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Effect of continuous recombinant human endostatin pumping combined with TP chemotherapy on serum malignant molecules and angiogenesis molecules in patients with advanced ovarian cancer

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ABSTRACT

Objective: To study the effect of continuous recombinant human endostatin pumping combined with TP chemotherapy on serum malignant molecules and angiogenesis molecules in patients with advanced ovarian cancer. **Methods:** 78 patients with advanced ovarian cancer who were treated in our hospital between July 2011 and December 2015 were selected and divided into observation group and control group (n=39) according to the single-blind randomized control method. Before treatment and after 4 cycles of treatment, electrochemical luminescence immunity analyzer was used to detect serum tumor marker levels; RIA method was used to determine serum apoptosis molecule levels; enzyme-linked immunosorbent assay (ELISA) was used to detect the serum angiogenesis molecule levels. **Results:** Before treatment, differences in serum levels of tumor markers, apoptosis molecules and angiogenesis molecules were not statistically significant between two groups of patients (P>0.05). After 4 cycles of treatment, serum carbohydrate antigen 125 (CA125), carbohydrate antigen 153 (CA153), human epididymis protein 4 (HE4), carcinoembryonic antigen (CEA), human chorionic gonadotropin (β-HCG), Bcl-2, Survivin, Bag-1, angiogenin-2 (Ang-2), vascular endothelial growth factor (VEGF) and basic fibroblast growth factor (bFGF) levels of observation group were significantly lower than those of control group (P<0.05) while Bax level was significantly higher than that of control group (P<0.05). **Conclusions:** Continuous recombinant human endostatin pumping combined with TP chemotherapy can decrease the malignant degree of advanced ovarian cancer and inhibit angiogenesis.

1. Introduction

Ovarian cancer is one of the most common malignant tumors of female reproductive organs, patients are without obvious early clinical manifestation, and they have been in moderate-advanced stage when they are with significant discomfort and receive inspection in hospital, and have missed the opportunity of surgery[1,2]. Conservative treatment is the main way to extend survival time and optimize the quality of life in patients with advanced ovarian cancer, intravenous chemotherapy is with the most wide clinical application, and it can not only kill tumor cells, but can also cause different degree of damage to normal tissues and organs. Conventional chemotherapy combined with other targeted anticancer drug treatment is the new treatment for patients with advanced cancer that is highly praised by many scholars[3]. Recombinant human endostatin, as the antitumor vascular targeted drug, has been successfully applied in advanced non-small cell lung cancer, metastatic gastric cancer and other malignant tumors[4], but there is no clear report at present about the effect of recombinant human endostatin on malignant molecules in patients with advanced ovarian cancer. In the following study, the effect of continuous recombinant human endostatin pumping combined with TP chemotherapy on serum malignant molecules and angiogenesis molecules in patients with advanced ovarian cancer was analyzed.

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2. Materials and methods

2.1. General information

78 patients with advanced ovarian cancer who were treated in our hospital between July 2011 and December 2015 were selected as the research subjects, and the included patients themselves signed the informed consent. According to the single-blind randomized control method, the patients were divided into observation group and control group (n=39). Observation group included 21 male cases and 18 female cases, they were 43–78 years old, and the body weight was 45–71 kg and (56.27±9.15) kg in average; control group included 20 male cases and 19 female cases, they were 41–76 years old, and the body weight was 44–75 kg and (55.98±9.07) kg in average. Two groups of patients were not statistically different in age, gender and weight distribution (P>0.05). Inclusion criteria: (1) diagnosed with advanced ovarian cancer by histopathology; (2) with primary ovarian cancer; (3) diagnosed for the first time and never receiving systemic treatment before; (4) with normal cognitive function and could cooperate with the whole treatment and inspection. Exclusion criteria: (1) with primary malignant tumor diseases of other tissue viscera; (2) with severe heart, liver and kidney dysfunction; (3) with aplastic disease, myelodysplastic syndrome and other bone marrow hematopoietic dysfunction; (4) quitting the treatment and with incomplete clinical data.

