

Effect of Kang' ai Injection (康艾注射液) on Serum Level of Soluble Interleukin-2 Receptor and Vascular Endothelial Growth Factor in Patients with Esophageal Carcinoma during Radiotherapy

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ABSTRACT **Objective:** To observe the effect of Kang' ai Injection (康艾注射液, KAI) on serum level of soluble interleukin-2 receptor (sIL-2R) and vascular endothelial growth factor (VEGF) in patients with esophageal carcinoma (EC) during radiotherapy (RT), and to investigate its synergistic effect with RT and its influence on immunological function of the body. **Methods:** One hundred and seventy patients with EC, who had missed the chance of surgical operational therapy, were assigned to the treated group (90 cases) and the RT group (80 cases), and at the same time a control group consisting of 80 inpatients without tumors was set up. Patients in the RT group were treated with RT alone but KAI was given additionally to those in the treated group, with 50 ml given once per day via intravenous dripping, 15 days as one course, and 2 courses administered in total. The immediate therapeutic efficacy and changes of serum sIL-2R and VEGF levels were observed, and the effect of KAI on patients' quality of life (QOF) was evaluated by Karnofsky scoring. **Results:** In 16 patients of the treated group it was completely remission (CR), in 54 partially remission (PR), in 18 it was stabilized disease (SD) and in 2 progressive disease (PD), with the total effective rate (CR+PR) as 77.8%, while in those of the control group it was 12, 46, 18, 4 and 72.5%, respectively, the immediate therapeutic efficacy in the treated group was somewhat better than that in the RT group, but showed no statistical significance ($P>0.05$). Serum levels of sIL-2R and VEGF in all the patients before treatment were higher than those in the control group, which were decreased after treatment in both groups ($P<0.05$), but the improvement in the treated group was better than that in the RT group, showing significant difference ($P<0.05$), and patients' QOF improved more significantly in the former as well (62.2% vs 40.0%, $P<0.05$). **Conclusion:** KAI in combination with RT in treating patients with EC could enhance the immunological function of patients, improve their QOF and enhance their sensitivity to RT.

KEY WORDS soluble interleukin-2 receptor, vascular endothelial growth factor, esophageal carcinoma, radiotherapy, Kang' ai Injection, sensitivity enhancing

Radiotherapy (RT) is an important approach for treatment of esophageal carcinoma (EC), especially for patients who have missed the chance of surgical section. The efficacy of RT is not satisfactory for its 1-year, 3-year and 5-year survival rates being merely 50%, 20%, and 10% respectively⁽¹⁾.

Researches in recent years showed that better efficacy can be obtained by combining RT with Chinese herbal medicines for EC that incapable to be resected. Kang' ai Injection (康艾注射液, KAI) is a Chinese herbal preparation consisting of Ginseng, milkvetch root and kushenin, which has been applied as an auxiliary treatment for tumor. In this study, the clinical efficacy of the combined treatment with RT and KAI in treating EC was observed, and the serum levels of soluble interleukin-2 receptor (sIL-2R) and vascular endothelial growth factor (VEGF) were detected in order to explore the effect of KAI in enhancing the efficacy of RT, inhibiting the tumor as

well as its influence on the immune function of the organism and quality of life (QOL).

METHODS

General Materials

The 170 patients of EC were inpatients of the authors' hospital from March 2001 to March 2005, who were incapable to accept with surgical resection with their confirmed diagnosis by endoscopic biopsy. All of them were proved to be normal in functions of heart, liver and kidney before treatment and their Karnofsky scoring (KPS) was over 50 scores. They

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were assigned to two groups. The 90 patients in the treated group were 54 males and 36 females; aged 39—68 years with the median age of 51; staging of disease⁽²⁾ showed that 5 patients were of stage II, 31 of stage III and 54 of stage IV; KPS was 51—60 scores in 28, 61—70 scores in 35, 71-80 scores in 17 and over 80 in 10.

The 80 patients in the RT group were 48 males and 32 females; aged 35—63 years with the median age of 47; 3 patients were of stage II, 28 of stage III and 49 of stage IV; KPS was 51—60 scores in 25, 61—70 scores in 31, 71—80 scores in 16 and over 80 in 8. No statistical difference was shown between the two groups in clinical data such as age, sex, condition of disease and KPS, and so they were comparable ($P>0.05$).

