Loss of TSLC1 expression in lung adenocarcinoma: Relationships with histological subtypes, sex and prognostic significance

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The *TSLC1* (tumor suppressor in lung cancer 1) gene is a tumor suppressor recently identified through functional complementation in a lung adenocarcinoma cell line A549. In this study we immunohistochemically examined the loss of TSLC1 expression in 93 cases of surgically resected lung adenocarcinoma, and investigated its correlation with clinicopathological parameters, including histological subtypes of tumors. The prognostic significance of loss of TSLC1 expression was analyzed by univariate and multivariate analyses, in parallel with other prognostic markers such as p53, p27, and Ki-67. In non-cancerous lung tissue, TSLC1 was weakly positive in bronchial and bronchiolar epithelial cells, type II pneumocytes and bronchial glands. Overall, TSLC1 was negative in 60 of 93 lung adenocarcinomas. TSLC1 was mainly localized in the cytoplasm of the cells, but cell membrane staining was also observed, especially at sites of cell–cell adhesion. TSLC1-negative tumors were more frequently observed in male cases (41/54 cases, 70.0%) than in female cases (19/39 cases, 48.7%) ($P < 0.01$). Notably, TSLC1 expression was preserved in a non-invasive, bronchiolo-alveolar histological pattern of tumor cells ($P < 0.0001$). Survival analyses showed that loss of TSLC1 expression was associated with lower patient survival in univariate and multivariate analyses ($P < 0.05$ and $P = 0.059$, respectively). Subset analyses further showed that the prognostic impact of loss of TSLC1 was significant for male patients ($P = 0.0089$), but not for female patients. We conclude that TSLC1 is expressed in a subset of lung adenocarcinomas, especially in those with bronchiolo-alveolar spread pattern. Loss of TSLC1 is associated with lower patient survival, supporting its role as a tumor suppressor. (*Cancer Sci* 2005; 96: 480–486)

TSLC1 (tumor suppressor in lung cancer 1) is a tumor suppressor gene on chromosome 11q23.2 recently identified by functional complementation.[1,2] *TSLC1* encodes a member of the immunoglobulin protein superfamily and shares significant homology with the neural cell adhesion molecule genes (*NCAM*)*-1* and *NCAM-2*.[1,2] Structural homology with NCAM-1 and NCAM-2 suggests that TSLC1 mediates cell–cell interaction.[2,3] Although inactivating mutation of the *TSLC1* gene is rare, loss of heterozigosity (LOH) on 11q23.2 is found in 42%, 33% and 17% of primary non-small cell lung cancer, hepatocellular carcinoma and pancreatic carcinomas, respectively.[1,2] Also, *TSLC1* promoter hypermethylation is observed in ~40% of primary non-small cell lung cancer,[4] as well as in 15–30% of tumors from liver,[1] stomach,[5,6] pancreas,[1,7] breast,[8] nasopharynx[9] and prostate,[10] and in 35–60% of cervical neoplasias.[11]

Although first identified in a lung adenocarcinoma cell line, the clinicopathological implications of TSLC1 abnormalities in lung adenocarcinoma have only been investigated in a limited number of surgically resected lung adenocarcinomas.[4,12,13] To further explore the role of TSLC1 in the development and progression of lung adenocarcinoma, we immunohistochemically examined a series of 93 surgically resected lung adenocarcinomas for loss of TSLC1, and investigated its correlation with clinicopathological parameters and patient survival. Also, we investigated the relationships between loss of TSLC1 and histological growth pattern of tumors, as loss of cell adhesion molecules is associated with tumor cell invasion.[14] Furthermore, we compared the loss of TSLC1 with other well-known immunohistochemical prognostic parameters, such as p53, p27 and Ki-67.

Materials and Methods

Patients and tissues

We examined a series of 93 lesions of small lung adenocarcinomas (maximum diameter 3 cm or less) with Institutional Review Board approval. The specimens were obtained from the patients who underwent pneumonectomy or lobectomy without preoperative chemotherapy or radiotherapy. All of the T1 cases (52 cases) operated during the same period were examined in the study. All samples were collected from the surgical pathology files at Tokyo Metropolitan Komagome Hospital, Tokyo, Japan, between 1977 and 1990. The patients comprised 58 men and 35 women. The patients' ages

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ranged from 32 to 89 years, with an average of 60.3 years. The observation periods ranged from 1 to 163 months, with a median follow-up period of 66.2 months. The patients with lung adenocarcinoma were staged according to the tumor-node-metastasis system adopted by the American Joint Committee on Cancer and the International Union Against Cancer.[15] The cases consisted of 46 stage I (28 stage IA, 18 stage IB), six stage II (one stage IIA, five stage IIB) and 40 stage III (25 stage IIIA, 15 stage IIIB) cases and one stage IV case.

