

# C-reactive protein, procalcitonin, interleukin-6, vascular endothelial growth factor and oxidative metabolites in diagnosis of infection and staging in patients with gastric cancer

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## Abstract

**AIM:** The current study was to determine the serum/plasma levels of VEGF, IL-6, malondialdehyde (MDA), nitric oxide (NO), PCT and CRP in gastric carcinoma and correlation with the stages of the disease and accompanying infection.

**METHODS:** We examined the levels of serum VEGF, IL-6, PCT, CRP and plasma MDA, NO in 42 preoperative gastric cancer patients and 23 healthy subjects. There were infection anamneses that had no definite origin in 19 cancer patients.

**RESULTS:** The VEGF levels (mean±SD; pg/mL) were 478.05±178.29 and 473.85±131.24 in gastric cancer patients with and without infection, respectively, and these values were not significantly different ( $P>0.05$ ). The levels of VEGF, CRP, PCT, IL-6, MDA and NO in cancer patients were significantly higher than those in healthy controls and the levels of CRP, PCT, IL-6, MDA and NO were statistically increased in infection group when compared with non-infection group ( $P<0.001$ ).

**CONCLUSION:** Although serum VEGF concentrations were increased in gastric cancer, this increase might not be related to infection. CRP, PCT, IL-6, MDA and NO have obvious drawbacks in the diagnosis of infections in cancer patients. These markers may not help to identify infections in the primary evaluation of cancer patients and hence to avoid unnecessary antibiotic treatments as well as hospitalization. According to the results of this study, IL-6, MDA, NO and especially VEGF can be used as useful parameters to diagnose and grade gastric cancer.

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## INTRODUCTION

Adenocarcinoma of the stomach (ACS) is the second most

common cancer worldwide. Efforts directing at prevention, early detection and intensive therapy have been effectively proved early diagnosis saves lives. There are 2 distinct biological and etiologic subtypes of ACS: epidemic-intestinal type and endemic-diffuse-infiltrative type. Diet and environment are important factors for the epidemic form of ACS, which is associated with chronic atrophic gastritis and intestinal metaplasia of gastric mucosa. Genetic instability, inactivation of tumour suppressor genes, whereas activation of oncogenes, expression of growth factors, cytokines and angiogenic factors promote tumour progression and invasion<sup>[1-5]</sup>.

Angiogenesis in tumours, as well as in a number of physiological (vasculogenesis, wound repair, *etc.*) and pathological (diabetic retinopathy, macular degeneration, rheumatoid arthritis, cancers and psoriasis) conditions, is induced by a number of factors, of which the most prevalent and only endothelium-specific one is vascular endothelial growth factor VEGF, also referred to as vascular permeability factor<sup>[6-9]</sup>. VEGF is a potent angiogenic mediator described in a wide variety of tumours, and it stimulates angiogenesis and increases vascular permeability<sup>[10]</sup>. It has been reported to be synthesized and secreted by gastro-intestinal tract adenocarcinomas. Recent studies have revealed a relationship between the increased expression of VEGF and tumour growth, distant metastasis, and the poor prognosis of patients with gastric carcinoma<sup>[11-13]</sup>. Experimentally, hypoxia and ischemia are known to induce angiogenesis in various cultured tumour cells by increasing VEGF<sup>[14-17]</sup>. Increased risk of malignancy is associated with chronic inflammation caused by chemical and physical agents<sup>[18]</sup>, and autoimmune and inflammatory reactions of uncertain etiology<sup>[19]</sup>. Gastric cancer is a global health problem. Several factors are suspected to play a role in gastric carcinogenesis, including effects of diet, exogenous chemicals, intragastric synthesis of carcinogens, genetic factors, infectious agents and pathological conditions in the stomach (such as gastritis). Studies have shown that high intake of smoked, salted and nitrated foods and carbohydrates and low intake of fruits, vegetables and milk significantly increase the risk for gastric cancer<sup>[20,21]</sup>. Dietary factors, such as consumption of salted and nitrated food, are believed to be primarily responsible for the high incidence and mortality rates of gastric cancer in Turkey. Clinical and epidemiological studies have suggested a link between gastric cancer and concurrent or previous infection with a bacterium or virus. *Helicobacter pylori* (*H. pylori*) is believed to play a role in about 60% of gastric cancer cases. Gastric carcinogenesis is a typical infection and inflammation-associated pathological alteration in which *H. pylori* plays a critical role. Current knowledge of the detailed mechanisms underlying the interplay between biological modulators and lesions induced by *H. pylori* is still incomplete<sup>[22]</sup>. Immune response to *H. pylori* involves a complex network of inflammatory mediators including chemokines [*e.g.* interleukin (IL)-8], pro-inflammatory cytokines (*e.g.* IL-1, IL-6, tumor necrosis factor  $\alpha$ ) and immunosuppressive peptides (*e.g.* IL-10)<sup>[23]</sup>. It is believed that chronic infection with *H. pylori* leads to alterations of the cell cycle, including increased epithelial

