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Drug concentrations in axillary lymph nodes after lymphatic chemotherapy on patients with breast cancer

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Abstract

Background Lymph node status is one of the decisive prognostic factors in breast cancer. Chemotherapy targeting regional lymphatic tissues has emerged as a promising therapy for the treatment of malignancies with a high tendency to disseminate lymphatically. The present study determined the drug concentrations in axillary lymph nodes after lymphatic chemotherapy (LC) in patients with breast cancer and compared the results with those receiving intravenous chemotherapy (VC) to investigate whether LC could improve the accumulation of anticancer drug in regional lymph nodes.

Methods Sixty patients with breast carcinoma confirmed by preoperative puncture-biopsy were divided into two groups at random. The LC group (n = 30) received a subcutaneous injection of 4 ml of carboplatin-activated carbon suspension, containing 20 mg of carboplatin, adjacent to the primary tumour. The VC group (n = 30) received an intravenous administration of an equal dose of aqueous carboplatin. At 1, 12, 24, 36 and 48 hours after administration, modified radical mammectomies were performed on 12 patients at each time point, with 6 from each group. Axillary lymph nodes were removed for pathological examination. The platinum concentrations in nodes were determined by Zeeman atomic absorption spectrometry.

Results A total of 275 axillary lymph nodes were resected, with 154 in the LC group and 121 in the VC group. Of the 275 lymph nodes, 136 (49.5%) from 23 patients (38.3%) had histopathologically detected metastases. At 1, 12, 24, 36 and 48 hours after injection, the carboplatin concentrations in the LC group were 11.82 ± 3.50, 23.58 ± 7.34, 18.22 ± 4.93, 16.70 ± 5.15 and 14.62 ± 4.29 μg/g (means ± SD), respectively, whereas those in the VC group were 0.06 ± 0.02, 0.11 ± 0.05, 0.10 ± 0.02, 0.05 ± 0.02 and 0 μg/g, respectively. Significant differences were found in each corresponding comparison ($P <$ 0.001). Lymph node metastasis was uncorrelated with drug concentration ($P >$ 0.05).

Conclusion LC can effectively and continuously improve the drug concentrations in axillary lymph nodes in patients with breast cancer, in comparison with VC.

Keywords: breast carcinoma, drug treatment, lymph node

Introduction

Regional lymph nodes are a common site for the postoperative relapse of advanced breast carcinoma. To improve the prognosis, axillary lymph nodes are routinely resected in radical mammectomy. However, there are several paths through which lymph drains from the breast to adjacent tissues, as shown in recent studies on sentinel lymph node [1,2]. Radical mammectomy fails in excising lymph nodes located in the mediastinum, internal mammary or hepatic portal. A similar problem can also be found in cancers of the digestive tract, which also tend to metastasize lymphatically.

During recent years, some gastroenterologists have administered preoperative chemotherapy targeting regional lymph nodes for gastric, oesophageal, rectal or peritoneal cancer by using special drug carriers such as activated carbon or liposomes [3-6]. Their results showed promise for the prevention of metastasis and recurrence after radical resection. The new method for the treatment of likely metastasis to regional lymphatic tissue is called lymphatic chemotherapy (LC) or medicinal lymph node dissection. In the present study we administered carboplatin-activated carbon suspension (CP-CH) to patients with breast carcinoma and determined the platinum concentrations in

CP-CH = carboplatin-activated carbon suspension; CP-Sol = carboplatin solution; LC = lymphatic chemotherapy; VC = intravenous chemotherapy.

axillary lymph nodes at different intervals. The results were compared with those obtained after intravenous chemotherapy (VC) with an equal dose of carboplatin solution (CP-Sol) to discover whether LC could effectively improve the anticancer drug accumulation in regional lymphatic tissues.

