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Effect of Xiao Chaihu Tang combined with intravenous chemotherapy on tumor markers and immune function in patients with advanced breast cancer

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ABSTRACT

Objective: To study the effect of Xiao Chaihu Tang combined with intravenous chemotherapy on tumor markers and immune function in patients with advanced breast cancer. **Methods:** 76 patients with advanced breast cancer treated in our hospital between May 2012 and November 2015 were collected and divided into the combined treatment group (n=34) who accepted Xiao Chaihu Tang combined with intravenous chemotherapy and the control group (n=42) who accepted intravenous chemotherapy alone according to different treatment, and the treatment cycle was 3 months for both groups. Before treatment and 3 months after treatment, ELISA method was used to detect serum levels of broad-spectrum tumor markers and breast cancer-specific tumor markers; flow cytometer was used to detect cellular immune function index levels, and turbidimetric immunoassay was used to detect humoral immune function index levels in peripheral blood. **Results:** Before treatment, differences in serum tumor marker levels as well as cellular immunity and humoral immunity index levels in peripheral blood were not statistically significant between two groups of patients (P>0.05); after 3 months of treatment, broad-spectrum tumor markers carcinoembryonic antigen (CEA), carbohydrate antigen 153 (CA153) and carbohydrate antigen 125 (CA125) levels in serum of combined treatment group were lower than those of control group, and breast cancer-specific tumor markers insulin-like growth factor-1 (IGF-1), midkine (MK), soluble E-cadherin (sEC) and thymidine kinase 1 (TK1) levels were lower than those of control group (P<0.05); $CD3^+$ and $CD4^+$ T lymphocyte levels as well as $CD4^+/CD8^+$ ratio in peripheral blood of combined treatment group were higher than those of control group while $CD8^+$ T lymphocyte level was lower than that of control group, and immunoglobulin G (IgG), immunoglobulin A (IgA) and immunoglobulin M (IgM) levels in peripheral blood were higher than those of control group (P<0.05). **Conclusions:** Xiao Chaihu Tang combined with intravenous chemotherapy can decrease the severity of advanced breast cancer and optimize the body's immune function.

1. Introduction

Breast cancer is the most common malignant tumor disease in female, the advanced patients that cannot detect it in time have missed the operation opportunity and can only choose conservative treatment[1,2]. Intravenous chemotherapy is the most common clinical conservative treatment, it reduces the malignant degree of tumor cells, but it also causes a certain degree of damage to the normal tissues and organs, and because of the severe immunosuppression, some patients even die of secondary infection[3]. Xiao Chaihu Tang is composed of radix bupleuri, pinellia ternata, ginseng, radix scutellariae, jujube and other Chinese patent drugs, it can harmonize the lesser yang, and it was mostly used for the treatment of malaria, chronic hepatitis and acute pancreatitis, etc[4]. Modern pharmacological studies have confirmed that Xiao Chaihu Tang has the effects such as anti-inflammation and immune regulation, and therefore, many scholars have recommended it for the adjuvant treatment of malignant tumor. In

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the following study, the effect of Xiao Chaihu Tang combined with intravenous chemotherapy on tumor markers and immune function in patients with advanced breast cancer was analyzed.

2. Materials and methods

2.1. General information

The treatment process and laboratory test results of 76 patients with advanced breast cancer treated in our hospital between May 2012 and November 2015 were retrospectively analyzed, the patients themselves signed the informed consent, and the research process was approved by the hospital ethics committee. According to different treatment, 76 patients were divided into the combined treatment group (n=34) who accepted Xiao Chaihu Tang combined with intravenous chemotherapy and the control group (n=42) who accepted intravenous chemotherapy alone. Combined treatment group were 37–78 years old, and the pathological types were as follows: 12 cases with invasive ductal carcinoma, 9 cases with invasive lobular carcinoma, 6 cases with medullary carcinoma, 4 cases with tubular carcinoma and 3 cases with others; control group were 38–76 years old, and the pathological types were as follows: 17 cases with invasive ductal carcinoma, 11 cases with invasive lobular carcinoma, 8 cases with medullary carcinoma, 4 cases with tubular carcinoma and 2 cases with others. Two groups of patients were not statistically different in age and tumor types (P>0.05).

2.2. Inclusion and exclusion criteria

Inclusion criteria: (1) tumor types and stages were confirmed by histopathology; (2) with primary breast cancer; (3) diagnosed for the first time and never accepting any other treatment before; (4) ≤80 years old; (5) accepting the entire treatment and with complete clinical data. Exclusion criteria: (1) with severe heart, liver and kidney dysfunction; (2) associated with autoimmune diseases; (3) associated with systemic infectious diseases; (4) pregnant or breastfeeding women.

