



Effects of oxaliplatin, leucovorin and fluorouracil on serum tumor markers, VEGF, CRP and matrix metalloproteinases in patients with advanced esophageal cancer

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ABSTRACT

Objective: To investigate the effects of oxaliplatin, leucovorin and fluorouracil on serum tumor markers, VEGF, CRP and matrix metalloproteinases in patients with advanced esophageal cancer. **Methods:** From March 2012 to March 2017 a total of 248 patients with advanced esophageal cancer were selected as the study subjects. According to random data table, they were divided into control group ($n=123$) and observation group ($n=125$) according to random data table. The control group was treated with cisplatin combined with fluorouracil, leucovorin chemotherapy, and patients in the observation group received oxaliplatin, leucovorin and fluorouracil chemotherapy, all patients were treated for 2 cycles. The changes of serum tumor markers, VEGF, CRP and matrix metalloproteinase levels in the two groups before and after treatment was compared. **Results:** Before treatment, there was no significant difference of the levels of serum CA125, CA19-9, CEA, VEGF, CRP, MMP-2 and MMP-9 between the control group and the observation group. Compared with the group before treatment, the levels of CA125, CA19-9, CEA, VEGF, CRP, MMP-2 and MMP-9 in the two groups were significantly lower. After treatment, the level of CA125, CA19-9, CEA, VEGF, CRP, MMP-2 and MMP-9 in the observation group was significantly lower than those of the control group. **Conclusion:** Oxaliplatin, leucovorin and fluorouracil chemotherapy can effectively reduce the levels of serum tumor markers, VEGF, CRP and matrix metalloproteinase in patients with advanced esophageal cancer, it has important clinical value.

1. Introduction

Esophageal carcinoma is one of the most common malignant tumors in clinic. China is a high incidence area of esophageal cancer. It is characterized by high malignancy and occult[1]. The clinical treatment include surgery, radiotherapy and chemotherapy, surgery is the preferred solution, but because of early disease is not easy to find, most patients are diagnosed at advanced stage, surgery has missed the best period, therefore, the treatment of advanced esophageal cancer is mainly chemotherapy[2,3]. Cisplatin, oxaliplatin, leucovorin and fluorouracil are effective drug for

the treatment of esophageal cancer, single drug chemotherapy is poor, studies have shown that the combined treatment of drugs has significantly improved the efficiency of chemotherapy for esophageal cancer[4,5]. The purpose of this study was to investigate the efficacy of oxaliplatin, leucovorin, and fluorouracil in combination with chemotherapy for advanced esophageal cancer.

2. Research subjects and methods

2.1 General information

A total of 248 patients with advanced esophageal cancer admitted to the Enshi Tujia and Miao Autonomous Prefecture Hospital from March 2012 to March 2017 were selected as research subjects. All patients meet the screening criteria of this study. The criteria were as follows: (1) All of them were in accordance with the diagnostic

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criteria for esophageal cancer[6], by pathology or cytology and other diagnostic diagnosis; (2) pathological staging belongs to III-IV; (3) are newly diagnosed patients, previous history of no cancer treatment; (4) expected survival ≥ 3 months; (5) patients blood routine test, electrocardiogram and liver and kidney function of the patients all accorded with the standard of chemotherapy (6) after admission, complete clinical data, voluntary treatment; (7) all patients and their families are informed consent and signed informed consent. Exclusion criteria: (1) associated with severe liver and kidney dysfunction, blood diseases and autoimmune diseases; (2) combined with other tumors, while receiving treatment; (3) not receiving treatment, or treatment of self-shedding cases, failed to complete the treatment by the course of treatment; (4) incomplete clinical data. All patients were divided into control group ($n = 123$) and observation group ($n = 125$) according to the random data table method. Among them, there were 112 cases of male patients and 11 cases of female patients in the control group. The age was 42-69 years old. pathological type: 109 cases of squamous cell carcinoma, adenocarcinoma in 16 cases; clinical stage: stage III 71 cases, stage IV 54 cases. There were no significant differences in sex, age, pathological type and tumor stage between the two groups ($P > 0.05$), comparable. This study is in accordance with the standards of the Hospital Ethics Committee and is subject to permission.