cell replication, increased rate of cell death (apoptosis) and production of oxidants. For instance, reactive oxygen species (ROS) could be elevated and cause oxidative DNA lesions in gastric epithelial cells during *H pylori*-elicited inflammation<sup>[24]</sup>. This, in combination with depletion of antioxidant defenses, may predispose to carcinogenesis by increasing the likelihood of DNA mutagenesis. Accumulation of mutations may then lead to metaplasia, dysplasia and gastric cancer. The level of oxidative DNA lesions was significantly higher in patients with chronic atrophic gastritis or gastric cancer than in normal gastric tissues, indicating that a progressive accumulation of oxidative DNA lesions could play a major role in gastric carcinogenesis<sup>[24-28]</sup>. Lipid peroxidation is initiated by free-radical attack of membrane lipids, generating large amounts of reactive products such as malondialdehyde (MDA), which have been implicated in tumour initiation and promotion.

Cytokines have been proposed to play an important role in *H pylori*-associated gastric inflammation and carcinogenesis, but the exact mechanism remains unclear. Several studies have indicated that infection with *H pylori* induced the expression and production of various cytokines in gastric mucosa, epithelial cells, or macrophages<sup>[29,30]</sup>. IL-6, but not other cytokines, has recently been reported to be induced by *H pylori* infection in gastric epithelial cells<sup>[31]</sup>. In addition, IL-6 has a strong activity in stimulating the growth of human gastric cancer cell lines. These findings suggest that IL-6 may have a potential role in the pathogenesis of gastric cancer<sup>[32]</sup>.

Recognized interactions and cytokine pathways mediated by surrounding stromal cells and between stromal cells and tumour play important roles in gastric carcinogenesis. VEGF and IL-6 are associated with the disease status of gastric carcinoma<sup>[33]</sup>. However, their relationship remains unclear. IL-6 produced in malignant tumours and inflammatory tissues has a stimulatory effect on tumour growth and a direct angiogenic activity<sup>[34]</sup>. Reactive oxygen species (ROS) and certain inflammatory cytokines always elevate during the human carcinogenic process. However, the biological significance of the interplay between ROS and inflammatory cytokines remains elusive. IL-6 is a pleiotropic cytokine capable of not only inducing growth and differentiation but also modulating cellular apoptosis in many cell types<sup>[35-37]</sup>.

In normal gastric mucosa, nitric oxide (NO) inhibits acid secretion, stimulates mucus and bicarbonate secretions, elevates mucosal blood flow and accelerates the healing of ulcers<sup>[38,39]</sup>. In some tumour tissues, NO has been found to enhance tumour angiogenesis and to induce vasodilatation, thus accelerating tumour growth<sup>[40]</sup>. NO may differentially affect tumour progression depending on the levels and timing of release. NO can also decrease adhesion molecule expression, increase vascular permeability and angiogenesis<sup>[41]</sup>. In addition to causing the initial mutagenic event leading to disease, NO has also been implicated, seemingly paradoxically, both in tumour reduction/prevention and in tumour promotion.

