The pemetrexed-containing treatments in the
non-small cell lung cancer, is -/low thymidylate
synthase expression better than +/-high
thymidylate synthase expression: a meta-analysis

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Abstract

Background: The predictive value of thymidylate synthase (TS) for clinical sensitivity to pemetrexed-containing chemotherapy in patients with non-small cell lung cancer (NSCLC) remains controversial. This meta-analysis is performed to provide an assessment of whether expression variations of TS are associated with objective response in patients with NSCLC treated with pemetrexed-containing chemotherapy.

Methods: An electronic search was conducted using the databases MEDLINE, EMBASE and CNKI, from inception to June 10th, 2013. A systemic review of the studies on the association between TS expression in NSCLC and objective response of pemetrexed-containing regimen was performed. Pooled odds ratios (OR) for the response rate were calculated using the software Revman 5.0.

Results: There were a total of 526 patients in the eight studies that met our criteria for evaluation. +/-high expression of TS was found in 269 patients (51.1%), and -/low expression for this gene was found in 257 (48.9%) patients. The objective response rate for pemetrexed-containing chemotherapy was significantly higher in patients with -/low expression TS expression (OR = 0.45; 95% CI, 0.29–0.70; p = 0.0004). Although patients with -/low expression of TS have a longer median overall survival time and progression free survival time than those with +/-high expression of TS, the difference was not statistically significant.

Conclusions: -/low expression of TS was associated with higher objective response in NSCLC patients treated with pemetrexed-containing chemotherapy. TS may be a suitable marker of sensitivity to pemetrexed-based chemotherapy in patients with NSCLC.

Keywords: Thymidylate synthase, Pemetrexed, Lung cancer, Meta-analysis

Background

Lung cancer is the most-common cause of cancer-related mortality worldwide and non-small cell Lung cancer (NSCLC) accounts for more than 85% of primary lung cancers and approximately two-thirds of NSCLC patients are diagnosed at an advanced stage [1-3]. Platinum-based chemotherapy is appropriate for selected patients who have a good performance status [4,5]. But the approach of treating patients with a platinum-containing regimen may have reached a plateau in terms of efficacy [6]. Most patients receiving front-line chemotherapy may experience disease progression and need second-line therapy [7]. One of several treatments for NSCLC as the second line therapy is pemetrexed, which is increasing its therapeutic scope from second-line therapy to first-line and maintenance therapy [8-10].

Pemetrexed is a multitargeted antifolate agent, inhibiting at least three of the enzymes involved in DNA synthesis and folate metabolism: thymidylate synthase (TS), dihydrofolate reductase (DHFR) and glycinamide

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Thymidylate synthase expression in NSCLC has attracted considerable attention because of its potential role as a promising predictor for response to pemetrexed-based chemotherapy.

A number of studies have explored the relationship between thymidylate synthase expression and overall response rate in NSCLC patients, but clinical data about TS expression and its predictive value in NSCLC patients receiving pemetrexed-containing chemotherapy are still inconclusive. There are published reports supporting that significantly higher response rates were associated with TS-negativity compared with TS-positivity in patients with NSCLC especially in those with nonsquamous NSCLC treated with pemetrexed-based Chemotherapy [3,17,18]. However, there are also reports of studies failed to find such an association [6,19-22]. Recently, several studies have demonstrated that high TS expression may be useful to predict survival after complete resection in p-stage I adenocarcinoma of the lung [23-25]. Whereas Zheng and colleagues found that patients with high TS expression actually had significantly increased overall survival (OS) when compared to patients with low expression [26]. To determine whether TS expression is associated with objective response in NSCLC patients treated with pemetrexed-containing therapy, we reviewed published studies and carried out a meta-analysis.

Methods
Search strategy
The search was performed by consulting the electronic database MEDLINE, EMBASE and CNKI f or all relevant papers published from the earliest publication date included in the database onward to June 10th 2013. Searches included the terms TS OR thymidylate synthase and lung cancer. The results were then hand searched for eligible studies. No language restrictions were imposed. The references of retrieved articles were also screened for relevant articles and two authors (L Wang and R Wang) conducted all searches independently.

Eligibility criteria
The following criteria for eligibility among studies were set before collecting articles: (1) utilized pemetrexed-containing chemotherapy for patients with pathologically proven NSCLC, (2) measured TS with immunohistochemistry (IHC) or real-time reverse transcriptase PCR (RT-PCR); (3) presented the data of objective response according to TS status.

