The interventional effect of new drugs combined with the Stupp protocol on glioblastoma: A network meta-analysis

Li Mei¹, Song Xiangqi², Zhu Jun¹, Fu Aijun¹, Li Jianmin¹, Chen Tong¹

1 Department of Neurosurgery, North China University of Science and Technology Affiliated Hospital, Tangshan 063000, Hebei Province, China

2 Department of Neurosurgery, People’s Hospital of Suning County, Cangzhou 062350, Hebei Province, China

Corresponding Author: Chen Tong,
Department of Neurosurgery, North China University of Science and Technology Affiliated Hospital, Tangshan 063000, Hebei Province, China
Tel: +86 15032915731
Email: ct.1973@163.com

Highlights
1 The article was used the network meta-analysis to evaluated the effects of various new drugs.
2 We revealed that each of the 7 protocols were all deemed superior to the Stupp protocol alone.
3 We presented a pathway for further investigation and provided encouraging data for future treatment of these neoplasms.
Abstract

Objective: New therapeutic agents in combination with the standard Stupp protocol (a protocol about the temozolomide combined with radiotherapy treatment with glioblastoma was research by Stupp R in 2005) were assessed to evaluate whether they were superior to the Stupp protocol alone, to determine the optimum treatment regimen for patients with newly diagnosed glioblastoma.

Patients and Methods: We implemented a search strategy to identify studies in the following databases: PubMed, Cochrane Library, EMBASE, CNKI, CBM, Wanfang, and VIP, and assessed the quality of extracted data from the trials included. Statistical software was used to perform network meta-analysis.

Results: The use of novel therapeutic agents in combination with the Stupp protocol were all shown to be superior than the Stupp protocol alone for the treatment of newly diagnosed glioblastoma, ranked as follows: cilengitide 2000 mg/5/week, bevacizumab in combination with irinotecan, nimotuzumab, bevacizumab, cilengitide 2000 mg/2/week, cytokine-induced killer cell immunotherapy, and the Stupp protocol. In terms of serious adverse effects, the intervention group showed a 29% increase in the incidence of adverse events compared with the control group (patients treated only with Stupp protocol) with a statistically significant difference (RR = 1.29; 95%CI 1.17–1.43; \( P < 0.001 \)). The most common adverse events were thrombocytopenia, lymphopenia, neutropenia, pneumonia, nausea, and vomiting, none of which were significantly different between the groups except for neutropenia, pneumonia, and embolism.

Conclusions: All intervention drugs evaluated in our study were superior to the Stupp protocol alone when used in combination with it. However, we could not conclusively confirm whether cilengitide 2000 mg/5/week was the optimum regime, as only one trial using this protocol was included in our study.

Keywords: Glioblastoma; Temozolomide; Bevacizumab; Cilengitide; Targeted therapy
1. Introduction

Glioblastoma (GBM), an invasive solid tumor, is the most common primary tumor of the brain [1, 2]. The standard treatment is radiotherapy (RT) plus concomitant and adjuvant therapy, with six cycles of temozolomide (TMZ) following surgical removal of the maximum safe scope, which is currently the internationally accepted treatment protocol (also was the Stupp protocol). However, despite the availability of advanced treatment, patients with GBM have a poor prognosis, with average survival of only 14.6–16 months and a 5-year survival rate of less than 10%, thus representing a significant treatment challenge [3-5]. GBM tumors are characterized by increased expression of vascular endothelial growth factor (VEGF) and epidermal growth factor receptor (EGFR), neoangiogenesis, and epidermal growth factor receptor gene amplification, among other factors. Recent research has tended towards the exploration of integrated molecular targeted drugs (e.g., bevacizumab [BEV], cilengitide, vandetanib, nimotuzumab, and cellular immune drugs) with the Stupp protocol and evaluating the subsequent effectiveness and safety of these approaches [6-11]. Nevertheless, the treatment outcomes of new drugs in large, randomized controlled trials and studies to contrast the efficacy of a variety new drugs in combination with the Stupp protocol are lacking, as clinical studies frequently compare the effectiveness of the new drugs combined with the Stupp with used it alone. Therefore, this study used a network meta-analysis approach, with the principle of direct and indirect comparison [12, 13], to evaluate whether treatment with new drugs in combination with the Stupp protocol is superior to the Stupp protocol alone in patients with newly diagnosed GBM.