2.2 Treatment

The patients in the control group were given cisplatin, leucovorin combined with fluorouracil chemotherapy regimen, the first day of treatment intravenous infusion of cisplatin injection (Nuo Xin, Jiangsu Hausen Pharmaceutical Co., Ltd. production, Chinese medicine Zhunzi H20010813, specifications model 6 mL: 20 mg \times 5 bottles), the specific dosage of 20 mg/m² into the 500 mL concentration of 0.9% sodium chloride injection intravenous infusion of 2 h, during the first day to the 5th day of treatment plus fluorouracil injection (Tianjin Jinyao Pharmaceutical Co., Ltd. production, the Chinese medicine Zhunzi H12020959, specifications 10 mL: 0.25 g), intravenous infusion dosage, according to body surface area of 750 mg/m² dissolved in 20 mL concentration of 0.9% sodium chloride injection, The time of intravenous infusion should not be less than 6-8 h, once a day; leucovorin injection (Chongqing Pharmaceutical Co., Ltd. production, pharmaceutical Zhunzi H20010615, drug specifications: 10 mL: 0.1 g) 200 mg/m² dissolved in 0.9% sodium chloride 250 mL injection dosing infusion solution (Note: infusion solution pH should not be less than 6.5), intravenous infusion of 2 h, once a day, last for 5 d since the first day of treatment. During the treatment of patients also received antiemetic, protect liver and protecting stomach and other symptomatic treatment, the treatment cycle of 21 d, with 2 cycles. The patients in the observation group were treated with oxaliplatin, leucovorin and fluorouracil. The use of leucovorin and fluorouracil

was the same as that of the control group. Oxaliplatin (manufactured by Zhejiang Haizheng Pharmaceutical Co., Ltd., H20093487, Mg/s), the dose by body surface area of 100 mg/m² added into 5% glucose solution 250 mL, intravenous infusion of 2-6 h, administration in the first day of treatment, once 21 d after administration. Treatment time is the same with the control group.

2.3. Observation indexes

Patients were collected with fasting venous blood before treatment and 2 cycles of treatment respectively, centrifuge to take serum, serum levels of tumor markers, vascular endothelial growth factor (VEGF), C-reactive protein (CRP) and matrix metalloproteinase (MMP) were measured. The serum tumor markers include: carbohydrate antigen 125 (CA125), carbohydrate antigen 19-9 (CA19-9) and carcinoembryonic antigen (CEA) levels, using electrochemical luminescence detector, the method for the electrochemical luminescence method; VEGF (MMP-2) and matrix metalloproteinase-9 (MMP-9) were detected by enzyme-linked immunosorbent assay (ELISA), and the corresponding detection kit (purchased from Shanghai enzyme biotechnology Co., Ltd.), The specific steps in strict accordance with the operating instructions.

2.4. Statistical analysis

Use SPSS 17.0 software for processing and analysis of data, level of serum biochemical indexes in accord with the normal distribution, the sample mean is expressed in (Mean \pm SD), before and after treatment, the number of samples between the two groups was compared with the t test, the statistical results $P < 0.05$ says that the difference was significant, statistically significant.

3. Results

3.1 Comparison of serum tumor markers before and after treatment

Before treatment, serum CA125, CA19-9 and CEA levels in the two groups were not significantly different ($P > 0.05$). After treatment, the levels of CA125, CA19-9 and CEA in the control group and the observation group were (9.89 \pm 2.94) U/mL, (10.02 \pm 2.78) U/mL, (1.69 \pm 0.43) ng/mL, (8.71 \pm 2.16) U/mL, (9.25 \pm 1.65) U/mL and (1.51 \pm 0.29) ng/mL respectively, were significantly lower than those before treatment, the difference was significant ($P < 0.05$). The level of observation group after treatment was significantly lower than the control group, the difference was significant ($P < 0.05$). As shown in Table 1.

Table 1.

Comparison of serum tumor marker levels between the two groups before and after treatment.