Quality assessment
Two investigators (L Wang and J Zhang) independently assessed the quality of each study using the Newcastle–Ottawa Quality Assessment Scale. Discrepancies were resolved by consensus. The Newcastle–Ottawa Quality Assessment Scale involves assessing three categories – patient selection, study comparability and outcome–based on eight items. Stars awarded to high-quality elements are used to compare study quality in a qualitative manner. Four items in the selection category, two items in the comparability and three items in the outcome category; a maximum of two stars can be given for comparability; a study can be awarded one star for each item in these categories. The scoring system was recommended by the Cochrane Non-randomized Studies Methods Working Group [27,28].

Statistical analysis
Two independent reviewers extracted the required information using pre-determined forms. Data on objective response rate were analyzed. The data were entered into the Cochrane Collaboration software (RevMan Version 5.0 for Windows; the Cochrane Collaboration, Oxford, UK) and the Cochran’s test was used to assess the heterogeneity of included studies. For the heterogeneity tests, a P-value below 0.05 was considered to indicate significance. If the test of heterogeneity was significant \((p < 0.05, I^2 > 50\%)\) the random-effect model would be used, otherwise the fixed model would be used. Publication bias was estimated by examining the relationship between the treatment effects and the standard error of the estimate (S.E log OR) using a funnel plot. Several additional sensitivity analyses (chemotherapy regimens and TS measurement methods) were also performed to further detect and evaluate clinical heterogeneity.

Results
Selection of studies
The search strategy identified 613 potentially relevant articles, 178 of which were excluded after the titles were reviewed. A total of 435 studies were included for abstract review after the first exclusion. Among the 435 studies, 56 of them were not relevant to clinical chemotherapy (only illustrating the TS expression and its clinicopathological correlation to NSCLC) and 98 of them were not relevant to NSCLCS (studying TS in small cell lung cancer, neuroendocrine, malignant pleural mesothelioma, thymic tumors, primary colorectal cancer, and so on). There were also 109 review articles in the 435 studies above. So 263 articles were excluded and 172 studies were extracted for full text review after careful
abstract review. We excluded 164 studies due to lack of sufficient information or methods discrepancies. After completing the selection process, data from a total of eight studies involving 526 patients (Figure 1) systematically was analyzed. All of them studied the association between TS expression and response to pemetrexed-containing chemotherapy [6,22-31].

The Newcastle–Ottawa Scale, composing of eight items that assess patient selection, study, comparability and outcome, was used to conduct the quality assessments for the eight studies. This scale has been adopted in other non-randomized studies [28,32,33]. Studies which met five or more of the eight criteria were given higher quality scores. A summary of the studies which scored highly is shown in Table 1. Characteristics of the eligible studies are presented in Table 2. Bias assessment was evaluated by funnel plot analysis shown in Figure 2 and the heterogeneity in the 8 studies was not significant statistically (\( p = 0.23 \)).

**Objective response**

All studies reported data on tumor objective response (Table 2), which included complete and partial tumor responses, stable disease and progression disease. Because no heterogeneity was found across studies (\( \chi^2 = 9.39, p = 0.23; I^2 = 25\% \)), the fixed-effects model was used. Pooled data from these eight studies showed an overall objective response rate of 19.3\% for TS +/high expression (\( n = 269 \)) and 30.0\% for TS -/low expression (\( n = 257 \)). These results indicate a statistically significant favorable clinical outcome for patients with -/low TS expression. The pooled odds ratio from the eight studies was 0.45 (OR = 0.45; 95% CI, 0.29–0.70; \( p = 0.0004 \); Figure 3).

Subgroup analysis was conducted based on chemotherapy regimen. Six studies described clinical results with patients treated with pemetrexed monotherapy or pemetrexed plus gemcitabine and three clinical studies described results of patients treated with pemetrexed plus cisplatin or carboplatin. For the regimen of pemetrexed monotherapy or pemetrexed plus gemcitabine, the overall objective response rate for patients with -/low TS expression was higher than that for patients with +/high TS expression (OR = 0.54, 95\% CI 0.33–0.91, \( p = 0.02 \)). For patients treated with pemetrexed plus cisplatin or carboplatin, a significantly better objective response rate was also observed in patients with -/low TS expression (OR = 0.27, 95\% CI 0.10–0.70, \( p = 0.007 \)). There existed no heterogeneity between two treatment subgroups (\( I^2 = 12\% \)).

