Chapter III

- Hyperthermic isolated regional perfusion of the limb with carboplatin

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Introduction

Hyperthermic isolated regional perfusion of the limb (ILP) is a technique which has the possibility to deliver high concentrations of chemotherapeutic agents to an extremity, whereas only low drug concentrates are reached in the systemic circulation. Since Luck introduced melphalan (L-phenylalanine mustard) in 1956 as an active agent to inhibit growth of malignant melanoma in mice, it has been the preferred agent in ILP for intransit metastases of melanoma.\(^1\) Although ILP with melphalan is effective in the treatment of the patients with intransit metastases of melanoma, with a complete response rate of 50%, it has no effect in the adjuvant setting.\(^2-4\) Newer drugs which might be more effective than melphalan in the treatment of melanoma by ILP have therefore been sought. Cisplatin is one of the most active drugs in cancer treatment, however it has serious dose-limiting toxicity such as neuro-, nephro-, and ototoxicity.\(^5\) Therefore ILP with cisplatin was evaluated as an alternative to overcome these limitations of systemic treatment in the treatment of recurrent melanoma of the limb. However the effectiveness of cisplatin in the so called therapeutic perfusion setting and for local control of tumor growth for the treatment of locally advanced melanoma and sarcoma of the extremities with delayed excision is not impressive.\(^6-10\) And the local neurological side effects of ILP with cisplatin in the perfused limb are still present in the form of a irreversible peripheral sensory and motor neuropathy.\(^7\) Therefore in several institutions cisplatin was abandoned as a cytostatic agent in the perfusion setting. There are however several other institutions that still use and advocate cisplatin in ILP.\(^11-13\) Carboplatin is a second generation platinum analogue with similar efficacy in most tumors but with less neuro-, oto- and nephrotoxicity when compared to cisplatin when applied systemically.\(^14,15\) A feasibility study was initiated to study the effect of carboplatin, as an agent for ILP with respect to the local tumoricidal effect, as well as the local treatment morbidity.

Patients and Methods

Between November 1991 and February 1993 two patients (patient 1 and patient 2) with locally advanced recurrent and intransit metastases of melanoma of the lower
limb (Stage IIIA, MD Anderson staging) and one patient (patient 3) with a locally advanced primary irresectable soft tissue sarcoma of the lower limb participated in a feasibility study of ILP with carboplatin. None of the patients had distant metastases at time of the perfusion. Patients were treated with intent of preserving the affected extremity. The maximum follow up period was 95 months. Patient data are summarized in Table 1. Since the systemic dosage of carboplatin used clinically is 4 times the cisplatin dosage, a fourfold higher dose compared to cisplatin was used for the limb perfusion. Based upon an earlier dose finding study where a maximum dose of cisplatin of 30 mg/L limb volume was used in ILP, an initial carboplatin dosage was chosen of 125 mg/L limb volume.

<table>
<thead>
<tr>
<th>Patient No.</th>
<th>Age (yrs)/Sex</th>
<th>Tumor type and staging*</th>
<th>Previous therapy</th>
<th>Perfusion level</th>
<th>Limb vol (l)</th>
<th>Total carboplatin in mg</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>57/F</td>
<td>melanoma stage IIIA</td>
<td>tumor excision + ILP with melphalan + actinomycin-D</td>
<td>popliteal</td>
<td>5.1</td>
<td>625 mg</td>
</tr>
<tr>
<td>2</td>
<td>72/M</td>
<td>melanoma stage IIIA</td>
<td>tumor excision</td>
<td>femoral</td>
<td>12</td>
<td>1500 mg</td>
</tr>
<tr>
<td>3</td>
<td>52/M</td>
<td>low grade myxoid liposarcoma</td>
<td>None</td>
<td>iliacal</td>
<td>13</td>
<td>1625 mg</td>
</tr>
</tbody>
</table>

