

Original Article

Efficacy and safety of chemotherapy with or without ifosfamide in primary osteosarcoma treatment: a systemic review of randomized controlled trials

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Abstract: Background: Currently, osteosarcoma is mainly treated with a combination of methotrexate, cisplatin, doxorubicin and/or ifosfamide. However, it is still unclear whether ifosfamide should be included in the preoperative chemotherapy. This study is to systemically and comprehensively compare the efficacy and safety of clinical trials with or without ifosfamide in treating pediatric and adult osteosarcoma. Methods: We searched Cochrane Central Register of Controlled Trials (CENTRAL), Medline, Embase and Clinical trial.gov database. In addition, we searched reference lists of relevant articles and conference proceedings databases. Randomized controlled trials (RCTs) or quasi-controlled clinical trials comparing the efficacy and safety of preoperative chemotherapy including ifosfamide to treatment without ifosfamide in treating primary osteosarcoma. Two reviewers independently conducted the study selection. Two reviewers performed the data extraction and analysis independently. Results: Compared with regimen without ifosfamide, the regimen which includes ifosfamide in preoperative chemotherapy does not improve the Overall Survival (HR: 0.98, 95% CIs: 0.91-1.06, P=0.6) and Event-free Survival (RR: 1.05, 95% CIs: 0.90-1.22, P=0.57). Moreover, the introduction of ifosfamide into the preoperative chemotherapy may lead to higher frequency of toxicity-related events. Conclusions: Given the efficacy and safety, ifosfamide should not be recommended to be included in preoperative chemotherapy for treating primary osteosarcoma.

Keywords: Osteosarcoma, ifosfamide, chemotherapy, systemic review

Introduction

Osteosarcoma is a malignant bone tumor, with an approximate incidence of 3/100,000 [1]. Prior to the chemotherapy era, osteosarcoma was mainly treated by amputation alone, and the survival rate is merely 20% [2]. Due to the introduction of neoadjuvant and adjuvant chemotherapy, the cure rate for osteosarcoma has remarkably increased to 60-70% [3]. The efficacy of therapeutic agent's methotrexate (MTX), doxorubicin (DOX), and cisplatin (CDP) in osteosarcoma have been separately demonstrated by multiple studies [4-10].

Currently, the majority of treatment protocols are based on a combination of 4 drugs: DOX, CDP, MTX and/or ifosfamide (IFO) [11-19]. However, the role of each drug in the combination has not been well established. Specifically, it is not clear whether IFO is an essential part of a multidrug combination.

It has been shown that IFO is effective in treating recurrent or metastatic osteosarcoma [20-23]. Further, one study supports the idea to include IFO into neoadjuvant chemotherapy for treating primary osteosarcoma [17].

Since morbidity and toxicity related to chemotherapy cannot be neglected, further optimization of the combination is essential to achieve better clinical outcomes. This need prompts us to conduct this study to systemically analyze existing studies on the role of IFO in the combined chemotherapy for treating primary osteosarcoma.

Materials and methods

Literature searching

A protocol that specified the method was conducted in advance. MEDLINE/PubMed, Cochrane Central Register of Controlled Trials

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(CENTRAL), EMBASE/Ovid, and Clinicaltrial.gov database were searched electronically. The search strategy included MeSH and/or free text words, with the MeSH terms of 'osteosarcoma' and 'ifosfamide'. Search strategies were combined with the Cochrane Highly Sensitive Search Strategy to identify randomized trials. No restrictions were placed on years. Reports not found in aforementioned database, either published or unpublished, were searched by hand in reference lists of relevant publications.

Selection of studies

The trials included should:

- Be randomized controlled trials (RCTs) or quasi-controlled clinical trials comparing the efficacy and safety of preoperative chemotherapy with IFO to chemotherapy without IFO in treating osteosarcoma.
- Have outcome variables including overall survival (OAS), event-free survival (EFS), response rate (RR) and toxicity.
- Have patients diagnosed with primary osteosarcoma without metastasis.