2. Patients and Methods

2.1. Inclusion and exclusion criteria

The inclusion and exclusion criteria for this study were based on the PICO strategy (P: patient, I: intervention, C: comparison, O: outcome). The inclusion criteria were as follows: 1) patients aged 18 years and older, with newly diagnosed and histopathologically confirmed GBM multiforme, Karnofsky status ≥ 50, and World Health Organization (WHO) performance scales
≤ 2; 2) in terms of intervention, treatment criteria were new drugs in combination with the Stupp protocol on the Stupp alone. In this study, the Stupp protocol was defined as TMZ (75 mg/m²/d, with a maximum of 49 days from the first day of RT until the last) with concomitant RT (total dose: 60 Gy, 30 × 2 Gy) and adjuvant TMZ (150–200 mg/m²/d, 1–5 d/28 d, total of 6 cycles); 3) randomized clinical trials, with research from China having being carried out only in hospitals of Grade 3A and above, sample size of ≥30, and only the most recent publication by a given author; 4) information on the following outcome indicators in the original text or available from the study authors: average overall survival, overall survival rate, and Kaplan-Meier survival curve. For our analysis, the overall survival rate with the longest follow-up period was selected.

Patients were excluded if they had a history of serious hematology, cardiopulmonary, liver, kidney, or other systemic disease; other primary or metastatic tumors than GBM; other intracranial diseases (e.g., intracranial hemorrhage or brain abscess); pregnant or breastfeeding women; previous treatment with radiotherapy, chemotherapy, or immunotherapy; or serious chemotherapy drug allergy.

2.2. Search strategy

The following key words, full text, and MeSH terms were used to search the PubMed, Cochrane Library, EMBASE, CNKI, CBM, VIP, and Wanfang databases for publications dated on or before October 8, 2016: glioma, glioblastoma, temozolomide, bevacizumab, carmustine, nimustine, chemotherapy, drug therapy, targeted therapy, antiepileptic, anticonvulsant, radiotherapy, radiation therapy, randomized controlled trial, non-randomized controlled trial, random*, non-random*. We also searched the references of related documents, magazines, journals, and meeting abstracts, and searched the WHO International Clinical Trials Registry Platform (WHO ICTRP) to identify trials that were ongoing or complete but not yet published, as well as trials included in relevant systematic reviews or meta-analysis published 2–3 years previously.
2.3. Study selection and quality assessment

The inclusion and exclusion criteria were applied by two systematic reviewers, who independently screened the literature retrieval results and read the full articles, using the Cochrane Quality Evaluation Method to assess randomized trials included in the present study. To avoid bias, differences were discussed by the two reviewers or agreed with a third party if consensus was not met.

2.4. Data extraction

The information extracted from the all included trials included the baseline characteristics (age; surgical status: complete removal, partial removal, biopsy, et al.; Karnofsky performance status [KPS]/WHO performance scores; steroid usage) of all patients and the number of patients per treatment group; the drugs, doses, and course of treatment; and the indicators of clinical outcome (average overall survival, overall survival rate, and hazard ratio with 95% confidence interval [CI]).

2.5. Statistical analysis

For heterogeneity, $X^2$ and $I^2$ statistics were used to analyze the data identified in our study. For values of $P < 0.1$ and $I^2 > 20\%$, results were considered to have significant heterogeneity and the random effects model was used; otherwise a fixed effects model was adopted.