*Staging according to M.D. Anderson staging system

Table 1: Patient characteristics, perfusion characteristics and carboplatin dosage
All patients had normal pre-operative kidney function. To prevent postoperative renal failure due to systemic leakage, hyperhydration was started one day before ILP and was continued up to 5 days postoperatively. Hyperhydration consisted out of 2 l 0.9% NaCl intravenously superfluous to the patients’ habitual fluid intake. The perfusion technique used at the Groningen University Hospital is based on the technique developed by Creech et al.\textsuperscript{17} All patients were operated under general anaesthesia. After a skin incision, the iliac, femoral or popliteal vessels were exposed and collateral vessels were clipped. The patients were heparinized (3.3mg/kg body weight) and catheters were inserted in the artery and vein, and both were connected to an extracorporeal circuit. The extremity was completely isolated from the central circulation with the aid of an Esmarch bandage. The perfusate consisted out of 250 ml red blood cell concentrate, 250 ml Isodex\textsuperscript{®} (NPBI, Emmer-Compascuum, The Netherlands) in 0.9% NaCl, 30 ml NaHCO\textsubscript{3} 8.4% and 25 mg heparine (2500 IU) (Braun Melsunger AG, Melsunger, Germany). Leakage to the systemic circulation was assessed by injecting I\textsuperscript{131} - albumin into the isolated circulation and continuously monitoring of radioactivity was performed by means of a NAC-scintillation detector, which was placed over the heart.\textsuperscript{18} The leakage was indicated by extent of cross circulation between the perfused extremity and the circulation in the rest of the body. All perfusions were performed under mild hyperthermic conditions (39 - 40°C). If the muscle temperature had reached a temperature of 38°C and after leakage to the systemic circulation was excluded, the carboplatin 125 mg/L limb volume (Bristol Myers Squibb Company, Woerden, The Netherlands) was administered to the perfusate over a 10 min period. The flow rate of the perfusion fluid in the perfusion circuit was approximately 500 ml/min. After 1 hour of perfusion, the extremity was flushed with 500 ml Isodex\textsuperscript{®} in 0.9% NaCl and 250 ml red blood cell concentrate. Both catheters were removed and the vessels were repaired. The heparin was then neutralized with protamine sulfate and a fasciotomy was performed to prevent a compartment syndrome. Before and during the perfusion, 10 ml perfusate samples were collected at 10 min intervals to determine the ultrafiltrated platinum (fPt) levels as previously extensively published.\textsuperscript{19} The leakage into the systemic circulation was also determined by measuring the patients fPt plasma concentrations at the end of each perfusion procedure both before and after restoration of the circulation to the perfused limb and during the 7 days following
the perfusion. For fPt perfusate elimination kinetics, data were subjected to logarithmic regression analysis (concentration = A.e\(^{-k.t}\)). The areas under the concentration vs. time curves (AUC) were calculated using the model independent trapezoidal rule and covers the perfusion period (t = 0 - 60 min). Tissue biopsies were taken for fPt determination in both melanoma patients at the end of the ILP, when the normal limb circulation was restored.

All three patients were followed up post operatively by history, physical examination, and routine blood counts and blood chemistry analysis were performed on a daily basis on days 1 - 7, and 14 and 21 days postoperatively. Total serum protein and albumin was controlled one day preoperatively and postoperatively. Six weeks after perfusion electrodiagnostic evaluations (electromyogram (EMG) and nerve conduction study) were performed to investigate nerve toxicity of the carboplatin perfusion in patients 1 and 3. Unfortunately patient 2 refused the neurological examination. The local limb perfusion toxicity was graded according to the criteria described by Wieberdink et al.

Eight weeks after perfusion, patient 3 was readmitted for a so called delayed resection of a locally advanced soft tissue sarcoma of the lower limb. The study was approved by the local Medical Ethical Committee of the Groningen University Hospital and all patients gave informed consent.

## Results

All three perfusions could be performed under standard perfusion conditions with minimal \(^{131}\)-albumin leakage (0-5%) (Table 2) and without any acute systemic toxicity or morbidity. The local toxicity consisted in all 3 patients of a grade II toxicity of the perfused limb which is characterized by erythema, edema or loss of sensation which resolved spontaneously in all patients. Patient 2 developed an abscess in the groin area of the perfused limb 3 weeks after the femoral perfusion with superficial inguinal node dissection. Except for a marked decrease in total serum protein and albumin 1 day postoperatively, there were no major biochemical changes or myelosuppression. The total serum protein decreased from a mean of 67.3 ± 2.5 g/L preoperatively to 47.7 ± 4.5 g/L (P<0.04 paired Student’s t-test) one day postoperatively. Serum albumin decreased from a mean of 46.3 ± 4.2
Table 2: Leakage, toxicity and neurological follow up

