of study conclusions. Information collected from these publications included first author, year of publication, targeted treatment, chemotherapy regimens, number of centers, number of patients, patient characteristics, study design (blinded or not), and the outcomes. Outcomes collected from these studies included response rate, median PFS and OS, hazard ratios for PFS and OS (HRPFS or HR OS) and their 95% confidence intervals (CIs), and adverse events. In addition, patient character-
istics collected from these studies included median age, the percentage of female, percentage of stage IV patients, ECOG performance status, and whether EGFR expression as entry criteria.

When HRs were not reported in collected papers, we computed HRs and its confidence intervals assuming an exponential distribution of the survival curve. In the estimation of HRs, we applied the published methodology [19] on the graphic software package Engauge to estimate the logarithm transformed HR and variance from the Kaplan–Meier curves.

6. Statistical analysis
Analyses were performed in intention-to-treat (ITT) population. We first tested the statistical heterogeneity between trials (meaningful differences between studies) using the chi-squared Q-test based on the fixed-effect model. The clinical trials were considered heterogeneous when the P value of the chi-squared Q-test was less than 0.10, or when I² was greater than 50%. When the analyses showed heterogeneity between different clinical trials, a random effect model was applied to accommodate the heterogeneity [20]. The pooled odds ratios for response rate (OR_{ORR}), HRs for PFS and OS (HR_{PFS} or HR_{OS}) were calculated. We decided to present three primary measures to show the treatment effect from different angles because PFS and OS can better describe the efficacy of a targeted drug than response rate. In addition, it is not uncommon to detect discrepancy between a clear benefit in PFS and a vague benefit in OS for lung cancer patients [21–23]. Furthermore, we estimated and tested the difference of treatment effect between bevacizumab combined with chemotherapy and other targeted drugs using the meta-regression model. The crude and risk-adjusted 95% confidence interval were reported when the models included/excluded patient characteristics. To demonstrate whether the progression free survival was associated with stable disease (SD) or objective response rate (ORR) to the medication, or both, we performed the additional analysis of logarithm transformed outcomes (HR_{PFS}) against use of bevacizumab and ORR controlling for patient characteristics (median age, mean ECOG performance score) and study design (chemotherapy type for the

Figure 3. Response rate, PFS, OS of Bevacizumab versus Gefitinib in NSCLC patients with different EGFR status.
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control group). Similarly, logarithm transformed HR_{OS} was modeled against HR_{PFS} and bevacizumab.

In addition to the above tests, we performed imputation study to test the influence of each individual study using the leave-one-out strategy [20]. Finally, we performed the funnel plot as well as Begg's and Egger's tests to examine potential publication bias.

We performed subgroup analysis in this study based on patient treatment status using the meta-regression models. Chemotherapy-naive patients were defined as those with no prior chemotherapy and no previous treatment with EGFR-targeted drugs or monoclonal antibodies. Previously-treated patients were defined as patients progressed or recurred after at least one previous chemotherapy regimen.

All the analyses were performed using STATA 11.0.

The study was written according to the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) statement [24].

**Results**

The flowchart of our study is shown in Figure 1. From 1,329 published papers and abstract that we found, 967 were excluded from this study based on our inclusion/exclusion criteria. In addition, 309 articles were further excluded if they were already review papers or comments. Among the 53 articles that were left from the above exclusion criteria, five articles were excluded since they were duplicate reports. Finally, 15 additional articles were excluded since they did not report outcomes relevant to our study. Our final sample included 15,650 patients collected from 30 randomized clinical trials.

Among the 30 multi-center randomized clinical trials [9–17,25–45] we included in this study, 13 were double-blinded trials. All of these studies were published in peer-reviewed journals except one that published as an abstract in ASCO annual meeting. Six of the clinical trials applied bevacizumab 15 mg/kg every 3 weeks combined with targeted treatment, four of them applied cetux-
Figure 5. Results of meta-regression. A: ln(HR PFS) – ln(OR ORR), in chemotherapy-naïve patients; B: ln(HR PFS) – ln(OR ORR), in previously-treated patients; C: ln(HR OS) – ln(HR PFS), in chemotherapy-naïve patients; D: ln(HR OS) – ln(HR PFS), in previously-treated patients.

