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Effect of Renshen Yangying Decoction Combined with Chemotherapy on Traditional Chinese Medical Symptoms and T Cell Subsets in Patients with Cervical Cancer of Ganyu Qizhi Type

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Keywords

Renshen Yangying decoction, Chemotherapy, Cervical cancer, Traditional Chinese medical syndrome, T cell subsets

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Abstract

Background To confirm the effects of the combination of Renshen Yangying decoction and chemotherapy on traditional Chinese medical symptoms and T cell subsets in patients with cervical cancer (CC) of Ganyu Qizhi type.**Methods:** Patients with CC of Ganyu Qizhi type admitted to our hospital (n=96) were selected and evenly divided into control and observation groups. The patients in control group received chemotherapy, while those in observation group were additionally treated with Renshen Yangying decoction. The clinical efficacy, traditional Chinese medical symptom (TCMS) score, serum tumor markers, changes of serum T lymphocyte subsets and adverse effects were compared.**Results:** The distribution of clinical efficacy grade in all patients after treatment was statistically significant, and the clinical benefit rate (CBR) of observation group was higher than that of control group. The TCMS score, the serum carcinoembryonic antigen (CEA), squamous cell carcinoma antigen (SCCA), and cancer antigen 125 (CA125) were also decreased after treatment, and those were decreased in observation group. Levels of serum $CD4^+$ and $CD4^+/CD8^+$, and natural killer (NK) cell percentage were higher, yet serum $CD8^+$ cell percentage was lower after treatment, and in patients of observation group, levels of serum $CD4^+$ and $CD4^+/CD8^+$, and NK cell percentage were higher, yet serum $CD8^+$ cell percentage was lower. The incidence of adverse effects in the observation group was lower than that in the control group.**Conclusion:** Renshen Yangying decoction combined with chemotherapy in the treatment of CC of Ganyu Qizhi type can alleviate the clinical symptoms, reduce serum tumor markers, and improve the cellular immunity, with significant

clinical efficacy and good safety on patients.

1 Introduction

Cervical cancer (CC) is a prevalent gynecological malignancy in clinical practice, whose clinical manifestations mainly include anal swelling, irregular vaginal bleeding, vaginal discharge, and so on, resulting in the systemic failure and threat the life of patients if not treated in time (1). Chemotherapy has been seen as a major tool for the treatment of CC in clinical practice, which can effectively prolong the patients' survival and delay the progression of lesions. However, some toxic effects have also been elicited by the application of chemotherapy, which brings damage to the normal cells in the body and increases the risks of adverse effects, such as immunosuppression and damage on the function of liver and kidney, therefore affecting the clinical efficacy (2, 3). The advantage of adjuvant treatment on tumors using traditional Chinese medicine (TCM) has been evidenced, along with the discussion on the effects of TCM on enhancing the immunity of the body and reducing the risks of adverse effects in patients with tumors (4). Renshen Yangying decoction, which originated from *Treatise on Three Categories of Pathogenic Factors*, is a representative formula for benefiting Qi and blood, which can also nourish the Qi, replenish the blood, benefit the heart and calm the mind (5). Nevertheless, the clinical efficacy of Renshen Yangying decoction on the treatment of CC in clinical practice remains to be further addressed, and in our study, we here investigate the clinical effects of Renshen Yangying decoction on traditional Chinese medical symptoms, serum tumor marker levels and immune function of patients with CC, with the hope to provide reference for the treatment of CC in clinical practice.

2 Materials and methods

2.1 Ethics statement

The Ethics Committee has approved our study, and all participated patients have also signed the written informed consent prior to our study as well.

2.2 Subjects

The random number table method was used to evenly divide a total number of 96 patients with CC of Ganyu Qizhi Type between January, 2018 to June, 2020 into observation group and control group. No statistical significance between these two groups was reported on the basic data, including age, course of disease, tumor grade, and pathological classification and the data were comparable. All information of patients was available in Table 1. All patients enrolled complied to the criteria as listed.