Histological study

Each tumor was histologically evaluated according to the WHO histological classification[16] or the classification by Noguchi *et al*.[17] According to the WHO scheme, tumors were subclassified into five major subtypes: (i) acinar ($n = 3$; men, 2; women, 1); (ii) papillary ($n = 16$; men, 11; women, 5); (iii) bronchiolo-alveolar ($n = 0$); (iv) solid ($n = 16$; men, 11; women, 5); and (v) mixed ($n = 58$; men, 34; women, 24). Thirty of 58 cases of mixed subtype adenocarcinomas had a component of bronchiolo-alveolar subtype, corresponding to type C according to the classification by Noguchi *et al*.[17] In this series, there was no adenocarcinoma with a pure bronchiolo-alveolar growth pattern, which corresponds to type A or type B according to the classification by Noguchi *et al*.[17] Histological differentiation of tumors was graded according to the criteria described in the Japanese General Rule for Clinical and Pathological Record of Lung Cancer.[18] Sixty-three cases were graded as moderately to poorly differentiated (men, 35; women, 28) and 30 cases were graded as well-differentiated (men, 23; women, 7). Lung adenocarcinoma was also evaluated histologically for lymph node metastasis, pleural infiltration, blood vessel invasion and lymphatic vessel invasion. For the evaluation of pleural infiltration and blood vessel invasion, elastica van Gieson stain of the sections was utilized routinely to identify the elastic fibers of the lung, pleura and blood vessels.

Antibodies

Rabbit polyclonal antibody to TSLC1 was raised against the C-terminal epitope of TSLC1 and has been described elsewhere.[19] Mouse monoclonal antibodies against Ki-67 (MIB1) was purchased from Dako Cytomation (Copenhagen, Denmark). Antibodies to p27 and p53 (DO7) were purchased from BD Transduction Laboratories (Lexington, KY, USA) and Novocastra Laboratories (Newcastle upon Tyne, UK), respectively.

Immunohistochemistry

All the resected specimens were fixed in 10% buffered formalin and embedded in paraffin. Sections (4 μm thick) were cut, and deparaffinized through graded alcohol and xylene. After antigen retrieval with autoclave treatment in 10 mM citrate buffer, pH 6.0, endogenous peroxidase was blocked with 3% hydrogen peroxide in methanol for 20 min. The sections were washed three times with cold 0.01 M phosphate-buffered saline (PBS). After blocking with 10% normal rabbit serum, the sections were incubated for 16 h at 4°C with rabbit polyclonal antibody against TSLC1 (diluted at 1:800), mouse monoclonal antibody against human Ki-67 (MIB1, diluted at 1:100), mouse monoclonal antibody against p53 (DO7, diluted at 1:100) or antibody against p27 (diluted at 1:100). The sections were incubated with biotinylated goat antirabbit IgG or rabbit antimouse IgG, and reacted with the streptavidin–biotin peroxidase reagent (LSAB2 Kit, Dako Cytomation). Finally, the reaction was visualized with a chromogen, diaminobenzidine in 3% hydrogen peroxidase. Sections were then counterstained with hematoxylin, dehydrated and mounted.

Assessment of immunohistochemical staining

Cells with cytoplasmic immunoreactivity for TSLC1 with or without membranous immunoreactivity were considered as positively immunostained cells for TSLC1. The labeling index (LI) for each protein was determined in each adenocarcinoma or each component. LI was determined by calculating the percentage of positively immunostained cells in 2000 cells. In some smaller lesions, all the cells in the entire area of the lesion were counted. Immunohistochemical results were subdivided into two categories, positive and negative with cut-off values of 30% of tumor cells for TSLC1, 20% for p27[20] and 20% for p53,[21] and 30% for Ki67.[20] Faint staining of cells was not considered to be positive. As there was no significant difference in staining intensity of TSLC1 immunohistochemistry, intensity of TSLC1 immunohistochemistry was not incorporated into the assessment.

Statistical analysis

Calculations were performed using StatviewJ-4.02 (Abacus Concepts, Berkeley, CA, USA) software. Differences in mean values between the groups were analyzed by the Student's *t*-test. Differences in frequency were analyzed by the χ^2-test. Disease-related survival was measured from the date of surgical resection to the date of death due to lung adenocarcinoma or the date when the patients were last known to be alive. Survival curves were estimated by the Kaplan–Meier method, and the differences in disease-related survival between subgroups were compared by the log-rank test. The Cox proportional hazards model was used for multivariate survival analysis. A result was considered significant if the P-value was < 0.05.

