Clinicopathological Significance of Large Tumor Suppressor (LATS) Expression in Gastric Cancer

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ABSTRACT

Purpose: The aims of this study were to evaluate the expression of the large tumor suppressor (LATS) genes LATS1 and LATS2 by immunohistochemical staining of gastric cancer, and to evaluate the clinicopathological significance of LATS expression and its correlation with overall survival (OS).

Materials and Methods: LATS1 and LATS2 expression in a tissue microarray was detected by immunohistochemistry, using 264 gastric cancer specimens surgically resected between July 2006 and December 2009.

Results: Low expression of LATS1 was significantly associated with more advanced American Joint Committee on Cancer (AJCC) stage (P=0.001) and T stage (P=0.032), lymph node (LN) metastasis (P=0.040), perineural invasion (P=0.042), poor histologic grade (P=0.007), and diffuse-type histology by the Lauren classification (P=0.033). Low expression of LATS2 was significantly correlated with older age (≥65, P=0.027), more advanced AJCC stage (P=0.001) and T stage (P=0.001), LN metastasis (P=0.004), perineural invasion (P=0.004), poor histologic grade (P<0.001), and diffuse-type histology by the Lauren classification (P<0.001). Kaplan-Meier survival analysis revealed significantly poor OS rates in the groups with low LATS1 (P=0.037) and LATS2 (P=0.037) expression.

Conclusions: Expression of LATS1 or LATS2 is a significant marker for a good prognosis in patients with gastric cancer.

Keywords: Stomach neoplasms; Tumor suppressor genes; LATS1 protein, human; LATS2 protein, human

INTRODUCTION

Gastric cancer is the fourth most common cancer and the second leading cause of cancer deaths worldwide, although its incidence and mortality rate have been decreasing for several decades [1,2]. In Korea, gastric cancer is the most frequent cancer in men, and the fourth most common cancer in women [3].

Currently, surgical resection including radical gastrectomy with lymph node (LN) dissection is the only curative treatment method for stomach cancer. In recent years, despite many remarkable advances in diagnostic and therapeutic techniques, the outcomes of patients...
with gastric cancer have shown only minor improvements. Patients diagnosed with locally advanced or metastatic gastric cancers have a very poor prognosis, and treatment primarily entails chemotherapy. These patients develop high-grade toxicity from aggressive chemotherapeutic regimens and experience severe deterioration in quality of life. Some patients may choose to receive only best supportive care.

The poor outcomes in patients with locally advanced or metastatic gastric cancer are thought to be associated with the heterogeneous pathogenesis of gastric cancer, which involves numerous different genetic mutations and molecular signaling pathways. In gastric cancer as well as many other malignancies, molecular signaling pathways have been a recent focus of investigation, and some of these pathways are being targeted with novel diagnostic tools and therapeutic agents. Accordingly, it is necessary to investigate the specialized molecular pathways and molecules associated with tumorigenesis and tumor progression.

Large tumor suppressor (LATS) is a serine/threonine-protein kinase originally isolated from Drosophila [4,5]. The LATS gene family, comprising LATS1 and LATS2, is a core component of the Hippo pathway, which is an essential regulator of homeostasis [6]. In mammals, the Hippo pathway has been reported to generate a tumor suppressor signal that inhibits cell proliferation and promotes apoptosis. Inactivation of the Hippo pathway — which is regulated by MST1/2, SAV1, LATS1/2, MOB, and yes-associated protein (YAP) — results in cell growth, enlargement of organ size, and malignant tumor formation [7-10]. LATS is involved in several important lives sustaining processes, including cell proliferation, apoptosis, cell migration, transcriptional regulation, and maintenance of genetic stability [11,12].

Abnormal expression or mutation of LATS has been found to be involved in malignant transformation and pathological progression of cervical squamous cell carcinoma, breast cancer, and hepatic malignancies [13-15]. However, the expression of LATS in gastric cancer and its implications have received little attention. LATS1 expression is decreased in gastric cancer than in normal gastric epithelium and adenomas, and its expression is significantly lower in gastric cancer with LN metastasis than that without LN metastasis [16].