Chronic infection and inflammation have long been recognized as risk factors for a variety of human cancers. Procalcitonin (PCT), a 116 amino acid propeptide of calcitonin, has been proposed as a new diagnostic marker of severe infections. PCT is presumably synthesized in tissues other than thyroid C-cells, which are the source of calcitonin (CT) in normal physiology. At the same time, PCT is excreted by numerous cancers and might well be a useful biological marker for the follow-up of productive tumours. Enhanced production of calcitonin-like peptides was also seen in patients with different malignancies<sup>[42,43]</sup>. Another infection marker, C-reactive protein (CRP), is most widely used as a marker of ongoing infection in clinical practice. The use of CRP values to diagnose infection in cancer patients was often difficult, because the underlying malignancy also induced CRP

production in hepatocytes<sup>[42,44,45]</sup>. Infections in cancer patients are milder than sepsis, and they are often associated with tumour necrosis and obstructions. Although the underlying cancer seems to disturb PCT and CRP, there may be quantitative differences and these markers did not help to identify infections and stage of cancer.

In this study, we investigated the circulating levels of VEGF and IL-6 which are markers directly related with tumour, and MDA and NO which are oxidative stress markers, and determined the correlation between these markers and infection markers (PCT and CRP) in cancer patients with abdominal pain, vomiting, malnutrition, weight loss, and increasing tendency of infection.

## MATERIALS AND METHODS

### Materials

Forty-two gastric cancer patients admitted to Firat University Hospital between 2001 and 2002 were included in the study. Serum VEGF, IL-6, PCT, CRP and plasma MDA, NO (nitrite/nitrate) levels were measured in 42 patients with gastric cancer, and 23 healthy subjects. The patients aged 46-78 years (average age 62). Nineteen were men, and 23 were women. Characteristics and demographic data of the study population are shown in Table 1. None of the patients received chemotherapy or radiation therapy before surgery. Each patient underwent the following procedures: chest radiography, stomach imaging (ultrasound or CT scan), endoscopy, other organ computed tomography (CT) scan, mammography, breast ultrasound, clinical diagnosis and hematological and biochemical profiles. The clinicopathological parameters were studied for prognostic values including age, tumour size, histological type, lymph node involvement, vascular involvement, distant metastasis, and serosal invasion. The pathologic diagnosis and classification were made according to the Union Internationale Contre le Cancer (UICC) TNM clinical classification for gastric cancer<sup>[46]</sup>. The histological diagnosis was based on morphological examination of hematoxylin and eosin-stained, routinely processed specimens. Tumour resection was carried out in all the patients (including 42 preoperative patients who subsequently underwent surgical resection for cure, and 4 patients with inoperable tumours). The control group consisted of 23 healthy controls (male: 11, female: 12, median age: 61 years (38-82 years)). ‘ ‘Healthy’ ’ was defined as being free from diabetes mellitus, arteriosclerosis and acute illness and without hospitalization for any illness during the previous 2 years.

**Table 1** Characteristics of studied populations and demographic data

	No. of subjects	Median age (yr) (interquartile range)	Sex (M/F)
Healthy control	23	61 (38-82)	11/12
Gastric cancer	42	62 (46-76)	19/23
Histologic type			
Adeno carcinoma	34	61 (46-76)	18/16
Signet-ring cell carcinoma	4	51 (48-62)	3/1
Mucinous carcinoma	4	56 (51-58)	1/3
Stage			
I	12	60 (46-74)	7/5
II	16	56 (51-64)	8/8
III	10	65 (62-72)	4/6
IV <sup>1</sup>	4	70 (68-76)	2/2

M, male; F, female. <sup>1</sup>Distant metastatic sites: peritoneal (2), hepatic (1), multiple sites (1).

