Clinical Research Paper

Intra-arterial hepatic bio-chemotherapy for the treatment of melanoma patients with liver metastasis

A phase II clinical study

Chuan-Liang Cui, Zhi-Hong Chi, Xiang-Qing Yuan, Hong-Yun Lian, Lu Si and Jun Guo*

Department of Melanoma and Renal Cancer; The School of Clinical Oncology; Peking University; Beijing Institute for Cancer Research; Beijing, P. R. China

Key words: intra-arterial hepatic bio-chemotherapy, liver neoplasm/metastatic, melanoma, efficacy

Background and Objective: The therapeutic effect on melanoma metastasizing to liver is poor. Researches have demonstrated that hepatic intra-arterial bio-chemotherapy can improve the treatment efficacy of metastatic melanoma. This study was to investigate hepatic intra-arterial bio-chemotherapy for the treatment of patients with liver metastasis from melanoma. Methods: Twenty-one patients with liver metastasis from melanoma were treated with hepatic intra-arterial infusion of dacarbazine (250 mg/m^2) from the first to the fifth day, and fotemustine (100 mg/m^2) at the sixth and fourteenth day, followed by adoptive transfer of autologous cytokine-induced killer cells and administration of interleukin 2 and 150 ug granulocyte/macrophage colony stimulating factor for 10–12 days. The overall treatment was repeated every 28 days. The overall survival, response and toxicity were analyzed. Results: Seventeen of twenty-one patients were valuable. One achieved complete remission (CR), one achieved partial remission (PR), six had stable disease (SD) and nine had progression disease (PD). The disease control rate was 47.06% (8/17), with a median progression free survival (PFS) of three months and a median overall survival (OS) of six months. Treatment related complications were mainly myelosuppression (grade III–IV), occurring in 38.1% (8/21) patients. Conclusions: Hepatic intra-arterial chemotherapy can improve the disease control rate of progressive melanoma. It tends to prolong the PFS and OS with tolerable toxicity in patients with liver metastasis from melanoma.

Malignant melanoma is originated from neural crest melanocytes. It has high malignancy, early metastasis and poor prognosis. Around 20% of malignant melanoma patients have liver metastasis, whose median survival time is only four to seven months and one-year survival rate is only 10%. Systemic chemotherapy for patients with liver metastasis is not desirable with an effective rate of only 30%.

As liver metastasis progresses rapidly, effective management of liver metastasis may improve the prognosis of malignant melanoma patients. Hepatic intra-arterial infusion can increase the local concentration of chemotherapy drugs and decrease systemic toxicity. Additionally, tumors with liver metastasis are mostly supplied by the hepatic artery, so combined embolization can further prolong the contact time between the drug and the tumor. When the artery is perfused with fotemustine, the drug concentration can be eight to 47 times higher in liver than in other normal tissues, accompanied with a decrease of the area under the curve (AUC) of serum by about 50%. A retrospective study from M.D. Anderson Hospital and Tumor Institute has demonstrated that hepatic intra-artery chemembolization is currently an effective strategy for melanoma with liver metastasis, which results an effective rate of about 38%.

Given the favorable efficacy of adoptive transfer of autologous cytokine-induced killer (CIK) cell in combination with lymphodepleting chemotherapy for patients with melanoma metastasizing to liver, we designed the phase II clinical trial to investigate the impact of hepatic intra-arterial bio-chemotherapy on the treatment of patients with liver metastases from melanoma.

Data and Methods

Selection of subjects. Twenty-one patients with liver metastases from melanoma were enrolled in the study from July, 2003 to November, 2007. There were eight male and 13 female subjects in the study, ranging in age from 24 to 70 years (median, 53 years). According to the scoring scale of Eastern Cooperative Oncology Group (ECOG), the performance status scores were the patients were 0–3, with an average value of 1.45 ± 0.93. One patient scored three. The primary sites of melanoma included skin (8/21, 38.1%), eyes (8/21), gastrointestinal tract (4/21, 19%) and an unknown site (1/21, 5%). All primary sites were dissected by surgery and confirmed by pathology examination.