2.3. Treatment methods

Control group of patients received intravenous chemotherapy alone, specifically as follows: 500 mg/m^2 each of cyclophosphamide (Hainan Jinrui Pharmaceutical Co., LTD., approved by H20113420) and fluorouracil (GrandPharma Huangshi Feiyun Pharmaceutical Co., LTD., approved by H20051137), by intravenous injection, on d 1 and d 8; adriamycin (Shanxi Pude Pharmaceutical Co., LTD., approved by H14023143) 30 mg/m^2, by intravenous injection, on d 1, 30 d as one treatment cycle, for continuous three cycles of treatment. Combined treatment group received Xiao Chaihu Tang combined with intravenous chemotherapy, specifically as follows: Xiao Chaihu Tang (10 g each of radix bupleuri, rhizoma alismatis, radix scutellariae and atractylodes, 15 g of dangshen, 20 g of tuckahoe, 5 g of cassia twig and 5 pieces of jujube), decocted with slow fire to 200–300 mL, 1 dose every day, divided into two and taken orally in the morning and evening, and 3 months as a course of treatment.

2.4. Observation indexes

2.4.1. Serum tumor markers

Before treatment and 3 months after treatment, 2 mL of peripheral venous blood was extracted from two groups of patients at the same point in time and centrifuged to get supernatant, detecting: (1) the broad-spectrum tumor markers: ELISA method was used to detect serum carcinoembryonic antigen (CEA), carbohydrate antigen 153 (CA153) and carbohydrate antigen 125 (CA125); (2) the breast cancer-specific tumor markers: ELISA method was used to detect insulin-like growth factor-1 (IGF-1), midkine (MK), soluble E-cadherin (sEC) and thymidine kinase 1 (TK1).

2.4.2. Immune function

2 mL of peripheral venous blood was extracted from two groups of patients in the same way, and flow cytometer (Beckman Coulter Commercial Enterprise Co., LTD., model Gallios) was used to determine the levels of cellular immunity indexes in it, including $CD3^+$, $CD4^+$ and $CD8^+$ T lymphocyte levels as well as $CD4^+/CD8^+$ ratio. The turbidimetric immunoassay was used to determine the levels of immunoglobulin G (IgG), immunoglobulin A (IgA) and immunoglobulin M (IgM) in peripheral blood.

2.5. Statistical analysis

SPSS18.0 software was used for statistical processing, measurement data in the study was in terms of mean ± standard deviation, comparison before and after treatment was by paired t test, comparison between two groups after treatment was by routine t test and P<0.05 indicated statistical significance in differences.

3. Results

3.1. Serum broad-spectrum tumor markers

Comparison of serum broad-spectrum tumor markers CEA, CA153 and CA125 levels between two groups of patients is as follows: before treatment, differences in serum CEA, CA153 and CA125 levels were not statistically significant between two groups of patients (P>0.05); after 3 months of treatment, serum CEA, CA153 and CA125 levels of both groups were significantly lower than those before treatment, and differences within same group were statistically significant before and after treatment (P<0.05); after 3 months of treatment, serum CEA, CA153 and CA125 levels of combined treatment group were lower than those of control group, and differences between groups were statistically significant after treatment (P<0.05), shown in Table 1.

Table 1

Comparison of serum broad-spectrum tumor marker levels before and after treatment ($\bar{x}\pm s$).

Groups	n	CEA (μg/L)		CA153 (U/mL)		CA125 (U/mL)	
		Before treatment	After treatment	Before treatment	After treatment	Before treatment	After treatment
Combined treatment group	34	4.38±0.52	0.76±0.09	56.38±6.19	23.12±2.74	154.28±20.49	23.76±3.98
Control group	42	4.45±0.59	2.41±0.35	57.25±6.58	39.85±4.76	157.74±16.84	84.18±9.76
t		0.182	7.492	0.167	8.293	0.218	9.862
P		>0.05	<0.05	>0.05	<0.05	>0.05	<0.05

3.2. Breast cancer-specific tumor markers

Comparison of serum breast cancer-specific tumor markers IGF-1, MK, sECand TK1 levels between two groups of patients is as follows: before treatment, differences in serum IGF-1, MK, sEC and TK1 levels were not statistically significant between two groups of patients (P>0.05); after 3 months of treatment, serum IGF-1, MK, sEC and TK1 levels of both groups were significantly lower than those before treatment, and differences within same group were statistically significant before and after treatment (P<0.05); after 3 months of treatment, serum IGF-1, MK, sEC and TK1 levels of combined treatment group were lower than those of control group, and differences between groups were statistically significant after treatment (P<0.05), shown in Table 2.