Besides, 80 inpatients without malignant tumors selected from the same department during the same period were allocated into the control group, who were 46 males and 34 females, aged 28—56 years, with median age of 35, and their KPS was 51—60 scores in 16, 61—70 scores in 25, 71—80 scores in 29 and over 80 in 10.

Treatment

All the patients received RT according to the following protocol: routine fractionated radiotherapy was applied firstly for 4 weeks as the preceding 2/3 therapeutic course by giving 40 Gy in 20 divisions, which was followed by a late course accelerated hyper-fractionated three-dimensional conformal radiotherapy in the subsequent 1/3 course by giving 1.5 Gy every time, twice a day with a total of 15—21 Gy given in 5—7 days. The total dosage reached 55—61 Gy, given in 30—34 divisions and the total course lasted for 33—35 days.

For the treated group, while it was receiving RT, KAI (made by Changbai Mountain Pharmaceutical Co. Ltd.) was given additionally, which consisted of ginseng, milkvetch root and kushenin. It was added into 250 ml of 5% glucose solution or normal saline for intravenous dripping once a day for 15 successive days as one therapeutic course, and the next course would begin after a 5-day interval, with two courses administered altogether.

For the control group, neither KAI or RT was given.

Determination of Serum Concentrations of sIL-2R and VEGF

Fasting whole blood (4 ml) of the subjects was

collected on the morning of the 1st and 35th day of treatment. The blood sample was centrifugated to get the serum, which was preserved at -20 °C for testing within one week. The serum concentration of sIL-2R and VEGF was determined using double antibody sandwich ELISA with the test kit purchased from Jingmei Biotech Company and GB Co. USA, respectively. It was conducted strictly according to the instruction.

Evaluation of Patients' QOF

KPS was adopted, and QOF was evaluated as improved when KPS increased by 10 scores or more; as deteriorated when KPS decreased by 10 scores or more and as stabilized when KPS was changed by less than 10 scores.

Efficacy Evaluation and Classification of Adverse Reaction

According to the standard promulgated by WHO⁽³⁾, the efficacy was classified into 4 grades: complete remission (CR), partial remission (PR), stabilized disease (SD) and progressive disease (PD), the total effective rate was calculated by (Cases of CR+ Cases of PR)/Total Cases × 100%.

The toxic and adverse reactions, with appearance of nausea, vomiting, lassitude, decrease of leukocyte, cavities, radiation pneumonia, etc. was classified to 5 grades as the follows: Grade 0: none, grade I : mild, grade II : moderate, grade III : severe and grade IV : so severe as to threaten patient' s life⁽⁴⁾.

Statistical Analysis

With the software package SPSS 12.0, the measurement data was analyzed by *t*-test and the enumeration data with χ^2 analysis, with $P<0.05$ regarded as significant difference.

RESULTS

Comparison of Immediate Effects between Groups

See Table 1. The total effective rate in the treated group was 77.8%(70/90) and in the RT group 72.5%(58/80). Comparison between them showed that the former was somewhat higher, but with no significance in statistical analysis ($P>0.05$).

Table 1. Comparison of Immediate Effects between the Two Groups [Case(%)]

Group	Case	CR	PR	SD	PD
Treated	90	16(17.8%)	54(60.0%)	18(20.0%)	2(2.2%)
RT	80	12(15.0%)	46(57.5%)	18(22.5%)	4(5.0%)

Changes of Serum Levels of sIL-2R and VEGF

As shown in Table 2, before treatment, the serum levels of sIL-2R and VEGF in both groups of patients were higher than those in the control group respectively ($P < 0.05$), which were lowered significantly after treatment in both groups ($P < 0.05$), but still higher than those in the control group ($P < 0.05$). The lowering was more significant in the treated group than that in the RT group ($P < 0.05$).

Table 2. Changes of Serum Levels of sIL-2R and VEGF among Groups ($\bar{x} \pm s$)

Group	Case	Time	sIL-2R (U/ml)	VEGF ($\mu\text{g/L}$)
Control	80		225.3 \pm 107.2	157.6 \pm 21.4
Treated	90	BT	618.5 \pm 211.3*	357.6 \pm 31.9*
		AT	288.5 \pm 111.7 * Δ Δ	238.1 \pm 27.3* Δ Δ
RT	80	BT	609.6 \pm 213.1*	341.2 \pm 24.7*
		AT	351.8 \pm 143.5* Δ	297.8 \pm 29.7* Δ

Notes: * $P < 0.05$, compared with the control group; $\Delta P < 0.05$, compared with BT in the same group; $\Delta\Delta P < 0.05$, compared with the RT group after treatment; BT: before treatment; AT: after treatment

Comparison of KPS between the Two Groups

As shown in Table 3, the improvement rate of KPS in the treated group was higher than that in the RT group (62.2% vs 40.0%), showing significant difference between them ($P < 0.05$).