Consistency checks were also performed before direct and indirect evidence was merged. The final part of the network meta-analysis strategy was an extension of the traditional head-to-head meta-analysis approach, and involved the identification of the best intervention, given the effect relationship between any two interventions. Based on this principle, network meta-analysis was identified using Stata version 13.0 statistical software.

3. Results

3.1. Literature retrieval and network chart construction

A total of 3409 items were retrieved (3375 articles from the electronic databases and the remainder from manual searches). Of these, 3272 articles were excluded after reading of the titles and the abstracts, and elimination of duplicate articles. Following full-text reading, only
19 studies remained, and when inclusion and exclusion criteria were applied, 7 studies were identified for inclusion in our analysis (Supplementary Fig. 1). In the network chart shown in Fig. 1, a connection between two interventions indicates evidence of a direct comparison, dots represent the sample size, and the thickness of the line represents the number of studies.

3.2. Characteristics of trials

Among the 7 studies included in our analysis, one was a three-arm trial that was integrated as a two-arm trial. The characteristics of all studies are shown in Table 1. Sample sizes in the treatment and control groups were 1437 and 1446, respectively, and all trials implemented intention-to-treat analysis.

3.2.1. Evaluation of quality

A Cochrane bias risk assessment was used to evaluate the quality of all articles, primarily from the following aspects: random allocation, allocation concealment, blinding method, data integrity, selective reports, and other bias. Although only two of the selected trials were blinded, the quality of all trials was considered high as the results were objective and had little effect on it (showed in Table 2, and the references of 25-27 was included in this table).

3.2.2. Heterogeneity

As shown in Fig. 2, heterogeneity was observed among the included studies \( (P < 0.1, I^2: 48\%) \) using the random effects model. However, there were slight differences in the types of patients and study design between all trials. The subgroup analysis of these factors showed a lack of heterogeneity \( (P > 0.1, I^2 < 20\%) \) using the fixed effects model (seen in Fig 3a, 3b).

3.2.3. Consistency

Given that a star network diagram (Fig. 1) was developed in our analysis and that there was no direct and indirect evidence to merge, consistency analysis was not required.

3.3. Network meta-analysis results

3.3.1. 24-months overall survival

The size of the area under the curve in the rank chart figure indicates the degree of interventional effect. The outcome of the network meta-analysis (Fig. 4) revealed the combinations of different treatment interventions with the Stupp protocol were all superior to
the Stupp protocol alone in patients with newly diagnosed GBM, in the following order: cilengitide 2000 mg/5/week; bevacizumab with irinotecan, nimotuzumab, bevacizumab, or cilengitide 2000 mg/2/week, cytokine-induced killer cells (CIK) immunotherapy, and the Stupp protocol.

3.3.2. Adverse effects

The toxic effects reported in trials in our study were recorded and graded according to the National Cancer Institute Common Terminology Criteria for Adverse Events (CTCAE), version 3.0. However, significant heterogeneity was observed among the recorded details of adverse events in each study. Three of the studies (Chinot OL [14], Nabors OL [15], Kong DS [3]) reported only the number of serious adverse effects, divided into subgroups to evaluate toxicity (Supplementary Fig. 2), and revealed that the treatment group increased the incidence of adverse events by 29% compared with the control group with a statistically significant difference (relative risk [RR]: 1.29, 95% CI: 1.17–1.43; P<0.001). Moreover, the Gilbert MR [16] study recorded the incidence of side effects during both the concomitant and the adjuvant phases. The most common hematologic and non-hematological toxicities observed were thrombocytopenia, neutropenia, lymphopenia, dizziness, vicious, and vomiting. However, only neutropenia was statistically significant between the two interventions. Among the remaining trials (see Table 1 in the Supplementary Appendix), the most common events reported were as for the Gilbert MR [16] study; the incidence of side effects in these studies, except for pneumonia and embolism events, were not statistically significant.

