



# Serum LAG-3 and DKK-1 levels in patients with gastric cancer and their correlation with clinical pathological characteristics

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## ABSTRACT

**Objective:** To study the correlation of serum LAG-3 and DKK-1 levels with cancer cell proliferation, invasion, angiogenesis and other clinical pathological characteristics in patients with gastric cancer. **Methods:** 48 patients who were diagnosed with early gastric cancer in our hospital between June 2014 and October 2016 were selected as the gastric cancer group of the research, 50 healthy volunteers who received physical examination in our hospital during the same period were selected as the control group of the research, serum was collected to determine the levels of lymphocyte activation gene-3 (LAG-3), Dickkopf-1 (DKK-1) and angiogenesis molecules, and the gastric cancer tissue and the tissue adjacent to carcinoma were collected to determine the expression of proliferation and invasion-related molecules.

**Results:** Serum LAG-1, DKK-1, angiogenin-1 (Ang-1), Ang-2, vascular endothelial growth factor (VEGF) and basic fibroblast growth factor (bFGF) levels of gastric cancer group were significantly higher than those of control group ( $P < 0.05$ ), and EPHA2, LOXL2, PCNA, Akt, CyclinD1, MYH-9, CXCR7, KDM1A and CatB mRNA expression in gastric cancer tissue were significantly higher than those in the tissue adjacent to carcinoma ( $P < 0.05$ ); serum Ang-1, Ang-2, VEGF and bFGF levels as well as EPHA2, LOXL2, PCNA, Akt, CyclinD1, MYH-9, CXCR7, KDM1A and CatB mRNA expression in gastric cancer tissue of patients with gastric cancer were positively correlated with serum LAG-3 and DKK-1 levels.

**Conclusion:** Serum LAG-3 and DKK-1 levels are valuable to diagnose early gastric cancer and can assess the cancer cell proliferation, invasion, angiogenesis and other clinical pathological characteristics in gastric cancer tissue.

## 1. Introduction

Gastric cancer is one of the most common malignant tumors of digestive system, and its occurrence and development is the progressive evolution process involving multiple genes[1]. The disease outcome of early gastric cancer is ideal after surgical resection and the survival rate is high, the surgical resection rate of middle-advanced gastric cancer is low, the prognosis is poor and the mortality is high. However, the diagnostic rate of early gastric cancer is low and the majority of patients with gastric cancer have developed middle-advanced disease when diagnosed, which is associated with the lack of effective clinical screening indexes for early gastric cancer. Lymphocyte activation gene-3 (LAG-3) and

Dickkopf-1 (DKK-1) are two new screening indexes for malignant tumors discovered in recent years, the former has inhibitory effect on T cell activation and proliferation and is able to participate in the immune escape of cancer cells, and the latter has inhibitory effect on Wnt/ $\beta$ -catenin pathway and is able to participate in the regulation of the malignant biological behavior of cancer cells[2–4]. In the following study, the correlation of serum LAG-3 and DKK-1 levels with cancer cell proliferation, invasion, angiogenesis and other clinical pathological characteristics in patients with gastric cancer was analyzed.

## 2. Materials and methods

### 2.1. Research subjects

48 patients who were diagnosed with early gastric cancer in our

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**Table 2**Proliferation-related gene mRNA expression in gastric cancer tissue and tissue adjacent to carcinoma ( $\bar{x}\pm s$ ).

Groups	n	EPHA2	LOXL2	PCNA	Akt	CyclinD1
Gastric cancer group	48	2.92±0.51	2.31±0.38	3.28±0.58	1.98±0.25	3.94±0.66
Control group	50	1.08±0.22	1.01±0.14	0.97±0.12	1.05±0.17	0.95±0.07
t		18.592	13.582	22.571	9.285	35.281
P		<0.05	<0.05	<0.05	<0.05	<0.05

expression in gastric cancer tissue were positively correlated with serum LAG-1 and DKK-1 levels.

### 3.3. Invasion-related gene mRNA expression in gastric cancer tissue and tissue adjacent to carcinoma

Analysis of invasion-related genes MYH-9, CXCR7, KDM1A and CatB mRNA expression in gastric cancer tissue and tissue adjacent to carcinoma was shown in Table 3: MYH-9, CXCR7, KDM1A and CatB mRNA expression in gastric cancer tissue were significantly higher than those in the tissue adjacent to carcinoma. Differences were statistically significant in MYH-9, CXCR7, KDM1A and CatB mRNA expression in gastric cancer tissue and tissue adjacent to carcinoma ( $P<0.05$ ). Pearson test showed that MYH-9, CXCR7, KDM1A and CatB mRNA expression in gastric cancer tissue were positively correlated with serum LAG-1 and DKK-1 levels.