**Table 1 Quality of the studies used in the meta-analysis**

| Studies          | Selection (stars) | Comparability (stars) | Outcome (stars) |
|------------------|-------------------|-----------------------|-----------------|
| Bepler et al. [6] | 4                 | 2                     | 3               |
| Chang et al. [30] | 3                 | 2                     | 3               |
| Chen et al. [21]  | 4                 | 2                     | 3               |
| Igawa et al. [31] | 3                 | 2                     | 3               |
| Park et al. [22]  | 3                 | 2                     | 3               |
| Sun et al. [17]   | 4                 | 2                     | 3               |
| Takezawa et al. [18] | 3          | 2                     | 3               |
| Wang et al. [19]  | 3                 | 2                     | 3               |

The Newcastle–Ottawa Quality Assessment Scale is composed of three categories, selection, comparability and outcome. Stars are awarded to high-quality elements and are used to compare study quality in a quantitative manner. There are four items in the Selection category and three items in the Outcome category; a study can be awarded one star for each item in these categories. A maximum of two stars can be given for comparability.
| Study          | All pts | TS detection method | Chemotherapy regimen       | Ethnicity | TS expression | Evaluable for response | Disease stage | ECOGPS | TS high/+ | TS low/- | OR (pts) | Total pts | OR (pts) | Total pts |
|----------------|---------|---------------------|-----------------------------|-----------|---------------|-------------------------|--------------|--------|-----------|----------|----------|-----------|----------|-----------|
| Bepler et al. [6] | 52      | RT-PCR              | pemetrexed + gemcitabine    | Caucasian | 35            | 35                      | I-III        | NR     | 5         | 17       | 7        | 18        |
| Chang et al. [30] | 110     | IHC                 | pemetrexed                  | Asian     | 55            | 52                      | advanced or reoccurrence | 0-4 | 23         | 41       | 4        | 11        |
| Chen et al. [21] | 268     | IHC                 | pemetrexed                  | Asian     | 49            | 42                      | IIIB-IV      | NR     | 3         | 20       | 5        | 22        |
| Park et al. [22] | 98      | IHC                 | pemetrexed                  | Asian     | 98            | 88                      | IIB,IIA-IV   | NR     | 5         | 54       | 5        | 34        |
| Takezawa et al. [18] | 24      | IHC                 | pemetrexed + platinum       | Asian     | 24            | 24                      | IIIB-IV      | 0-1    | 1         | 12       | 6        | 12        |
| Wang et al. [19] | 38      | RT-PCR              | pemetrexed + platinum       | Asian     | 38            | 38                      | IIIB-IV      | 0-1    | 2         | 10       | 11       | 28        |
| Igawa et al. [31] | 104     | IHC                 | pemetrexed                  | Asian     | 54            | 54                      | IIIB-IV or reoccurrence | 0-3 | 0          | 23       | 5        | 31        |
| Sun et al. [17]  | 285     | IHC                 | pemetrexed                  | Asian     | 149           | 149                     | IIIB-IV      | 0-1    | 9         | 75       | 21       | 74        |
|                |         |                     | pemetrexed + platinum       | Asian     | 44            | 44                      | IIIB-IV      | 0-1    | 4         | 17       | 13       | 27        |

Abbreviations: TS thymidylate synthase, NR no report, IHC immunohistochemistry, RT-PCR real-time reverse transcriptase PCR, NSCLC non-small cell lung cancer, PTS patients.
(Figure 4). Although the objective response rate difference did not reach statistical significance between patients treated with platinum-free and platinum containing subgroup, −/low TS expression patients treated with pemetrexed plus cisplatin or carboplatin might be more likely to achieve complete or partial response. (Figure 4).

Immunohistochemistry (IHC) was used in 6 studies and real-time reverse transcriptase PCR (RT-PCR) was used to detect TS in 2 studies and. In IHC subgroup, objective response rate in TS −/low expression patients was significantly higher than that in TS +/high expression patients (OR = 0.44; 95% CI, 0.27–0.71; \( p = 0.0009 \)). In Real-time reverse transcriptase PCR subgroup, there was a trend that TS −/low predicted better objective response rate, but the difference did not reached statistical significance (OR = 0.52; 95% CI,0.18–1.54 \( p = 0.24 \)). We noted no evidence of heterogeneity between TS detection method subgroups in this meta-analysis (Chi\(^2\) = 8.33, \( p = 0.22 \); \( I^2 = 28\% \)) (Figure 5).