The aims of the present study were to confirm the expression of LATS1 and LATS2 in gastric cancer, to assess the association between expression of LATS1/2 and clinicopathological factors as well as the overall survival (OS) of patients, and to evaluate whether LATS can be used as a potential prognostic factor in patients with gastric cancer.

**MATERIALS AND METHODS**

**Patient selection and tissue samples**

Data from 264 patients who underwent surgical resection for gastric cancer at Soonchunhyang University Cheonan Hospital between July 2006 and December 2009 were retrospectively analyzed. Patient medical records were reviewed for clinicopathological information, including age; sex; tumor location; tumor, node, metastasis (TNM) stage; tumor differentiation; presence of lymphatic, vascular, or perineural invasion; and Lauren classification. Survival data were obtained from patients' medical records. All hematoxylin and eosin (H&E) stained slides were independently re-examined by 2 pathologists to confirm the diagnosis and other pathological features. The pathologists selected the most representative sections from each gastric cancer sample. Tumor stages and grades were re-classified according to the Seventh Edition of the...
American Joint Committee on Cancer (AJCC) Staging Manual. We excluded patients who presented with other critical medical conditions or had received neoadjuvant chemotherapy, or cases in which tissue blocks were unavailable.

**Construction of tissue microarrays (TMAs)**

TMAs were constructed by reviewing H&E stained slides and selecting one representative formalin-fixed paraffin-embedded archival block for each case. The most representative tumor area was carefully marked on the H&E stained slide. Tissue cores (2-mm thick) were extracted from individual formalin-fixed paraffin-embedded blocks (donor blocks) and re-arranged into recipient paraffin blocks (TMA blocks), using a trephine apparatus (Super Bio Chips Laboratories, Seoul, Korea). In addition, normal gastric mucosa specimens were included in 26 cases, using the same procedure. One section from the TMA block was stained with H&E for tissue confirmation.

**Immunohistochemistry (IHC) for expression of LATS1 and LATS2**

Expression of **LATS1** and **LATS2** was detected by IHC. Tissue sections 4 μm thick extracted from the TMA blocks were transferred to poly-L-lysine-coated glass slides and incubated in a dry oven at 60°C for 1 hour. These sections were then de-waxed in xylene (3 changes), re-hydrated in a graded series of decreasing ethanol concentration, and rinsed in Tris-buffered saline solution (pH 7.4). Endogenous peroxidase activity was inactivated with 5% hydrogen peroxide in methanol at 37°C for 15 minutes. For antibody staining, antigen retrieval was performed using a microwave treatment in an epitope retrieval solution (pH 6.0) for 20 minutes. The tissue sections were incubated with a primary antibody in a humidified chamber at 4°C for 16 hours. The primary antibodies were a rabbit polyclonal antibody against **LATS1** (1:100 dilution; Abcam, Cambridge, UK), and a rabbit polyclonal antibody against **LATS2** (1:200 dilution; Abcam). A secondary antibody was then applied using a Bond Polymer Refine Detection kit (Leica, Wetzlar, Germany). Diaminobenzidine was used as the chromogen and the tissue sections were counterstained using Mayer’s hematoxylin solution. Positive controls, consisting of cases with known reactivity for the antibody, and negative controls obtained by omitting the primary antibody, were also included.

**Immunohistochemical assessments**

IHC staining was separately evaluated by 2 pathologists, and in the rare instances in which there was a discrepancy in their judgments, the 2 investigators reviewed the slides together using a multi-head microscope and reached a consensus. Semi-quantitative IHC scores were assigned for assessment of both the intensity and extent of staining. The intensity of staining was scored on a scale of 0 to 3, corresponding to negative, weak, moderate, and strong positivity (Figs. 1 and 2). The extent of staining was also scored on a scale of 0 to 3 according to the percentage of cells (0%, ≤10%, >10% and ≤50%, or >50%, respectively) that stained positive for each protein. The product of the intensity and extent scores was used as the final score (i.e., 0, 1, 2, 3, 4, 6, or 9). The IHC results were classified as follows: scores of 0–1 indicated low expression of **LATS1**, and scores of 2–9 indicated high expression of **LATS1**. For **LATS2**, scores of 0–3 were defined as low expression and others were defined as high expression. Similar semi-quantitative scoring systems have been successfully used for other TMA evaluations [17].