The following articles were excluded:

- Review articles, cohort studies, case-control studies and other kinds of observational studies.
- Repetitive publication (only the well-described one was included).
- Have patients with prior treatment of osteosarcoma.
- Have patients with medical contraindications to the drugs included in the protocol.
- Have patients with osteosarcoma with metastasis.

Selection of studies was independently conducted by two reviewers. Any study seemingly meeting the aforementioned criteria on the basis of the title and/or abstract, was further investigated in full-text articles. Reasons for excluding any study were clearly stated. Disagreements between reviewers were resolved by discussion. Microsoft Excel was employed to manage the articles.

Primary and secondary outcomes

Primary outcomes:

- (1) Overall survival (calculated from the first day of chemotherapy to death or the last follow-up examination).
- (2) Event-free survival (calculated from the first day of chemotherapy to recurrence, death from all causes, the appearance of secondary tumors, or the last follow-up examination).

Secondary outcomes:

- (1) Response rate (calculated as classical response rates or percentage of achieved necrosis [24] or based on surgical margins).
- (2) Toxicities (any treatment-related adverse events).

Risk of bias assessment

Risk of bias assessment was conducted according to the Cochrane Collaboration's tool 2011 on the following seven domains: (1) random sequence generation, (2) allocation concealment, (3) blinding of participants and personnel, (4) blinding of outcome assessment, (5) incomplete outcome data, (6) selective reporting and (7) other bias [25]. Each domain was judged as 'high risk', 'unclear risk' or 'low risk'. The risk of bias, bias in each domain for each study was assessed by one reviewer and checked by another reviewer. Disagreements between reviewers were resolved by discussion.

Data extraction

A customized data extraction form was developed in advance by the reviewers. The following items were included: characteristics of participants (age, sex, metastatic status, primary or secondary disease), interventions (drugs, cumulative dose, dose intensity, and duration of chemotherapy), outcome measures and length of follow-up. Data extraction was performed by one reviewer using standardized forms and checked by another reviewer. Disagreement between reviewers was resolved by discussion.

Data analysis

Review Manager 5.1 (The Nordic Cochrane Centre, The Cochrane Collaboration, Copenhagen, Denmark) was employed for data analysis.

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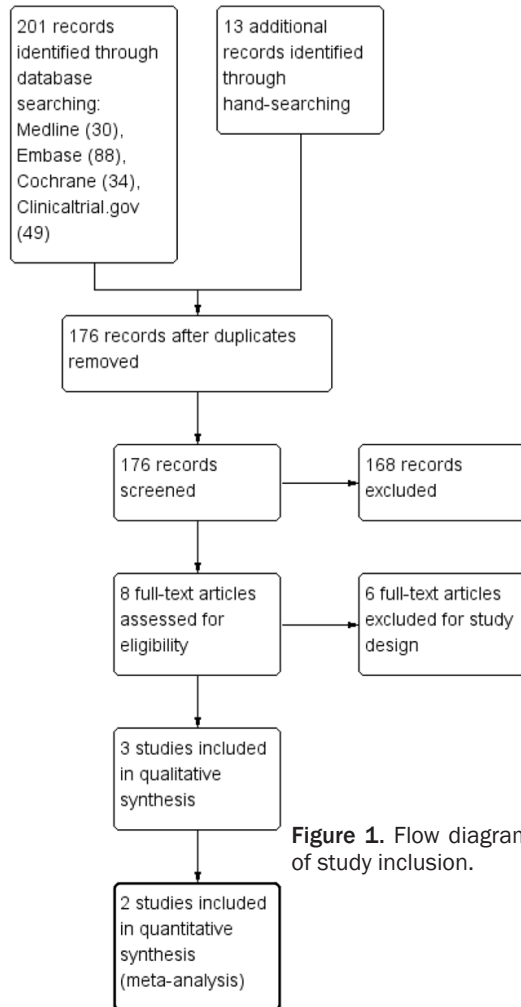


Figure 1. Flow diagram of study inclusion.

sis. The combined results for dichotomous outcomes were expressed as risk ratio (RR) or hazard ratio (HR) and 95% confidence intervals (95% CIs). The outcomes were analyzed based on allocated intervention, irrespective of compliance with the protocol, in an 'intention-to-treat' manner. Due to the anticipated variability in included studies, a random effects model was applied in the meta-analysis. The statistical significance of the hypothesis test was set at $P < 0.05$ (two-tailed z-tests). Statistical heterogeneity was explored by test for heterogeneity (I^2 test) at the level of $\alpha = 0.10$. Sensitivity analysis was conducted to test the stability of the results.