Group	n	Treatment time	CA125 (U/mL)	CA19-9 (U/mL)	CEA (ng/mL)
Control group	123	Before treatment	12.58 \pm 4.91	12.57 \pm 4.14	2.76 \pm 1.18
		After treatment	9.89 \pm 2.94*	10.02 \pm 2.78*	1.69 \pm 0.43*
Observation group	125	Before treatment	12.61 \pm 5.07	12.59 \pm 4.35	2.79 \pm 1.04
		After treatment	8.71 \pm 2.16**	9.25 \pm 1.65**	1.51 \pm 0.29**

Note: * compared with the same group before treatment $P < 0.05$, ** compared with the level of the control group after treatment $P < 0.05$.

3.2. Comprison of serum VEGF, CRP levels before and after treatment

The levels of serum VEGF and CRP in the two groups before and after treatment were shown in Table 2. The levels of VEGF and CRP in the control group and the observation group were similar before treatment, the difference was not statistically significant ($P>0.05$). The levels of VEGF and CRP in the control group after treatment were (353.94 ± 44.87) ng/L and (8.76 ± 2.42) mg/L, the observation group level (269.54 ± 39.84) ng/L and (5.75 ± 1.44) mg/L, compared with the group before treatment, the two levels were significantly decreased ($P<0.05$); comparison between groups after treatment. The observation group VEGF and CRP were significantly lower than the control group, the difference was significant ($P<0.05$).

Table 2.

Comparison of serum VEGF and CRP levels between the two groups before and after treatment.

Group	n	Treatment time	VEGF (ng/L)	CRP (mg/L)
Control group	123	Before treatment	415.22±57.03	11.84±3.95
		After treatment	353.94±44.87*	8.76±2.42*
Observation group	125	Before treatment	411.76±58.09	12.01±4.35
		After treatment	269.54±39.84**	5.75±1.44**

Note: *compared with the same group before treatment $P<0.05$, indicated compared with the level of the control group after treatment $P<0.05$.

3.3. Comprison of the serum MMP-2 and MMP-9 levels before and after treatment

The changes of serum MMP-2 and MMP-9 levels before and after treatment in the two groups were shown in table 3. Before treatment, there was no significant difference between the two groups, and there was no statistical significance ($P>0.05$). After treatment, MMP-9, MMP-2 level of the observation group were (65.74 ± 8.86) ng/mL and (299.62 ± 141.29) ng/mL, the index level within the group before treatment was significantly decreased ($P<0.05$), and significantly lower than the control group after treatment (87.36 ± 7.99) ng/mL and (460.76 ± 175.24) ng/mL, the difference was statistically significant ($P<0.05$).

Table 3.

Comparison of serum MMP-2 and MMP-9 levels before and after treatment.

Group	n	Treatment time	MMP-2 (ng/mL)	MMP-9 (ng/mL)
Control group	123	Before treatment	125.28±15.17	691.44±218.51
		After treatment	87.36±7.99*	460.76±175.24*
Observation group	125	Before treatment	124.64±15.53	694.91±232.07
		After treatment	65.74±8.86**	299.62±141.29**

Note: *compared with the same group before treatment $P<0.05$, indicated compared with the level of the control group after treatment $P<0.05$.

4. Discussion

Esophageal cancer is one of the most common malignant tumors of the digestive tract. It is characterized by progressive dysphagia in the lower middle esophagus. Squamous cell carcinoma mainly accounts for more than 90% of the total esophageal cancer in our country[7]. The pathogenesis of esophageal cancer is still unclear, it is generally believed that the occurrence is related with the living environment,

long-term bad habits (such as smoking, drinking and eating lots of nitrite amine food etc.) and genetic factors (genetic susceptibility and lack of anti-cancer factors) and other relevant[8,9]. The 5 year survival rate of patients with advanced esophageal cancer is only 35%, while the 5 year survival rate of early esophageal cancer is 85%. Therefore, early diagnosis and treatment is very important[10]. The treatment of esophageal cancer is mainly based on surgery, supplemented by radiotherapy, chemotherapy and endoscopy. The purpose is to improve the survival rate of patients with esophageal cancer. But because of the occult characteristics of esophageal cancer, most patients already belongs to the late inoperable, therefore, chemotherapy plays an important role in the treatment of advanced esophageal cancer, has become the only effective solution to the prevention and treatment of systemic metastasis[11].