Results

TSLC1 in non-tumorous lung tissue and lung adenocarcinoma cells

Figure 1 shows the representative results of TSLC1 immunostaining. In the non-neoplastic lung tissue, TSLC1 expression was weakly positive in bronchial and bronchiolar epithelial cells (Fig. 1a) and reactive type II pneumocytes (Fig. 1b). The staining intensity varied from cell to cell. Type I pneumocytes, lymphocytes, fibroblasts and endothelial cells were all negative. Inflammatory cells, most likely representing plasma cells, were occasionally positive for TSLC1 (Fig. 1c).

In adenocarcinoma cells, TSLC1 immunoreactivity was observed in 33 of 93 cases examined (33.3%). TSLC1 immunoreactivity was mainly observed in the cytoplasm of lung adenocarcinoma cells. Membranous staining was often present, especially at the cell–cell border of epithelial

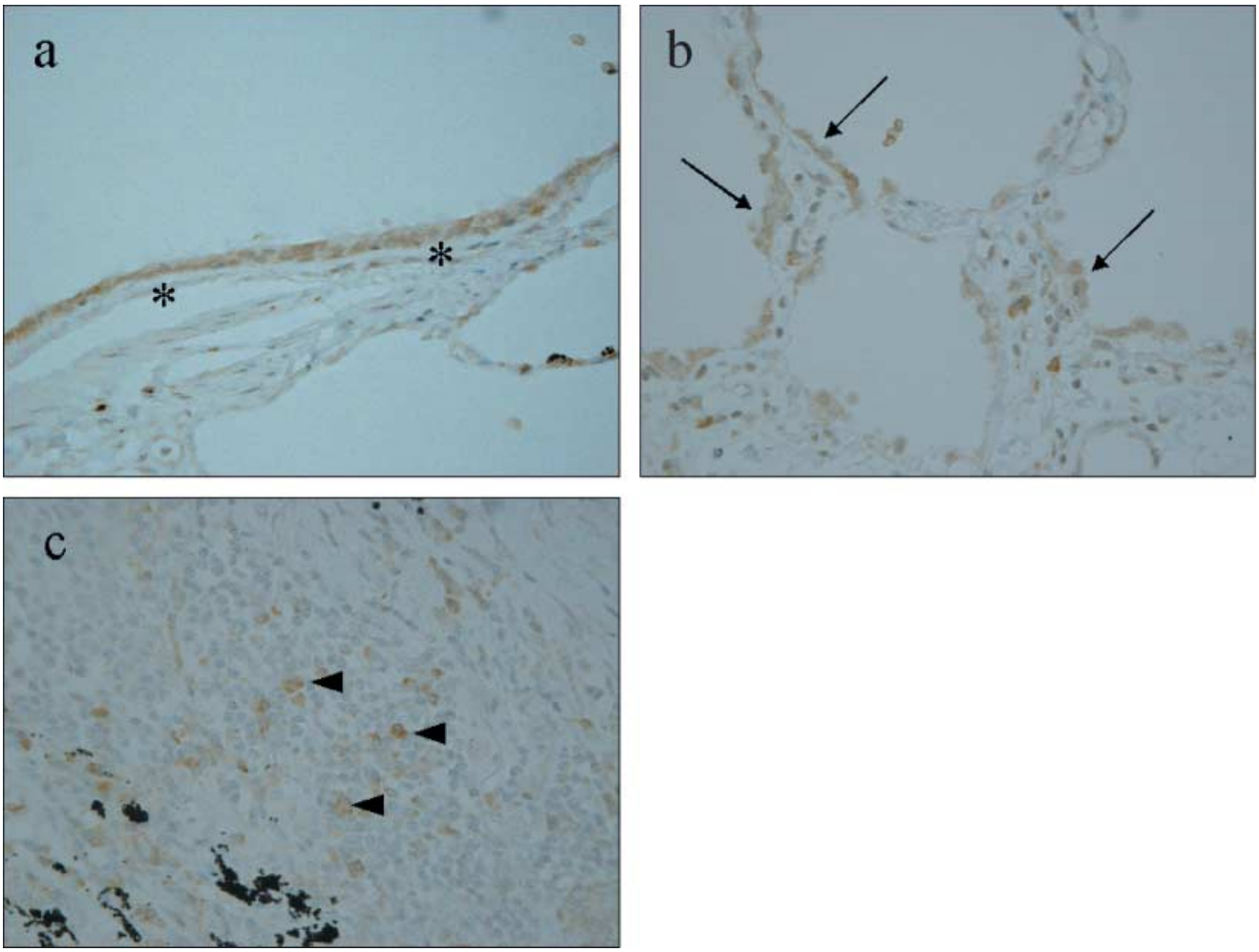

Fig. 1. Immunohistochemical findings for tumor suppressor in lung cancer 1 (TSLC1) in lung tissue. (a) Positive staining of the bronchiolar epithelial cells (asterisk) and (b) reactive type II pneumocytes (arrow). (c) Occasional positivity for TSLC1 was seen in inflammatory cells, most representing plasma cells (arrowhead). Magnification ×40 (objective).

cancer cells (Fig. 2a). As shown in Fig. 2b, TSLC1 immunoreactivity was classified as negative in the remaining 60 cases examined.

Correlation between loss of TSLC1 expression and histological subtypes