3.3. Cellular immunity indexes

Comparison of cellular immunity indexes $CD3^+$, $CD4^+$ and $CD8^+$T lymphocyte levels as well as $CD4^+/CD8^+$ ratio in peripheral blood between two groups of patients is as follows: before treatment, differences in $CD3^+$, $CD4^+$ and $CD8^+$ T lymphocyte levels as well as $CD4^+/CD8^+$ ratio in peripheral blood were not statistically significant between two groups of patients (P>0.05); after 3 months of treatment, $CD3^+$ and $CD4^+$T lymphocyte levels as well as $CD4^+/CD8^+$ ratio of both groups were lower than those before treatment while $CD8^+$ T lymphocyte levels were higher than those before treatment, and differences within same group were statistically significant (P<0.05); after 3 months of treatment, $CD3^+$ and $CD4^+$T lymphocyte levels as well as $CD4^+/CD8^+$ ratio in peripheral blood of combined treatment group were higher than those of control group while $CD8^+$T lymphocyte level was lower than that of control group, and differences between groups were statistically significant (P<0.05), shown in Table 3.

3.4. Humoral immunity indexes

Comparison of humoral immunity indexes IgG, IgA and IgM levels in peripheral blood between two groups of patients is as follows: before treatment, differences in IgG, IgA and IgM levels in peripheral blood were not statistically significant between two groups of patients (P>0.05); after 3 months of treatment, IgG, IgA and IgM levels in peripheral blood of both groups were significantly lower than those before treatment, and differences within same group were statistically significant before and after treatment (P<0.05); after 3 months of treatment, IgG, IgA and IgM levels in peripheral blood of combined treatment group were higher than those of control group, and differences between groups were statistically significant after treatment (P<0.05), shown in Table 4.

Table 2

Comparison of serum breast cancer-specific tumor marker levels before and after treatment ($\bar{x}\pm s$).

Groups	n	IGF-1 (ng/mL)		MK (pg/mL)		sEC (ng/mL)		TK1 (pmol/L)	
		Before treatment	After treatment	Before treatment	After treatment	Before treatment	After treatment	Before treatment	After treatment
Combined treatment group	34	214.28±25.93	54.72±6.09	453.28±50.19	173.29±20.75	2.74±0.35	0.56±0.06	5.37±0.62	0.73±0.09
Control group	42	209.76±28.34	113.95±15.47	462.17±52.64	315.62±35.88	2.81±0.34	1.34±0.17	5.46±0.59	2.15±0.27
t		0.214	9.283	0.372	11.382	0.382	6.283	0.162	6.394
P		>0.05	<0.05	>0.05	<0.05	>0.05	<0.05	>0.05	<0.05

Table 3

Comparison of cellular immunity index levels in peripheral blood before and after treatment ($\bar{x}\pm s$).

Groups	n	$CD3^+$		$CD4^+$		$CD8^+$		$CD4^+/CD8^+$	
		Before treatment	After treatment	Before treatment	After treatment	Before treatment	After treatment	Before treatment	After treatment
Combined treatment group	34	56.19±6.45	53.17±6.09	38.47±4.19	35.27±4.38	23.12±0.27	25.38±3.09	1.72±0.19	1.57±0.21
Control group	42	56.07±6.12	48.69±5.15	38.56±4.08	30.53±3.65	23.09±0.28	30.63±4.28	1.71±0.18	1.26±0.17
t		0.582	7.498	0.182	8.124	0.167	7.394	0.125	6.832
P		>0.05	<0.05	>0.05	<0.05	>0.05	<0.05	>0.05	<0.05

Table 4

Comparison of humoral immunity index levels in peripheral blood before and after treatment (g/L, $\bar{x}\pm s$).

Groups	n	IgG		IgA		IgM	
		Before treatment	After treatment	Before treatment	After treatment	Before treatment	After treatment
Combined treatment group	34	9.32±0.98	7.48±0.87	1.14±0.18	0.93±0.10	2.12±0.25	1.35±0.19
Control group	42	9.24±0.97	4.12±0.56	1.17±0.19	0.45±0.06	2.09±0.27	0.74±0.09
t		0.182	7.823	0.153	6.182	0.168	5.983
P		>0.05	<0.05	>0.05	<0.05	>0.05	<0.05

4. Discussion

Patients with advanced breast cancer are already with distant metastasis and unable to accept radical operation, and the appropriateness of conservative treatment will directly decide the patients' survival time and quality of life[5]. Intravenous chemotherapeutic medication is the most conventional treatment, which aims at reducing the tumor cell activity within breast and the surrounding metastases, and inhibiting its further invasion and metastasis. Intravenous chemotherapy reaches various tissues and organs through the blood circulation system, it has the advantage of extensive efficacy, but the intravenous medication concentration is low in target organs, the killing effect on tumor cells is limited, it can also produce side effects to normal tissues and organs, and hematopoietic function inhibition and immune dysfunction are the two main side effects of intravenous chemotherapy[6,7]. How to improve the therapeutic effect of intravenous chemotherapy without increasing the adverse reactions is the focus of current clinical research, many scholars suggest using Chinese patent drugs as the adjuvant drugs for intravenous chemotherapy, and Xiao Chaihu Tang is a new antitumor Chinese patent medicine that has received wide clinical attention.