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Table 2. Crude and risk-adjusted hazard ratio of BEV comparing to C/E/G.

| patients     | Response variable | Treatment group | Number of trials | Crude          | Adjusted        |
|--------------|-------------------|-----------------|------------------|----------------|-----------------|
|              |                   |                 |                  | HR Crude       | HR Adjusted     |
|              |                   |                 |                  | 95% CI         | 95% CI          |
| Chemotherapy-naïve | HR PFS          | Bev             | 3                | 0.753 (0.570, 0.996) | 0.847* (0.687, 1.043) |
|               |                  | C/E/G           | 18               | 1 -            | 1 -             |
| Previously-treated | HR PFS        | Bev             | 2                | 0.758 (0.482, 1.191) | 0.680* (0.492, 0.942) |
|               |                  | C/E/G           | 6                | 1 -            | 1 -             |
| Chemotherapy-naïve | HR OS            | Bev             | 2                | 0.774 (0.617, 0.972) | 1.151** (0.828, 1.600) |
|               |                  | C/E/G           | 18               | 1 -            | 1 -             |
| Previously-treated | HR OS          | Bev             | 2                | 0.985 (0.658, 1.475) | 1.262** (0.927, 1.710) |
|               |                  | C/E/G           | 6                | 1 -            | 1 -             |

*HR Adjusted was adjusted by ln(OR ORR).  
**HR Adjusted was adjusted by ln(HR PFS).
bevacizumab (400 mg/m², initial dose followed by 250 mg/m² every week), six of them applied erlotinib 150 mg/d, and the other fourteen clinical trials applied gefitinib 250 mg/d (Table 1). The patient level analyses showed that patient median age varied from 58 to 71, percent of female varied from 12% to 69.8%, and 65–100% of patients having cancer stage higher than 3 in different trials. Individual results of included trials were summarized in figure 2.

Among the 30 clinical trials included in the meta-analysis, 25 reported hazard ratios for PFS and OS (HR_PFS and HR_OS) and the corresponding 95% confidence intervals (CIs). For other 5 trials, 3 reported the HR_PFS directly and 2 reported the HR_OS directly. In terms of the efficacy for patients treated with gefitinib (2 trials [15,17] for EGFR-mutated patients among 14 clinical trials), meta-analysis showed that pooled OR_ORR in EGFR-mutated patients was 4.862 (95%CI: 3.064, 7.715; I² = 20.2%; Figure 3) compared to 1.199 (95%CI: 1.003, 1.434; I² = 43.3%) in EGFR untested patients (P < 0.001). Pooled HR_PFS in EGFR-mutated patients (0.379, 95%CI: 0.235, 0.611; I² = 74.2%) was smaller than that in EGFR untested patients (0.896, 95%CI: 0.738, 1.087; I² = 79.1%, P = 0.001). In addition, pooled HR_OS in EGFR-mutated patients was 1.046 (95%CI: 0.509, 2.149; I² = 63.0%), compared to 1.005 (95%CI: 0.924, 1.093; I² = 38.5%) in EGFR untested patients (P = 0.914). Therefore, in the following comparison, we compared bevacizumab with other targeted drugs (gefitinib, erlotinib and cetuximab) in EGFR untested patients. However, in terms of HR_OS, the comparison was made in both EGFR-mutated and EGFR untested patients.