TSLC1 is supposed to function as a cell adhesion molecule,[2,3] and loss of TSLC1 may predispose the cancer cells to stromal invasion and metastasis. Therefore, we next investigated the possible correlation between loss of TSLC1 expression and histological subtypes that were classified according to the invasive or non-invasive mode of tumor spread (Table 1). We found that 30 of 93 cases had a bronchiolo-alveolar spread pattern that corresponded to non-invasive growth of tumor cells. Strikingly, 28 of 30 bronchiolo-alveolar components were positive for TSLC1 expression, while non-bronchiolo-alveolar components of the same tumors, including a scattered growth component, were frequently TSLC1-negative (15 of 30; Fig. 2c,d; Table 1) ($P < 0.0001$). The relationship between TSLC1 immunohistochemistry and histological subtype of the tumors is also shown in Table 1. TSLC1-negative tumors were more frequently observed in the solid subtype than in the mixed subtype ($P < 0.01$) or in the non-bronchiolo-alveolar component of mixed subtype adenocarcinoma ($P < 0.01$).

Correlation between loss of TSLC1 expression and clinicopathological factors in lung adenocarcinoma

The data are summarized in Table 2. TSLC1-negative adenocarcinomas were more frequently observed in men than in women ($P < 0.01$). None of the other clinicopathological factors examined was significantly correlated with loss of TSLC1.

Relationship between loss of TSLC1 and p27, p53 or Ki67

The data are summarized in Table 3. TSLC1-negative tumors showed higher Ki-67 LI (LI = 21.3, SE = 2.59) than TSLC1-positive tumors (LI = 11.8, SE = 2.57) ($P < 0.05$). Loss of TSLC1 expression in lung adenocarcinomas was not correlated with p53 or p27 expression.