4. Discussion

Glioma, derived from the neuroglial cells, is the most common primary intracranial tumor. It has been into four grades by the WHO, with GBM, a grade IV astrocytoma, classed as the most malignant [17-20]. Recent studies on the treatment of GBM that focused on aspects of molecular biology/pathology have contributed to significant treatment progress. We identified 7 trials for analysis, and showed that the effective of included treatment measures (cilengitide, bevacizumab, bevacizumab with irinotecan, nimotuzumab, and CIK cells, in combination with
the Stupp protocol) were better than the Stupp protocol alone. However, our study included only one trial of cilengitide 2000 mg/5/week, nimotuzumab, and CIK, so the ranking of these trials cannot be considered of clinical relevance.

Treatment with bevacizumab with or without irinotecan is used as initial treatment for relapsed GBM and has been shown to improve prognosis. On this basis, some researchers have combined this treatment with the Stupp protocol whether has a better valid than itself in patients with newly diagnosed GBM [21-23]. The rank chart generated in this study indicated that this approach (bevacizumab with or without irinotecan, in combination with the Stupp protocol) was superior to the Stupp protocol alone, and that bevacizumab together with irinotecan was the optimum treatment. In addition, we also found that the Stupp protocol combined with cilengitide 2000 mg/2/week was an effect treatment measure. was Although we only included one trial of cilengitide 2000 mg/5/week, the higher dose of this drug was shown to be superior to the standard regimen of 2000 mg/2/week. This conclusion—the range of tolerance in these patients that the higher dose of cilengitide is superior to the lower dose—is consistent with the safe execution of the applicable randomized phase II trial [24].

From this perspective, the cilengitide regimens (2000 mg/5/week or 2000 mg/2/week) were also both superior to the Stupp protocol. However, whether cilengitide 2000 mg/5/week is the best available treatment requires further exploration. In terms of the incidence of side effects, the large variation in information available in our study and it is unknown whether the same patients had a variety of adverse events; thus, to avoid overestimation, we grouped side effects for analysis and showed that the incidence of the serious adverse events with bevacizumab, CIK, and both doses of cilengitide treatment was significantly higher than those reported for the Stupp protocol. Furthermore, the most common hematologic and non-hematological toxicities associated with cilengitide 2000 mg/2/week, nimotuzumab, bevacizumab, and irinotecan were thrombocytopenia, neutropenia, lymphopenia, leucopenia, embolism, pneumonia, dizziness, vicious, and vomiting. Except for the incidence of embolism and pneumonia, however, these effects had no statistical significance.
5. Conclusion

Our meta-analysis showed that treatment with the various new drugs included in our analysis in combination with the Stupp protocol was superior to the Stupp protocol alone. However, it is difficult to draw clinical conclusions from our findings because of the low number of studies evaluated. Moreover, differences in the data available for adverse effects were observed between each trial. These limitations should be addressed in future studies on treatment ranking and the merged analysis of effectively and safety.
Acknowledgements

Funding: This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Conflict of Interest: Author Mei Li has no conflict of interest. Author Xiang-Qi Song has no conflict of interest. Author Jun-Zhu has no conflict of interest. Author Ai-Jun Fu has no conflict of interest. Author Jian-Min Li has no conflict of interest. Author Tong Chen has no conflict of interest.

Ethical approval: This article does not contain any studies with human participants or animals performed by any of the authors.
References


25. Chauffert B, Feuvret L, Bonnetain F, Taillandier L, Frappaz D, Taillia H, et al. Editor's choice: Randomized phase II trial of irinotecan and bevacizumab as neo-adjuvant and adjuvant to temozolomide-based chemoradiation compared with


Figure legends

Figure 1 Network chart
Figure 2 Analysis of heterogeneity
Figure 3 a. Heterogeneity in patient type. b. Heterogeneity in trial type
Figure 4 Rank chart