Table 3. Comparison of KPS between the Two Groups [Case(%)]

Group	Case	Improved	Stabilized	Deteriorated
Treated	90	56(62.2%)*	26(28.9%)	8(8.9%)
RT	80	32(40.0%)	20(25.0%)	28(35.0%)

Note: * $P < 0.05$, compared with the RT group

Toxic and Adverse Reactions to RT

The rate of WBC lowering by two grades and over two grades were 29.3%, 12.1% in the treated group and 36.7%, 21.6% in the RT group respectively; the occurrence rate of radiation pneumonia by two grades and over two grades were 26.7%, 12.4% and 35.5 \pm 5%, 16.9% respectively, the treated group showed that the occurrence of toxic and adverse reactions was slightly lower than that in the RT group, but the difference was of no statistical significance ($P > 0.05$). And the comparison of other adverse reactions between the two groups also showed no significant difference.

DISCUSSION

Soluble interleukin 2 receptor (sIL-2R) is P55 protein sloughed out from the surface of activated T lymphocytes, and drifts in the serum and urine, its existence marking the activation of lymphocytes⁽⁶⁾. As a transport protein of interleukin-2 (IL-2), sIL-2 could conjugate with IL-2 to prolong its half-life in blood. It is also an immuno-suppressor, showing the blocking factor-like action, could neutralize the IL-2 of T lymphocytes to reduce the auto-secretory effect of organism⁽⁶⁾. For this reason, it could reflect to some extent the immunological function of organism. Moreover, some studies have proved that the serum level of sIL-2 was significantly elevated in patients with malignant tumor, and so it could be regarded as a mark of tumor's active proliferation⁽⁷⁾.

VEGF is a new mark for angiogenesis in tumor, and a vascular endothelial growth factor with the highest specificity and the strongest potent for inducing angiogenesis. It has been proved that there exists the expression of VEGF and its receptor in some malignant solid tumor like pulmonary carcinoma, which is related with the number of vessels in tumor^(8,9). Besides, the evidently elevated serum concentration of VEGF has also been found in patients with several solid tumors or with metastatic tumor. Therefore, serum level of VEGF is also regarded as a biological mark of some malignant tumors' proliferative activity⁽¹⁰⁾.

KAI is a Chinese herbal preparation manufactured through fine extraction with modern technique, which could be used in combination with radio- and/or chemo-therapy, showing an inhibitory action on cancer cells and able to reduce the toxic and adverse reactions due to radio- and chemo-therapy. The active components of ginseng are mainly panaxans, ginsenosides, etc. which could induce stromal cells in hemopoietic micro-environment to secrete interleukin-3 (IL-3), granulocyte colony stimulating factor (GM-CSF), etc. and induce differentiation of hemopoietic progenitor^(11,12). Milkvetch root, which contains multiple ingredients such as flavones, astragalus polysaccharides and astragaloside, could raise the activity of T-lymphocyte subsets and NK cells in tumor patients so as to improve their immunity. Astragalus polysaccharides could also promote the hemopoietic function by stimulating the karyocyte to secrete hemopoietic cytokines like granulocyte colony-stimulating factor (G-CSF) and GM-CSF⁽¹³⁾. The main ingredients of flavescent sophora root, matrine and oxymatrine

could stably inhibit the proliferation of endothelial cells induced by tumor cells⁽¹⁴⁾.

Results in the study showed that KAI tends to enhance the clinical efficacy in assisting RT for treatment of EC, could evidently raise QOF of tumor patients, effectively reduce the toxic and adverse reactions and increase patients' sensitivity to RT. Moreover, it could obviously lower the serum levels of sIL-2R and VEGF. So, it is held that KAI could, to certain extent, enhance the immunological function of organism, inhibit the proliferative activity, invasion and metastasis of tumor, and improve the prognosis of patients. But it is necessary to have further long-term and large sampled clinical observation and basic study.

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