Results

Search and study inclusion

The original electronic searches of MEDLINE, EMBASE, Cochrane Central Register of Con-

trolled Trials and Clinicaltrial.gov database identified 30, 88, 34, and 49 potentially eligible articles, respectively. A total of 13 studies were obtained by hand-searching of references of relevant articles. After removing duplicate studies, 176 studies remained. After screening the titles, abstracts and no published results, a total of 8 studies were considered eligible and their full texts were retrieved. Two studies were excluded for not including osteosarcoma patients in the trials [26, 27]. Two studies were excluded because they were not RCTs [28, 29]. One study [30] was excluded because it was preliminary data report of one included study [31]. However, we used this article for reference when we were assessing the included study [31]. Another study was excluded for enrolling patients with metastatic osteosarcoma [32]. Finally, 2 studies were included for this systemic review and further meta-analysis [31, 33] (**Figure 1**).

Description of studies

A total of 2 studies were included for meta-analysis and systemic review [31, 33]. The characteristics of the included trials were summarized in **Table 1**. Both studies were RCTs with a relatively large sample size (at least 246 patients). Also, both studies compared the safety and efficacy between 3-drug regimen (MTX, DOX and CDP) and 4-drug regimen (MTX, DOX, CDP and IFO) in preoperative chemotherapy for treating primary osteosarcoma. Both studies [31, 33] included all important pre-specified outcomes. Also, both studies assessed patients with localized osteosarcoma.

Risk of bias assessment

No included studies were completely free of potential bias. The presence of selection bias (random sequence generation and/or allocation concealment) could not be ruled out from both studies. Further, one study [33] may have attrition bias due to potential incomplete outcome data. The detail of risk of bias assessment on each study was shown in **Figures 2** and **3**. Generally speaking, the included studies were of good quality and reported reliable results. In addition, both studies conducted reliable statistical analysis and handled the loss of follow-up in a proper way.

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Table 1. Characteristics of studies included

Study	Design	Nation	Sex	Age (years)	Metastatic status	Intervention	Median follow-up (months)	Outcomes
Meyers 2008	RCT	USA	361 (M), 301 (F)	1-30 (median: 13)	Localized	Regimen A (CDP, DOX and MTX); Regimen B (IFO, CDP, DOX and MTX)	92	EFS, OAS, RR, Toxicities
Ferrari 2012	RCT	Italy	146 (M), 100 (F)	4-39 (median: 14)	Localized	Regimen A (IFO given postoperatively when pathological response to CDP, DOX and MTX was poor); Regimen B (IFO given in the primary phase of chemotherapy with CDP, DOX and MTX)	66	EFS, OAS, RR, Toxicities

RCT, randomized clinical trials; M, male; F, female; OAS, overall survival; EFS, event-free survival; RR, response rate.

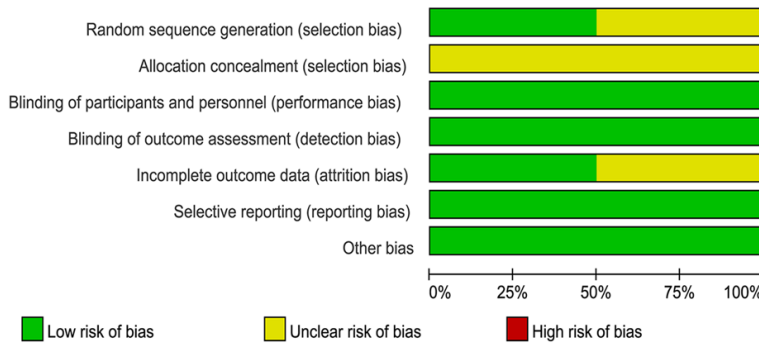


Figure 2. Risk of bias graph.