Materials and methods

Patients and study design

From June 2002 to August 2003 a total of 60 female patients with breast carcinoma confirmed by preoperative puncture-biopsy were enrolled in this study. The ages of patients ranged from 35 to 71 years (mean 49.2 years). The pathological diagnoses were as follows: ductal ($n = 28$), lobular ($n = 18$), medullary ($n = 6$), mucinous ($n = 4$), tubular ($n = 3$) and papillary ($n = 1$) carcinoma, respectively. According to the TNM classification (*AJCC Cancer Staging Manual*, 6th edition, 2002), 11 patients fell into the T_1 category, 38 into T_2, 8 into T_3 and 3 into T_4. Axillary lymph node metastases were present in 23 patients; of these, 17 were classified as N1, 5 as N2 and 1 as N3. Distant metastases occurred in four patients. Clinical grades consisted of grade I ($n = 9$), grade II ($n = 40$), grade III ($n = 7$), and grade IV ($n = 4$). Locations of tumours were as follows: subnipple ($n = 5$), outer-upper ($n = 21$), outer-lower ($n = 14$), inner-upper ($n = 11$) and inner-lower ($n = 9$) field. The inclusion criteria included the following: World Health Organization status of 0 or 1, without previous chemotherapy or radiation therapy, white blood cell count greater than 4000/μl and platelet count greater than 100,000/μl, no abnormality of cardiac, renal or liver function. The patients who met these criteria were divided into two groups in accordance with a prospectively generated randomization schedule, with 30 patients in each group, to receive either LC or VC as preoperative adjuvant treatment. The study protocol was approved by our institutional review board as being in accordance with the Declaration of Helsinki. Informed consent was obtained from every patient before enrolment.

Preparation of CP-CH

Activated carbon granules (CHR-30) were ground in a ball-mill and sifted with a 100-mesh sieve. The 100-mesh passing fraction, 81% of which was smaller than 40 μm, was used for this preparation as activated carbon particles. After the moisture had been removed from the activated carbon particles by dry heating at 120°C for 2 hours, 375 mg of activated carbon particles was sealed in a glass tube, which was sterilized under two atmospheres of pressure for 10 minutes and stored in a refrigerator. Just before use, 100 mg of carboplatin dissolved in 20 ml of normal saline was added to the activated carbon particles. The mixture was shaken for 10 minutes to allow sufficient adsorption of carboplatin on the activated carbon particles, resulting in CP-CH [6].

Determinations of tissue platinum

Patients in the LC group received a subcutaneous injection of 4 ml of CP-CH, containing 20 mg of carboplatin, adjacent to the primary tumour. The VC group received an intravenous injection of 4 ml of CP-Sol containing 20 mg of carboplatin. Modified radical mammectomies were performed 1, 12, 24, 36 and 48 hours later on 12 patients, with 6 from each group each time. Axillary lymph nodes were excised entirely, one of which was picked at random and split into two parts: one part was sent for platinum examination, and the remaining part and the rest of the lymph nodes were sent for histopathological observation. The lymph node for platinum examination was cut to a fragment weighing 200 mg and placed in a 5 ml screw-capped tube. The fragment was left to stand overnight with 4 ml of 10% hydrochloric acid and 70% nitric acid (1:4, by volume). The following day, tissue was digested for 2 hours at 90°C in an electric frying pan positioned in a fume hood. The solubilized tissue was dried and mixed with 0.5 ml of Milli-Q water. The solution was subjected to graphite-furnace Zeeman atomic absorption spectrometry. Platinum analysis was performed as described previously [7]. The concentration of carboplatin was determined by multiplying the concentration of platinum by 1.903.

Statistical analysis

Concentrations are shown as means ± SD. Results were tested for significant differences by Fisher's test and Student's *t*-test. $P < 0.05$ was considered to indicate statistical significance.

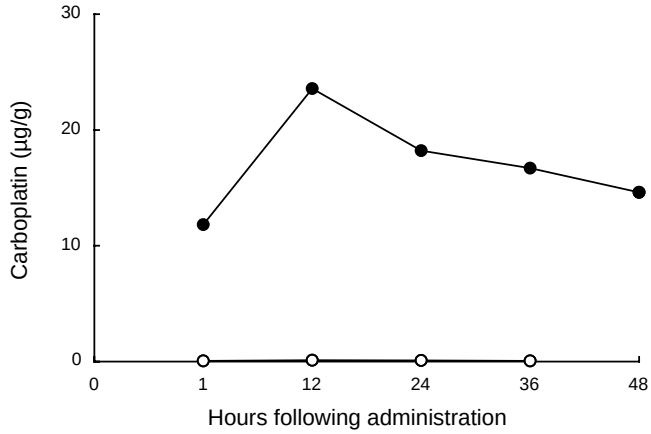
Results

A total of 275 axillary lymph nodes were resected during operations, with 154 in the LC group and 121 in the VC group. Of the 275 lymph nodes, 146 (53.1%) from 23 patients (38.3%) had histopathologically detected metastases. Of these 146 metastasized nodes, 80 were from 13 patients in the LC group and 66 were from 10 patients in the VC group. No statistical difference in case distribution was found ($P > 0.05$) between the two groups.

