Expression of MALAT1 gene in esophageal carcinoma and its relationship with tumor progression

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Objective: To investigate the expression of MALAT1 gene in esophageal carcinoma and its relationship with tumor progression. Methods: A total of 98 patients with primary esophageal cancer who underwent radical operation for esophageal cancer in our hospital between August 2015 and December 2017 were selected as the research subjects, and the tissue specimens obtained during operation were divided into esophageal cancer tissues and tissues adjacent to carcinoma. The differences in the expression of MALAT1 gene, esophageal cancer proliferation-related genes and esophageal cancer invasion-related genes in different esophageal cancer tissues were compared, and correlation analysis was used to further evaluate the inner link between MALAT1 gene expression in esophageal cancer tissues and tumor condition. Results: MALAT1 mRNA expression in esophageal cancer tissues was higher than that in adjacent tissues; proliferation-related genes FOXA1, MACC1, MAGE-A1, Nucleostemin and ABCE1 mRNA expression were higher than those in adjacent tissues whereas KLF4 mRNA expression was lower than that in adjacent tissues; invasion-related genes PAR-2, PEBP4, UHRF1 and Nestin mRNA expression were higher than those in adjacent tissues whereas TSLC1 mRNA expression was lower than that in adjacent tissues. Pearson test showed that the MALAT1 gene expression in esophageal cancer was directly correlated with the proliferation and invasion activity of cancer cells. Conclusion: The expression of MALAT1 gene abnormally increases in esophageal cancer tissues, and the proliferation and invasion activity of cancer cells increase with the increase of the gene expression.

1. Introduction

Primary esophageal cancer is a clinical tumor disease with extremely high malignancy, many patients have missed the optimal timing of treatment due to early atypical manifestation, gene therapy may be the efficient way to optimize the outcome of patients with middle-advanced esophageal cancer, but the treatment practice depends on finding more genes closely associated with disease progression and artificially regulating the gene expression to block the key pathways of cancer cell proliferation and invasion[1,2]. MALAT1 gene was first discovered in non-small-cell lung cancer, and it was found to be able to promote the proliferation, invasion and metastasis of lung cancer cells[3,4]. Currently, more and more scholars believe that MALAT1 gene is also involved in the occurrence and development of esophageal cancer, but there are few studies on relevant molecules. In this study, the expression levels of MALAT1 gene in esophageal cancer and paracancerous tissues were compared, and the internal relationship between the expression level of MALAT1 gene and the severity of patients' condition was further determined so as to clarify the role of MALAT1 gene in esophageal cancer.

2. Data and methods

2.1 Case data

According to the following inclusion and exclusion criteria, the patients who underwent radical operation for esophageal cancer in Yulin First Hospital between August 2015 and December 2017 were selected as the study subjects. A total of 98 cases were chosen,
including 52 males and 46 females who were 54-79 years old. Esophageal cancer and paracancerous tissues were collected during operation.

2.2 Inclusion and exclusion criteria

Inclusion criteria: (1) diagnosed with esophageal cancer by pathological results; (2) having not received chemoradiotherapy or other anti-tumor treatment before operation; (3) ≤80 years old; (4) signing the informed consent form. Exclusion criteria: (1) with history of malignant tumor disease; (2) combined with autoimmune diseases or having taken hormone drugs or immunomodulatory drugs.

2.3 Gene expression detection

Esophageal cancer tissues and paracancerous tissues were collected and the gene expression was detected by fluorescence quantitative PCR. Firstly, total RNA in the tissue was extracted, the RNA precipitation was washed with 75% ethanol and then the RNA purity and concentration were measured. The reverse transcription system was configured according to the RNA concentration to synthesize cDNA; cDNA was taken and the mRNA expression of genes MALAT1, FOXA1, MACC1, MAGE-A1, Nucleostemin, KLF4, ABCE1, PAR-2, PEBP4, UHRF1, Nestin and TSLC1 were calculated after amplification.

2.4 Statistical methods

MALAT1 gene expression, esophageal cancer proliferation-related gene expression and esophageal cancer invasion-related gene expression were all recorded into SPSS 23.0. Group t test was used for comparison between groups, Pearson test was used for correlation analysis, and statistical value P was calculated. \( P < 0.05 \) meant statistically significant difference.

3. Results

3.1 MALAT1 gene expression

Comparison of MALAT1 gene expression in different esophageal tissues was as follows: MALAT1 mRNA expression in esophageal cancer tissue was \((109.37±13.28)\), MALAT1 mRNA expression in adjacent tissue was \((77.84±9.12)\) and MALAT1 mRNA expression in esophageal cancer tissues was higher than that in adjacent tissues. The difference was statistically significant in MALAT1 mRNA expression in different esophageal tissues \((P<0.05)\).