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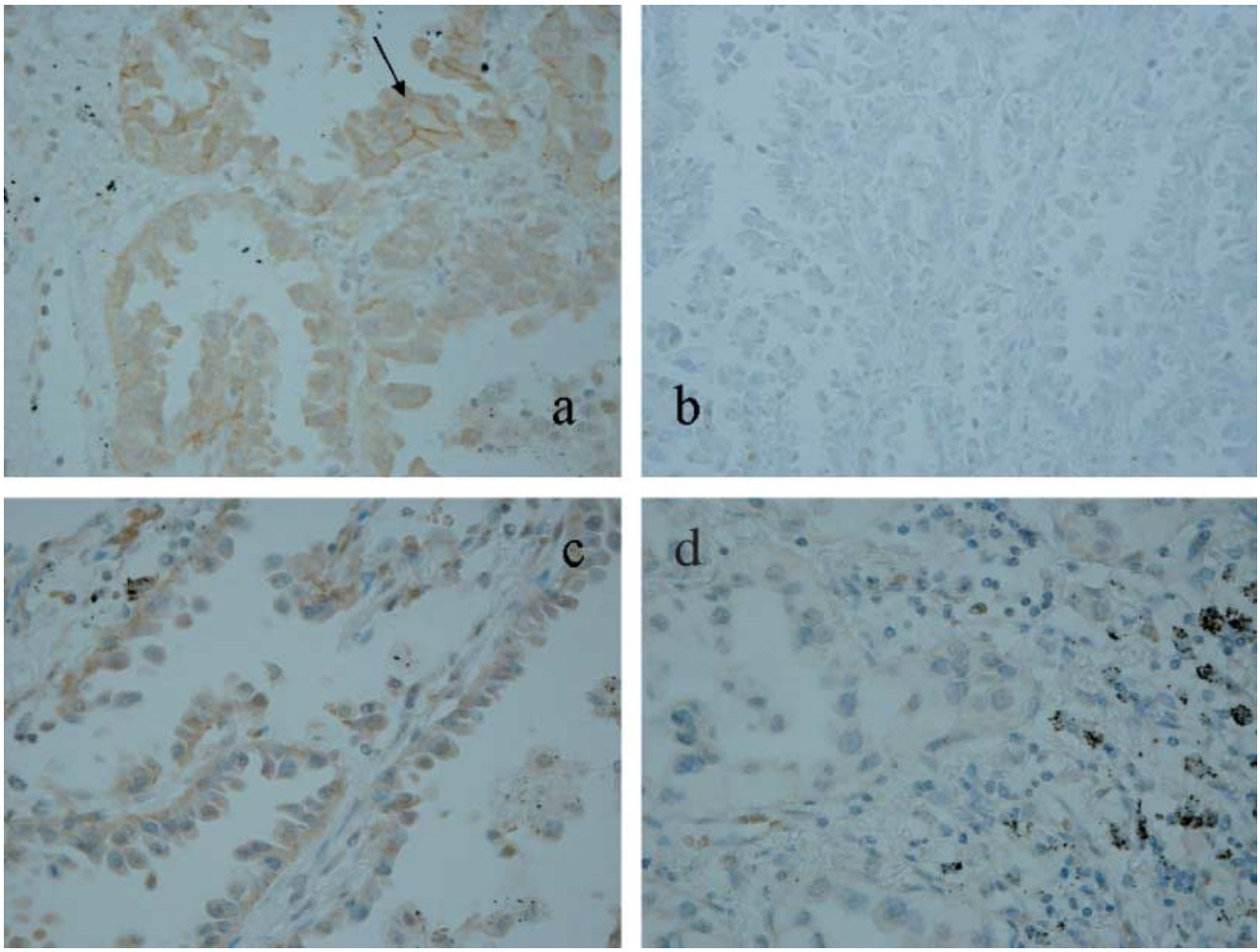

Fig. 2. Immunohistochemical findings for tumor suppressor in lung cancer 1 (TSLC1) in lung adenocarcinoma. (a) Adenocarcinoma positive for TSLC1. Besides cytoplasmic staining, membranous staining was observed (arrow). (b) Papillary adenocarcinoma negative for TSLC1 expression. (c,d) A lung adenocarcinoma showing (c) TSLC1 expression in the BAC component and (d) loss of TSLC1 expression in the non-BAC component. Magnification ×40 (objective).

Table 1. Loss of tumor suppressor in lung cancer 1 (TSLC1) expression and histological subtypes or components in lung adenocarcinoma

Histological subtype or component	*n*	TSLC1 expression	
		Positive (%)	Negative (%)
Acinar	3	2(66.7)	1 (33.3)
Papillary	16	5 (31.3)	11 (68.8)
Solid	16	1 (6.3)	15 (93.8)*,**
Mixed	58	25 (43.1)	33 (56.9)*,**
BA components in mixed subtype tumors	30	28 (93.4)	2 (6.7)**,***
Non-BA components in mixed subtype tumors with BA components	30	15 (50)	15 (50)**,***

*$P < 0.05$, **$P < 0.01$, ***$P < 0.0001$. BA, bronchiolo-alveolar.

Prognostic significance of loss of TSLC1 expression in lung adenocarcinoma

Next, we examined the prognostic significance of loss of TSLC1 expression by using the Kaplan–Meier method (Fig. 3). TSLC1-negative cases showed a worse outcome compared to TSLC1-positive cases, with statistical significance of $P < 0.05$. Moreover, subset analyses further showed that prognostic impact of loss of TSLC1 was significant for male patients ($P = 0.0089$), but not for female patients.

The prognostic significance of immunoreactivity of p27, p53 or MIB1, and each clinicopathological factor was also examined. Cases with negative p27 immunoreactivity, higher stage (stage II, III or IV), lymph node metastases, pleural infiltration and blood vessel invasion showed worse outcome in this series (Table 4).