The time from dissection of the primary site to occurrence of liver metastasis varied from 0–204 months. The longest interval was 16 years after the surgery. Among the 21 patients, 11 had liver metastasis...
Intra-arterial hepatic bio-chemotherapy for the treatment of melanoma patients with liver metastasis

alone and the other 10 showed extra-hepatic metastasis, including lung (ten cases), lymph nodes (five cases), skin (three cases), bone (two cases), pancreas (two cases), pelvic cavity (one case), spleen (one case), adrenal gland (one case) and mammary gland (one case). All patients underwent dissection of the primary site of melanoma. Six patients were previously treated with systemic chemotherapy of dacarbazine, cisplatin or paclitaxel, and five with immunotherapy induced mainly by interleukin (IL)-2 and interferon (Table 1).

**Therapy protocol.** All patients showed no contraindications of chemotherapy and the metastatic areas accounted for less than 50% of the livers. No ascites was observed. Consent forms for interventional operation, chemotherapy and CIK cell therapy were obtained from the patients. The hepatic intra-arterial infusion pump (Celsite ZMPLATOPIX) was initiated via the femoral artery. One patient had inguinal lymph node metastasis, so the catheter was implanted via the left subclavian artery instead. The lymphodepleting chemotherapy was administered with specific procedures as follows: intra-arterial infusion of dacarbazine 250 mg/m² was administered from day 1 to day 5; then 4-h intra-arterial infusion of fotemustine 100 mg/m² was administered on day 6 and day 14, followed by adoptive transfer of autologous cytokine-induced killer (CIK) cells on day 7, day 14 and day 16. Before cells were infused for the first time, phenergan (25 mg) and dexamethasone (5 mg) were used for pretreatment. After autologous cells were infused, intravenous bolus of IL2 (two million unit per day) and subcutaneous injection of granulocyte/macrophage colony stimulating factor (GM-CSF) (150 μg per day) were administered for 10 to 12 days. If reinfusion of CIK cells showed no obvious adverse effects at the first time, the dose of subsequent pretreatment of phenergan was reduced to 25 mg per day, a cycle of 28 days. Amplification of CIK cells was performed using GM-CSF 150 μg per day for 5 days. The blood cell separator (CS-3000 plus) was used to collect peripheral blood mononuclear cells. The circulatory blood volume was 8000 mL. Mononuclear cells were cultured in the RPMI-1640 medium supplemented with 10% serum extracted from the patients and incubated for 1 h in the incubator with 5% CO₂ at 37°C. Suspended cells in culture bottles were sucked, followed by addition of CIK complete medium containing 1000 U/mL ILγ. After cells were cultured for 24 h, the mouse anti-human CD3 monoclonal antibody (100 ng/mL), IL2 (1000 U/mL) and IL1α (1000 U/mL) were added into the culture medium. Cell growth conditions were observed daily, and the medium was changed every two days with addition of fresh IL2. On day 7, 14 and 16 of cell culture, CIK cells were collected.

**Measurement of immunophenotypes of CIK cells:** the amount of separated mononuclear cells was more than 2.5 x 10⁸. Before infusion, the fluorescein isothiocyanate (FITC) conjugated mouse anti-human CD3 monoclonal antibody and phycoerythrin (PE) conjugated mouse anti-human CD 56 monoclonal antibody were used for double staining for CIK cells. The percentages of CD 3⁺CD56⁺ cells should be over 80%, and the total infused cells per week should be more than 1.6 x 10¹⁰ to guarantee therapeutic efficacy.

**Evaluation and follow-up.** The CT scan was performed on the target lesions whose diameters were greater than 1 cm, such as multiple lesions. Five lesions from one organ and ten lesions from the whole body were chosen. After one or two cycles of treatment, CT was performed again to evaluate the efficacy. The efficacy was confirmed 4 weeks later. The efficacy evaluation was performed according to the Response Evaluation Criteria in Solid Tumors (RECIST). The disease control rate was calculated as the proportion of patients presenting complete response (CR), partial response
Results

After treatment, the erythrocyte sedimentation rate (CSR) were measured before and after treatment.

Statistical analysis. The primary end point of this study was overall survival (OS), which was defined as time from diagnosis of liver metastases to death. The secondary end point was progression free survival (PFS), which was defined as time from starting the treatment for liver metastasis to disease progression.