Table 1 The general information of patients

Group	Case	Age (Year)	Course of disease (Month)	Tumor grade (Case)			Pathological classification (Case)	
				Stage Ⅰ	Stage Ⅱ	Squamous carcinomas	Adenocarcinoma	Squamous cell adenocarcinoma
Control group	48	50.84±7.16	10.75±4.14	22	26	35	11	2
Study group	48	51.15±6.72	10.59±3.82	25	23	37	8	3
$\chi^2/t/Z$		-0.336	0.253		-0.609		2.764	
P		0.715	0.801		0.542		0.251	

Inclusion criteria: a) those who met the diagnostic criteria of CC in clinical obstetrics and gynecology (6); b) those who were diagnosed with CC by pathological examination; c) those who were diagnosed with CC of Ganyu Qizhi Type in accordance with the diagnostic and therapeutic criteria for Chinese medical evidence (7); d) those with age ranging from 40 to 60 years old.

Exclusion criteria: a) those with serious organic lesions of vital organs on the body, including heart, liver, kidney, lung and bone marrow; b) those with cognation dysfunction; c) those who had taken other medications within the last 1 month; d) those who were pregnant or lactating; e) those with hematological diseases.

2.3 Treatment regimens

Patients in the control group were given basic treatment, such as water-electrolyte balance maintenance, nutritional support and conventional chemotherapy. 50 mg/m^2 cisplatin (Nanjing Pharmaceutical Factory Co., Ltd., Approval number for National Medical Products Administration (NMPA): H20103216) was added into 1500 ml of 0.9% saline solution, while 140 mg/m^2 paclitaxel (Chenxin Pharmaceutical Co., Ltd., Approval number for NMPA: H20057404) was added into 500 ml of 0.9% saline solution. These chemotherapeutic agents were administered intravenously. The treatment was continued for 7 days as one course, with the interval for 4 weeks and 3 courses in total.

In addition to the therapy applied to control group, the patients in the observation group were treated with Renshen Yangrong decoction, including 10 g Ginseng pieces, 10 g Dang Gui (*Radix Angelicae Sinensis*), 30 g Huang Qi (*Radix Astragali*), 6 g Gan Cao pieces (*Radix Glycyrrhizae*), 15 g Bai Zhu (*Rhizoma Atractylodis Macrocephalae*), 12 g Fu Ling (*Poria*), 15 g Bai Shao (*Radix Paeoniae Alba*), 10 g Yuan Zhi (*Radix Polygalae*), 30 g Shu Di Huang (*Radix Rehmanniae Preparata*), 5 g of Wu Wei Zi (*Fructus Schisandrae*),10 g of Chen Pi (*Pericarpium Citri Reticulatae*), 6 g Rou Gui (*Cortex Cinnamomi*), 10 g E Jiao (*Corii Asini Colla*), 30 g Huang Jing (*Rhizoma Polygonati*), and 6 g of Di Long (*Pheretima*). All components were decocted and 300 ml of total volume was taken The enrolled patients were administered with one dose (2 times per day in the morning and evening) from the first day of chemotherapy to the end.

2.4 Observational index

(I) Clinical efficacy: After treatment, the clinical efficacy of patients in both groups was evaluated using the criteria for the evaluation on the efficacy of solid tumors (6), which was detailed as follows: complete remission (CR): the disappearance of all lesions for over 4 weeks; partial remission (PR): the shrinkage of the tumor was over 50%; stable disease (SD): the increase of the tumor was less than 25% or the decrease of the tumor was less than 50%; and progression disease (PD): the increase of the tumor was over 25% or new lesions formed. The clinical benefit rate (CBR) was calculated in accordance with the formula: CBR = cases of (CR+PR) / total cases × 100%.