At 1, 12, 24, 36 and 48 hours after local injection of CP-CH, the mean carboplatin concentrations in axillary lymph nodes were 11.82 ± 3.50, 23.58 ± 7.34, 18.22 ± 4.93, 16.70 ± 5.15 and 14.62 ± 4.29 μg/g, respectively, whereas the concentrations were 0.06 ± 0.02, 0.11 ± 0.05, 0.10 ± 0.02, 0.05 ± 0.02 and 0 μg/g, respectively after intravenous injection of CP-Sol (Fig. 1). The difference was significant in each corresponding comparison ($P < 0.001$).

Among the 60 lymph nodes that were sent for platinum determination, 19 (31.7%) had histopathologically detected metastases. Of these 19 nodes, 11 belonged to the LC group with the mean carboplatin concentration of

Figure 1



Mean carboplatin concentrations in axillary lymph nodes at different times after administration. Filled circles represent concentrations after administration of carboplatin-activated carbon suspension; open circles represent concentrations after administration of carboplatin solution.

15.48 ± 6.10 μg/g. Of the remaining 41 nodes without metastasis, 19 were from the LC group with the mean carboplatin concentration of 17.24 ± 7.82 μg/g. No significant difference was found ($P > 0.05$) between them.

Discussion

Regional lymphatic tissues are often involved in advanced breast carcinoma. Lymph nodes adjacent to the primary tumour are susceptible to metastases, which usually lead to recurrences and distant metastases after curative resection. However, the traditional therapies for breast carcinoma are not satisfactory in solving this problem. Mediastinal, parasternal or parahepatic lymph nodes, which are likely to be affected by metastases because they receive lymph drainage from breast tissues, cannot be resected in radical mammectomy. Even lymph nodes and lymphatic vessels located in the axilla can rarely be dissected entirely. Furthermore, the destruction of lymphatic tissues during operation can bring about the dissemination of tumour cells in the operating field. Radiotherapy cannot effectively improve the result either, because of the limitation of the radiation field. VC supplies an extensive distribution of anticancer drugs. Unfortunately, high-dose anticancer drugs are often associated with acute toxicity, such as myelosuppression and immunosuppression, as well as with cumulative cardiotoxicity. As a result, only limited quantities of drugs are thereby distributed to lymphatic tissues.

LC, as a potential method for solving this problem, has been proved to have preventive effects on lymphatic metastases of gastroenteric cancers [3-6]. In the process, activated carbon and liposomes have been most extensively explored for their excellent characteristic of transferring anticancer drugs to regional lymphatic tissues after local administration [8].

We used activated carbon to deliver carboplatin in the present study. Activated carbon particles have numerous micropores that can adsorb large amounts of anticancer agents, such as mitomycin C, methotrexate, peplomycin or carboplatin. Once injected into the connective system, they are taken up selectively by lymphatic vessels and delivered to neighbouring lymph nodes. The lymph nodes then turn black, which simplifies their identification during radical lymphadenectomy [9].

Our results confirmed that subcutaneous injection of CP-CH adjacent to the primary tumour could provide a specific and continuous delivery of carboplatin to axillary lymph nodes. Carboplatin concentrations after regional administration of CP-CH were markedly higher than those after intravenous administration of CP-Sol at each sampling time. The mean drug levels at 1, 12, 24 and 36 hours after administration of CP-CH were 197, 214, 182 and 334 times higher than the corresponding drug levels after administration of CP-Sol.

On the other hand, drug determination at different sampling times indicated that CP-CH evidently slowed down the clearance of carboplatin in lymph nodes. It is well known that both prolonging the duration and elevating the concentration of carboplatin improves its anticancer efficacy [10].

The mean carboplatin concentration of lymph nodes without metastasis (17.24 μg/g) was slightly higher than that of lymph nodes with metastasis (15.48 μg/g), but the difference was not statistically significant. The result indicated that metastatic tumour cells in nodes exerted little influence on the accumulation of CP-CH at lymph nodes.

No haematological or non-haematological toxicities were detected in this experiment owing to the small dose of carboplatin used on its own. No evidence of local toxicity such as drug allergy, ulceration or erosion of skin was observed from CP-CH injection to operation, except that there were some carbon particles and inflammatory cells gathering at the injection sites, as detected pathologically, suggesting that the clinical application of CP-CH could be safe.

Conclusions

In conclusion, our study shows that LC could effectively and continuously improve concentration of anticancer drug in regional lymph nodes adjacent to the primary tumour in comparison with intravenous administration. Further investigations are warranted to reveal whether preoperative LC is useful in preventing the recurrence of cancer and in improving survival after mammectomy.

Competing interests

None declared.

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