2.2. Treatment methods

Control group received conventional TP chemotherapy, specifically as follows: docetaxel (Dandong Yichuang Pharmaceutical Co., LTD., approved by H20110138) 75 mg/m^2, by intravenous injection, d1; cisplatin (Guizhou Hanfang Pharmaceutical Co., LTD., approved by H20020273) 60 mg, by intravenous injection, d1–d4, 21 d as a cycle, for at least 4 cycles of treatment. Based on TP chemotherapy, observation group of group received continuous recombinant human endostatin pumping treatment, specifically as follows: recombinant human endostatin (Shandong Simcere-Medgenn Bio-pharmaceutical Co., LTD., approved by S20050088) in normal saline 200 mL, by continuous intravenous pumping with portable infusion pump, 3–4 h every day, 21 d as one cycle, for at least 4 cycles of treatment. TP chemotherapy regimen was the same as that of the control group.

2.3. Serum indexes

Before treatment and after 4 cycles of treatment, 2 mL of peripheral venous blood was extracted from two groups of patients at the same point in time, let stand at room temperature and centrifuged at low speed to get supernatant, and the following indexes were detected: (1) tumor markers: electrochemical luminescence immunity analyzer (Beijing Biolot Technology Development Co., LTD., article number B3811) was used to detect tumor markers carbohydrate antigen 125 (CA125), carbohydrate antigen 153 (CA153), human epididymis protein 4 (HE4), carcinoembryonic antigen (CEA) and human chorionic gonadotropin (β-HCG) levels; (2) apoptosis molecules: RIA kit (Thermo Fisher Company, the article number TH829) instructions were followed to detect apoptosis molecule levels, including Bcl-2, Bax, Survivin and Bag-1; (3) angiogenesis molecules: ELISA method was used to detect angiogenin-2 (Ang-2), vascular endothelial growth factor (VEGF) and basic fibroblast growth factor (bFGF) levels.

2.4. Statistical analysis

The data in the study were input in software SPSS23.0, measurement data was in terms of $\bar{x}\pm s$, comparison before and after treatment was by paired t test, comparison groups was by routine t test and P<0.05 indicated statistical significance in differences.

3. Results

3.1. Serum tumor marker levels

Comparison of serum tumor markers CA125, CA153, HE4, CEA and β-HCG levels between two groups of patients is as follows: before treatment, differences in serum CA125, CA153, HE4, CEA and β-HCG levels were not statistically significant between two groups of patients (P>0.05); after 4 cycles of treatment, serum CA125, CA153, HE4, CEA and β-HCG levels of both groups were significantly lower than those before treatment, and differences within same group before and after treatment were statistically significant (P<0.05); after 4 cycles of treatment, serum CA125, CA153, HE4, CEA and β-HCG levels of observation group were significantly lower than those of control group, and differences between groups after treatment were statistically significant (P< 0.05), shown in Table 1.

Table 1

Comparison of serum tumor marker levels before and after treatment (n=39, $\bar{x}\pm s$).

Groups	Time	CA125 (U/mL)	CA153 (U/mL)	HE4 (pmol/L)	CEA (ng/mL)	β-HCG (mU/mL)
Observation group	Before treatment	243.81±29.75	85.48±9.11	24.38±3.09	30.26±3.98	23.17±2.86
	After treatment	43.28±5.19$^{*\#}$	23.18±2.95$^{*\#}$	2.94±0.35$^{*\#}$	3.18±0.45$^{*\#}$	4.28±0.52$^{*\#}$
Control group	Before treatment	241.84±25.83	86.17±9.05	25.16±2.87	31.21±3.75	23.21±2.79
	After treatment	92.74±10.18*	51.72±5.98*	9.24±0.98*	10.56±1.78*	13.15±1.98*

Compared with same group before treatment, $^{*}P$<0.05; compared with control group after treatment, $^{\#}P$<0.05.