In terms of efficacy for chemotherapy-naive patients, a higher pooled OR_ORR was found in trials applying bevacizumab (2.741, 95%CI: 2.046, 3.672; I² = 0.0%) than those applying other targeted drugs (OR = 1.255, 95%CI: 1.117, 1.410; I² = 48.9%) for chemotherapy-naive patients (P < 0.001, Figure 4). The pooled HR_PFS was found to be lower in trials applying bevacizumab (HR = 0.643, 95%CI: 0.561, 0.743; I² = 0.0%) than those applying other targeted drugs (HR = 0.875, 95%CI: 0.779, 0.982; I² = 78.3%, P = 0.001). In addition, the pooled HR_OS was found to be lower in trials applying bevacizumab (HR = 0.790, 95%CI: 0.674, 0.926; I² = 0.0%) than those applying other targeted drugs (HR = 0.969, 95%CI: 0.889, 1.057; I² = 50.2%, P = 0.027). Analysis for previously-treated patients showed that pooled OR_ORR, HR_PFS, and HR_OS were similar in trials applying bevacizumab versus other targeted drugs. For example, the OR_ORR was 2.008 (95%CI: 1.184, 3.404; I² = 13.8%) and 2.704 (95%CI: 1.349, 5.424; I² = 82.4%) for the two groups, respectively (P = 0.503); pooled HR_PFS was 0.624 (95%CI: 0.524, 0.742; I² = 0.0%) and 0.831 (95%CI: 0.698, 0.989; I² = 79.7%), respectively (P = 0.022). And the pooled HR_OS was 0.936 (95%CI: 0.780, 1.124; I² = 11.6%) and 0.916 (95%CI: 0.799, 1.031; I² = 64.3%), respectively (P = 0.855).

In chemotherapy-naive patient, a meta-regression analysis showed that the overall lnHR_PFS was negatively associated with the lnOR_ORR (β = −0.251, P = 0.001; Figure 5 and Table 2). The subgroup analyses based on patient treatment status showed that the treatment of bevacizumab for previously-treatment patients was statistically different from those of other targeted drugs in terms of disease progression (P = 0.027). For HR_OS, we found similar results for both chemotherapy-naive patients and previously-treated patients (β = 0.374, P = 0.009; and β = 0.685, P = 0.020, Figure 5 and Table 2). Trials applying bevacizumab were marked in red and grey shaded areas with the confidence band for the regression line. The size of the circles represented the weight of each trial in the regression procedure.

The Begg’s funnel tests were conducted to demonstrate the influence of publication bias (figure 6). The p-values were 0.301, 0.159 and 0.851, respectively.

Discussion

Our meta-analyses showed that compared to other commonly used targeted drugs, chemotherapy with bevacizumab significantly improved patients’ response rate, PFS and OS. The above findings were similar to previous findings [46]. In addition, bevacizumab provided significantly higher OR ORR, lower HR_PFS, and lower HR_OS among chemotherapy-naive patients, and lower HR_PFS among previous treated patients. It was also found that in EGFR-mutated patients, gefitinib significantly improved OR ORR and reduces HR_PFS. However, in general patients with EGFR status untested, bevacizumab showed a clear benefit in OR ORR, HR_PFS, as well as HR_OS, compared with gefitinib. These findings were consistent with previous publications [30].

Generally, mechanism of action of anticancer drugs was causing cancer cell death or blocking cancer cell growth. Objective response rate (ORR), which refers to the proportion of CR+PR, reflects the treatment effect by causing cancer cell death. On the other hand, SD reflects the treatment effect by blocking cancer cell death or blocking cancer cell growth.
growth. Our meta-regression models were performed to decompose the two treatment mechanisms among NSCLC patients by introducing ln(OR ORR) together with the bevacizumab indicator into the model. In these models we identified differences between the two types of targeted drugs in the contribution of blocking cell growth by estimating the adjusted bevacizumab effect, controlling the effect on contribution of killing tumor cells (OR ORR).

From the results (table 2), we found that in previously-treated patients, although bevacizumab was not outstanding in promoting beneficial events such as CR and PR, it surpassed other targeted drugs in maintaining the pharmacodynamic effect. This finding was consistent with the mechanism of bevacizumab which was slowing down the vessel growth instead of causing cell death. As we can see in figure 5, several trials with treatment group applying bevacizumab (marked in red) fell below the regression line, indicating that there are other factors contributing to the prolongation of PFS in spite of the elevation of ORR. The contribution of SD in PFS time is greater in the treatment group than in the control group.