Table 2. Tumor suppressor in lung cancer 1 (TSLC1) immunoreactivity and clinicopathological factors in lung adenocarcinoma

Clinicopathological factor	*n*	TSLC1		*P*-value
		Positive (%)	Negative (%)	
Sex				
Male	54	13 (24.1)	41 (76.0)	0.0017
Female	39	20 (51.3)	19 (48.7)	
Average age (years)		59.3 ± 1.6	61.5 ± 2.2	0.60
Stage				
I	47	20 (42.6)	27 (57.4)	0.15
II, III, IV	46	13 (28.3)	33 (71.7)	
Lymph node metastasis				
+	37	10 (27.0)	27 (73.0)	0.86
–	56	23 (41.1)	33 (58.9)	
Lymphatic vessel invasion				
+	71	23 (32.4)	48 (67.6)	0.54
–	22	10 (45.5)	12 (54.5)	
Blood vessel invasion				
+	72	23 (31.9)	49 (68.1)	0.19
–	21	10 (47.6)	11 (52.4)	
Pleural infiltration				
+	55	17 (30.9)	38 (69.1)	0.26
–	38	16 (42.1)	22 (57.9)	
Pleural dissemination				
+	13	4 (30.8)	9 (69.2)	0.70
–	80	29 (36.3)	51 (63.8)	
Intrapulmonary metastasis				
+	10	1 (10)	9 (90)	0.074
–	83	32 (38.6)	51 (61.4)	

Table 3. Correlations between tumor suppressor in lung cancer 1 (TSLC1) immunoreactivity and p27, p53 and Ki-67 immunoreactivities in lung adenocarcinoma

	p27		p53		Ki-67
	+	–	+	–	Average LI (%)
TSLC1					
+	19	14	17	16	10.4*
–	31	29	31	29	22.1*

+, Positive cases; –, negative cases; LI, labeling index. *$P < 0.05$.

Loss of TSLC1 and these factors were further examined for prognostic significance by Cox's proportional hazards model. Higher stage was not included in variables as it was obviously dependent on lymph node metastasis by definition. Negative TSLC1 expression retained its marginal prognostic significance with a *P*-value of 0.0594 and hazard ratio of 2.265 (Table 5). In male cases, higher stage and lymph node status showed worse outcomes. Loss of TSLC1 and lymph node metastasis were examined for prognostic significance by Cox's proportional hazards model. Negative TSLC1 expression retained its prognostic significance with a *P*-value of 0.0445 and hazard ratio of 2.285 in male cases.

Discussion

Tumor suppressor in lung cancer 1 is a tumor suppressor gene recently identified through the suppression of tumor

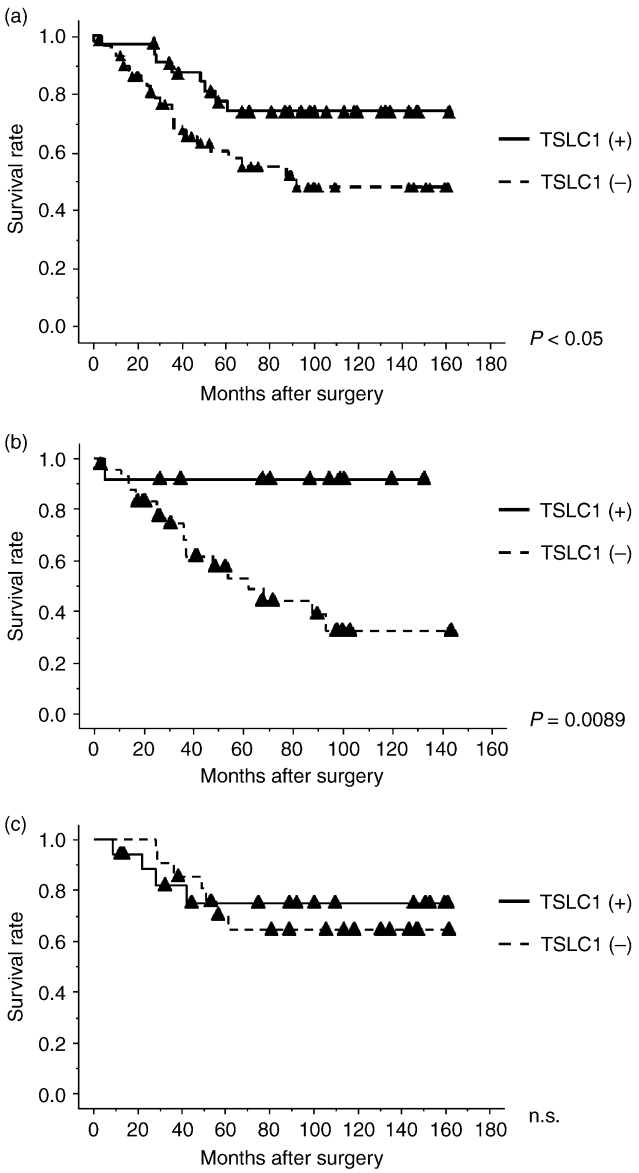

Fig. 3. Kaplan–Meier survival curves of lung adenocarcinoma cases. Survival curves were stratified in terms of tumor suppressor in lung cancer 1 (TSLC1) expression in (a) total cases, (b) male cases and (c) female cases. Censored cases were indicated by tick marks on the curves. n.s., not significant.