g/L preoperatively to 32.0 ± 4.0 g/L (P<0.002 paired Student’s t-test) one day postoperatively. This resulted in the development of edema in the perfused limb postoperatively for which intravenous albumin was administered postoperatively in all three patients to correct the serum albumin. Physical examination of the first two patients, both melanoma intransit metastases, revealed clinically a complete response making delayed excision unnecessary. A CT scan of the liposarcoma of patient 3 showed a 25% reduction of the tumor size which allowed a complete macroscopical marginal resection after 8 weeks. However microscopically vital tumor was observed in at least 50% of the resected material with focal necrosis. The patient received adjuvant radiation therapy (60 Gy) at the primary tumor site, which was well tolerated. Clinically all 3 patients showed signs of sensory and motor neuropathy in the perfused limb. Electrodiagnostic evaluations were performed in order to assess the local neurotoxicity of the carboplatin in patients 1 and 3. Patient 1 was tested both pre- and postoperatively because this patient underwent an ILP with melphalan of the same limb one year previously. Preoperative nerve conduction velocities of the perfused left leg (peroneal nerve, F
response, H reflex) were all slightly slower then on the right side. The sensory sural nerve amplitude was also lower on the left side (left 4µV, right 16µV). The control EMG and nerve conduction study postoperatively showed a further decrease in the conduction velocity of the peroneal nerve and the amplitude from the sural nerve decreased by 2µV. The postoperative EMG and nerve conduction study of patient 3 revealed that the right (perfused limb) extensor hallucis muscle and abductor hallucis muscle showed partial denervation, and the sural nerve also showed a lower amplitude compared with the left leg.

In all three patients the affected limb could be preserved. After a complete initial clinical response to the perfusion, patient 1 developed local recurrences over the next few years and the first being within 1 year postperfusion. All of these local recurrences and intransit metastases were treated by local excision only. This patient is still alive 95+ months after perfusion with carboplatin. Patient 2 also showed a complete initial clinical response but within 1.5 year a local recurrence appeared at the primary tumor site which was excised. This patient went on to develop lung metastases 44 months after perfusion and died 56 months after perfusion. Patient 3 did not develop a local recurrence after radiation therapy but died from lung metastases 31 months after perfusion. Systemic plasma concentrations of fPt, determined at the end of perfusion before restoration of the normal circulation, were also found to be relatively low (<0.1 - 0.98 mg/L). Systemic plasma fPt concentrations remained negligible in all patients during the 7 days following the perfusion. Perfusion data and pharmacokinetic parameters are summarized in Table 1 and Table 3. Concentrations of fPt in tissue biopsies, taken from patients 1 and 2 of the skin and the anterior tibial muscle at the anterolateral part of the lower extremity during the fasciotomy after ILP, showed a great variation in concentration between the skin and muscle, with especially high fPt concentrations in the skin. In patients 1 and 2 the concentrations in the skin were respectively 27.6 ug fPt g\(^{-1}\) tissue and 25.0 ug fPt g\(^{-1}\) tissue compared to the concentrations in the muscle which were respectively 9.0 ug fPt g\(^{-1}\) tissue and 7.6 ug fPt g\(^{-1}\) tissue.
<table>
<thead>
<tr>
<th>Patient</th>
<th>t½ (min)</th>
<th>Cmax (mg/L)</th>
<th>C\textsubscript{t=60} (mg/L)</th>
<th>AUC ((mg/L)/min)</th>
<th>systemic fPt concentration (mg/L)</th>
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<tbody>
<tr>
<td>1</td>
<td>104</td>
<td>488</td>
<td>344</td>
<td>24070</td>
<td>&lt;0.1</td>
</tr>
<tr>
<td>2</td>
<td>30</td>
<td>824</td>
<td>113</td>
<td>9995</td>
<td>0.98</td>
</tr>
<tr>
<td>3</td>
<td>50</td>
<td>620</td>
<td>250</td>
<td>20378</td>
<td>&lt;0.1</td>
</tr>
</tbody>
</table>

Table 3: Pharmacokinetic parameters of fPt in the perfusate during 60 min ILP with 125 mg/L carboplatin extremity (assuming first order kinetics) and systemic fPt concentration after 60 min ILP.