We presented three primary measures (ORR, PFS and OS) to show the treatment effect of different targeted drugs. Response rate is greatly affected by the original volume of the solid tumor, average duration of administration, and the clinical stage of patients, while PFS and OS time can be greatly affected by the frequency of follow-up. These are possible reasons of having only one clinical trial (E4599) with significant overall survival benefit. Another possible reason of the negative findings in overall survival was not able to detect for patients in second-line or third-line treatment, which suggests that patients may be more likely to show the advantage in chemotherapy-naïve patients was mainly attributed to the elevation of ORR and prolongation of PFS. In addition, compared with other targeted drugs mentioned, chemotherapy with bevacizumab significantly improved patients’ response rate, PFS and OS, especially for chemotherapy-naïve patients.

Selection of target is essential in targeted therapies; therefore whether EGFR is mutated or not is of great significance in clinical decision. However, a considerable number of patients are unable to provide adequate tissue samples for accurate genotyping in practice. Our study showed that the benefit from bevacizumab was independent of EGFR status among a relatively large number of patients especially for those of first-line treatment. Such an effect was not able to detect for patients in second-line or third-line treatment, which suggests that patients may be more likely to show better response to the anti-angiogenic drug at early stage. Based on these findings, we would recommend early use of bevacizumab.

Limitations exist in this study. First, our meta-analysis cohort is heterogeneous regarding chemotherapies of the controls, and this may lead to unreliable findings. To address this issue, we performed an imputation study with leave-one-out strategy. The imputation analysis showed that the results had only slight difference when any single trial was removed from the meta-analysis, which indicates robustness of our study. Secondly, our analysis included a number of steps to minimize the potential for publication bias, including the Begg's test and Egger's test. The symmetrical distributions presented in Funnel plot showed a small number of outliers, which may result from the limit of published language. Third, with limited data information, our study was not able to control for heterogeneity of EGFR status in testing the treatment effect of different medications. However, literature shows that bevacizumab is an anti-VEGF mAb with a high affinity for VEGF [47]; therefore the treatment effect would not differ from the EGFR status of patients. In addition, when gefitinib was used, patients with EGFR mutated were found to have better treatment effects than those with unknown EGFR status (compared of both patients with EGFR mutation and those without EGFR mutation) [15,34]. Given the fact that we found better treatment effect of bevacizumab comparing to gefitinib for patients with unknown EGFR status, we believe bevacizumab should show better treatment effect than gefitinib for patients without EGFR mutation.

Our study included clinical trials with only slightly different enrollment criteria and patient demographics. However patient characteristics (age, gender, ECOG performance status) were found not to be balanced between groups in a small number of trials. Such patient level difference may lead to heterogeneity in the meta-analysis. We carefully included aggregated patient characteristics into our meta regression level to control for heterogeneity in our study. Inconsistency of chemotherapies of the control group did exist in this analysis, which could not be eliminated due to the study background. Further analysis with Bayesian method might solve this problem [48].

Finally, the clinical trials collected in this study show high heterogeneity. Due to the relative small sample size, our analysis may not be considered as strong evidence of treatment effect as other meta-analysis although we controlled for patient characteristics as well as study design. A large RCT(s) or individual-patient data meta-analysis may be needed in the future to further examine the treatment difference.

In conclusion, we found from this meta-analysis study that for chemotherapy-naïve patients, the advantage of bevacizumab in HR OS is mainly due to the elevation of ORR and prolongation of PFS. In addition, compared with other targeted drugs mentioned, chemotherapy with bevacizumab significantly improved patients’ response rate, PFS and OS, especially for chemotherapy-naïve patients.

**Supporting Information**

**Table S1 PRISMA Checklist.**

(DOC)

**Author Contributions**

Conceived and designed the experiments: JLC NQZ. Performed the experiments: JLC MZ. Analyzed the data: JLC NQZ. Contributed reagents/materials/analysis tools: NQZ. Wrote the paper: JLC TSL XYC.