Peripheral venous blood samples were collected in routine biochemical test tubes for VEGF, IL-6, PCT and CRP analysis and in EDTA.K3 tubes for MDA and NO (nitrite/nitrate) determination. Plasma and serum tubes were centrifuged at 3 500 r/min for 10 min at 4 °C. The serum and plasma were separated and stored at -70 °C until further processing.

### Methods

Serum VEGF levels were determined by a quantitative sandwich enzyme immunoassay technique (Quantikine R&D Systems, Minneapolis, Minn.) according to the manufacturer's instructions. The system used a solid phase monoclonal antibody and an enzyme-linked polyclonal antibody rose against recombinant human VEGF. For each analysis 100 µL of serum was used. All the analyses and calibrations were performed in duplicate. The calibrations on each microtiter plate included recombinant human VEGF standards. Results were calculated from a standard curve generated by a four-parameter logistic curve-fit and expressed in pg/mL. The coefficient of variation of interassay determinations reported by the manufacturer varied from 6.2% to 8.8% when the VEGF concentrations ranged between 50 and 1 000 pg/mL. Human IL-6 levels were determined by enzyme linked immunosorbent assay (ELISA) kits (Bender MedSystems, MedSystems Diagnostics, Vienna, Austria) according to the manufacturer's guidelines.

Measurement of plasma MDA levels, MDA, the end product of lipid peroxidation, reacted with tiobarbutiric acid (TBA) to produce a fluorescence product, which was measured spectrophotometrically. Thus, plasma MDA levels were measured as an index of lipid peroxidation according to the TBA spectrophotometric method that was modified from the methods of Satoh and Yagi<sup>[47]</sup>. 1, 1, 3, 3, tetraethoxypropane was used as standard, and plasma MDA levels were expressed as nanomoles per milliliter.

The measurement of plasma NO was difficult because this radical was poorly soluble in water and had a short half-life in

tissue (10-60 s), but its half-life might be as long as 4 min in the presence of oxygen. For these reasons, the determination of NO itself was difficult and required the handling of radioisotopes. In spite of this, the end products, nitrate and nitrite, were preferentially used in clinical biochemistry. Nitrite, a stable end-product of NO, was measured in plasma by using the spectrophotometric Greiss reaction<sup>[48]</sup>. One thousand µL experimental samples of deproteinised plasma was reacted with 500 µL N-naphthylethylenediamine, 10 g/L sulfanilamide for 45 min at room temperature and analyzed by spectrophotometry at 545 nm. Concentrations were determined by comparison with sodium nitrite. The lower limit of detection was 0.2 µmol/L.

The concentration of CRP in serum was determined by turbidimetric assay using Alfawassermann kits. Leukocyte counts were determined using a Beckman Coulter cell counter (Coulter Corporation, Miami, USA). Serum PCT concentrations were measured by immunoluminometric assay using Brahms kits, according to the manufacturer's protocol (LumiTest; Brahms Diagnostica, Berlin, Germany). PCT levels were found to be <0.08 ng/mL in plasma from 23 healthy controls.

Statistical analysis was carried out using the Statistical Package for the Social Sciences (SPSS). The results were expressed as mean±SE. Mann-Whitney *U*-test and Pearson's rank correlation test were used to compare levels of parameters among the patients. A *P* value <0.05 was considered statistically significant. Variables found to be significant at a level of *P*<0.05 were considered eligible for multivariate regression analysis.

### RESULTS

Our findings on the assessed parameters in gastric cancer and healthy control subjects, and the differences between these parameters in infection and non-infection groups of cancer patients are shown in Table 2. Mean serum/plasma levels of IL-6, NO\*, MDA, CRP and VEGF in patients with gastric cancer were significantly higher than those in the healthy

**Table 2** Mean levels of VEGF, IL-6, PCT, CRP, MDA and NO in controls, cancer patients and in infection and non-infection groups of cancer patients