3.2. Serum apoptosis molecule levels

Comparison of serum apoptosis molecules Bcl-2, Bax, Survivin and Bag-1 levels between two groups of patients is as follows: before treatment, differences in serum Bcl-2, Bax, Survivin and Bag-1 levels were not statistically significant between two groups of patients (P>0.05); after 4 cycles of treatment, serum Bcl-2, Survivin and Bag-1 levels of both groups were significantly lower than those before treatment while Bax levels were significantly higher than those before treatment, and differences within same group before and after treatment were statistically significant (P< 0.05); after 4 cycles of treatment, serum Bcl-2, Survivin and Bag-1 levels of observation group were significantly lower than those of control group while Bax level was significantly higher than that of control group, and differences between groups after treatment were statistically significant (P<0.05), shown in Table 2.

3.3. Serum angiogenesis molecule levels

Comparison of serum angiogenesis molecules Ang-2, VEGF and bFGF levels between two groups of patients is as follows: before treatment, differences in serum Ang-2, VEGF and bFGF levels were not statistically significant between two groups of patients (P>0.05); after 4 cycles of treatment, serum Ang-2, VEGF and bFGF levels of both groups were significantly lower than those before treatment, and differences within same group before and after treatment were statistically significant (P<0.05); after 4 cycles of treatment, serum Ang-2, VEGF and bFGF levels of observation group were significantly lower than those of control group, and differences between groups after treatment were statistically significant (P< 0.05), shown in Table 3.

4. Discussion

The treatment of patients with advanced ovarian cancer is the clinical research focus, combined platinum/paclitaxel chemotherapy is the most common intravenous chemotherapy, but the recurrence rate is higher than 85% after disease remission[5]. To explore the more effective new therapy that patients can tolerate is a research hotspot of oncology, the molecular targeting treatment develops rapidly at present, and the vascular targeting drugs, in particular, have opened a new era for the treatment of patients with advanced cancer[6]. Recombinant human endostatin (endostar) is the multi-targeted vascular endothelial inhibitor independently developed in our country, which inhibits the migration of the endothelial cells that form blood vessels to achieve the purpose of inhibiting tumor angiogenesis, and blocks the blood and oxygen supply to the tumor cells to inhibit tumor proliferation. At present, the overall efficacy of recombinant human endostatin in treatment of a variety of advanced malignant tumors has been confirmed, but the influence on the expression of specific malignant molecules and angiogenesis molecules is less covered[7,8]. In the study, recombinant human endostatin combined with TP chemotherapy was used as a new treatment and applied to patients with advanced ovarian cancer, the changes in serum index levels before and after treatment were detected in order to define the molecular-level curative effect of recombinant human endostatin.

There is unusually high expression of a series of molecules in patients with ovarian cancer, it is an effective auxiliary treatment for disease diagnosis, and detecting their levels can objectively reflect the effect of drug treatment[9]. Both CA125 and CA153 are carbohydrate antigens, belong to the broad-spectrum tumor markers, and are highly expressed in malignant reproductive system tumor and digestive tract tumor. HE4 is an independent risk factor for the prognosis of ovarian cancer, and the high HE4 level mostly indicates poor treatment outcome[10]. CEA is a glycoprotein from the colorectal cancer tissue, it has currently been found to be highly express in a variety of other malignant tissues, but its specificity and sensitivity are not high, and it is an auxiliary index for disease diagnosis and illness detection[11,12]. β-HCG belongs to hormone markers, is secreted by gestational trophoblastic cells, can also be massively expressed when malignant reproductive system tumor occurs, and is an important index for the diagnosis of reproductive system tumor[13]. It was found in the study that after treatment, serum CA125, CA153, HE4, CEA and β-HCG levels of observation group were lower (P<0.05), indicating that adding recombinant human endostatin treatment on the basis of TP chemotherapy can macroscopically contain the malignant degree of tumor.

Pro-apoptotic/anti-apoptotic molecule expression imbalance in

Table 2

Comparison of serum apoptosis molecule levels before and after treatment (n=39, $\bar{x}\pm s$).