**References**

1. Jemal A, Siegel R, Ward E, Murray T, Xu JQ, et al. (2006) Cancer statistics. CA Cancer J Clin 56:106–30.
2. Boyle P, Levin B, editors. (2008) World cancer report 2008. Lyon: IARC.
3. National Comprehensive Cancer Network. Clinical Practice Guidelines in Oncology: Non-Small Cell Lung Cancer v.2.2009. Available from: http://www.nccn.org/professionals/physician_gls/pdf/lsclg.pdf. Accessed 2012 Sep 18.
4. Novello S, Le Chevalier T. (2003) Chemotherapy for non-small-cell lung cancer. Part 1: Early-stage disease. Oncol (Williston Park) 17(3): 357–364.

5. Xue D, Zhou YB. (2008) Advancement in research of molecular target treatment in NSCLC. Int J Intern Med 33(5): 419–425.

6. Grulich C. (2009) Targeted therapies and non-small-cell lung cancer: work in progress? Current Opinion in Oncology 18:132–134.

7. De Maio E, Tibaldi C, D’Incecco A, Bursi S, Barbara C, et al. (2010) Consequences of targeted treatments for second-line therapy. Annals of Oncology 21(7): 1249–1250.

8. Yang K, Wang YJ, Chen XR, Chen HN. (2010) Effectiveness and safety of bevacizumab for unresectable non-small-cell lung cancer: a meta-analysis. Clin Drug Investig 30(4): 229–41.

9. Nishino M, Hara T, Komishol K, Ichinose Y, Nishiwaki Y, et al. Randomized, open-label, multicenter phase II study of bevacizumab in combination with carboplatin and paclitaxel in chemotherapy-naïve Japanese patients with advanced or recurrent nonresectable non-small lung cancer (NSCLC). J Thorac Oncol. 2009; available from: http://meeting.ascopubs.org/cgi/content/abstract/27/11S/8036 Accessed 2012 May 26.

10. Herbst RS, Ansari R, Budin F, Flynn H, Hart L, et al. (2011) Efficacy of bevacizumab plus erlotinib versus erlotinib alone in advanced non-small-cell lung cancer after failure of standard first-line chemotherapy. [British Journal of Cancer] A double-blind, placebo-controlled, phase 3 trial. Lancet 377: 1846–1854.

11. Johnson DH, Fehrenbacher L, Novotny WF, Herbst RS, Nemunaitis JJ, et al. (2004) Randomized Phase II Trial Comparing Bevacizumab Plus Carboplatin and Paclitaxel With Carboplatin and Paclitaxel Alone in Previously Untreated Locally Advanced or Metastatic Non-Small-Cell Lung Cancer. J Clin Oncol 22:2184–2191.

12. Mitsudomi T, Morita S, Yatabe Y, Negoro S, Okamoto I, et al. (2010) Gefitinib and Paclitaxel With Carboplatin and Paclitaxel Alone in Previously Untreated Advanced Non–Small-Cell Lung Cancer: A Phase III Trial. J Clin Oncol 28:1316–1324.

13. Herbst RS, Ansari R, Bustin F, Flynn P, Hart L, et al. (2011) Gefitinib plus best supportive care in previously treated patients with refractory advanced non-small-cell lung cancer: results from a randomised, placebo-controlled, multicentre study [Resa Survival Evaluation in Lung Cancer]. Lancet 366: 1935–1943.

14. Kim ES, Hirsh V, Mok T, Socinski MA, Gervais R, et al. (2006) Gefitinib versus docetaxel in previously treated non-small-cell lung cancer (INTEREST): a randomised phase III trial. Lancet 368: 901–908.

15. Higashiyama T, Nishiwaki Y, Takahashi H, Uemura M, Nishio T, et al. (2007) Randomized Phase II Study of Gefitinib Compared With Placebo in Chemotherapy-Naïve Patients With Advanced Non-Small-Cell Lung Cancer and Poor Performance Status. J Clin Oncol 25(21): 2159–2165.

16. Rosell R, Robinet G, Szczesna A, Ramlau R, Constenla M, et al. (2008) Randomized Phase II Study of Bevacizumab Versus Placebo in Patients With Advanced Non-Small-Cell Lung Cancer (INVITAE): A Randomized, Phase II Study. J Clin Oncol 26(24): 4235–4260.