	Control (n=23)	Gastric cancer (n=42)	<i>P</i> value	Non-infection group (n=23)	Infection group (n=19)	<i>P</i> value
VEGF (pg/mL)	51.26±30.04	489.28±172.91 <sup>b</sup>	<0.000	462.84±165.46	521.28±180.72	NS
IL-6 (pg/mL)	1.41±0.34	4.68±2.00 <sup>b</sup>	<0.000	3.47±0.77	6.15±2.07 <sup>d</sup>	<0.000
PCT (ng/mL)	0.15±0.065	0.59±0.29 <sup>b</sup>	<0.000	0.42±0.19	0.79±0.27 <sup>d</sup>	<0.000
CRP (mg/L)	15.30±11.28	78.21±43.36 <sup>b</sup>	<0.000	57.83±16.45	102.89±52.71 <sup>d</sup>	<0.002
MDA(nmol/mL)	4.37±2.99	11.42±3.22 <sup>b</sup>	<0.000	9.85±2.12	13.32±3.35 <sup>d</sup>	<0.001
NO (µmol/L)	37.97±6.35	49.32±9.07 <sup>b</sup>	<0.000	45.79±9.16	53.59±7.06 <sup>d</sup>	<0.004

<sup>b</sup>*P*<0.000 vs control, <sup>d</sup>*P*<0.005 vs non-infection group, NS: not significant.

**Table 3** Mean serum levels of VEGF, IL-6, PCT, CRP, MDA and NO in different stages of cancer patients

	Stage I (n=12)	Stage II (n=16)	Stage III (n=10)	Stage IV (n=4)
VEGF (pg/mL)	331.92±106.45	471.85±113.57 <sup>b</sup>	580.87±94.64 <sup>d,g</sup>	802.14±128.57 <sup>d,l,h</sup>
IL-6 (pg/mL)	5.05±2.89	4.16±1.28	4.03±0.81	7.33±1.13 <sup>f</sup>
PCT (ng/mL)	0.63±0.37	0.56±0.32	0.57±0.23	0.67±0.22
CRP (mg/L)	82.08±61.92	77.75±38.21	62.50±21.41	107.75±30.27 <sup>e</sup>
MDA (nmol/mL)	8.99±2.39	11.25±2.66 <sup>a</sup>	12.34±2.03 <sup>b</sup>	17.07±2.06 <sup>d,n,f</sup>
NO (µmol/L)	41.11±6.36	48.34±7.06 <sup>a</sup>	54.84±3.82 <sup>d,g</sup>	64.16±3.72 <sup>d,l,f</sup>

<sup>b</sup>*P*<0.005, <sup>a</sup>*P*<0.05, <sup>d</sup>*P*<0.000 vs stage I, <sup>l</sup>*P*<0.000, <sup>n</sup>*P*<0.001, <sup>g</sup>*P*<0.05 vs stage II, <sup>f</sup>*P*<0.001, <sup>e</sup>*P*<0.05 vs stage III.

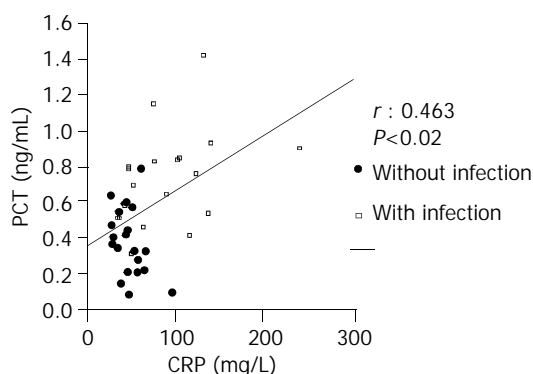
**Table 4** Pearson's rank correlation coefficients between each two of serum/plasma MDA, NO, VEGF and IL-6 levels in gastric cancer patients

MDA-NO	MDA-VEGF	MDA-IL-6	VEGF-IL-6	VEGF-NO	IL-6-NO
r=0.674 <i>P</i> <0.000	r=0.556 <i>P</i> <0.000	r=0.434 <i>P</i> <0.004	r=0.192 <i>PV</i> NS	r=0.535 <i>P</i> <0.000	r=0.534 <i>P</i> <0.000

NS.: not significant.

controls. Serum VEGF levels in patients with gastric cancer were fairly higher than those in healthy controls, although it was not statistically significant compared between gastric cancer patients with and without infections.