Median survival time and time to progression
Median survival data were available in five of 8 studies on the association between TS expression and response to pemetrexed-containing chemotherapy [6,17,21,30,31]. Patients with +/high expression of TS had a median overall survival(OS) time of 14.4 months; patients with −/low expression of TS had a median overall survival time of 19.0 months. Although those with −/low expression of TS have a longer median overall survival time than those with +/high expression of TS, the difference was not statistically significant (\( p = 0.239 \)) (Table 3).

Similar association was also found in five studies that provide sufficient data for progression free survival time (PFS) (7.6 months in patients with −/low expression of TS Vs 5.8 months in patients with +/high expression of TS, \( p = 0.13 \)) [6,17,21,30,31] (Table 4).

Discussion
In this meta-analysis, we evaluated the effects of expression variations in TS on the objective response rate to
pemetrexed-containing chemotherapy for NSCLC. Our goal was to test the hypothesis that -/low TS expression is associated with better objective response rate. Among the included studies, four of them reported a higher objective response in the TS -/low expression arm compared with the TS +/high expression arm [6,17,18,31]. While the rest four studies reported no statistically significant relationship between TS expression and response to chemotherapy [19,21,22,30]. We found that the objective response rate of patients with -/low TS expression was significantly higher than that in patients with +/high TS expression. We also conducted a trend that TS -/low predicted better median survival time and progression free survival time but without significant difference in patients receiving pemetrexed-containing chemotherapy.

Previous in vitro studies have shown that TS expression correlated with objective response of NSCLC treated with pemetrexed-containing chemotherapy Ozasa et al. [15] documented that the expression level of the TS gene was significantly correlated with the concentration of pemetrexed for 50% cell survival (IC50) in 11 non-small cell lung cancer cell lines, suggesting up-regulation of the expression of the TS gene may have an important role in the acquired resistance to pemetrexed. Wu et al. found down-stream of TS gene may serve as new biomarkers for predicting responsiveness to pemetrexed [13]. Similar results were also reported by Chiappori in small cell lung

| Study or Subgroup | High-positive | Low-negative | Odds Ratio M-H, Fixed 95% CI | Odds Ratio M-H, Fixed 95% CI |
|-------------------|---------------|--------------|-----------------------------|-----------------------------|
| 1.1.1 pemetrexed alone or pemetrexed plus gemcitabine subgroup | | | | |
| Breier et al. 2008 | 5 | 17 | 7 | 8.2% | 0.05 (0.05, 0.48) |
| chang et al. 2010 | 23 | 41 | 4 | 11 | 4.6% | 2.34 (0.67, 8.04) |
| chen et al. 2011 | 3 | 20 | 5 | 22 | 8.0% | 0.00 (0.01, 1.92) |
| igawa et al. 2012 | 0 | 23 | 5 | 19 | 7.0% | 0.10 (0.01, 1.98) |
| park et al. 2000 | 5 | 54 | 5 | 34 | 9.6% | 0.05 (0.01, 0.22) |
| sun et al. 2011 | 9 | 75 | 21 | 74 | 32.0% | 0.34 (0.15, 0.81) |
| Subtotal (95% CI) | 230 | 1 | 190 | 69 | 9.6% | 0.54 (0.33, 0.89) |
| Total events | 45 | 47 | | | | |
| Heterogeneity: Ch² = 4.48, df = 6 (P = 0.26), I² = 23%
| Test for overall effect Z = 2.33, P = 0.02 |

| Study or Subgroup | High-positive | Low-negative | Odds Ratio M-H, Fixed 95% CI | Odds Ratio M-H, Fixed 95% CI |
|-------------------|---------------|--------------|-----------------------------|-----------------------------|
| 1.1.2 pemetrexed plus cisplatin or carboplatin subgroup | | | | |
| sun et al. 2011 | 4 | 17 | 13 | 37 | 12.2% | 0.33 (0.09, 1.38) |
| Taniwaza et al. 2011 | 1 | 12 | 6 | 12 | 6.4% | 0.09 (0.01, 0.94) |
| wang et al. 2010 | 2 | 10 | 11 | 20 | 8.0% | 0.39 (0.07, 2.17) |
| Subtotal (95% CI) | 39 | 67 | 30.6% | 0.27 (0.10, 0.67) |
| Total events | 7 | 30 | | | | |
| Heterogeneity: Ch² = 1.98, df = 2 (P = 0.38), I² = 0%
| Test for overall effect Z = 2.65, P = 0.007 |