formation by the human lung adenocarcinoma cell line A549.[1] Inactivation of *TSLC1* through LOH or promoter hypermethylation is observed in non-small cell lung carcinomas and in various other carcinomas, including those of pancreas, stomach, breast, prostate and uterine cervix.[1,2,4–11] Although the function of TSLC1 is not yet fully clarified, its role in the regulation of cell adhesion and motility has been indicated by *in vivo* and *in vitro* observations.[2,3,19,22]

Uchino *et al*. previously reported that low TSLC1 expression related to advanced pathological stage, T (primary tumor) and N (regional lymph nodes) classification, and worse prognosis in lung adenocarcinoma.[12] Although their findings with regard to patient prognosis are similar to those of the present study, their conclusion was derived from a limited number of cases

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Table 4. Summary of relationships between immunohistochemical and clinicopathological variables and survival

	n	Median survival time (months)	*P*-value (log-rank test)
Sex			
Male	54	53.9	0.067
Female	39	65.7	
Lymph node metastasis			
+	37	41.6	<0.0001
–	56	81.1	
Stage			
I	47	82.5	0.0001
II, III, IV	46	43.6	
Lymphatic vessel invasion			
+	71	66.8	0.097
–	22	75.1	
Blood vessel invasion			
+	72	66.0	0.033
–	21	77.8	
Pleural infiltration			
+	55	63.2	0.018
–	38	74.1	
TSLC1 immunoreactivity			
Positive	33	65.2	0.03
Negative	60	55.5	
p27 immunoreactivity			
Positive	50	77.4	0.029
Negative	43	59.3	
p53 immunoreactivity			
Positive	48	48.1	0.151
Negative	45	67.2	
MIB1 immunoreactivity			
Positive	54	63.9	0.161
Negative	39	72.6	

TSLC1, tumor suppressor in lung cancer 1.

Table 5. A multivariate analysis on disease-related survival of 93 lung adenocarcinoma patients

Variables	Category	Risk ratio	*P*-value
Lymph node metastasis	– vs +	4.230	0.0004
p27	Positive vs negative	2.330	0.0218
TSLC1	Positive vs Negative	2.265	0.0594
Blood vessel invasion	– vs +	1.542	0.4699
Pleural infiltration	– vs +	1.291	0.5691

TSLC1, tumor suppressor in lung cancer 1.

($n = 38$), which might be insufficient to establish the prognostic significance of TSLC1. In the present study, we examined the loss of TSLC1 in a larger number of cases ($n = 93$) and investigated prognostic significance of loss of TSLC1 by both univariate and multivariate analyses. By using multivariate analysis, we sought to find whether the prognostic value of loss of TSLC1 was due to its possible association with other known prognostic parameters. We demonstrated that loss of TSLC1 is a useful prognostic indicator associated with better patient survival. Furthermore, loss of TSLC1 retained prognostic significance by multivariate analysis. Thus, loss of TSLC1 proved to be a useful prognostic factor independent of conventional clinicopathological parameters such as stage and lymph node metastasis, as well as molecular markers such as p27, p53 and Ki-67.

We analyzed small-sized lung adenocarcinomas with maximum diameters of 3 cm or less in the present study. Even in small-sized lung adenocarcinomas with maximum diameters 2 cm or less, the results of surgery still remain unsatisfactory with an average 5-year survival rate of 77.2%.[23] To improve treatment for patients with small-sized adenocarcinoma, reliable prognostic factors that identify a subset of patients at high risk of recurrence are certainly required; effective postoperative adjuvant therapy may then be given to the high-risk patient group. The present study showed the potential clinical usefulness of TSLC1 status in small-sized lung adenocarcinoma.

Fukami *et al*. reported that the promoter of the *TSLC1* gene was frequently methylated in non-small cell lung cancer.[4] They also reported that promoter methylation of *TSLC1* was found in 17% of pT1 cases and 80% of pT2 cases. In the present study, 55.6% (15/27 cases) of stage IA (pT1pN0M0) cases were negative for TSLC1 expression. Thus, there might be a mechanism other than promoter methylation underlying the inactivation of *TSLC1* in lung adenocarcinoma. In lung adenocarcinoma, chromosomal loss might be involved in inactivation of *TSLC1* as the chromosomal locus 11q23.2, where the *TSLC1* gene is located, is frequently lost in lung cancer.[1] Further experiments are warranted to determine the mechanisms underlying the downregulation of TSLC1 in lung adenocarcinoma.

