

Prognostic value of serum Tie-2 and vascular endothelial growth factor levels in cancer patients

M.M. SEKER, E. SANCAKTAR¹, D.O. ACIBUCU¹, A.K. FILIZ², K. DEVECI¹, A. BAHCECI, T. KACAN, N. BABACAN, S. YUCE³

Department of Medical Oncology, ¹Department of Biochemistry, ²Department of Physiology, ³Department of Ear & Nose & Throat, Cumhuriyet University Faculty of Medicine, Sivas, Turkey

Abstract. – BACKGROUND: Angiogenesis is a very essential process in tumor biology. Vascular endothelial growth factor (VEGF), angiopoietin and its receptor (TIE-2) are very important mediators for angiogenesis. In this trial, we aimed to analyze the role of these mediators on chemotherapy response and survival.

PATIENTS AND METHODS: Forty four cancer patients and 22 healthy controls were included in the study. Baseline serum samples were obtained from all participants and post-chemotherapy serum samples were obtained from the cancer patients. Serum vascular endothelial growth factor and TIE-2 levels were measured with quantitative enzyme-linked immunosorbent assay techniques.

RESULTS: The baseline serum vascular endothelial growth factor level was 187.5 and 120.2 pg/ml in cancer patients and the control group ($p = 0.006$). The baseline serum TIE-2 level was 615.9 and 242.5 pg/ml in the patients and control group ($p < 0.001$). There was a significant difference between patients' baseline and post-chemotherapy VEGF levels (111.9 pg/ml; $p < 0.001$) and patients' baseline and post-chemotherapy TIE-2 levels (344.5 pg/ml; $p < 0.001$). The overall survival rate was better in patients who had lower baseline VEGF and TIE-2 levels and whose TIE-2 level had decreased with chemotherapy.

CONCLUSIONS: Higher baseline TIE-2 and VEGF levels are related and worsen survival. Decreasing levels of TIE-2, but not VEGF, which, with chemotherapy, may be predictive for survival.

Key Words:

Tie-2, Vascular endothelial growth factor, Cancer, Prognosis.

esis is performed by several proteins, such as fibroblast growth factor, hypoxia inducible factor, vascular endothelial growth factor (VEGF), angiopoietin (ANG), and its receptor TIE-2, many investigators have focused on VEGF pathway^{1,2}. Agents, such as bevacizumab, sunitinib, and sorafenib, using this pathway are part of standard therapies for many malignancies, including colorectal cancer, non-small cell lung cancer, renal cell cancer, and hepatocellular cancer^{3,4}. However, physicians gain little or no benefit when using these agents⁵⁻⁷.

The ANG-TIE signaling pathway consists of three ligands (ANG-1, ANG-2 and ANG-4) and two tyrosine kinase receptors (TIE-1 and TIE-2)^{3,8,9}. TIE-1 has no known ligand, and the role of ANG-4 in tumorigenesis needs to be further investigated. ANG-1 is a TIE-2 agonist and ANG-2 is generally, but not always, a TIE-2 antagonist^{10,11}. The binding of ANG-1 to TIE-2 activates the Akt signaling pathway and results in blood vessel stability, endothelial cell survival, and decreased permeability¹²⁻¹⁴. This activation is correlated with increased invasive potential, lymph node, and distant metastasis in many cancers¹⁵⁻¹⁷.

VEGF is one of the most important mediators of ANG-TIE system⁴. In this trial, we aimed to analyze the importance of VEGF and TIE-2 levels in lung and colorectal cancer patients and the effects of conventional chemotherapeutic agents on VEGF and TIE-2 levels.

Introduction

Tumor angiogenesis, the generation of tumor associated vascular system, is one of the most current issues regarding tumor biology and cancer treatment. Although stimulation of angiogen-

Patients and Methods

This study includes cancer patients older than 18 years old who were treated at Cumhuriyet University Medical Faculty's Medical Oncology Department. Institutional Ethical Committee ap-

proval was obtained for this study. Patients' ages, genders, primary tumors, tumor stages, types of chemotherapy regimens, and complete blood count parameters tumor markers were recorded. Tumor stages were determined according to the TNM Classification of Malignant Tumours 7th edition (TNM) staging system.