**Statistical analysis**

Statistical analyses were conducted using the Statistical Package for the Social Sciences (SPSS) software for Windows version 19.0 (IBM Corp., Armonk, NY, USA). Associations
between LATS1 and LATS2 expression and patient clinicopathological parameters were assessed using Pearson’s χ² and Fisher’s exact tests. OS was defined as the duration from the date of surgery to the date of death or last follow-up. OS rates in relationship to LATS1
and LATS2 expression were calculated using the Kaplan-Meier method. To assess the differences between Kaplan-Meier curves, a log-rank test was performed. Cox proportional hazards modeling was used to investigate the significance of prognostic factors. Statistical significance was defined as a P-value of less than 0.05.

RESULTS

Clinicopathological characteristics of gastric cancer patients

Of the 264 patients with gastric cancer included in this study, 184 were men and 80 were women. The age at diagnosis (mean±standard deviation) was 60.20±12.59 years (range, 25–85 years). The cohort comprised 103 patients with early stage and 161 patients with advanced gastric cancer. In total, there were 121 for stage I, 44 for stage II, 91 for stage III, and 8 for stage IV tumors. At the time of diagnosis, 126 patients showed signs of LN metastasis and an additional 8 patients showed signs of distant metastasis.

LATS1/2 expression was evaluated in 26 normal gastric mucosa samples and 264 gastric cancer samples. In normal gastric mucosa, weak or moderate expression of LATS1/2 was observed. However, no LATS1/2 expression was seen in foveolar epithelium. Interestingly, gastric mucosa with intestinal metaplasia demonstrated strong expression of LATS1.

Expression of LATS1 was observed in 204 patients with gastric cancer (77.3%) and expression of LATS2 was observed in 77 patients with gastric cancer (29.2%) (Table 1). The correlations between LATS1/2 expression and the clinicopathological factors of the patients are presented in Tables 2 and 3. Low expression of LATS1 was significantly associated with more advanced AJCC stage (P=0.001) and T stage (P=0.032), LN metastasis (P=0.040), perineural invasion (P=0.042), poor histologic grade (P=0.007), and diffuse-type histology by the Lauren classification (P=0.033). Weak correlations between low LATS1 expression and lymphatic invasion (P=0.061) were also observed, although these did not reach formal statistical significance.

Low expression of LATS2 was significantly correlated with older age (≥65, P=0.027), more advanced AJCC stage (P=0.001) and T stage (P=0.001), LN metastasis (P=0.004), perineural invasion (P=0.004), poor histologic grade (P=0.001), and diffuse-type histology by the Lauren classification (P<0.001). Low LATS2 expression was also associated with lymphatic invasion (P=0.067) and distant metastasis (P=0.065), although these correlations did not reach statistical significance.

OS of gastric cancer patients with LATS1 and LATS2 expression

The follow-up period for patients ranged from 5 to 113 months (median interval, 60.7 months). During the follow-up period, 85 patients died. Low expression of LATS1 was significantly associated with poor OS rates, according to Kaplan-Meier analysis (P=0.037). The 5-year OS rate for patients with high expression of LATS1 was 71.2% compared with 56.9% for patients with low LATS1 expression. Patients with low LATS2 expression had significantly shorter OS rates than patients with high LATS2 expression (P=0.037). For LATS2, the 5-year OS rate was 76.5% vs. 64.4% for high expression vs. low expression, respectively (Fig. 3). In the Cox regression analysis, LATS2 expression was a significant factor in univariate OS analysis (LATS1, P=0.038; LATS2, P=0.038), but no significant difference was observed in multivariate OS analysis (Table 4).
DISCUSSION