Discussion

Regional isolation perfusion was introduced to deliver large doses of cytotoxic drugs to the local tumor area while avoiding systemic toxicity. Melphalan is still the most frequently used antitumor drug in ILP for melanoma.\textsuperscript{6,8,22,23} Even with a high remission rate (80%), a great deal (50%) of the melanoma patients fail to achieve a durable complete response.\textsuperscript{2,3,23} Therefore a search was initiated for alternative drugs or the application of combination of agents to be used in ILP. Several cytostatic agents have been used besides melphalan, such as actinomycin D and DTIC (imidazole carboxamide). The feasibility of cisplatin in ILP has also previously been investigated by several institutions, but with conflicting results. Some authors claim that the local severe toxicity and the equal or less effectiveness does not justify the use of cisplatin.\textsuperscript{6-10} But there are also other institutions that still use and advocate the use of cisplatin in ILP.\textsuperscript{11-13} Carboplatin, a second generation platinum compound, was attractive for clinical use because of its reduced nephro-, neuro-, and ototoxicity than cisplatin when administered systemically.\textsuperscript{24} It differs in the molecular structure from cisplatin by replacing the two \textit{cis} chloride atoms by
a cyclobutane dicarboxylate molecule.\textsuperscript{5} Platinum co-ordination complexes inhibit tumor growth by their effects on DNA replication. Once bifunctionally bound to DNA, the mode of action of carboplatin is not different from cisplatin, but the platinum induced DNA lesions (DNA interstrand cross links) are formed at a slower rate with carboplatin compared to cisplatin.\textsuperscript{6} Our clinical results show that application of carboplatin in ILP led to the development of, or an increase in the neuropathy in the perfused limbs of all three patients. In two patients this was confirmed by electrodiagnostic evaluation. Patient 1 already showed clinical signs of neuropathy in the affected limb which was confirmed by EMG and nerve conduction study prior to the carboplatin perfusion. This neuropathy was probably due to the previous perfusion with melphalan. The postoperative tests after the carboplatin perfusion of this patient showed no apparent denervation potentials, but increased disturbances in the motor and sensory conduction velocities compared to the first neurological tests were observed. Patient 2 and 3 had no clinical signs of a neuropathy preoperatively. However, the postoperative electrodiagnostic findings of patient 3 did show a new development of a neuropathy distally in the perfused limb. With the limitations of the small number of patients treated in this feasibility study it should be noted that with respect to the locoregional tumor control, the treatment regimen did not appear to be promising. Both melanoma patients who underwent perfusion with carboplatin developed local recurrency and/or intransit metastases within 1 year after perfusion, and histologically 50\% vital tumor was still observed after excision of the sarcoma from patient 3.

There has only been one case report and one study in which carboplatin has been used for isolated limb perfusion. Nakayama et al. reported minimal side effects after perfusing 1 patient with 450 mg carboplatin for melanoma of the lower limb.\textsuperscript{25} They found a complete clinical and histologic response of the tumor and in their follow up period of 10 months, no local recurrency was observed. This is in concordance with the findings of Ariyan et al. who performed ILP on 20 patients with carboplatin (400 mg/m\textsuperscript{2} body surface area) and the results were found to be effective and with very low toxicity with a median follow up of 25 months.\textsuperscript{11} Both studies however do not report on observed local neurotoxicity. In our study the carboplatin dosage was calculated on the limb volume and not on the body surface area, thus leading to a much larger carboplatin dosis administered (up to 4 times
as much). This could possibly explain the enhanced neurotoxicity seen in this series.

Next to the search for new agents for ILP, different combinations of drugs are being investigated. The combination of tumor necrosis factor α (TNFα) and melphalan has been used for the local perfusion of recurrent melanoma of the limbs and for extremity sarcomas.⁶⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻吸入四 hopeless the enhanced neurotoxicity seen in this series.

Next to the search for new agents for ILP, different combinations of drugs are being investigated. The combination of tumor necrosis factor α (TNFα) and melphalan has been used for the local perfusion of recurrent melanoma of the limbs and for extremity sarcomas.⁶⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻⁻吸入四 hopeless the enhanced neurotoxicity seen in this series.

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Nakayama\textsuperscript{26} and Ariyan\textsuperscript{11} the patients in this study received a higher dose of carboplatin which could account for the observed neurotoxicity.
Reference list