| Total (95% CI) | Total events | | |
|----------------|---------------|-----------------------------|-----------------------------|
| 269 | 257 | 100.0% | 0.46 (0.29, 0.72) |

| Study or Subgroup | High-positive | Low-negative | Odds Ratio M-H, Fixed 95% CI | Odds Ratio M-H, Fixed 95% CI |
|-------------------|---------------|--------------|-----------------------------|-----------------------------|
| 1.3 IHC | | | | |
| chang et al. 2010 | 23 | 41 | 4 | 11 | 4.6% | 2.34 (0.67, 8.04) |
| chen et al. 2011 | 3 | 20 | 5 | 22 | 8.0% | 0.00 (0.01, 1.92) |
| igawa et al. 2012 | 0 | 23 | 5 | 19 | 7.0% | 0.10 (0.01, 1.98) |
| park et al. 2000 | 5 | 54 | 5 | 34 | 9.6% | 0.05 (0.01, 0.22) |
| sun et al. 2011 | 13 | 92 | 26 | 101 | 40.6% | 0.32 (0.16, 0.63) |
| Taniwaza et al. 2011 | 1 | 12 | 6 | 12 | 9.2% | 0.09 (0.01, 0.94) |
| Subtotal (95% CI) | 248 | 21 | 241 | 84.2% | 0.44 (0.27, 0.71) |
| Total events | 45 | 59 | | | | |
| Heterogeneity: Ch² = 1.15, df = 6 (P = 0.10), I² = 45%
| Test for overall effect Z = 2.33, P = 0.009 |

| Total (95% CI) | Total events | | |
|----------------|---------------|-----------------------------|-----------------------------|
| 269 | 217 | 100.0% | 0.45 (0.25, 0.81) |

| Study or Subgroup | High-positive | Low-negative | Odds Ratio M-H, Fixed 95% CI | Odds Ratio M-H, Fixed 95% CI |
|-------------------|---------------|--------------|-----------------------------|-----------------------------|
| 1.3 R T PCR | | | | |
| Breier et al. 2008 | 5 | 17 | 7 | 10 | 8.0% | 0.65 (0.16, 2.68) |
| wang et al. 2010 | 2 | 10 | 11 | 20 | 7.7% | 0.39 (0.07, 2.17) |
| Subtotal (95% CI) | 27 | 46 | 15.1% | 0.52 (0.18, 1.54) |
| Total events | 7 | 19 | | | | |
| Heterogeneity: Ch² = 0.22, df = 1 (P = 0.64), I² = 0%
| Test for overall effect Z = 2.18, P = 0.03 |

| Total (95% CI) | Total events | | |
|----------------|---------------|-----------------------------|-----------------------------|
| 269 | 257 | 100.0% | 0.45 (0.25, 0.81) |

Figure 4 Subgroup analysis by chemotherapy regimen. In the subgroup of the patients treated with pemetrexed plus cisplatin or carboplatin, overall objective response rate in TS -/low patients was significantly higher than that in TS +/high patients (OR = 0.27, 95% CI 0.10–0.70, p = 0.007). For the patients treated with pemetrexed monotherapy or pemetrexed plus gemcitabine, the overall objective response rate for patients with -/low TS expression was significantly higher than that for patients with +/high TS expression (OR = 0.54, 95% CI 0.33–0.91, p = 0.02).
cancer cell line [34]. In our meta-analysis, the available data indicated that the quality of response to pemetrexed-containing chemotherapy was significantly higher in patients with -/low TS expression than those with +/high expression of TS, the difference was not statistically significant (p = 0.24).