The LATS gene family is one of the core components of the Hippo pathway, an emerging signaling pathway that is an essential regulator of homeostasis. In mammals, the Hippo pathway has been reported as a tumor suppressor signal that inhibits cell proliferation and promotes cell apoptosis. Inactivation of the Hippo pathway results in cell growth, enlargement of organ size, and tumorigenesis. In 1995, \textit{lats} was first identified by Xu et al. \[5\] as a tumor suppressor gene of \textit{Drosophila melanogaster}. Subsequently, Xu's study group also isolated mouse (\textit{Lats1}) and human (\textit{LATS1}) genes \[18\]. In addition, human \textit{Lats2} was isolated after the discovery of mouse \textit{Lats2} by Yabuta et al. \[19\], and the researchers suggested that \textit{LATS2} may also be a tumor suppressor gene like \textit{LATS1}. \textit{LATS1} and \textit{LATS2}, members of the family of \textit{LATS} proteins, play an important role in maintaining cellular homeostasis \[6\] including cell proliferation, cell apoptosis, cell migration \[11\], transcriptional regulation, and maintenance of genetic stability \[12\].

\textit{LATS1} and \textit{LATS2} have been implicated in a number of human malignant tumors. Abnormal expression or gene mutation of \textit{LATS1} contributes to malignant transformation and histologic progression in cervical squamous cell carcinoma \[13\]. Down-regulation of \textit{LATS1} and \textit{LATS2} is correlated in breast cancer with alterations of p53 function and cell migration \[20\]. \textit{LATS2} expression is lower in human prostate tumors than in normal prostate tissue, and \textit{LATS2} is a negative regulator of the androgen receptor \[21\]. Overexpression of \textit{LATS1} promotes YAP phosphorylation and inhibits tumorigenesis in human renal cell carcinoma \[22\]. It has also been reported that \textit{LATS1} contributes to better prognosis through negative regulation of YAP in non-small cell lung cancer (NSCLC) \[23\].

However, the expression of \textit{LATS} in gastric cancer and its implications have received little investigation. In a Chinese study, \textit{LATS1} expression was found to be downregulated and negatively associated with YAP in human gastric cancer \[16\]. The same study reported that

### Table 1. \textit{LATS1} and \textit{LATS2} expression in gastric cancer according to the scoring system

|      | \textit{LATS1} | \textit{LATS2} |
|------|---------------|---------------|
| **Scores** | **Intensity score** | **Extent score** | **Final score** | **Low expression** | **High expression** | **Total** |
|      | 0 | 18 (6.8) | 14 (5.3) | 31 (11.7) | 60 (22.7) | 264 (100) |
|      | 1 | 169 (64.0) | 9 (3.4) | 33 (12.5) | 135 (51.1) | 264 (100) |
|      | 2 | 73 (27.7) | 25 (9.5) | 57 (21.6) | 73 (27.7) | 264 (100) |
|      | 3 | 4 (1.5) | 4 (1.5) | 143 (54.2) | 216 (81.8) | 264 (100) |

Values are presented as number (%). 
LATS = large tumor suppressor.
LATS1 expression was lower in gastric cancer than in normal gastric epithelium and adenoma, and its expression was significantly decreased in gastric cancer with LN metastasis than that without LN metastasis [16].

In the present study, we hypothesized that decreased expression of LATS1 and LATS2 would be associated with a poor prognosis in patients with gastric cancer and that overexpression of LATS1 and LATS2 would be correlated with a better prognosis through inhibition of tumor progression. We found that overexpression of LATS1 and LATS2 was significantly associated with factors indicating a good prognosis. Positive expression of LATS1 was associated with lower AJCC stage, negative LN metastasis, absence of perineural invasion, well/moderately differentiated grade, and intestinal-type histology based on the Lauren classification. LATS2 expression was also associated with a number of clinicopathological factors. In addition, the