Groups	Time	Bcl-2 (ng/mL)	Bax (ng/mL)	Survivin (pg/mL)	Bag-1 (μg/mL)
Observation group	Before treatment	2.18±0.25	4.12±0.59	14.38±1.93	42.57±5.99
	After treatment	0.63±0.08*#	15.85±1.94*#	2.18±0.24*#	6.34±0.78*#
Control group	Before treatment	2.21±0.25	4.09±0.53	15.21±1.94	43.68±4.98
	After treatment	1.34±0.18*	8.17±0.92*	7.35±0.81*	15.19±1.76*

Compared with same group before treatment, *P<0.05; compared with control group after treatment, #P<0.05.

Table 3

Comparison of serum angiogenesis molecule levels before and after treatment (n=39, $\bar{x}\pm s$).

Groups	Time	Ang-2 (ng/mL)	VEGF (ng/mL)	bFGF (pg/L)
Observation group	Before treatment	89.53±9.11	324.39±39.76	15.38±1.92
	After treatment	10.17±1.86*#	40.17±4.95*#	4.57±0.73*#
Control group	Before treatment	88.76±9.45	330.17±37.83	15.76±1.89
	After treatment	34.59±4.52*	101.28±14.36*	9.23±0.98*

Compared with same group before treatment, *P<0.05; compared with control group after treatment, #P<0.05.

malignant tumor tissue is one of the fundamental mechanisms contributing to persistent tumor cell proliferation and metastasis, and detecting serum apoptosis molecule expression can further visually assess the tumor malignancy and clinical therapeutic effect[14]. Bcl-2 family plays a key role in the programmed cell death, and Bcl-2 inhibits cell apoptosis and is highly expressed in malignant tumor tissue; Bax can promote cell apoptosis, and it has been confirmed that its expression is inhibited in lung cancer, colorectal cancer, liver cancer and other malignant tumor tissues[15]. Survivin is a newly discovered anti-apoptotic gene, is mostly expressed in embryo and developing fetus tissue, and is also highly expressed in most malignant tumor tissues[16]. Bag-1 belongs to anti-apoptotic molecule, has independent anti-apoptotic effect, and can also increase the Bcl-2 function and form complexes with it to enhance the anti-apoptotic ability of cells[17]. In the study, the serum levels of above apoptosis molecules were tested, and it was found that compared with the control group of patients, observation group of patients were with lower serum anti-apoptotic molecules Bcl-2, Survivin and Bag-1 levels as well as higher pro-apoptotic molecule Bax level after 4 cycles of treatment ($P<0.05$), confirming that recombinant human endostatin can effectively restrain the anti-apoptotic molecule activity and contain the tumor cell proliferation.

Under physiological condition, angiogenesis appears only in embryonic development or tissue damage repair period, and abnormal angiogenesis is one of the specific symbols of malignant tumors[18]. VEGF is considered to be the strongest and most specific pro-angiogenesis regulatory factor, and research has confirmed that VEGF is highly expressed in ovarian cancer, and its content decreases with the realization of tumor treatment effect[19]. Both Ang-2 and bFGF have pro-angiogenesis activity, Ang-2 can induce angiogenesis and remodeling, and it is abundantly expressed in a variety of malignant tumor tissues; bFGF is a peptide factor with wide biological activity, and study in vitro has demonstrated that bFGF can stimulate cell proliferation and migration[20]. In the study, serum levels of angiogenesis molecules were detected, and it was found that compared with the control group of patients, observation group of patients were with lower serum Ang-2, VEGF and bFGF levels ($P<0.05$), indicating that continuous recombinant human endostatin pumping combined with TP chemotherapy can effectively contain tumor angiogenesis, which is the internal cause of decreased malignant molecule expression in the study.

To sum up, it is concluded as follows: continuous recombinant human endostatin pumping combined with TP chemotherapy can decrease the malignant degree of advanced ovarian cancer and inhibit angiogenesis, and it's worth popularization and application in clinical practice in the future.

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