(II) Traditional Chinese medical symptom (TCMS) score: The TCMS score was used for the evaluation on the clinical symptoms in patients of both groups, including vaginal bleeding, anxiety and irritability, chest pain, bitterness in mouth and dryness of throat, and generalized pain of the body. The score for each symptom ranges from 0 to 6, and higher score indicates the more evident clinical symptoms and the more severity on the condition.

(III) Serum tumor marker levels: Before and after treatment, 5 ml of fasting venous blood was collected from patients from both groups in the morning, and the supernatant was harvested via the centrifugation to determine the serum levels of carcinoembryonic antigen (CEA), cancer antigen 125 (CA125), and squamous cell carcinoma antigen (SCCA) using enzyme-linked immunosorbent assay as guided by the protocol of the manufacturer.

(IV) Changes of serum T lymphocyte subsets: Before and after treatment, 5 ml of venous blood was collected from patients in both groups in the morning,

and $CD4^+$, $CD8^+$, $CD4^+/CD8^+$, and natural killer (NK) cells were quantified with a flow cytometer.

(V) Adverse effects: the occurrence of adverse effects, including nausea and vomiting, loss of appetite, diarrhea, hair loss and leukopenia, in all patients were monitored.

2.5 Statistical analysis

All data here were analyzed using SPSS 20.0 (IBM, Endicott, NY, USA) and expressed as mean ± standard deviation (SD). Statistical significance, determined with chi-square test and *t* test, was defined when *P*-value was <0.05.

3 Results

3.1 Comparison on clinical efficacy

After treatment, the statistical significance has been evident concerning the distribution of clinical efficacy grades between patients in both groups ($p<0.05$; Table 2), and the CBR of patients in the observation group was evidently higher than that in the control group ($p<0.05$; Table 2).

Table 2. The comparison on the clinical efficacy [cases (%)]

Group	Case	CR	PR	SD	PD	CBR
Control group	48	5 (10.42%)	17 (35.42%)	15 (31.25%)	11 (22.92%)	22 (45.83%)
Observation group	48	10 (20.83%)	22 (45.83%)	10 (20.83%)	6 (12.50%)	32 (66.67%)
Z/χ^2 value			-2.156			4.233
P-value			0.031			0.040

3.2 Comparison on TCMS score

No statistical significance has been reported concerning the TCMS score in all patients prior to the treatment (Table 3, $p>0.05$). However, the TCMS score, including vaginal bleeding, anxiety and irritability, chest pain, bitterness and dryness in mouth and throat, and generalized pain of the body in all patients were decreased after treatment. Much lower scores were further evident in those of the observation group (Table 3, $p<0.05$).

3.3 Comparison on serum tumor marker levels

As illustrated in Table 4, no statistical difference has been reported between the two groups regarding CEA, SCCA and CA125 levels ($p>0.05$). After treatment, the levels in all patients were all significantly decreased, and those levels in the study group have been additionally confirmed to be lower as compared to the data of the control group ($p<0.05$).

3.4 Comparison on changes of serum T lymphocyte subsets

No statistical significance has been found in the comparison of serum $CD4^+$, $CD8^+$, $CD4^+/CD8^+$, and NK cell percentages in patients of both groups. After treatment, serum $CD4^+$, $CD4^+/CD8^+$, and NK cell percentages are increased yet serum $CD8^+$ cell percentage was decreased as compared to those before treatment (Table 5, $p<0.05$). Meanwhile, serum $CD4^+$, $CD4^+/CD8^+$, and NK cell percentages have been further confirmed to be increased yet serum $CD8^+$ cell percentage was decreased in the patients of observation group in comparison with those in control group (Table 5, $p<0.05$).

3.5 Comparison on the adverse effects

In Table 6, the total incidence rates of adverse effects in patients of the control and observation groups were 41.67% and 20.83% (Table 6).