As seen in Table 3, as the stage of the disease increased, slightly differences were observed in the levels of PCT and CRP. There were no differences in PCT levels among the 4 stages, whereas there was a statistically significant difference in CRP levels between stage 4 and stage 3 ( $P<0.001$ ). There was a statistically significant correlation between PCT and CRP in gastric cancer patients with or without infection ( $r=0.463$ ,  $P<0.02$ , Figure 1). IL-6 levels were significantly increased in stage 4 compared with stage 2 and stage 3. Levels of MDA and NO, the markers of oxidative stress, were increased in line with the stage of cancer. However, we had some problems in evaluation of the results in different cancer stage groups because of the small number of patients in stage 4 group. CRP, PCT, IL-6, NO and MDA levels were significantly higher in the cancer group with infection than in gastric cancer group without infection ( $P<0.0001$ ). When the cancer group was classified based on their stages, VEGF levels were proportionally increased with the stage of cancer and there were statistically significant differences between the groups. Of the nineteen cancer patients with infection, 4 were in stage 1, 4 in stage 2, 6 in stage 3 and 5 in stage 4. Consequently, there was no significant difference among the stages of cancer patients with infection. Statistically significant correlations were determined in gastric cancer patients between each two of MDA, NO, VEGF, IL-6, except for the correlation between VEGF and IL-6 ( $P>0.05$ ) (Table 4).



**Figure 1** Correlations between serum PCT and CRP in gastric cancer group with and without infection.

## DISCUSSION

Angiogenesis was required for tumour growth and progression and it was involved in metastasis<sup>[6,8]</sup>. The process could result from an imbalance between positive and negative angiogenic regulators released by both tumour cells and host cells<sup>[49]</sup>. Tumour vascularisation correlated directly with the prognosis of cancer patients in many carcinomas<sup>[1-4]</sup>. Gastric cancer is a major malignant disease. The development in new diagnostic techniques and mass screening have led to increased detection rates of patients with early-stage gastric cancer. However, even after curative resection of early gastric cancer, there are still various types of recurrences, and residual occult disease and distant micro metastasis. Gastric adenocarcinoma is associated with a high incidence of serosal invasion, direct invasion into the neighboring organs, peritoneal dissemination, lymph node metastasis, and liver metastasis. These lesions have led to a low resection rate and a poor prognosis, however a curative operation should be attempted to improve the 5-year survival rate<sup>[50]</sup>. Increased serum levels of VEGF in patients with various types of cancer and the relationship between tumour

development and these VEGF levels have been determined. In our study, we investigated the circulating levels of VEGF in gastric cancer patients as well as in healthy controls using a specific ELISA and found a statistically significant increase in VEGF levels directly proportional to the stage of cancer ( $P<0.001$ ). Significantly higher VEGF levels were found in cancer patients than in healthy controls, but the increased VEGF levels were not related to infection ( $P>0.05$ ). We concluded that VEGF was an important marker related with cancer.

IL-6 may play a role in tumour-related angiogenesis by inducing tumour cell proliferation and VEGF expression in tumour cells<sup>[51-54]</sup>. In the present study, plasma levels of IL-6 were significantly elevated in advanced gastric cancer patients. This result could be explained by IL-6 production in tumour cells and by inflammatory cytokines produced by stromal cells, which stimulated IL-6 production in various cells<sup>[52,53]</sup>. Serum IL-6 levels were significantly increased in cancer patients with infection than in those without infection ( $P<0.001$ ). It has been reported to be an independent prognostic factor by multivariate analysis in patients with metastatic disease<sup>[55]</sup>, and this result was confirmed by the present study, which found that plasma IL-6 was a predictor of metastasis and infection in patients with gastric cancer. Serum IL-6 levels in gastric carcinoma patients seemed to increase in a stage-related manner.