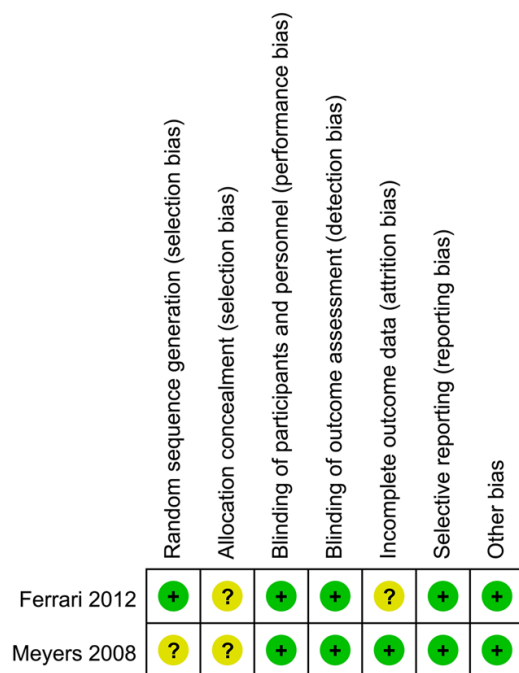


Figure 3. Risk of bias summary.

Effects of intervention

Overall survival (OAS): One study [31] reported 4-year and 6-year OAS, while the other [33]

reported 5-year OAS. For simplicity of meta-analysis, we used 6-year OAS from this study [31]. The pooled outcome showed that there was no statistical significance between two regimens in OAS (HR: 0.98, 95% CIs: 0.91-1.06, P=0.6) (Figure 4). The I² test yielded a value of 0, indicating that the statistical heterogeneity was really low enough to justify the meta-analysis. When using 4-year

OAS from Meyers study, the pooled outcome was similar to what we have reported.

Event-free survival (EFS): One study [31] reported 4-year and 6-year EFS, while the other reported 5-year EFS. For simplicity of meta-analysis, we used 6-year EFS from this study. The pooled outcome from these studies showed that there was no statistical significance between two regimens in EFS (RR: 1.05, 95% CIs: 0.9-1.22, P=0.57) (Figure 5). The I² test yielded a value of 46%. When using 4-year OAS from Meyers study, the pooled outcome was similar to what we have reported.

Response rate: These two studies [31, 33] used different definitions of response rate. One study [33] defined good responders (GRs) as the percentage of tumor necrosis was higher than 90%. The study conducted by Meyers and colleagues [31] were grading necrosis was according to the method of Huvos, as modified by CCG. However, two studies obtained similar results: no significant difference between two regimens.

Toxicities: Both studies [31, 33] reported 5 deaths, with multiple toxicity-related events including alanine aminotransferase (ALT) elevation, stomatitis and infection. For most of the

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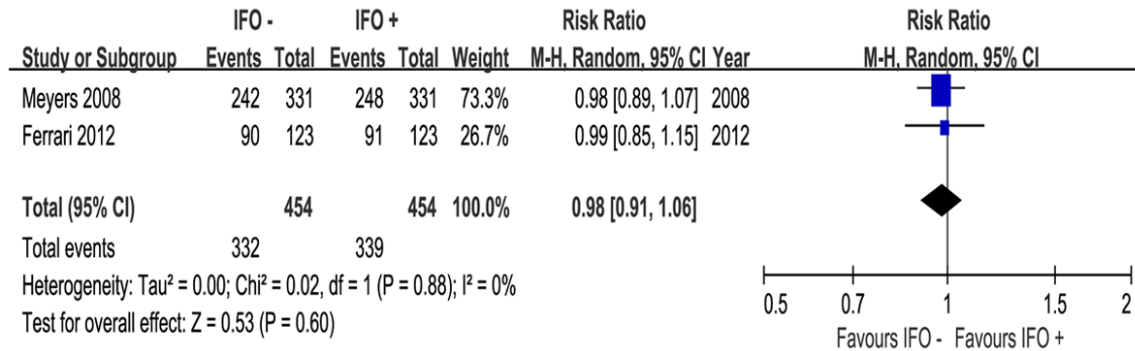


Figure 4. Meta-analysis comparing regimen without ifosfamide to regimen with ifosfamide in overall survival in treating osteosarcoma. IFO, ifosfamide.

