JOURNAL OF CANCER PREVENTION
VOL. 19, NO. 3, SEPTEMBER, 2014

http://dx.doi.org/10.15430/JCP.2014.19.3.231
pISSN 2288-3649 · eISSN 2288-3657

ORIGINAL ARTICLE

L1 Cell Adhesion Molecule Expression Is Associated With Pelvic Lymph Node Metastasis and Advanced Stage in Diabetic Patients With Endometrial Cancer: A Matched Case Control Study

Dong Hoon Suh[1], Min A Kim[2], Hee Seung Kim[3], Hyun Hoon Chung[3], Noh Hyun Park[3], Yong Sang Song[3,4,5], Soon-Beom Kang[6]

[1]Department of Obstetrics and Gynecology, Seoul National University Bundang Hospital, Seongnam, Korea, [2]Department of Pathology, Seoul National University College of Medicine, Seoul, Korea, [3]Department of Obstetrics and Gynecology, Seoul National University College of Medicine, Seoul, Korea, [4]Cancer Research Institute, Seoul National University College of Medicine, Seoul, Korea, [5]Major in Biomodulation, World Class University, Seoul National University, Seoul, Korea, [6]Women's Gynecologic Oncology Center, Department of Obstetrics and Gynecology, Konkuk University School of Medicine, Seoul, Korea

Background: Diabetic patients with endometrial cancer had more lymph node metastasis than non-diabetic patients with endometrial cancer. L1 cell adhesion molecule (L1CAM) could be possibly associated with lymph node metastasis in diabetic patients with endometrial cancer via epithelial-mesenchymal transition. We aimed to investigate the association between L1CAM expression and lymph node metastasis in diabetic patients with endometrial cancer.
Methods: We conducted a matched case control study of 68 endometrial cancer patients who comprise each 34 diabetic and non-diabetic patients. L1CAM expression was evaluated by immunohistochemistry using fresh formalin-fixed paraffin-embedded tissue block of the patients. The association between L1CAM expression and pelvic lymph node metastasis was assessed according to the presence of diabetes.
Results: Of the 68 patients, 13 (19.1%) were positive for L1CAM immunostaining. Positive rate of L1CAM expression in diabetic endometrial cancer patients was similar to that in non-diabetic endometrial cancer patients (14.7% vs. 23.5%, $P = 0.355$). Tumor recurred more frequently in patients with positive L1CAM expression than those with negative L1CAM expression (33.3% vs. 1.6%, $P = 0.019$). However, we failed to find any significant association between L1CAM expression and lymph node metastasis. Only for the diabetic patients (n = 34), patients with pelvic lymph node metastasis had more L1CAM expression than those without lymph node metastasis (50.0% vs. 3.6%, $P = 0.035$). Advanced stage was the only risk factor for recurrence that showed a significant association with L1CAM expression for the diabetic endometrial cancer patients ($P = 0.006$), as well as all the enrolled patients ($P = 0.014$).
Conclusion: L1CAM expression is associated with pelvic lymph node metastasis and advanced stage in diabetic patients with endometrial cancer.
(J Cancer Prev 2014;19:231-239)

Key Words: Neural cell adhesion molecule L1, Immunohistochemistry, Endometrial cancer, Diabetes mellitus, Lymphatic metastasis

INTRODUCTION

Endometrial cancer (EC) is the most common gynecological malignancy in the United States.[1] ECs commonly present in the early stage and almost two-thirds are diagnosed in stage I, according to the 2009 International Federation of Gynecology and Obstetrics (FIGO) staging system. However, once tumor recurs, treatment of recurrent EC remains a difficult clinical problem, due to the ineffectiveness of any combinations of chemotherapy, radiation therapy, and novel molecular targeted agents.[1] Lymph

Received July 28, 2014, **Revised** August 31, 2014, **Accepted** September 1, 2014

Correspondence to: Soon-Beom Kang
Department of Obstetrics and Gynecology, Konkuk University School of Medicine, 120-1 Neungdong-ro, Hwayang-dong, Gwangjin-gu, Seoul 143-729, Korea
Tel: +82-2-2030-8361, Fax: +82-2-2030-8363, E-mail: ksb308@gmail.com
This research was conducted when Soon-Beom Kang was a professor at Seoul National University College of Medicine.

node (LN) metastasis is one of the most important risk factors for recurrence of EC.[2] Patients with pelvic and paraaortic LN metastasis are designated as FIGO stage IIIC1 and IIIC2, respectively.[3]

Diabetes mellitus (DM) is known to increase the risk of EC.[4] Moreover, diabetic patients with EC were reported to have more LN metastasis and poorer prognosis than non-diabetic EC patients.[5] Therefore, reliable pre-or intra-operative predictors for LN metastasis could help tailor the treatment and improve the prognosis, especially in diabetic patients with EC.