| Table 2. Association with LATS1 expression and clinicopathological factors |
|-----------------------------|---------------------|---------------------|---------------------|
| Variables                  | Cases              | LATS1 expression    | P-value |
|                            |                    | Low (22.7)          | High (77.3)        | 0.706 |
| Sex                        |                    | 264                 | 60 (22.7)          | 204 (77.3) |
| Male                       | 184                | 43 (71.7)           | 141 (69.1)         | 0.384 |
| Female                     | 80                 | 17 (28.3)           | 63 (30.9)          | 0.633 |
| Age                        |                    | 165                 | 27 (45.0)          | 138 (67.6) |
| <65                        | 141                | 35 (58.3)           | 106 (52.0)         | 0.001 |
| ≥65                        | 123                | 25 (41.7)           | 98 (48.0)          | 0.032 |
| Location                   |                    | 132                 | 23 (38.3)          | 109 (53.4) |
| Upper                      | 36                 | 9 (15.0)            | 27 (13.2)          | 0.040 |
| Middle                     | 36                 | 6 (10.0)            | 30 (14.7)          | 0.876 |
| Lower                      | 192                | 45 (75.0)           | 147 (72.1)         | 0.061 |
| AJCC stage                 |                    | 165                 | 27 (45.0)          | 138 (67.6) |
| I/II                       | 99                 | 33 (55.0)           | 66 (32.4)          | 0.032 |
| T1/T2                      | 142                | 25 (41.7)           | 117 (57.4)         | 0.040 |
| T3/T4                      | 122                | 35 (58.3)           | 87 (42.6)          | 0.876 |
| LN metastasis              |                    | 132                 | 23 (38.3)          | 109 (53.4) |
| Absent                     | 132                | 37 (61.7)           | 95 (46.6)          | 0.676 |
| Present                    | 256                | 58 (96.7)           | 198 (97.1)         | 0.042 |
| Distant metastasis         |                    | 126                 | 35 (58.3)          | 91 (44.6) |
| Absent                     | 256                | 58 (96.7)           | 198 (97.1)         | 0.007 |
| Present                    | 8                  | 2 (3.3)             | 6 (2.9)            | 0.033 |
| Lymphatic invasion         |                    | 138                 | 25 (41.7)          | 113 (55.4) |
| Absent                     | 138                | 35 (58.3)           | 91 (44.6)          | 0.676 |
| Present                    | 126                | 58 (96.7)           | 184 (90.2)         | 0.042 |
| Vascular invasion          |                    | 237                 | 53 (88.1)          | 184 (90.2) |
| Absent                     | 237                | 7 (11.7)            | 20 (9.8)           | 0.007 |
| Present                    | 27                 | 198                 | 39 (65.0)          | 159 (77.9) |
| Histologic grade           |                    | 66                  | 21 (35.0)          | 45 (22.1) |
| WD/MD                      | 101                | 14 (23.3)           | 87 (42.6)          | 0.007 |
| PD/other                   | 163                | 46 (76.7)           | 117 (57.4)         | 0.033 |
| Lauren classification      |                    | 102                 | 17 (28.3)          | 85 (41.7) |
| Intestinal                 | 154                | 43 (71.7)           | 111 (54.4)         | 0.033 |
| Diffuse                    | 8                  | 0 (0)               | 8 (3.9)            | 0.033 |

Values are presented as number only or number (%). LATS = large tumor suppressor; AJCC = American Joint Committee on Cancer; LN = lymph node; WD = well differentiated; MD = moderately differentiated; PD = poorly differentiated.
expression of LATS1 and LATS2 was associated with T stage, and the proportion of LATS2-positive cases increased as the T stage decreased (P=0.007). LATS2 expression was observed in 40/103 cases with pT1 (51.9%), 15/42 with pT2 (19.5%), 14/75 with pT3 (18.2%), and 8/44 with pT4 (10.4%).

Expression of LATS1 and LATS2 was also significantly associated with stage (LATS1/AJCC stage, p=0.014; LATS2/AJCC stage, P=0.002). LATS1 overexpression was observed in 102 of 121 stage I patients (84.3%), and lack of expression of LATS2 was found in 74 of 91 stage III patients (81.3%), but of 8 patients with stage IV, none showed expression of LATS2.