Table 3. Comparison on TCMS score

Group		Control group	Observation group	*t* value	*P* value
vaginal bleeding	Before treatment	4.49±0.98	4.51±1.14	-0.092	0.927
	After treatment	3.32±1.24*	2.46±0.87*	3.933	<0.001
anxiety and irritability	Before treatment	4.44±1.04	4.39±1.21	0.217	0.829
	After treatment	2.85±0.96*	2.12±1.06*	3.537	0.001
chest pain	Before treatment	4.37±1.02	4.31±0.96	0.397	0.767
	After treatment	2.57±0.91*	1.92±1.03*	3.277	0.001
bitterness in mouth and dryness of throat	Before treatment	3.56±0.95	3.41±1.12	0.708	0.481
	After treatment	2.14±0.74*	1.43±0.83*	4.424	<0.001
generalized pain of the body	Before treatment	3.87±1.14	3.93±1.22	-0.249	0.804
	After treatment	2.64±0.94*	1.78±0.88*	4.627	<0.001

*p<0.05, vs. Before treatment

Table 4. Comparison of serum tumor marker levels

Group		Control group	Observation group	*t* value	*P* value
CEA (μg/L)	Before treatment	13.26±2.31	13.08±2.22	0.389	0.698
	After treatment	5.67±1.14*	4.22±0.96*	6.741	<0.001
SCCA (μg/L)	Before treatment	11.06±3.12	10.91±3.36	0.227	0.821
	After treatment	6.34±1.02*	4.71±1.21*	7.136	<0.001
CA125 (U/mL)	Before treatment	48.79±10.88	48.68±11.06	0.049	0.961
	After treatment	32.12±5.61*	25.87±6.17*	5.193	<0.001

*p<0.05, vs. Before treatment

Table 5. Comparison of serum T lymphocyte subsets

组别		Control group	Observation group	*t* value	*P* value
$CD4^+$ (%)	Before treatment	33.05±4.20	33.36±3.86	-0.377	0.707
	After treatment	37.74±3.94*	40.39±3.11*	-3.658	<0.001
$CD8^+$ (%)	Before	33.12±4.05	33.60±4.11	-0.576	0.566

	treatment				
	After treatment	29.12±3.82*	25.83±3.39*	4.463	<0.001
$CD4^+/CD8^+$	Before treatment	1.03±0.25	1.02±0.24	0.200	0.842
	After treatment	1.34±0.32*	1.61±0.33*	-0.407	<0.001
NK (%)	Before treatment	15.89±2.38	15.72±2.06	0.374	0.709
	After treatment	17.52±2.61*	21.07±3.17*	-5.990	<0.001

*$p<0.05$, vs. Before treatment

Table 6. The comparison on the adverse effects [cases (%)]

Group	Case	Nausea and vomiting	Diarrhea	Hair loss	Leukopenia	Total incidence rate
Study group	48	3 (6.25%)	4 (8.33%)	8 (16.67%)	5 (10.42%)	20 (41.67%)
Control group	48	1 (2.08%)	3 (6.25%)	4 (8.33%)	2 (4.07%)	10 (20.83%)
χ^2						4.848
P						0.028

Discussion

According to the belief of TCM, CC belongs to the category of "metrorrhagia and metrostaxis", "Gynecologic Abdominal Lumps", and "Multicolored leucorrhea". It can be further divided into four subtypes of Ganyu Qizhi (liver depression and Qi stagnation), Shi're Yundu (damp-heat with exuberance of virulence), Gansheng Yinxu (deficiency of Yin in liver and kidney) and Zhongqi Xiaxian (the failure of Qi to lift) (8). Among these subtypes, the type of Ganyu Qizhi is more prevalent due to the weakness of internal organs, the invasion of external pathogen, and the depression of the emotions. These adverse outcomes result in the blood stasis, toxin accumulation, liver regulation loss and liver Qi stagnation. Therefore, the weakness of the positive energy in the body, the stagnation of Qi and the blood stasis are the core factors for the pathogenesis, and the treatment thus should be focused on the support and the strengthening on the positive energy, the tonification of Qi, and the dredge on the liver (9). In Gao *et al*.'s study, the combination therapy of accelerated rehabilitation surgery nursing concept and ear acupoint pressing beanson was used to treat patients with CC, while Yan *et al*. adopted Renshen Yangyingdecoction combined with point moxibustion for the treatment on patients with CC (10, 11), which have both achieved better clinical efficacy, indicating that TCM has some certain advantages in the treatment of CC.