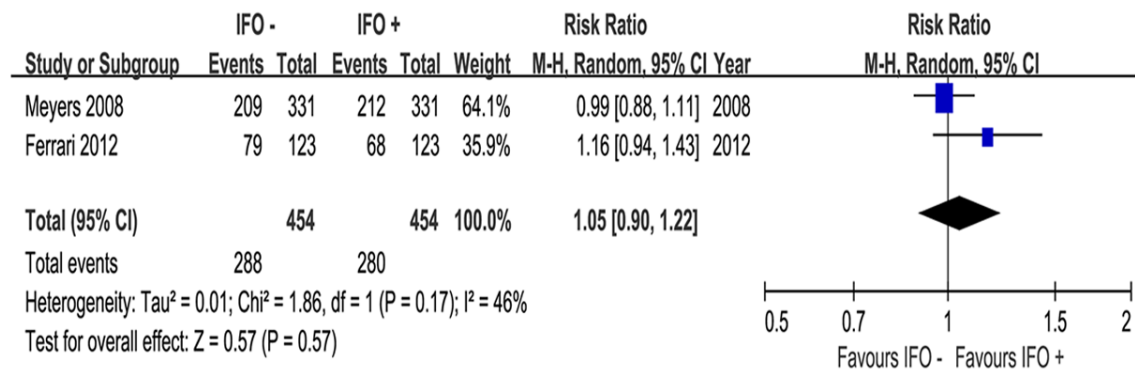


Figure 5. Meta-analysis comparing regimen without ifosfamide to regimen with ifosfamide in event-free survival in treating osteosarcoma. IFO, ifosfamide.

toxicities reported, there was no statistically significant difference between two regimens in this study. However, in the study conducted by Ferrari and colleagues [33], there was one death in regimen without IFO, while 3 in regimen with IFO. Further, there was significantly higher frequency of toxicity-related events including grade 4 leukopenia, thrombocytopenia, neutropenia, RBC and platelet transfusion ($P < 0.001$). In summary, it appeared that adding IFO into the preoperative chemotherapy may increase the incidence of toxicity-related events.

Discussion

As a result of the introduction of chemotherapy, the survival of children with osteosarcoma has improved dramatically from 20% to 50-60% [2]. Nowadays, single agent chemotherapy treatment of patients with osteosarcoma is considered to be inadequate. Currently, the majority

of treatment protocols are based on a combination of DOX, CDP, MTX and/or IFO. Currently, there are no randomized studies examining the role of DOX in adjuvant chemotherapy, and randomized studies of CDP have been conducted only on the basis of patient responses to other therapy [13]. A systemic review has been conducted to assess the role of MTX in treating osteosarcoma [34]. However, no eligible RCTs have been identified to make a firm conclusion whether MTX should be included in the chemotherapy. In spite of these efforts, the specific role of each drug has not been elucidated.

Specifically, IFO has been shown to have significant effects in primary or metastatic osteosarcoma [17, 21-23]. However, there is evidence that inclusion of IFO has no significant effects in treating primary osteosarcoma [31, 33]. Therefore, there is no consensus on whether and how IFO should be included in the preoperative chemotherapy [35]. This study is the

first systematic review evaluating the current state of evidence on the use of IFO in the preoperative chemotherapy of pediatric and adult patients with primary osteosarcoma.