17. Tashcher N, Chang A, Parikh P, Pereira JR, Cieana T, et al. (2009) Gefitinib plus best supportive care in previously treated patients with refractory advanced non-small-cell lung cancer: results from a randomised, placebo-controlled, multicentre study [Resa Survival Evaluation in Lung Cancer]. Lancet 366: 1935–1943.

18. Herbst RS, O’Neill VJ, Fehrenbacher L, Belani CP, Bonomi PD, et al. (2007) Randomized Phase III Trial Comparing Bevacizumab Plus Carboplatin and Paclitaxel Alone in Previously Untreated Advanced Non–Small-Cell Lung Cancer. J Clin Oncol 25(30): 4743–50.

19. Kudchadkar RA, Shaha AR, Mijalsky E, Gralow J, Dickler M, et al. (2009) Randomized Phase II Study of Epirubicin Plus Paclitaxel Plus Bevacizumab in Metastatic Non-Small Cell Lung Cancer: Results of the Randomized Multicenter Phase III Trial. J Clin Oncol 27(30): 5080–5087.

20. Caballero P, Rege SE, Carmona J, Taibeb ME, van der Zee J, et al. (2010) Randomized, Phase II Study of Bevacizumab Plus Carboplatin and Paclitaxel Versus Paclitaxel Alone in Advanced Non-Small-Cell Lung Cancer. J Clin Oncol 28(18): 2715–2723.

21. Mok TS, Wu YL, Yu CJ, Zhou C, Chen YM, et al. (2009) Randomized, Placebo-Controlled, Phase II Study of Sequential Erlotinib and Chemotherapy As First-Line Treatment for Advanced Non-Small-Cell Lung Cancer. J Clin Oncol 27(30): 5080–5087.

22. Goss, G., Ferry, D., Wierzbicki, R., Laurie, S., Thompson, J., et al. (2009) Randomized Phase II Study of Gefitinib Compared With Placebo in Chemotherapy-Naïve Patients With Advanced Non-Small-Cell Lung Cancer and Poor Performance Status. J Clin Oncol 27(15): 2253–2260.

23. Conroy T, Desmedt CE, Weil J, Mailliard B, Greil R, et al. (2008) Randomized Phase II Trial of Cetuximab Plus Best Supportive Care in Previously Untreated Locally Advanced or Metastatic Non-Small-Cell Lung Cancer. J Clin Oncol 26(24): 4235–4260.

24. Goss, G., Ferry, D., Wierzbicki, R., Laurie, S., Thompson, J., et al. (2009) Randomized Phase II Study of Gefitinib Compared With Placebo in Chemotherapy-Naïve Patients With Advanced Non-Small-Cell Lung Cancer and Poor Performance Status. J Clin Oncol 27(15): 2253–2260.

25. Herbst RS, Giaccone G, Scagliotti G, Rosell R, et al. (2004) Randomized Phase II Trial of Paclitaxel Plus Bevacizumab Versus Paclitaxel Alone in Advanced Non-Small-Cell Lung Cancer. J Clin Oncol 22(16): 3697–705.

26. Carbone DP, de Marinis F, Moinfar F, Naunheim KS, Wheatley K, et al. (1995) Randomized, placebo-controlled, phase III intergroup study of cisplatin plus vindesine versus cisplatin alone in advanced non–small-cell lung cancer: the West Japan Thoracic Oncology Group (WJTOG) Randomized Phase II Study of Cisplatin Plus Cisplatin and Paclitaxel Chemotherapy in Advanced Non–Small-Cell Lung Cancer. J Clin Oncol 23(12): 2542–50.

27. Novello S, Le Chevalier T. (2003) Chemotherapy for non-small-cell lung cancer. Part 1: Advanced disease. Oncology 17(3): 357–364.

28. Novello S, Le Chevalier T. (2003) Chemotherapy for non-small-cell lung cancer. Part 1: Early-stage disease. Oncol (Williston Park) 17(3): 357–364.

29. Novello S, Le Chevalier T. (2003) Chemotherapy for non-small-cell lung cancer. Part 1: Early-stage disease. Oncol (Williston Park) 17(3): 357–364.