Table 3 Median overall survival time in the studies

| Study          | Median survival time(months) | Median ratio | p value |
|----------------|------------------------------|--------------|---------|
| T5 +/high      | T5 -/low                     |              |         |
| Bepler et al. [6] | 27.8                         | 27           | 0.97    | 0.272   |
| Chang et al. [30] | 10.7                         | 9.5          | 1.42    | 0.688   |
| Chen et al. [21]  | 19.9                         | 21.4         | 2.14    | 0.09    |
| Sun et al. [17]  | 3.4                          | 4.8          | 1.41    | 0.01    |
| Igawa et al. [31] | 6.6                          | 14.7         | 1.71    | 0.04    |
| Pool           |                              |              |         |
|                | 14.4                         | 19.0         | 1.32(95% CI 0.92-2.72) | 0.24 |

Patients with +/high expression of TS who received pemetrexed-based chemotherapy had a median overall survival time of 14.4 months; patients with -/low expression of TS had a median overall survival of 19.0 weeks. Although patients with -/low expression of TS perform a longer median overall survival time than those with +/high expression of TS, the difference was not statistically significant (p = 0.24).

Table 4 Median progression free survival time in the studies

| Study          | Median progression free survival time(months) | Median ratio | p value |
|----------------|-----------------------------------------------|--------------|---------|
| T5 +/high      | T5 -/low                                     |              |         |
| Bepler et al. [6] | 3.4                          | 4.8          | 1.41    | 0.01    |
| Chang et al. [30] | 2.4                          | 1.85         | 1.85    | 0.407   |
| Chen et al. [21]  | 3.4                          | 4.8          | 1.41    | 0.01    |
| Sun et al. [17]  | 2                            | 4.1          | 2.05    | 0.001   |
| Igawa et al. [31] | 4.8                          | 5.8          | 3.63    | 0.03    |
| Pool           |                              |              |         |
|                | 5.8                          | 7.6          | 1.31(95% CI 0.43-2.19) | 0.13 |

Patients with +/high expression of TS who received pemetrexed-based chemotherapy had a median progression free survival time of 5.8 months; patients with -/low expression of TS had a median progression free survival time of 7.6 months. Although there was a trend that T5 -/low predicted better progression free survival time, the difference did not reach statistical significance (p = 0.13).

Intuitively, although there was a trend that TS -/low predicted better median overall survival time and progression free survival time, the difference did not reach statistical significance [25]. Recent studies have reported that intratumoral TS expression was significantly related to the prognosis in patients with mesothelioma [43], gastric cancer [44] and colorectal cancer [45-47]. For NSCLC, low TS mRNA level was associated with a better PFS in stage I and II patients [48]. In our analysis, most patients available for evaluating median OS and PFS suffered from advanced or recurrent lung cancer and that may be the possible reason why no significant difference was found between patients with -/low and +/high expression.

The study has many limitations. Only eight studies are eligible for our meta-analysis and the sample size analyzed in each group was relatively small. However, this is the first and initial meta-analysis of assessment whether TS expression is associated with objective response in patients with
NSCLC treated with pemetrexed-containing chemotherapy. More published studies will be helpful in clarifying whether this is a true association. There was no observed significant heterogeneity among the included studies ($I^2 = 25\%$), we further explored heterogeneity by conducting subgroup analyses. Although significant heterogeneity was not found either, heterogeneity in HCC subgroup was moderately high at 45%, which was mainly due to the diversity of regimen combinations and different populations. Publication bias is also a possible limitation because studies that report negative results are published less frequently than those reporting positive results or those consistent with prevailing theories [49]. However, we did not find that publication bias significantly influences our result of the meta-analysis.

Conclusion

In conclusion, this meta-analysis provided us with evidence that low TS expression is associated with a higher objective response rate for NSCLC patients treated with pemetrexed-containing chemotherapy. Our results may be useful in matching NSCLC patients with suitable drugs and predict response rate to pemetrexed-containing chemotherapy as well as for the further investigation of random clinical trial on patients receiving platinum-based or pemetrexed-based chemotherapy.

Competing interests

The authors have declared that no competing interests exist.

Authors' contributions

Conception and design: LW, RW and HC; Acquisition of data: all authors; Analysis and interpretation of data: all authors; Manuscript drafting: LW, JZ and HC; Acquisition of data: all authors; and predict response rate to pemetrexed-containing chemotherapy. Our results may be objective response rate for NSCLC patients treated with pemetrexed-containing chemotherapy. Our results may be


due to the diversity of regimen combinations and different populations. Publication bias is also a possible limitation because studies that report negative results are published less frequently than those reporting positive results or those consistent with prevailing theories [49]. However, we did not find that publication bias significantly influences our result of the meta-analysis.