In the present study, we used different cut-off values for positive expression of LATS1 and LATS2. Because the differences in the cut-off values are presumably the result of the different

| Variables                  | Cases   | LATS2 expression | P-value |
|---------------------------|---------|------------------|---------|
|                           |         | Low  | High        |         |
| Total                     | 264     | 187  | 77 (29.2)   |         |
| Sex                       |         |      |             | 0.694   |
| Male                      | 184     | 129  | 55 (71.4)   |         |
| Female                    | 80      | 58   | 22 (28.6)   |         |
| Age                       |         |      |             | 0.027   |
| <65                       | 141     | 108  | 33 (42.9)   |         |
| ≥65                       | 123     | 79   | 44 (57.1)   |         |
| Location                  |         |      |             | 0.614   |
| Upper                     | 36      | 26   | 10 (13.0)   |         |
| Middle                    | 36      | 23   | 13 (16.9)   |         |
| Lower                     | 192     | 138  | 54 (70.1)   |         |
| AJCC stage                |         |      |             | 0.001   |
| I/II                      | 165     | 105  | 60 (77.9)   |         |
| III/IV                    | 99      | 82   | 17 (22.1)   |         |
| Tumor depth               |         |      |             | 0.001   |
| T1/T2                     | 142     | 88   | 54 (70.1)   |         |
| T3/T4                     | 122     | 99   | 23 (29.9)   |         |
| LN metastasis             |         |      |             | 0.004   |
| Absent                    | 132     | 83   | 49 (63.6)   |         |
| Present                   | 132     | 104  | 28 (36.4)   |         |
| Distant metastasis        |         |      |             | 0.065   |
| Absent                    | 256     | 179  | 77 (100)    |         |
| Present                   | 8       | 8    | 0 (0)       |         |
| Lymphatic invasion        |         |      |             | 0.067   |
| Absent                    | 138     | 91   | 47 (61.0)   |         |
| Present                   | 126     | 96   | 30 (39.0)   |         |
| Vascular invasion         |         |      |             | 0.402   |
| Absent                    | 237     | 166  | 71 (92.2)   |         |
| Present                   | 27      | 21   | 6 (7.8)     |         |
| Perineural invasion       |         |      |             | 0.004   |
| Absent                    | 198     | 131  | 67 (87.0)   |         |
| Present                   | 66      | 56   | 10 (13.0)   |         |
| Histologic grade          |         |      |             | <0.001  |
| WD/MD                     | 101     | 56   | 45 (58.4)   |         |
| PD/other                  | 163     | 131  | 32 (41.6)   |         |
| Lauren classification     |         |      |             | <0.001  |
| Intestinal                | 102     | 55   | 47 (61.0)   |         |
| Diffuse                   | 154     | 124  | 30 (39.0)   |         |
| Mixed                     | 8       | 8    | 0 (0)       |         |

Values are presented as number only or number (%).
LATS = large tumor suppressor; AJCC = American Joint Committee on Cancer; LN = lymph node; WD = well differentiated; MD = moderately differentiated; PD = poorly differentiated.
dilutions of the antibodies, it is possible that there is a difference between the degree of LATS1 and LATS2 expression. Most normal gastric mucosal tissues were weakly positive for LATS1 and LATS2. However, the expression of LATS varies in gastric cancer; as shown in the previous results, overexpression was associated with good prognostic factors as well as with tumor suppressor function. However, with loss of expression, the tumor suppressor function is weakened, and it is associated with factors indicative of a poor prognosis. These findings suggest that activation of LATS acts as a tumor suppressor in gastric cancer. Further studies will be needed to confirm the tumor suppressive activity of these proteins in non-neoplastic gastric epithelial cells or to explain the manner in which expression of LATS1 and LATS2 plays an important role.

In addition, we investigated whether the expression of LATS was associated with survival in patients with both LATS1 and LATS2 expression. The number of patients with expression of...

Table 4. Univariate and multivariate analysis of factors in patients with gastric cancer by Cox regression analysis