To comprehensively evaluate the role of IFO in the treatment of primary osteosarcoma the optimal study design is an RCT in which the sole difference between the intervention and control group is using IFO or not. We identified two studies that compared regimen with IFO to regimen without IFO. Meyers and colleagues [31] recruited patients with primary high grade osteosarcoma and no prior treatment, and compared 4-drug regimen (MTX, DOX, CDP and IFO) to 3-drug regimen (MTX, CDP and DOX). Also, within each regimen, patients were randomly assigned with or without muramyl tripeptide (MTP). In Ferrari study [33], IFO was given postoperatively to patients with poor pathological responses to 3-drug combination (Regimen A), or given in preoperative 4-drug combination (Regimen B). In total, there were 246 patients with primary high grade osteosarcoma and no prior treatment.

The primary outcome of this meta-analysis is OAS and EFS. In Ferrari study [33], 5-year OAS and EFS were used, while 4-year and 6-year OAS and EFS in Meyers study [31]. To simplify the calculation, we extracted 6-year OAS and EFS from Meyers study for meta-analysis. Compared with regimen without IFO, the regimen including IFO in preoperative chemotherapy does not improve the OAS (HR: 0.98, 95% CIs: 0.91-1.06, P=0.6) and EFS (RR: 1.05, 95% CIs: 0.9-1.22, P=0.57). When 4-year data from Meyers study were used, the results in meta-analysis were similar. Therefore, at least in terms of OAS and EFS, there were no significant improvements by including IFO in the drug combination for treating osteosarcoma.

Further, previous studies of primary and metastatic osteosarcoma showed that the degree of necrosis observed with neoadjuvant chemotherapy can be used to predict overall survival [36-38]. In these included studies [31, 33], there were no significant difference between two regimens in terms of response rate. Since no meta-analysis were conducted due to different definitions of response rate used in these two studies, it will be interesting to see RCTs using a definition as recommended by either study for further comparison.

Toxicity is an essential parameter to assess the safety and efficacy of chemotherapy in treating osteosarcoma. The follow-up time in both studies were long enough (at least 66 months as the median) to assess the toxicity-related events. In Meyers study [31], we found toxicity-related description from its preliminary reports [30]. Principle toxicities were ALT elevation, stomatitis and infection with rare renal dysfunction. For most of the toxicities reported, there was no statistically significant difference between two regimens in this study. In contrast, Ferrari study [33] reported that there were significant higher incidences of hematological toxicity in regimen including IFO in preoperative treatment. Even comparison is made only considering patients who received IFO postoperatively, the hematological toxicity in 4-drug preoperative chemotherapy regimen was still with statistically higher incidence. The hematological toxicity reported in this study includes grade 4 leukopenia, grade 4 thrombocytopenia, RBC transfusion, PLT transfusion, G-CSF, neutropenic fever and hospitalization. In light of these results, although it is still premature to draw a conclusion that inclusion of IFO will increase toxicity incidence, we need be cautious about using IFO in preoperative chemotherapy for less adverse events.

Telling from primary outcome and secondary outcomes, the results did not favor the inclusion of IFO in preoperative chemotherapy. Generally speaking, these included studies were of good quality and reported most essential parameters for assessing the efficacy and safety of including IFO in preoperative chemotherapy. Also, these included studies had relatively large sample size and long enough follow-up time. Therefore, this systematic review does not support the argument that IFO should be included in preoperative chemotherapy for treating osteosarcoma, at least with the doses and schedules used in the included studies.

Admittedly, this study is not totally free of limitations. One possible limitation of this meta-analysis is the relatively poor performance in risk of bias assessment. The risk of bias in included studies was difficult to assess due to a lack of reporting. More studies with clear descriptions for risk of bias assessment will further advance our understanding in this topic. Another potential limitation is the selection bias which cannot be avoided by all systemic

reviews. Besides mining the database, we also conducted hand-searching to minimize such bias. In addition, the number of studies included in meta-analysis may be relatively small, which might yield bias, for instance, generalizability, confounder adjustment and cohort effects, in the outcomes.

In addition, although this systemic review did not support the inclusion of IFO in preoperative chemotherapy for treating primary osteosarcoma, there were clinical evidence to support the use of IFO postoperatively [17, 29, 39]. Therefore, it will be interesting to review related evidence and elucidate the role of IFO in postoperative chemotherapy.

Disclosure of conflict of interest

None.

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