In mixed subtype with bronchiolo-alveolar carcinoma (BAC), TSLC1 expression was more frequently retained in BAC than in non-BAC components. Ito *et al*. also reported similar findings.[13] These observations indicate that loss of TSLC1 expression is involved in tumor invasion in lung adenocarcinoma, as the BAC component is by definition a non-invasive component of lung adenocarcinoma. Yageta *et al*. recently reported that TSLC1 associates with DAL-1, a gene product of another lung tumor suppressor belonging to the protein 4.1 family.[19] They suggested that TSLC1 interacted with the actin filament through an anchoring protein, DAL-1, and participated in organizing the actin cytoskeleton and in constructing stable adhesion between adjacent cells. Loss of these functions of TSLC1 might lead to augmentation of tumor invasion in lung adenocarcinoma. In fact, TSLC-1 transfection was reported to suppress motility and invasion of esophageal squamous cell carcinoma a cell line lacking TSLC1 expression.[22] As for correlations with histological subtypes, the frequency of TSLC-1-negative tumors was strikingly high in the solid subtype of lung adenocarcinoma (15/16 cases, 93.8%) as compared to acinar (1/39 cases, 33.3%) and papillary carcinomas (11/16 cases, 68.8%). Thus, development of the solid subtype of lung adenocarcinoma might be more closely related than other subtypes to loss of TSLC1 function.

Loss of TSLC1 expression was also correlated to the sex of lung adenocarcinoma patients. Male cases lost TSLC1 expression more frequently than female cases. Moreover, prognostic significance of loss of TSLC1 was different between male and female cases. In male cases, prognostic significance of loss of TSLC1 was notable in univariate and multivariate analyses, while its prognostic significance was not confirmed in female cases. In the present cases, sex difference in histological factors might not

influence the prognosis, as the histological distribution did not differ significantly between sexes. Recently, insights into the sex difference in tumorigenesis of lung adenocarcinoma were obtained by mutational study of the *EGFR* gene.[24,25] The difference in the frequency of loss of TSLC1 expression and its prognostic significance might also reflect the sex differences in development of lung adenocarcinoma. Loss of TSLC1 might also be unique as a molecular prognostic marker of lung adenocarcinoma as no other markers have been reported to show sex differences to date. Loss of TSLC1 expression failed to show a significant correlation with the other clinicopathological factors, such as lymph node metastasis, pleural infiltration, pleural dissemination, intrapulmonary metastasis, blood and lymphatic vessel invasion. Additional molecular biological abnormalities other than loss of TSLC1 expression might be required for tumor cells to attain these clinicopathological characteristics related to tumor progression.

Kuramochi *et al*. reported that the volume of tumors formed in nude mice by injection of the A549 lung adenocarcinoma cells was substantially decreased by restoration of TSLC1 expression in A549 cells.[1] Ito *et al*. reported that an esophageal squamous cell carcinoma cell line (KYSE520) lacking TSLC1 expression showed significantly lower population of G_1 cells than cell clones expressing TSLC1, which were obtained from KYSE520 by *TSLC1* transfection.[22] In spite of these *in vitro* observations, the relationship between loss of TSLC1 expression and tumor proliferation has not been documented in clinical tumor samples. Our *in vivo* observation of the relationship between loss of TSLC1 expression and higher Ki-67 LI also indicates the tumor-suppressive role of the TSLC1 gene in lung adenocarcinoma. In addition, loss of TSLC1 expression was not correlated to p27 (Kip1) or p53 status in lung adenocarcinomas. The p27 protein is an inhibitor of G_1 cyclin-dependent kinases and a negative regulator of cell proliferation; p53 is also a negative regulator of cell proliferation. Functional loss of p27 or p53 is certainly involved in development of lung adenocarcinoma.[20,26] Thus, loss of TSLC1 might lead to an increase in tumor cell proliferation via a pathway independent of these cell cycle regulators.

In conclusion, loss of TSLC1 expression was frequently observed in lung adenocarcinoma, and loss of its expression was significantly correlated to the non-BAC component and proliferative activity of the tumors, suggesting the importance of tumor-suppressive function of TSLC1 in development of lung adenocarcinoma. Lung adenocarcinoma cases with high TSLC1 expression showed better prognosis than cases with loss of TSLC1 expression. Further investigations are needed to elucidate the mechanism by which TSLC1 is downregulated and how loss of TSLC1 influences proliferative activity and invasiveness of lung adenocarcinoma.

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