| Parameters                                      | Overall survival |
|-------------------------------------------------|------------------|
|                                                 | Univariate analysis | Multivariate analysis |
|                                                 | P-value | Hazard ratio (95% CI) | P-value | Hazard ratio (95% CI) | P-value |
| Gender (female vs. male)                        | 0.302    | 0.980 (0.586–1.638) | 0.938    | 0.980 (0.586–1.638) | 0.938    |
| Age (<65 vs. ≥65)                               | 0.021    | 1.688 (1.052–2.709) | 0.030    | 1.688 (1.052–2.709) | 0.030    |
| Tumor depth (T1/T2 vs. T3/T4)                   | <0.001   | 8.314 (2.973–23.247) | <0.001   | 8.314 (2.973–23.247) | <0.001   |
| LN metastasis (absent vs. present)              | <0.001   | 1.510 (0.511–4.460) | 0.456    | 1.510 (0.511–4.460) | 0.456    |
| Distant metastasis (absent vs. present)         | <0.001   | 3.016 (1.356–6.708) | 0.007    | 3.016 (1.356–6.708) | 0.007    |
| Histologic grade (WD/MD vs. PD/other)           | 0.002    | 1.071 (0.407–2.820) | 0.890    | 1.071 (0.407–2.820) | 0.890    |
| Lauren classification (intestinal vs. diffuse/mixed) | 0.003    | 1.535 (0.572–4.119) | 0.395    | 1.535 (0.572–4.119) | 0.395    |
| Lymphatic invasion (absent vs. present)         | <0.001   | 2.363 (0.905–6.770) | 0.079    | 2.363 (0.905–6.770) | 0.079    |
| Vascular invasion (absent vs. present)          | <0.001   | 1.784 (1.051–3.030) | 0.032    | 1.784 (1.051–3.030) | 0.032    |
| Perineural invasion (absent vs. present)        | <0.001   | 2.739 (1.572–4.761) | <0.001   | 2.739 (1.572–4.761) | <0.001   |
| LATS1 expression (high vs. low)                  | 0.038    | 1.039 (0.638–1.691) | 0.879    | 1.039 (0.638–1.691) | 0.879    |
| LATS2 expression (high vs. low)                  | 0.038    | 0.586 (0.306–1.119) | 0.105    | 0.586 (0.306–1.119) | 0.105    |

LATS = large tumor suppressor; LN = lymph node; WD = well differentiated; MD = moderately differentiated; PD = poorly differentiated; CI = confidence interval.

Fig. 3. Kaplan-Meier survival analysis with log-rank test. Low expression of LATS1 (A) and LATS2 (B) were significantly associated with poor OS, respectively. LATS = large tumor suppressor; OS = overall survival.
both LATS1 and LATS2 was 74/264 (28.0%), and there was a statistically significant correlation between survival and simultaneous LATS1/2 expression (P=0.016).

A potential limitation of this study was the lack of significant results in the subgroup and multivariate analyses. Thus, additional studies should be performed to support the recognition of LATS expression as an important prognostic factor.

In conclusion, the expression of LATS1 and LATS2 in patients with gastric cancer was found to be related to a number of clinicopathologic factors, and was associated with a good prognosis, including a higher survival rate. If detailed mechanisms underlying the loss or overexpression of LATS can be identified, this knowledge may have a positive effect on the treatment and clinical outcomes of various malignant tumors, including gastric cancer.