Results and adverse effects. Patients underwent the treatment for an average of 2.2 ± 1.2 cycles, and the longest one was five cycles. The main adverse effects resulting from chemotherapy included grade III and IV myelosuppression (38.1%, 8/21) and grade I and II hepatic toxicity (9/21, 42.85%). Grade I and II gastrointestinal reactions were controlled by medication. Five cases showed grade IV myelosuppression, with the major manifestation of thrombopenia (PLT < 25 x 10^9/L). The hemogram findings returned to normal after symptomatic treatment.

Evaluation of efficacy. Seventeen patients were suitable for evaluation, among which one achieved CR, one achieved PR, six achieved SD and nine were progressed. The disease control rate of these patients was 20%. The disease control rate was 47.06% (8/17). The patient who finally achieved CR showed liver metastasis after operation of plantar melanoma. Systemic chemotherapy of dacarbazine, cisplatin and etoposide, in conjunction with IL2 interferon for six cycles were given, followed by one time radio frequency treatment; subsequently, hepatic intra-artery embolization was performed when the disease progressed. The tumor shrunk significantly after two courses of chemotherapy, and was removed by surgery. No tumor cells were found by the pathological examination after operation. The patient achieved clinical CR. The patient who finally achieved PR had liver metastasis after operation for thumb melanoma. He was treated with dacarbazine and fotemustine using the hepatic intra-artery pump for two cycles before evaluating efficacy. After three cycles, efficacy was evaluated and the patient achieved CR (Table 1). Among nine patients whose disease progressed, three had stable liver lesions.

Discussion

The medium PFS was three months (ranged 1–12 months) and the medium OS was six months (ranged 3–20 months). Some patients did not reach the end point of the follow-up. The medium PFS of eight patients who achieved effective efficacy reached four months (ranged 2–12 months) and their medium OS reached nine and a half months (ranged 3–20 months). The median follow-up time was 10 months (ranged 3–21 months) until November 2007. The patient who achieved CR had tumor free survival and five patients survived with tumors.

The common metastatic sites of advanced melanoma included the skin, lung, liver, brain, and so on, of which liver metastasis accounted for around 20%. The prognosis of patients with liver metastasis is poor, with the median survival of four and half to five months, one-year survival of 10%, and predicted five-year survival of only 3%. Effects of conventional chemotherapy and immunotherapy are not favored by researchers, while the effective rates of dacarbazine, cisplatin, Carmustine, IL2 interferon alone, or in combination are only about 3% to 5%, which fail to prolong the survival.

In order to improve the treatment efficacy for patients with advanced melanoma, researchers have carried out a large number of studies. According to the report of Dudley et al. from National Cancer Institute of America, favorable effects were obtained by combining lymphodepleting chemotherapy with adoptive cell transfusion using tumor-infiltrating lymphocytes (TILs). Chemotherapeutic drugs were used to reduce the number of lymphocytes in the blood circulation, thus to enhance the antitumor effect of specific lymphocytes during adoptive cell transfer. IL2 was administered in the process of adoptive transfer to promote maturity and differentiation of tumor killer lymphocytes, and maintain their antitumor activity. Decrease of metastatic tumor mass was observed in 51% (18/35) patients in different metastatic sites, including liver, lung, brain, lymph nodes, and skin. Fifteen of them achieved PR with a period of two months to above two years. The result reveals promising application prospects of chemotherapy combined with acquired immunotherapy.

Fotemustine is a new drug belonging to the group of nitrosourea. It shows an effective rate of 24% for patients with diffuse melanoma, but a reaction rate of only 8.8% for melanoma patients with liver metastasis. Because of its short half-life period and obvious first-pass effect, the drug concentration of fotemustine is eight to 47 times higher in the liver than in normal tissues. The AUC for serum fotemustine is decreased by about 50% after hepatic arterial infusion of...
Intra-arterial hepatic bio-chemotherapy for the treatment of melanoma patients with liver metastasis

the drug.

Hepatic infusion of fotemustine increased the response rate for chemotherapy to around 40% (12/30) and that for liver lesions to about 60%. Patients had PFS of 11 months, OS of 14 months, and two-year survival rate of 29%. Additionally, the therapeutic efficacy was not found to be related to the primary site of melanoma. Fotemustine was as effective for liver metastasis from cutaneous melanoma as for that from ocular melanoma.

In order to further improve efficacy, researchers employed hepatic arterial chemotherapy together with nimustine, vincristine and cisplatin to treat melanoma. They observed that the liver tumor metastasizing from melanoma shrank obviously and no serious adverse effects appeared. Although the combined chemotherapy displays a favorable result, clinical trials with a large sample size are needed to verify the effect.