L1 cell adhesion molecule (L1CAM) is a type I membrane glycoprotein of the immunoglobulin superfamily, which is initially identified in the nervous system.[6] Apart from multiple regulatory functions of L1CAM in neuronal cells, such as cell-to-cell interactions, neuronal migration, and neurite fasciculation and myelination,[7] recent studies have demonstrated that L1CAM was aberrantly expressed in a variety of human cancers, including EC.[8-10] Overexpression of L1CAM in tumor cells was reported to increase cell motility, enhance the growth rate, and promote cell transformation and tumorigenicity, as well as form metastases.[6] High L1CAM expression was shown to be associated with recurrent disease and short survival in EC.[11] However, there was no study that evaluated the association of L1CAM expression in EC with LN metastasis.

A mechanism how DM negatively affects the prognosis of EC remained to be determined. Based on a small number of studies, which provided indirect evidence for possible association of L1CAM expression with DM,[12-14] we hypothesized that L1CAM expression might play a crucial role in LN metastasis in diabetic patients with EC. In this study, we aimed to investigate the association between L1CAM expression and LN metastasis in diabetic patients with EC.

MATERIALS AND METHODS

1. Patients and tissue samples

We reviewed medical records of 470 patients, who underwent surgery for EC between January 1990 and June 2010. Approval of the Institutional Review Board of Seoul National University Hospital was obtained in advance. Of the 470, 46 were selected for diabetic group if the patient had DM at the time of diagnosis of EC. Of the 46, key paraffin-embedded tissue blocks of 11 patients were not available, and we retrieved 35 paraffin-embedded tissues from the archival files of the Department of Pathology. Each diabetic patient in diabetic group was matched to one non-diabetic patient in the non-diabetic group for age at diagnosis, preoperative body mass index, histological type, grade, and stage. Except one whose tissue blocks were not available, 34 in the non-diabetic group were included. Finally, 34 patients with paraffin-embedded tissue in each group were included in the