In our current study, patients in the control group were treated with chemotherapy alone, while those in the observation group were additionally received Renshen Yangying decoction. The corresponding results presented that after treatment, there existed significant differences in the distribution of clinical efficacy grades between the patients in these two groups. The TCMS score was significantly lower than that before treatment, whilst the CBR was evidently higher, suggesting that the treatment using Renshen Yangying decoction combined with chemotherapy can effectively alleviate the clinical symptoms of CC of Ganyu Qizhi type, with a better clinical efficacy as well. Paclitaxel is a novel anti-microtubule drug, which can inhibit the formation of spindle apparatus during mitosis, while cisplatin is a cell cycle non-specific drug acting on the bases of DNA strands and leading to the obstruction of DNA replication.

Both of these processes can prevent tumor cells from dividing and proliferating, thus exerting anti-tumor efficacy. In the medicinal parts of Renshen Yangying decoction, the ginseng and *Radix Astragali* have the effects on tonifying the vital energy. *Rhizoma Atractylodis Macrocephalae* can remove dampness, induce diuresis, benefit Qi, and stop sweating. *Poria* can strengthen the spleen and promote the excretion. *Radix Angelicae Sinensis*, *Radix Rehmanniae Preparata*, and *Radix Paeoniae Alba* are effective in nourishing and tonifying the blood. *Fructus Schisandrae* and *Radix Polygalae* are capable to calm the mind and nourish the heart. *Cortex Cinnamomi* possesses the efficacy of assisting Yang and warming the kidney. *Pericarpium Citri Reticulatae* regulates Qi, activates the spleen, removes stagnation and eliminates distension. *Rhizoma Polygonati* moistens the lung and nourishes the kidney. *Corii Asini Colla* is able to nourish Yin and tonify blood, while *Pheretima* can clear the heat and promote the circulation. *Radix Glycyrrhizae* tablets can strengthen the spleen and harmonize all the medicines (12, 13). The entire formula is effective in nourishing the spleen and kidney, clearing heat and detoxifying toxins, activating blood circulation, resolving blood stasis and clearing ligaments, and benefiting Qi and nourishing Yin.

CA125 is a polymeric glycoprotein with antigenic properties, CEA is an embryonic carcinogen, and SCCA is a serine protease inhibitor, all of which are common tumor markers for the determination on the severity of CC, and their levels in the serum tend to increase with the development of CC (14, 15). According to the results of this study, after treatment, the serum levels of CEA, SCCA, and CA125 were significantly lower in both groups than before treatment, and the serum levels of CEA, SCCA, and CA125 in the observation group were significantly lower than those in the control group, which showed that the application of Renshen Yangying decoction combined with chemotherapy on patients with CC of Ganyu Qizhi type can help retard the development of CC and reduce the serum tumor marker levels. In line with the results of the modern pharmacological studies, in the formula of Renshen Yangying decoction, Angelica polysaccharide, the main component of *Radix Angelicae Sinensis*, and limonene, the major component of *Pericarpium Citri Reticulatae*, can help repress the proliferation and differentiation of malignant cells. Additionally, the polysaccharide components in *Radix Codonopsis*, *Radix Astragali*, *Radix Rehmanniae Preparata* and *Poria* have anti-tumor and antioxidant effects. The nucleotide components in *Pheretima* can promote the apoptosis of tumor cells, which thus effectively delays the development of cancer (16). Therefore, Renshen Yangying decoction combined with chemotherapy can effectively prevent the deterioration of patients with CC and reduce the levels of serum tumor markers.