Infections are associated with elevated malondialdehyde in gastric mucosa which is a lipid peroxidation product. The infection of gastric mucosa stimulates influx of polymorphonuclear leukocytes, leading to the generation of reactive oxygen and nitrogen species. Cell membranes, rich in polyunsaturated fatty acids, are readily attacked by these compounds, producing fatty acid radicals and lipid hydroperoxides, which can decompose in complex ways, yielding species that are more radical and a wide range of compounds, notably aldehydes. Of these, MDA and 4-hydroxynonenal were most common<sup>[24,28,56]</sup>. MDA formed by the breakdown of prostaglandin endoperoxides<sup>[56]</sup>, is a strongly genotoxic carbonyl compound that can react directly with DNA to produce a variety of adducts. Hypoxia generally takes place in tumours because of the increased oxygen requirement of proliferative cells and the increased tissue oncotic pressure. Hypoxia also induces VEGF and enhances lipid peroxidation and angiogenic potential of tumour cells. In our study, levels of MDA, the end product of lipid peroxidation, were higher both in patients with gastric cancer compared to healthy controls and in patients with infection compared to cancer patients without infection ( $P<0.000$ ,  $P<0.001$ , respectively). Plasma MDA levels were also evaluated significantly when metastasis and invasion to other tissues occurred ( $P<0.001$ ).

VEGF stimulates NO production by endothelial cells *in vitro* and *in vivo*. To date, experiments examining the role of NO in cancer initiation and progression showed that NO played a complex role. NO could be involved in initiating diseases by damaging DNA<sup>[39-41]</sup> and additionally, a much larger amount of NO released by cells in response to cytokines and endotoxins was synthesized by the inducible form of NOS and could mediate some of the cytotoxic and cytostatic effects of the immune system<sup>[57]</sup>. NO could also have a protective role. It was essential for the tumoricidal activity of immune cells<sup>[41,58-60]</sup>. Conversely, NO has been shown to have tumour-promoting effects<sup>[60]</sup>. In this study, we demonstrated that plasma NO levels were increased in patients with gastric cancer. In cancer patients, NO levels were significantly higher in infection group than in non-infection group and were higher in stage 4 compared with stages 1 and 2.

PCT was a specific marker of infection and CRP an acute-phase reactant that serves as a pattern recognition molecule in the innate immune system<sup>[42]</sup>. CRP has been traditionally thought as a bystander marker of vascular inflammation, which

did not play a direct role in the inflammatory process. However, recent evidences suggest that CRP may contribute directly to the proinflammatory state. CRP could stimulate monocyte release from inflammatory cytokines such as IL-1b, IL-6, and TNF- $\alpha$  and might directly act as a proinflammatory stimulus to phagocytic cells<sup>[44,45,61]</sup>. PCT and CRP levels were significantly increased in cancer patients with infection compared to gastric cancer patients without infection and were significantly higher in all gastric cancer groups compared to control group ( $P < 0.0001$ ). There was no statistically significant difference in PCT and CRP levels between different stages of cancer. PCT is an important determinant used in the diagnostic evaluation of fever and infections. Increased PCT levels in gastric cancer patients with infection can be used as a useful parameter for pre-diagnosis and follow-up. Therefore, CRP and PCT are two important markers for the diagnosis of infection in patients with cancer rather than for the diagnosis of cancer.

This study demonstrated that the plasma/serum levels of VEGF, IL-6, MDA and NO in patients with stage IV cancer were significantly higher than those in patients with stages I and II/III cancer, and that increased plasma levels of IL-6, MDA, NO and especially VEGF, might be useful in identifying metastatic gastric cancer patients, thus allowing more effective treatment strategies to be implemented. Moreover, our results support the existence of interactions between angiogenesis and the inflammatory system, which contributes to tumour metastasis and progression.

In conclusion, levels of MDA, IL-6, NO and especially VEGF can be used as important parameters for clinical diagnosis and follow up of cancer cases. CRP is a useful marker in the diagnosis of infections that frequently occur in cancer cases.

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