Conclusion

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Authors’ contributions

Conception and design: LW, RW and HC; Acquisition of data: all authors; Analysis and interpretation of data: all authors; Manuscript drafting: LW, JZ and HC; Manuscript revising: all authors; final approval of this version: all authors. All Authors read and approved the final manuscript.

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References

1. Jemal A, Siegel R, Ward E, Hao Y, Xu J, Thun MJ: Cancer statistics, 2009. CA Cancer J Clin 2009, 59(4):225–249.
2. Socinski MA, Schell MJ, Peterson A, Bakri K, Yates S, Gitten R, Unger P, Lee MF, Hsiao YM, Huang CF, Huang YH, Yang WJ, Chan HW, Chang JT: Genetic determinants of pemetrexed responsiveness and nonresponsiveness in non-small cell lung cancer cells. J Thorac Oncol 2008, 3(11):1343–1351.
3. Wu MF, Hsiao YM, Huang CF, Huang YH, Yang WJ, Chan HW, Chang JT, Ho JL: Enzyme inhibition, polyglutamation, and the effect of LY231514 (MTA) on purine biosynthesis. Semin Oncol 1999, 26(2 Suppl 6):62–47.
4. Adjei AA: Pemetrexed (Alimta): a novel multitargeted antifolate agent. Expert Rev Anticancer Ther 2003, 3(2):145–156.
5. Wu MF, Hsiao YM, Huang CF, Huang YH, Yang WJ, Chan HW, Chang JT, Ho JL: Genetic determinants of pemetrexed responsiveness and nonresponsiveness in non-small cell lung cancer cells. J Thorac Oncol 2010, 5(8):1118–1126.
6. Sun JM, Han J, Ahn JS, Park K, Ahn MJ: Phase III trial of pemetrexed versus docetaxel in patients with non-small-cell lung cancer previously treated with chemotherapy. J Clin Oncol 2004, 22(9):1589–1597.
7. Scagliotti GV, Parikh P, von Pawel J, Biesma B, Vansteenkiste J, Manegold C, Saravolatz L, Gatzemeier U, Digumarthy R, Zhang J, Lee JS, Mellermdaard A, Park K, Patil S, Rolksi J, Goksel I, Manegold C, Simons L, Sugarman KP, Gandara D: Phase III study comparing cisplatin plus gemcitabine with cisplatin plus pemetrexed in chemotherapy-naive patients with advanced-stage non-small-cell lung cancer. J Clin Oncol 2008, 26(21):3543–3551.
8. Uramoto H, Onitsuka T, Shimokawa H, Hanagiri T, Yamauchi H, Nagai K, Yamaguchi H, Nishio K, Nakagawa K: Thymidylate synthase expression and outcome and quality of life of advanced non-small cell lung cancer. Br J Cancer 2010, 101(3):494–500.
9. Ozasa H, Oguri T, Uemura T, Miyazaki M, Maeno K, Sato S, Ueda R: Pemetrexed induces both intrinsic and extrinsic apoptosis through ataxia telangiectasia mutated/p53-dependent and -independent pathways. Mol Carcinog 2011, 50:1118–1126.
non-small cell lung carcinoma: the association with treatment efficacy of pemetrexed. Lung Cancer 2011, 74(1):132–138.

22. Park CK, Kim KS, Oh IJ, Tieden IM, Choi YD, Kwak YS, Kim YI, Lim SC, Kim YC. Efficacy of pemetrexed in relapsed non-small cell lung cancer and thymidylate synthase expression. Tuberc Respir Dis 2009, 67(3):191–198.

23. Nakagawa T, Tanaka F, Otake Y, Yanagihara K, Miyahara R, Matsuo K, Takata T, Yamada T, Fukushima M, Wada H. Prognostic value of thymidylate synthase expression in patients with p-stage I adenocarcinoma of the lung. Lung Cancer 2002, 35(2):165–170.

24. Zheng Z, Chen T, Li X, Hauru E, Sharma A, Bepler G. DNA synthesis and repair genes RRM1 and ERCC1 in lung cancer. N Engl J Med 2007, 356(6):800–808.

25. Millet GA, Flores SA, Marks G, Reed JB, Herbst RJ. Circumcision status and risk of HIV and sexually transmitted infections among men who have sex with men: a meta-analysis. JAAMA 2008, 300(14):1674–1684.

26. Schoenleber SJ, Kurtz DM, Talwalkar JA, Roberts LR, Gores GJ. Prognostic role of vascular endothelial growth factor in hepatocellular carcinoma: systematic review and meta-analysis. Br J Cancer 2009, 100(9):1385–1392.