## 4. Discussion

Early gastric cancer screening is of important value for increasing the resection rate and improving the disease outcomes. At present, there are no effective indexes for clinical early gastric cancer screening. LAG-3 and DKK-1 are the new malignant tumor markers discovered in recent years. LAG-3 is the negative costimulatory molecule in immunoglobulin family, and its combination with ligand MHC-II-like molecules can inhibit T lymphocyte maturation, differentiation and proliferation, and then lead to the immune escape of cancer cells. In the malignant tumor tissues, metalloproteinases AMAD10 and AMAD17 may act on LAG-3 and make it break and released into the blood circulation, and detecting serum LAG-3 level can assess tumor malignancy[5]. DKK-1 is a member of Dickkopf family, which can on the one hand, be combined with Wnt LRP5/6 to affect the formation of receptor complexes and thereby impede Wnt signaling pathway, and on the other hand, be directly combined

with the membrane receptors to promote cell proliferation and migration[6]. In order to define the value of LAG-3 and DKK-1 for early gastric cancer screening, serum LAG-3 and DKK-1 levels were compared between patients with early gastric cancer and healthy volunteers, and the results showed that serum LAG-1 and DKK-1 levels of gastric cancer group were significantly higher than those of control group ( $P<0.05$ ). It means that serum LAG-1 and DKK-1 levels have increased significantly in early gastric cancer patients, and measuring the serum LAG-1 and DKK-1 levels has a certain value for early screening of gastric cancer.

In the development and change of gastric cancer, the angiogenesis as well as cancer cell proliferation and invasion in lesions is the important malignant biological behaviors, and also the important factors causing illness development. The angiogenesis in tumor tissue is regulated by Ang-1, Ang-2, VEGF, bFGF and a variety of pro-angiogenesis factors, and these cytokines can not only exert biological effect in local tumor lesions, but can also be massively secreted into the blood circulation. Both Ang-1 and Ang-2 belong to angiogenin, the former has maintaining effect on continuous growth after angiogenesis, and the latter can directly promote the occurrence and growth of new blood vessels[7]; VEGF is the factor with the most powerful effect on promoting angiogenesis, and can act on endothelial cells and promote its division, proliferation and forming lumen[8]; bFGF can promote fibroblast proliferation and participate in the formation of complete vascular structures[9]. In the study, analysis of above serum pro-angiogenesis molecule levels in patients with early gastric cancer proved that serum Ang-1, Ang-2, VEGF and bFGF levels of gastric cancer group were significantly higher than those of control group ( $P<0.05$ ) and positively correlated with serum LAG-3 and DKK-1 levels. It means that the abnormally elevated serum LAG-3 and DKK-1 levels in early gastric cancer patients can be used to evaluate the biological process of tumor angiogenesis.

The angiogenesis in gastric cancer tissue can provide the necessary nutrients for cancer cell proliferation and invasion. Gastric cancer

**Table 3**Invasion-related gene mRNA expression in gastric cancer tissue and tissue adjacent to carcinoma ( $\bar{x}\pm s$ ).

Groups	n	MYH-9	CXCR7	KDM1A	CatB
Gastric cancer group	48	3.38±0.59	2.92±0.47	2.49±0.41	3.10±0.52
Control group	50	1.05±0.16	1.02±0.15	0.97±0.07	1.08±0.19
t		23.472	18.595	14.285	21.228
P		<0.05	<0.05	<0.05	<0.05

cell proliferation is regulated by EPHA2, LOXL2, PCNA, Akt, CyclinD1 and other molecules, and the invasion process is regulated by MYH-9 CXCR7, KDM1A, CatB and other molecules. EPHA2 is a member of the receptor tyrosine kinases EPH family that can promote the activation of Akt and increase the expression of CyclinD1[10]; LOXL2 is a member of lysyloxidase family that can increase the cyclin PCNA expression and promote DNA replication[11]. Highly expressed CyclinD1 and PCNA in cells can work together to promote the cell cycle progression and cell proliferation[12,13]. MYH-9 can be combined with S100A4 to increase the polarity of cells, and promote the directional movement of cells[14]; CXCR7 can be combined with ligand CXCL12 to promote the epithelial-mesenchymal transition and migration of cancer cells[15,16]; CatB has acidic proteolytic enzyme activity, and can degrade extracellular matrix and promote cell invasion[17]; KDM1A is a histone-specific demethylase that can induce the expression of transcription factors Snail and Slug and promote the epithelial-mesenchymal transition of cells[18]. In the study, analysis of above proliferation and invasion molecule expression in gastric cancer tissue proved that EPHA2, LOXL2, PCNA, Akt, CyclinD1, MYH-9, CXCR7, KDM1A and CatB mRNA expression in gastric cancer tissue were significantly higher than those in the tissue adjacent to carcinoma ( $P<0.05$ ) and positively correlated with serum LAG-3 and DKK-1 levels. This means that the abnormally elevated serum LAG-3 and DKK-1 levels in early gastric cancer patients can be used to evaluate the cancer cell proliferation and invasion in gastric cancer tissue.

Serum LAG-3 and DKK-1 levels significantly increase in patients with early gastric cancer, and serum LAG-3 and DKK-1 content detection has diagnostic value for early gastric cancer; besides, serum LAG-3 and DKK-1 levels can be used to assess cancer cell proliferation, invasion, angiogenesis and other clinical pathologic features in gastric carcinoma tissue.

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