Footnotes: Stupp: (the Stupp protocol) TMZ (75 mg/m²/d, with a maximum of 49 days from the first day of RT until the last) with concomitant RT (total dose: 60 Gy, 30 × 2 Gy) and adjuvant TMZ (150–200 mg/m²/d, 1–5 d/28 d, total of 6 cycles); CIK: cytokine-induced killer cell immunotherapy added to the Stupp protocol; BEV+IRI-RT+TMZ+BEV+IRI-BEV+IRI: bevacizumab + irinotecan combined with the Stupp protocol; BEV: BEV added to the Stupp protocol; nimotuzumab: nimotuzumab added to the Stupp protocol; cilengitide/2000mg/2/week: cilengitide/2000mg/2/week added to the Stupp protocol; cilengitide/2000mg/5/week: cilengitide/2000mg/5/week added to the Stupp protocol.
**Figure 2** Analysis of heterogeneity

<table>
<thead>
<tr>
<th>Id</th>
<th>RR (95% CI)</th>
<th>Weight</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kong DS</td>
<td>0.99 (0.74, 1.34)</td>
<td>11.30</td>
</tr>
<tr>
<td>Nabors BL</td>
<td>0.92 (0.84, 1.00)</td>
<td>10.36</td>
</tr>
<tr>
<td>Nabors BL</td>
<td>0.79 (0.55, 1.07)</td>
<td>4.42</td>
</tr>
<tr>
<td>Chauffert B</td>
<td>0.82 (0.43, 1.62)</td>
<td>8.03</td>
</tr>
<tr>
<td>Stupp R</td>
<td>0.86 (0.59, 1.24)</td>
<td>19.29</td>
</tr>
<tr>
<td>Clinton DL</td>
<td>0.82 (0.42, 1.61)</td>
<td>21.97</td>
</tr>
<tr>
<td>Gilbert NR</td>
<td>1.04 (0.94, 1.03)</td>
<td>15.47</td>
</tr>
<tr>
<td>Westphal M</td>
<td>0.87 (0.61, 1.23)</td>
<td>9.15</td>
</tr>
<tr>
<td>Overall (I-squared = 48.0%, p = 0.092)</td>
<td>0.88 (0.77, 1.00)</td>
<td>100.00</td>
</tr>
</tbody>
</table>

NOTE: Weights are from random effects analysis.
**Figure 3 a.** Heterogeneity in patient type

![Figure 3 a. Heterogeneity in patient type](image)

**Figure 3 b.** Heterogeneity in trial type

![Figure 3 b. Heterogeneity in trial type](image)

**Figure 4** Rank chart

![Figure 4](image)
By reading title, abstract, repetition, there are rest of 137

By reading the full-text rest of 19

Finally 7 trials for Network Meta-Analysis
### Table 1. Overview of literature included