Cisplatin and fluorouracil chemotherapy is standard treatment guide, but the research pointed out that the relapse rate of patients after scheme treatment is high, remedial plan less, therefore need to explore more effective chemotherapy[12]. The study found that in the treatment of cisplatin and fluorouracil combined with leucovorin, can further improve the total effective rate and the total control rate of esophageal cancer[13,14]. The main reason is that leucovorin is a biochemical regulator of fluorouracil, can effectively enhance the inhibition of fluorouracil on DNA synthesis, thereby enhancing the overall effect of chemotherapy[15]. Oxaliplatin belongs to the third generation of platinum compounds, and fluorouracil has a significant synergistic effect, has a strong inhibitory effect on cisplatin-resistant cell lines, colorectal cancer cell lines and fluorouracil-resistant tumors. A large number of studies have confirmed that oxaliplatin has a significant effect on colorectal cancer, gastric cancer and esophageal cancer[16-18]. This study from the serum tumor marker research, VEGF, CRP and matrix metalloproteinase, compared with oxaliplatin combined with cisplatin leucovorin, fluorouracil chemotherapy efficacy in patients with advanced esophageal cancer.

Serum tumor markers are important serological markers for tumor diagnosis and curative effect evaluation. The level of serum markers is closely related to the occurrence and development of tumor. CA125, CA19-9 and CEA are clinically more common serum tumor markers, and their levels are very low in normal tissues, and their serum levels are significantly elevated at the time of tumorigenesis[19,20]. The results of this study show that both chemotherapy regimens can effectively reduce the levels of serum CA125, CA19-9 and CEA in patients with advanced esophageal cancer, indicating that both regimens are effective regimens for advanced esophageal cancer treatment, and the effect of oxaliplatin was significantly better than that of cisplatin, which may be related to the strong inhibitory effect of oxaliplatin on cisplatin-resistant cell lines and the synergistic effect with fluorouracil.

VEGF is a specific growth factor, which can induce angiogenesis and inhibit tumor cell apoptosis, and its high level of expression can promote the further development and migration of tumor[21,22]. CRP is an acute phase reactive protein synthesized by the liver during infection and injury, which is part of the nonspecific immune mechanism of the body. It is highly expressed in cancer patients, and indirectly indicates that there is an inflammatory response in cancer patients, which can be used as an important indicator for cancer treatment and prognosis evaluation[23]. MMP-2 and MMP-9 levels were also significantly higher in cancer patients except that the above indexes were abnormally elevated,

The cancer patient in addition to the above indicators of abnormal

increase, MMP-2 and MMP-9 levels are abnormally high expression of MMP-2 and MMP-9, belonging to the matrix metalloproteinase (MMPs) protein family, MMPs is mainly composed of cancer cell invasion and metastasis process, combined with extracellular matrix receptors on the surface, the high level of expression, indicates that the higher the degree of malignancy of cancer cells to the surrounding normal cell infiltration and invasive ability[24,25]. The results showed that the two chemotherapy regimens can effectively reduce the serum levels of VEGF, CRP, MMP-2 and MMP-9, and oxaliplatin chemotherapy is better for the target regulation. The results of this study reveal that oxaliplatin chemotherapy scheme can effectively reduce the level of VEGF, CRP, promote the apoptosis of cancer cells, improve inflammation in patients with cancer microenvironment, and down-regulation of MMP-2 and MMP-9 level, to reduce the degradation of cancer cells to the surrounding extracellular matrix and basement membrane, and thus inhibit the invasion and metastasis of cancer cells. Oxaliplatin, cisplatin and fluorouracil have synergistic effects, but oxaliplatin plays a more synergistic role and can promote the apoptosis of cancer cells. However, the specific mechanism of oxaliplatin remains to be further explored.

In summary, oxaliplatin and cisplatin plus leucovorin, fluorouracil chemotherapy regimens are effective in the treatment of advanced esophageal cancer, but the curative effect of oxaliplatin is better for advanced esophageal cancer compared with cisplatin, Specifically, it can further reduce the level of serum tumor markers, down regulate the levels of VEGF, MMP-2 and MMP-9, and improve the inflammatory microenvironment of cancer patients, and so on, which has important clinical value.

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