REFERENCES

1. Bertuccio P, Chatenoud L, Levi F, Praud D, Ferlay J, Negri E, et al. Recent patterns in gastric cancer: a global overview. Int J Cancer 2009;125:666-673.
2. Bosetti C, Bertuccio P, Malvezzi M, Levi F, Chatenoud L, Negri E, et al. Cancer mortality in Europe, 2005–2009, and an overview of trends since 1980. Ann Oncol 2013;24:2657-2671.
3. Oh CM, Won YJ, Jung KW, Kong HJ, Cho H, Lee JK, et al. Cancer statistics in Korea: incidence, mortality, survival, and prevalence in 2013. Cancer Res Treat 2016;48:436–450.
4. Justice RW, Zilian O, Woods DF, Noll M, Bryant PJ. The Drosophila tumor suppressor gene warts encodes a homolog of human myotonic dystrophy kinase and is required for the control of cell shape and proliferation. Genes Dev 1995;9:534-546.
5. Xu T, Wang W, Zhang S, Stewart RA, Yu W. Identifying tumor suppressors in genetic mosaics: the Drosophila lats gene encodes a putative protein kinase. Development 1995;121:1053-1063.
6. Visser S, Yang X. LATS tumor suppressor: a new governor of cellular homeostasis. Cell Cycle 2010;9:3892-3903.
7. Oh HJ, Lee KK, Song SJ, Jin MS, Song MS, Lee JH, et al. Role of the tumor suppressor RASSF1A in Mst1-mediated apoptosis. Cancer Res 2006;66:2562-2569.
8. Zhao R, Li L, Lei Q, Guan KL. The Hippo-YAP pathway in organ size control and tumorigenesis: an updated version. Genes Dev 2010;24:862-874.
9. Zhao R, Wei X, Li W, Udan RS, Yang Q, Kim J, et al. Inactivation of YAP oncoprotein by the Hippo pathway is involved in cell contact inhibition and tissue growth control. Genes Dev 2007;21:2747-2761.
10. Zhou D, Conrad C, Xia F, Park JS, Payer B, Yin Y, et al. Mst1 and Mst2 maintain hepatocyte quiescence and suppress hepatocellular carcinoma development through inactivation of the Yap1 oncogene. Cancer Cell 2009;16:425-438.
11. Xia H, Qi H, Li Y, Pei J, Barton J, Blackstad M, et al. LATS1 tumor suppressor regulates G2/M transition and apoptosis. Oncogene 2002;21:1233-1241.
12. Zeng Q, Hong W. The emerging role of the hippo pathway in cell contact inhibition, organ size control, and cancer development in mammals. Cancer Cell 2008;13:188-192.
14. Morinaga N, Shirata Y, Yanagita Y, Koida T, Kimura M, Asao T, et al. Molecular analysis of the h-warts/LATS1 gene in human breast cancer. Int J Oncol 2000;17:1125-1129.
PUBMED
15. Li H, Wolfe A, Septer S, Edwards G, Zhong X, Abdulkarim AB, et al. Deregulation of Hippo kinase signalling in human hepatic malignancies. Liver Int 2012;32:38-47.
PUBMED | CROSSREF
16. Xu ZP, Zhu JS, Zhang Q, Wang XY. A breakdown of the Hippo pathway in gastric cancer. Hepatogastroenterology 2011;58:1611-1617.
PUBMED | CROSSREF
17. Kim KJ, Jung HY, Oh MH, Cho H, Lee JH, Lee HJ, et al. Loss of ARID1A expression in gastric cancer: correlation with mismatch repair deficiency and clinicopathologic features. J Gastric Cancer 2015;15:201-208.
PUBMED | CROSSREF
18. Tao W, Zhang S, Turenchalk GS, Stewart RA, St John MA, Chen W, et al. Human homologue of the Drosophila melanogaster lats tumour suppressor modulates CDC2 activity. Nat Genet 1999;21:177-181.
PUBMED | CROSSREF
19. Yabuta N, Fujii T, Copeland NG, Gilbert DJ, Jenkins NA, Nishiguchi H, et al. Structure, expression, and chromosome mapping of LATS2, a mammalian homologue of the Drosophila tumor suppressor gene lats/warts. Genomics 2000;63:263-270.
PUBMED | CROSSREF
20. Furth N, Ben-Moshe NB, Pozniak Y, Porat Z, Geiger T, Domany E, et al. Down-regulation of LATS kinases alters p53 to promote cell migration. Genes Dev 2015;29:2325-2330.
PUBMED | CROSSREF
21. Powzaniuk M, McElwee-Witmer S, Vogel RL, Hayami T, Rutledge SJ, Chen F, et al. The LATS2/KPM tumor suppressor is a negative regulator of the androgen receptor. Mol Endocrinol 2004;18:2011-2023.
PUBMED | CROSSREF
22. Yu T, Bachman J, Lai ZC. Evidence for a tumor suppressor role for the large tumor suppressor genes LATS1 and LATS2 in human cancer. Genetics 2013;195:1193-1196.
PUBMED | CROSSREF
23. Lin XY, Zhang XP, Wu JH, Qiu XS, Wang EH. Expression of LATS1 contributes to good prognosis and can negatively regulate YAP oncoprotein in non-small-cell lung cancer. Tumour Biol 2014;35:6435-6443.
PUBMED | CROSSREF