27. Checl ML, Checl JK, Davies E, Kiefer D. Effect of antagonists vs agonists on in vitro fertilization outcome. Clin Exp Obst Gynecol 2004, 31(4):257–259.

28. Chang MH, Ahn JS, Lee J, Kim KH, Park YH, Han J, Ahn MJ, Park K. The efficacy of pemetrexed as a third- or fourth-line therapy and the significance of thymidylate synthase expression in patients with advanced non-small cell lung cancer. Lung Cancer 2010, 69(3):323–329.

29. Wang S, Byeye S, Wada M, Otani S, Maki S, Takakura A, Kato T, Fukushima M, Sato Y, Masuda N. Pemetrexed for previously treated patients with non-small cell lung cancer and differences in efficacy according to thymidylate synthase expression. Chemotherapy 2012, 58(4):313–320.

30. Stang A. Critical evaluation of the Newcastle-Ottawa scale for the assessment of the quality of nonrandomized studies in meta-analyses. Eur J Epidemiol 2010, 25(9):609–605.

31. Krygwo M, Tsimoupi L, Vrekoussis T, Martin-Hirsch P, Arbyn M, Martin-Hirsch P, Prendiville W, TDG, Corbet W. Efficacy of pemetrexed in relapsed non-small cell lung cancer and thymidylate synthase expression. Tuberc Respir Dis 2009, 67(3):191–198.

32. Park CK, Kim KS, Oh IJ, Tieden IM, Choi YD, Kwak YS, Kim YI, Lim SC, Kim YC. Efficacy of pemetrexed in relapsed non-small cell lung cancer and thymidylate synthase expression. Tuberc Respir Dis 2009, 67(3):191–198.

33. Nakagawa T, Tanaka F, Otake Y, Yanagihara K, Miyahara R, Matsuo K, Takata T, Yamada T, Fukushima M, Wada H. Prognostic value of thymidylate synthase expression in patients with p-stage I adenocarcinoma of the lung. Lung Cancer 2002, 35(2):165–170.

34. Shih C, Chen VJ, Gossett LS, Gates SB, MacKellar WC, Habeck LL, Shackelford PV. Prognostic and predictive value of thymidylate synthase expression in primary colorectal cancer. Anticancer Res 2010, 30(2):645–651.

35. Qiu LX, Tang QY, Bai JL, Qian XP, Li RT, Liu BR, Zheng MH. Predictive value of thymidylate synthase expression in advanced colorectal cancer patients receiving fluoropyrimidine-based chemotherapy: evidence from 24 studies. Int J Cancer 2008, 123(10):2384–2389.

36. Kurosu K, Sakaida E, Sekine I, Tanabe N, Tagawa M, Tatsumi K. In vitro sensitivity to platinum-derived drugs is associated with expression of thymidylate synthase and dihydropteroamide dehydrogenase in human lung cancer. Oncol Rep 2000, 15(6):1533–1539.

37. Kitazono-Saitoh M, Takiguchi Y, Kitazono S, Ashinuma H, Kitamura A, Tada Y, Kurosu K, Sakaeda E, Sekine I, Tanabe N, Tagawa M, Tatsumi K. Interaction and cross-resistance of cisplatin and pemetrexed in malignant pleural mesothelioma cell lines. Oncol Rep 2012, 28(1):33–40.

38. Ibe T, Shimizu K, Nakano T, Kakegawa S, Kiyokawa H, Nakajima T, Kita K. Takeyoshi I. High-grade neuroendocrine carcinoma of the lung shows increased thymidylate synthase expression compared to other histotypes. J Surg Oncol 2010, 102(1):11–17.

39. Monica V, Scagliotti GV, Ceppi P, Righi L, Gambi A, Lo Iacono M, Saviozzi S, Valente M, Novello S, Papotti M. Differential thymidylate synthase expression in different variants of large-cell carcinoma of the lung. Clin Cancer Res 2009, 15(24):7547–7552.

40. Hou J, Lambers M, den Hamer B, den Bakker MA, Hoogsteden HC, Grosveld F, Hegmans J, Aerts J, Philipsen S. Expression profiling-based subtyping identifies novel non-small cell lung cancer subgroups and implicates putative resistance to pemetrexed therapy. J Thorac Oncol 2012, 7(3):105–114.