Xiao Chaihu Tang is the main prescription for treatment of Shaoyang disease in Treatise on Febrile Diseases, and radix bupleuri is bitter and flat and harmonizes channel qi; radix scutellariae clears pathogenic heat; ginseng and radix liquiritiae support the vital energy and resist pathogenic factors; ginger and jujube harmonize stomach Qi and engender liquid. All these medicines work altogether to achieve the effect of relieving pathogenic factors, harmonizing Shaoyang, smoothing the upper heater and producing engender liquid, and they can optimize the overall state of patients with malignant tumor and improve the effect of intravenous chemotherapy[8]. Tumor markers are the specific indexes secreted into the blood by tumor cells, and their serum levels can objectively reflect the tumor malignancy and clinical therapeutic effect[9,10]. CEA, CA153 and CA125 belong to broad-spectrum tumor markers, and their expression levels have already been detected in serum of patients with gastric cancer, liver cancer, colorectal cancer and other malignant tumors[11]. In the study, the serum levels of above broad-spectrum tumor markers were detected, and it was found that compared with before treatment, serum CEA, CA153 and CA125 levels of both groups decreased after treatment (P<0.05); compared with control group, combined treatment group were with lower CEA, CA153 and CA125 levels (P<0.05), confirming that after adding Xiao Chaihu Tang therapy, the overall disease in patients with advanced breast cancer is optimized.

There are also many markers directly related to breast cancer in the serum, IGF-1 can work with estrogen to promote breast cancer, and studies have confirmed that IGF-1 levels are positively correlated with the risk of breast cancer. MK is highly specific in breast cancer tissue, and high expression of MK mostly indicates high malignant degree of tumor[12,13]. sEC can maintain the epithelial cell polarity and intercellular junction, and the sEC expression increases when malignant tumor occurs. Studies have pointed out that TK1 abnormally increases in malignant tumor cells and is released into the blood, and it is regarded as one of the new targets for breast cancer treatment[14,15]. In the study, the levels of above breast cancer-specific tumor markers were detected, and it was found that compared with control group, combined treatment group were with lower serum IGF-1, MK, sEC and TK1 levels (P<0.05), further confirming that the Xiao Chaihu Tang can reduce the disease severity in patients with advanced breast cancer, and the realization of the specific function is directly related to the efficacy of radix bupleuri, pinellia ternata, ginseng, radix scutellariae and other Chinese patent medicines.

Large-dose and long-term intravenous chemotherapy can directly inhibit the patients' immune function, some patients even develop severe systemic infectious disease during chemotherapy, and the mortality is high[16]. It is believed that Xiao Chaihu Tang has the effect of regulating immune function, and can activate macrophages and increase the interleukin-1 generation, increase the activation of T and B lymphocytes, increase antibody generation, and ultimately enhance the patients' immune function. The body's immune function is specifically divided into cellular immunity and humoral immunity, both are suppressed during chemotherapy in patients with malignant tumor, and therefore, to identify the effect of Xiao Chaihu Tang on the immune function of patients with advanced breast cancer needs to confirm both cellular immune function and humoral immune function[17]. It was found in the study that compared with control group, combined treatment group were with higher $CD3^+$ and $CD4^+$T lymphocyte levels as well as $CD4^+/CD8^+$ ratio, and lower $CD8^+$T lymphocyte level in peripheral blood after 3 months of treatment (P<0.05), indicating that Xiao Chaihu Tang can optimize the cellular immune function in patients with advanced breast cancer after intravenous chemotherapy. At the same time, IgG, IgA and IgM levels in peripheral blood of combined treatment group were higher than those of control group (P<0.05), further illustrating that the Xiao Chaihu Tang can enhance the humoral immune function in patients with advanced breast cancer after intravenous chemotherapy, eventually avoid the occurrence of infection and other immune function decline-related complications, and ensure the realization of the anticancer effect of drugs.

To sum up, it is concluded as follows: Xiao Chaihu Tang combined with intravenous chemotherapy can decrease the severity of advanced breast cancer and optimize the body's immune function, and it's worth popularization and application in clinical practice in the future.

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