3.2 Changes of proliferation gene in esophageal cancer

Comparison of proliferation genes FOXA1, MACC1, MAGE-A1, Nucleostemin, KLF4 and ABCE1 mRNA expression in esophageal cancer tissues and adjacent tissues was as follows: FOXA1, MACC1, MAGE-A1, Nucleostemin and ABCE1 mRNA expression in esophageal cancer tissues were higher than those in adjacent tissues whereas KLF4 mRNA expression was lower than that in adjacent tissues. The differences were statistically significant in proliferation-related genes FOXA1, MACC1, MAGE-A1, Nucleostemin, KLF4 and ABCE1 mRNA expression in different esophageal tissues \((P<0.05)\), shown in Table 1.

3.3 Changes of invasion gene in esophageal cancer

Comparison of invasion genes PAR-2, PEBP4, UHRF1, Nestin and TSLC1 mRNA expression in esophageal cancer tissues and adjacent tissues was as follows: compared with the invasion genes in adjacent tissues, PAR-2, PEBP4, UHRF1 and Nestin mRNA expression in esophageal cancer tissues were significantly higher whereas TSLC1 mRNA expression was significantly lower. The differences were statistically significant in invasion gene expression in esophageal cancer tissues and adjacent tissues \((P<0.05)\), shown in Table 2.

3.4 Correlation analysis

After Pearson test, the MALAT1 mRNA expression in esophageal cancer tissues was positively correlated with FOXA1, MACC1, MAGE-A1, Nucleostemin, ABCE1, PAR-2, PEBP4, UHRF1 and Nestin mRNA expression, and negatively correlated with KLF4 and TSLC1 mRNA expression \((P<0.05)\), shown in Table 3.

4. Discussion

MALAT1 belongs to the non-coding gene IncRNA, which was first found in non-small-cell lung cancer. Currently, it has been found that MALAT1 is highly expressed in a variety of tumor tissues and is involved in the malignant progression of tumors[5,6]. Studies have confirmed that MALAT1 can up-regulate the epithelial mesenchymal transition activity of tumor cells and play a competitive endogenous

<table>
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<th>Tissue types</th>
<th>n</th>
<th>FOXA1</th>
<th>MACC1</th>
<th>MAGE-A1</th>
<th>Nucleostemin</th>
<th>KLF4</th>
<th>ABCE1</th>
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<tr>
<td>Adjacent tissue</td>
<td>98</td>
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<th>UHRF1</th>
<th>Nestin</th>
<th>TSLC1</th>
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Table 3.
Correlation of MALAT1 gene expression with cancer cell proliferation and invasion activity in esophageal cancer tissues.

<table>
<thead>
<tr>
<th>Indexes</th>
<th>Determination coefficient r</th>
<th>P</th>
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<tr>
<td>FOXA1</td>
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<td>Nestin</td>
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<td>&lt;0.05</td>
</tr>
<tr>
<td>TSLC1</td>
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messenger role. Now more scholars believe that abnormal MALAT1 gene expression is of great importance in the malignant progress of gastrointestinal tumor, the gene expression in esophageal cancer tissues and tissues adjacent to carcinoma was compared in this study and it was found that compared with that in adjacent tissues, MALAT1 mRNA expression in esophageal cancer tissues increased sharply, proving that the highly expressed MALAT1 gene is involved in the occurrence and development of esophageal cancer. The internal relationship between MALAT1 gene expression and specific malignant behaviors of esophageal cancer cells will be further elaborated below.