<table>
<thead>
<tr>
<th>Study</th>
<th>Year</th>
<th>Patient</th>
<th>Intervention group</th>
<th>Control Group</th>
<th>Number of patient</th>
<th>HR(95% CI)</th>
<th>Outcomes</th>
<th>Analysis method</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chinot OL</td>
<td>2014</td>
<td>Glioblastoma</td>
<td>Bevacizumab + Temozolomide</td>
<td>The Stupp</td>
<td>458/463</td>
<td>0.88(0.76-1.02)</td>
<td>24-Overall survival (from randomized to death)</td>
<td>ITT</td>
</tr>
<tr>
<td>Gilbert MR</td>
<td>2014</td>
<td>Glioblastoma</td>
<td>Bevacizumab + Temozolomide</td>
<td>The Stupp</td>
<td>309/312</td>
<td>1.13(0.93-1.37)</td>
<td>24-Overall survival (from randomized to death)</td>
<td>ITT</td>
</tr>
<tr>
<td>Chauffert B</td>
<td>2014</td>
<td>Unresectable Glioblastoma</td>
<td>Bevacizumab + Irinotecan added to the Stupp</td>
<td>The Stupp</td>
<td>60/60</td>
<td>———</td>
<td>24-Overall survival (from randomized to death)</td>
<td>ITT</td>
</tr>
<tr>
<td>Stupp R</td>
<td>2014</td>
<td>MGMT methylate Glioblastoma</td>
<td>Cilengitide 2000mg/2/week</td>
<td>The Stupp</td>
<td>272/273</td>
<td>1.02(0.81-1.29)</td>
<td>24-Overall survival (from randomized to death)</td>
<td>ITT</td>
</tr>
<tr>
<td>Authors</td>
<td>Year</td>
<td>Tumor Type</td>
<td>Treatment</td>
<td>Study</td>
<td>Event Rate</td>
<td>HR (95% CI)</td>
<td>24-Month Survival (%)</td>
<td>ITT</td>
</tr>
<tr>
<td>-------------</td>
<td>------</td>
<td>----------------</td>
<td>------------------------------------------</td>
<td>-------</td>
<td>------------</td>
<td>-------------</td>
<td>------------------------</td>
<td>-----</td>
</tr>
<tr>
<td>Westphal M</td>
<td>2015</td>
<td>Glioblastoma</td>
<td>Nimotuzumab + Temozolomide</td>
<td>The Stupp</td>
<td>71/71</td>
<td>0.953 (0.669-1.359)</td>
<td>24-Overall survival (from randomized to death)</td>
<td>ITT</td>
</tr>
<tr>
<td>Nabors BL</td>
<td>2015</td>
<td>unMGMT Glioblastoma</td>
<td>Cilengitide 2000mg/5/week</td>
<td>The Stupp</td>
<td>88/89</td>
<td>0.858 (0.612-1.204)</td>
<td>24-Overall survival (from randomized to death)</td>
<td>ITT</td>
</tr>
<tr>
<td>Nabors BL</td>
<td>2015</td>
<td>unMGMT Glioblastoma</td>
<td>Cilengitide 2000mg/2/week</td>
<td>The Stupp</td>
<td>88/89</td>
<td>0.686 (0.484-0.972)</td>
<td>24-Overall survival (from randomized to death)</td>
<td>ITT</td>
</tr>
<tr>
<td>Kong DS</td>
<td>2016</td>
<td>Glioblastoma</td>
<td>CIK + Temozolomide</td>
<td>The Stupp</td>
<td>91/89</td>
<td>0.693 (0.512-0.937)</td>
<td>24-Overall survival (from randomized to death)</td>
<td>ITT</td>
</tr>
</tbody>
</table>

**Footnotes:** The Stupp: TMZ (75 mg/m²/d, with a maximum of 49 days from the first day of RT until the last) with concomitant RT (total dose: 60 Gy, 30 x 2 Gy) and adjuvant TMZ (150–200 mg/m²/d, 1–5 d/28 d, total of 6 cycles); ITT: Intent-to-treatment
Table 2. Quality assessment of identified literature

<table>
<thead>
<tr>
<th>Included study</th>
<th>Random</th>
<th>Allocation concealment</th>
<th>Blind</th>
<th>Complete data</th>
<th>Selective report</th>
<th>Other bias</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chinot OL 2014 [14]</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
<td>Unclear</td>
</tr>
<tr>
<td>Gilbert MR 2014 [16]</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
<td>Unclear</td>
</tr>
<tr>
<td>Chauffert B 2014 [25]</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>Unclear</td>
</tr>
<tr>
<td>Stupp R 2014 [26]</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>Unclear</td>
</tr>
<tr>
<td>Nabors BL 2015 [15]</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>Unclear</td>
</tr>
<tr>
<td>Westphal M 2015 [27]</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>Unclear</td>
</tr>
<tr>
<td>Kong DS 2016 [3]</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>Unclear</td>
</tr>
</tbody>
</table>

Footnotes: Yes is used these methods which was listed in the table; No represents not used these; Unclear is not know whether have other bias, this is not mentioned in the original and no link to the author.