Currently, dacarbazine is the first choice of doctors using chemotherapy to treat melanoma. Its combination with fotemustine can increase the drug sensitivity. In our study, the disease control rate was 47.06% (8/17) and one patient achieved CR. Another patient with multiple lesions showed 75% decrease of metastatic tumors after three cycles of treatment.

Hepatic intra-arterial bio-chemotherapy increases the disease control rate by increasing the drug concentration after a change of the drug administration approach. The combination of dacarbazine with fotemustine not only kills tumor cells, but also depletes lymphocytes, thus to improve the immune tolerance of the body. Therefore CIK cells could exert better immunotherapeutic functions.

In our study, six patients were still alive by the end of the follow-up. The medium PFS and OS for patients were three and six months, compared with 11 months and 14 months, respectively reported by Peter et al. Shorter survival in our study may be caused by patient selection. Most of our patients had multi-organ metastasis and had a large tumor burden. In addition, the dose used in our study was lower. Moreover, some patients did not reach the end point.

In our previous study, when chemotherapy alone or chemotherapy combined with IL2 and interferon was used for patients (n = 5) with liver metastasis, the PFS was about one and a half months and the survival time was only four months. Reports from M.D. Anderson Cancer Center and ECOG found that the survival time for patients with melanoma metastasizing to liver was four and a half months to five months after chemotherapy or immune therapy alone. We observed significantly higher median PFS and OS in the 17 patients compared with the results from Albert et al., PFS and OS of eight patients who achieved efficacy were further prolonged. However, it is reported that, although intra-arterial chemotherapy combined with systemic chemotherapy could increase the treatment efficacy (21.7 vs. 8%) for eye melanoma patients with liver metastasis, the survival condition was not improved. The OS was 369 d vs. 349 d.

The adverse effects of hepatic intra-arterial bio-chemotherapy were similar to those of systemic therapy, including myelosuppression, liver toxicity and gastrointestinal reactions. Gastrointestinal reactions and liver toxicity were mostly minor to moderate. The rate of grade III–IV myelosuppression was 38.1% (8/21), mainly thrombocytopenia. Six patients received systemic chemotherapy of different regimens for two to four cycles, which might be the reason of myelosuppression. Our ratios were similar to the reported results of grade III–IV myelosuppression, 36% of neutropenia and 15% of thrombocytopenia. During treatment, no side effects caused by the placement of an intra-arterial catheter appeared, such as catheter dislocation, leakage and thrombosis. The placement of catheter did not influence the life quality of patients. Khayat et al. also reported similar findings.

Liver metastasis of some patients in our study was under control, but the extra-hepatic metastases continued to progress. Khayat et al. noted that the extra-hepatic progression rate could reach as high as 46.1%. The intra-arterial hepatic pump was not successfully placed in some patients due to their vessel abnormality. This problem needs to be solved in the future.

For metastatic melanoma, the initial site of metastases, disease-free survival before distant metastases, and the initial staging of the disease are three independent prognostic factors. However, for liver metastasis from melanoma, reported prognostic factors vary in different studies. Disease-free survival before distant metastases and the level of serum LDH are generally accepted as the main independent factors for survival of this disease. Studies with a large number of samples have shown that the survival of melanoma patient with liver metastasis is not correlated with age, gender, extra-hepatic metastasis, and the treatment regimen. Because of the small sample size, we did not analyze the prognostic factor for survival.

Currently used isolated hepatic infusion (IHP) is an effective local management for liver metastases. It is the priority strategy for patients with highly invasive hepatic metastasis (multiple tumors, big size and fast progression). Recent clinical trials exhibit that the anti-tumor efficacy of IHP is about 60%, with PFS of about 10–11 months. But IHP-associated mortality rate is 5%. Compared with IHP, hepatic intra-arterial infusion is simpler, better tolerated and cheaper, thus it becomes the first choice for patients with liver metastases from melanoma in China.

In conclusion, hepatic intra-arterial bio-chemotherapy has a potent therapeutic effect for melanoma metastasizing to liver, and may improve the outcome of those patients without serious side effects.

References


Intra-arterial hepatic bio-chemotherapy for the treatment of melanoma patients with liver metastasis