Modern medical research has suggested the close relation between the tumorigenesis and the dysregulation on the autoimmune system within the body (17). T cell subsets, those important cells for tumor immune surveillance and the basis for maintaining the body's cellular immune function, can be divided into $CD4^+$ and $CD8^+$ subsets according to the different differentiation antigens on the surface of $CD3^+$ cells, of which $CD4^+$ cells induces humoral and cellular immunity mainly through the secretion of lymphokines. $CD8^+$ cells can inhibit antibody production by B cells, thus reducing the body's immunity (18). NK cells are those non-specific killer cells and act as the body's natural barrier against tumors. Here, it was seen that after treatment, serum $CD4^+$ and CD4+/$CD8^+$ levels, and NK cell percentage in both groups were significantly higher, and serum $CD8^+$ cell percentage was evidently lower, with the degree of improvement in the observation group evidently better than that in the control group. These data thus suggest that the application of Renshen Yangying decoction combined with chemotherapy on patients with CC of Ganyu Qizhi type can help regulate the serum T lymphocyte subsets and enhance their immunity. The polysaccharide in *Radix Codonopsis*, *Radix Astragali*, and *Poria* can improve the biological activity of NK cells and macrophages, enhance the non-specific immunity of the body, and

maintain the stability of immunity by regulating the level of T-lymphocyte subpopulation in the body, thus improving the anti-infection ability of patients. The extract of *Radix Rehmanniae Preparata* can increase the level of leukocytes and promote the biosynthesis of lymphocyte, which thus enhances the immunity of the body, while the major constituting part of *Corii Asini Colla* can exert immuno-protective effects by inhibiting apoptosis of immune cells. The protein and peptide components in *Pheretima* can improve the phagocytosis of macrophages, promote the transformation of body lymphocytes, and enhance the immunity of the body (19). Therefore, the application of Renshen Yangying decoction combined with chemotherapy on the treatment of patients with CC of Ganyu Qizhi type can effectively improve the level of T lymphocyte subpopulation and enhance the immunity of patients. In addition, the results of our present study showed that the incidence of adverse effects in the observation group was significantly lower than that in the control group, which thus suggested that the application of Renshen Yangying decoction combined with chemotherapy for patients with CC of Ganyu Qizhi type can reduce the risk of adverse effects in patients and has a good safety profile. Radiotherapy treatment is based on the principle of attacking the pathogen with poison, and dispersing nodules and stasis, which may lead to internal invasion of heat and poisonous pathogen and even results in the injured Yin, the depleting qi and blood, and even some adverse effects such as nausea and vomiting, diarrhea, hair loss and leukopenia. On the opposite, Renshen Yangying decoction is a medicine that can reduce the toxic effects of chemotherapy and thus effectively reduce the occurrence of adverse effects in the body (20).

In conclusion, the application of Renshen Yangying decoction combined with chemotherapy in the treatment of CC of Ganyu Qizhi type can effectively reduce the clinical symptoms of patients, lower serum tumor marker levels, and improve immune function, along with significant clinical efficacy and good safety.

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Not applicable.

Conflict of Interest

The authors declare no conflicts of interest.

Author contributions

Conceptualization, Z.J.Y and X.Q.H; Data curation, Z.J.Y; Formal analysis, X.Q.H; Methodology, Z.J.Y; Writing-Original draft, X.Q.H and Z.J.Y; Writing-review and editing, X.Q.H and Z.J.Y; All authors have read and agreed to the published version of the manuscript.

Ethics Approval and Consent to Participate

The study was approved by the Medical Ethics Committee, and the patients were informed and consented.

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Availability of Data and Materials

The data presented in this study are available on request from the corresponding author.

Supplementary Material

Not applicable

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