The malignant degree of esophageal cancer cells is relatively high, which is closely related to their vigorous proliferation activity. The molecular mechanism is the imbalance in the expression of pro-proliferation genes/ anti-proliferation genes, which eventually leads to the unlimited proliferation potential of tumor cells, the increasing tumor volume and the increasing tumor load. It has been proven at the cytological level that FOXA1 is a gene closely related to the proliferation of esophageal cancer cells, and it is believed that negative regulation of FOXA1 expression can significantly inhibit the proliferation of esophageal cancer cell line KYSE150[7,8]. MACC1 is a new gene closely related to the metastasis of colon cancer. Recent studies have found that MACC1 is also related to the disease progression of cervical cancer, lung cancer and other malignant tumors, and regulates the process of cell proliferation and apoptosis by activating the P38/PI3K/AKT signaling pathway[9,10]. Study has shown that silencing MACC1 expression can significantly inhibit the proliferation of esophageal cancer cell line Eca109. MAGE-A1 gene has little expression in normal tissues and has been found to be abnormally highly expressed in various malignant tumor tissues and play an active role in regulating cell cycle[11]. Nucleostemin, a p53-binding protein, is highly expressed in stem cells, and its expression decreases sharply when differentiation begins. Studies showed that specific inhibition of Nucleostemin gene expression in tumor tissues resulted in significantly lower cell proliferation rate[12]. KLF4 is a newly discovered zinc finger transcription factor, which is involved in the regulation of cell cycle and apoptosis. DNA damage can effectively stimulate KLF4 expression. KLF4 is differentially expressed in different tumor tissues, and its increased expression in breast cancer cells is closely related to prognosis, while it plays a negative regulatory role in the proliferation of human esophageal cancer cells[13]. ABCE1 gene can block the antiviral pathway of cells and inhibit their apoptosis process, and it hinders apoptosis to enhance the proliferation and invasion ability of cancer cells[14,15]. In this paper, the expression levels of the above proliferation-related genes were significantly different in different esophageal tissues. The mRNA levels of PAR-2, PEBP4, UHRF1 and Nestin in esophageal cancer tissues were higher and the mRNA level of KLF4 was lower, which is consistent with the vigorous proliferation activity of esophageal cancer cells. Correlation analysis further showed that MALAT1 gene expression in esophageal cancer tissues was positively correlated with FOXA1, MACC1, MAGE-A1, Nucleostemin and ABCE1 mRNA expression, and negatively correlated with KLF4 mRNA expression, it confirms that the MALAT1 gene expression is highly consistent with the cancer cell proliferation activity, and in other words, the highly expressed MALAT1 gene is a sign of strong proliferation of esophageal cancer cells.

The high malignancy of esophageal cancer is also reflected in the invasiveness of tumor cells. Most patients diagnosed with primary esophageal cancer can undergo distant metastasis in a short term if not treated in time. The molecular mechanism is that the expression of pro-invasion genes in cancer cells increases abnormally while the expression of anti-invasion genes decreases or is even lost. PAR-2, as a cell membrane surface receptor, is widely distributed in a variety of tissues and organs in the human body, and can be activated by multiple factors in the body to regulate the angiogenesis inside the tumor and promote the invasion and metastasis of tumor cells[16]. PEBP4 is mainly involved in the spermatogenesis, neurodevelopment, cell apoptosis and other processes under physiological state. Recent studies have found that its expression levels in ovarian cancer, breast cancer and prostate cancer are higher than that in healthy tissues, and inhibiting its over-expression can significantly reduce the invasion and metastasis ability of cancer cells[17,18]. UHRF1 is a nucleoprotein gene closely related to cell growth, which is not expressed in highly differentiated cells, but is abnormally highly expressed in a variety of juvenile tumor cells. Moreover, studies have found that UHRF1’s ability to regulate the invasion and metastasis of cancer cells significantly reduces after the expression levels of PTEN and other tumor suppressor genes are up-regulated[19,20]. Nestin is a newly discovered intermediate filament protein, which is involved in cytoskeleton formation and highly expressed in various malignant tumor tissues. Study shows that down-regulation of Nestin expression can significantly inhibit the invasion and metastasis ability of esophageal cancer cell line ECA109, which may be because that it inhibits the nuclear metastasis of β-catenin to down-regulate the expression of MMP-9, VEGF and other pro-angiogenesis genes[21]. TSLC1 is a tumor suppressor gene that is involved in many processes such as cell movement, signal transduction and immune regulation. After artificially established TSLC1 recombinant vector transfects esophageal cancer cells, it is found that the esophageal cancer cells.
over-expressing TSLC1 and their proliferation and invasion abilities all significantly decrease (22, 23). In this paper, the expression levels of the above invasion-related genes in different esophageal tissues were compared and it was found that the mRNA levels of PAR-2, PEBP4, UHRF1 and Nestin in esophageal cancer tissues were higher, while the mRNA level of TSLC1 was lower, which is consistent with the vigorous invasion activity of esophageal cancer. Correlation analysis further showed that the expression of MALAT1 gene in esophageal cancer tissues was positively correlated with the mRNA expression of PAR-2, PEBP4, UHRF1 and Nestin, and negatively correlated with the mRNA expression of TSLC1, which confirms that the high expression of MALAT1 gene is a marker of the high invasion and metastasis ability of esophageal cancer cells.

Therefore, it is concluded that there is abnormally high expression of MALAT1 gene in esophageal cancer tissues, and the specific expression of MALAT1 gene is positively correlated with the proliferation and invasion ability of esophageal cancer cells, which can be used as a reliable indicator to determine the malignancy degree of tumors macroscopically, and is also expected to be a new target of esophageal cancer gene therapy.

References