Both non-metastatic and metastatic patients were included in this study. Serum samples for baseline serum VEGF and serum TIE-2 levels were obtained 3-4 weeks after the radical surgery and before the first cycle of adjuvant chemotherapy in non-metastatic patients. In metastatic patients, serum samples for baseline serum VEGF and serum TIE-2 levels were obtained before first cycle of chemotherapy. All patients had received 4-6 cycles of chemotherapy. Serum samples for post-chemotherapy VEGF and TIE-2 had obtained one week after the end of 4-6 cycles of chemotherapy.

The control group consisted of age- and sex-matched healthy controls free of malign, inflammatory, and connective tissue diseases. Serum samples for baseline serum VEGF and serum TIE-2 were also obtained from the control group.

Sera was obtained from all subjects, and was separated immediately by centrifugation at 4,000 g for 10 min and frozen at -80 °C. Plasma concentrations of VEGF and TIE-2 were measured in duplicate with a quantitative enzyme-linked immunosorbent assay technique (Boster Biological Technology Co., Ltd., Wuhan, China) according to the manufacturer's guidelines.

Statistical Analysis

Statistical analysis was performed using SPSS for Windows version 17.0 (SPSS, Chicago, IL, USA). Baseline characteristics of groups were compared using χ^2 tests. Non-parametric tests (Mann-Whitney test for two independent samples and Wilcoxon test for two related samples) were used to compare the two groups. Kaplan-Meier survival analysis was carried out for progression

free survival (PFS) and overall survival (OS). Survival analysis was based on the date of diagnosis. A 2-sided p value < 0.05 was considered to be statistically significant.

Results

There were a total of 44 patients (36 male, 8 female) with non-small cell lung cancer and colorectal cancer and 22 healthy controls. The patients' median age was 60.5 (45-76) years. Twenty two (50.0%) patients had non-small cell lung cancer (NSCLC) and 22 patients (50.0%) had colorectal cancer (CRC). Twenty two (50.0%) patients were non metastatic at time of diagnosis and 22 patients (50.0%) were metastatic at the time of diagnosis. The patients' baseline VEGF and TIE-2 levels were higher than the control group's levels. After 4-6 cycles of chemotherapy, TIE-2 levels had decreased in 30 (68.2%) patients and VEGF levels had decreased in 34 (77.3%) patients. Also, the patients' baseline VEGF and TIE-2 levels were higher than post-chemotherapy VEGF and TIE-2 levels (Table I).

The median TIE-2 and VEGF levels were not different between NSCLC and CRC patients. However, there was a positive correlation between stage and TIE-2/VEGF levels. The median of the baseline TIE-2 levels was 491.0 ± 261.9 and 852.0 ± 408.3 pg/ml in non-metastatic and metastatic patients respectively ($p = 0.034$). The median of the baseline VEGF levels was 140.2 ± 405.5 and 250.7 ± 408.1 pg/ml in non-metastatic and metastatic patients respectively ($p = 0.011$).

The median follow up time was 26 months (12-88 months). During the follow up period, disease progression was detected in 26 (59.1%) patients, and 14 (31.8%) patients had died. Median progression free survival (PFS) was 24 months and median overall survival (OS) was 44 months. The median PFS was 20 months in patients whose baseline TIE-2 level was higher than 600 pg/ml

Table I. Patients' baseline and postchemotherapy VEGF and TIE-2 levels.

	Patients	Controls	p value
VEGF-baseline (pg/ml)	187.5 ± 312.9	120.2 ± 155.6	0.006
VEGF-postchemotherapy (pg/ml)	111.9 ± 203.6		
p value	< 0.0001		
TIE-2-baseline (pg/ml)	615.9 ± 388.9	242.5 ± 179.7	< 0.0001
TIE-2-postchemotherapy (pg/ml)	344.5 ± 245.7		
p value	< 0.0001		

and 24 months PFS was 63.6% in patients whose baseline TIE-2 was lower than 600 pg/ml ($p < 0.0001$). Also the PFS was 20 months in patients whose TIE-2 levels were not decreased with chemotherapy. The 24 months PFS was 60% in patients whose TIE-2 levels had decreased with chemotherapy ($p < 0.0001$). The median PFS was 20 months in patients whose baseline VEGF level was higher than 200 pg/ml and 24 months PFS was 63.6% in patients whose baseline VEGF was lower than 200 pg/ml ($p = 0.001$). The median PFS was 24 months both in patients whose VEGF levels had decreased with chemotherapy and in patients whose VEGF levels had not decreased. The median OS was 36 months in patients whose baseline TIE-2 level was higher than 600 pg/ml and 44 months PFS was 65.6% in patients whose baseline TIE-2 was lower than 600 pg/ml ($p = 0.014$). The OS was 26 months in patients whose TIE-2 levels had decreased with chemotherapy and 36 months OS was 75.0% in patients whose TIE-2 levels had not decreased with chemotherapy ($p < 0.0001$). The median OS was 36 months in patients whose baseline VEGF level was higher than 200 pg/ml and 44 months PFS was 63.6% in patients whose baseline VEGF was lower than 200 pg/ml ($p = 0.039$). The OS was 44 months in patients whose VEGF level had not decreased with chemotherapy and the 36 months OS was 51.1% in patients whose VEGF levels had decreased with chemotherapy ($p > 0.05$).

Discussion

Cancer is one of the most common cause of deaths all around the world and it is expected that the incidences of cancer-related deaths will be higher in the next two or three decades^{18,19}. Thus, many investigations have focused on cancer prevention or treatment. The treatment success is directly related to our knowledge about tumor biology. Angiogenesis is an essential process in most human malignancies. With the increasing usage of anti-angiogenic therapies, physicians want to predict both the effectiveness of their therapies and the prognosis.

There are conflicting results about the predictive and prognostic value of serum VEGF. The number of studies about the role of ANG-TIE system in cancer patients is increasing. Engin et al^{20,21} investigated the importance of baseline serum ANG-1, ANG-2 and TIE-2 levels in colon cancer and gastric cancer patients. They found that

ANG-2 and TIE-2 were higher in patients than in a healthy group. However, the ANG-1 levels were not different between groups. In Chin et al's study²², higher preoperative TIE-2 and VEGF levels have been found to be associated with a worsened survival. In our study, both the VEGF and the TIE-2 levels were higher in cancer patients than in the healthy group. Our report also showed that the VEGF and TIE-2 levels were correlated with the tumor stage. Both VEGF and TIE-2 levels were higher in metastatic patients. In Engin et al's study, ANG-2 was also higher in stage III colon cancer patients compared with stage II patients. Another remarkable finding from our study was that the type of tumor was not as important as tumor stage in terms of VEGF and TIE-2 levels as we did not find a significant difference between NSCLC and CRC patients.

To the best of our knowledge, this is the first and only study investigating the effects of chemotherapeutic agents in VEGF and TIE-2 levels and the survival effect of VEGF and TIE-2 response to the chemotherapeutic agents. At present, most anti-angiogenic therapies act by blocking the VEGF receptor. However, the number of studies about the agents blocking the ANG-TIE-2 system is increasing and results are promising. However, most of them are in early phases^{23,24}. We found that conventional chemotherapeutic agents other than VEGF antagonist can also decrease VEGF and TIE-2 levels. This study is compatible with the other works, and has demonstrated that higher baseline VEGF and TIE-2 levels are related with worsened PFS and OS^{15,22,25}. Additionally, this report also demonstrated that PFS and OS are better in patients whose TIE-2 levels had decreased with chemotherapy. This survival benefit, however, could not be detected in patients whose VEGF levels had decreased with chemotherapy.

Conclusions

Angiogenic process is very important in tumor biology, both for non-metastatic and metastatic cancer, and TIE-2 and VEGF are two important components of this process. Higher baseline TIE-2 and VEGF levels are also related to higher tumor stages and worsened survival. Decreasing levels of TIE-2, but not VEGF, with chemotherapy may be predictive of survival, and many conventional chemotherapeutic agents may influence angiogenic system, VEGF, and TIE-2 levels.

Conflict of Interest

The Authors declare that there are no conflicts of interest.

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