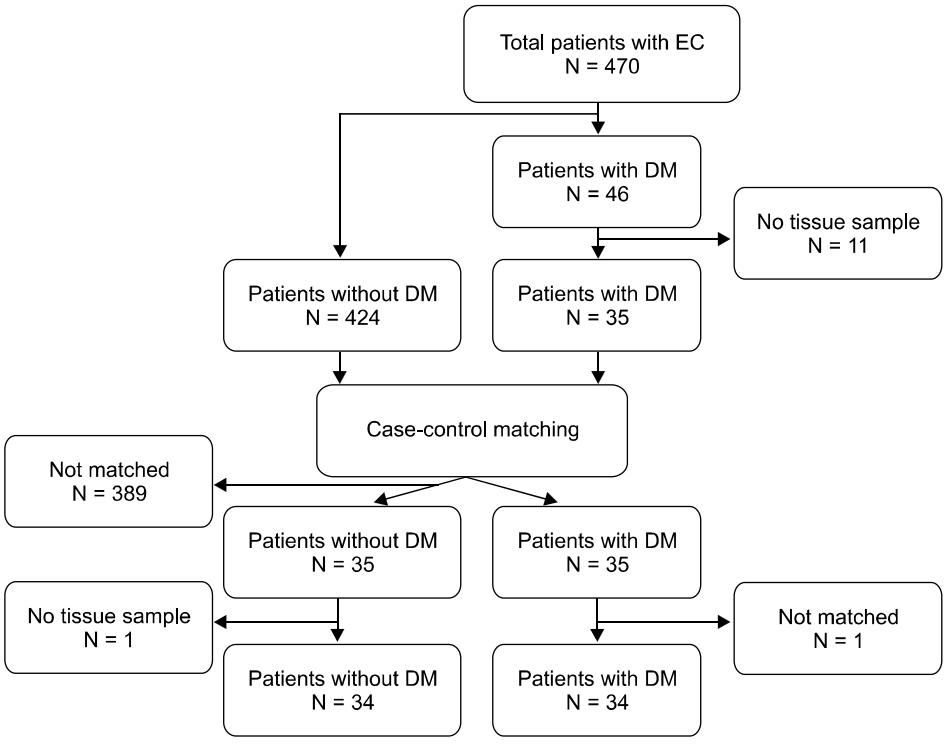

Figure 1. Process for a case-control patient matching: EC, endometrial cancer; DM, diabetes mellitus.

analysis (Fig. 1).

We collected patient information regarding age at diagnosis, preoperative body mass index, presence of DM, intraoperative findings, pathologic findings, including preoperative endometrial curettage, as well as tissue specimen obtained during the surgery, FIGO stage, and follow-up results, which are summarized in Table 1. The survival time and follow-up period were calculated from the date of surgery.

Table 1. Clinicopathologic characteristics

Characteristics	EC with DM	EC without DM	P
Number of patients	34	34	
Age (years), mean (range)	56.7 (31-81)	56.7 (34-76)	0.991
BMI (kg/m^2), mean (range)	28.2 (21.0-42.1)	26.4 (20.5-43.4)	0.086
Serum CA125 levels (U/ml), mean (range)	38.8 (3.2-336.5)	28.0 (4.4-307.0)	0.535
FIGO stage, n (%)			0.782
IA	24 (70.6)	26 (76.5)	
IB	3 (8.8)	1 (2.9)	
II	3 (8.8)	3 (8.8)	
IIIC	4[a] (11.8)	4[b] (11.8)	
Histological type, n (%)			0.259
Endometrioid	28 (82.4)	32 (94.1)	
Non-endometrioid	6 (17.6)	2 (5.9)	
Histological grade, n (%)			0.329
I	17 (50.0)	21 (61.8)	
II, III	17 (50.0)	13 (38.2)	
Myometrial invasion, n (%)			1.000
≤ 1/2	30 (88.2)	30 (88.2)	
> 1/2	4 (11.8)	4 (11.8)	
LVSI, n (%)			0.741
Positive	6 (18.2)	5 (15.2)	
Negative	27 (81.8)	28 (84.8)	
Primary tumor size (cm), mean (range)	3.0 (0.1-13.5)	2.2 (0.1-5.0)	0.104
Washing cytology, n (%)			0.493
Positive for malignancy	2 (5.9)	0 (0)	
Negative for malignancy	32 (94.1)	32 (100.0)	
Lymphadenectomy, n (%)			1.000[c]
PLND only	20 (58.8)	25 (73.5)	0.177[d]
PLND and PALND	12 (35.3)	7 (20.6)	
Not done	2 (5.9)	2 (5.9)	
Number of LN harvested, mean (range)	26.3 (0-50)	24.5 (0-57)	0.552
LN metastasis, n (%)			1.000
Positive	4 (12.5)	4 (12.5)	
Negative	28 (87.5)	28 (87.5)	
Paraaortic LN metastasis[e], n (%)			0.263
Positive	3 (25.0)	0 (0)	
Negative	9 (75.0)	7 (100.0)	
Recurrence, n (%)			1.000
Yes	2 (5.9)	1 (2.9)	
No	32 (94.1)	33 (97.1)	
L1CAM expression, n (%)			0.355
Positive	5 (14.7)	8 (23.5)	
Negative	29 (85.3)	26 (76.5)	

BMI, body mass index; DM, diabetes mellitus; EC, endometrial cancer; FIGO, International Federations of Gynecology and Obstetrics; LN, lymph node; LVSI, lymphovascular space invasion; PALND, paraaortic lymph node dissection; PLND, pelvic lymph node dissection.

[a]1 IIIC1 and 3 IIIC2.

[b]2 IIIC1 but, pathologic reports for paraaortic LN metastasis of the other 2 are not available.

[c]Comparison between lymphadenectomy and no lymphadenectomy.

[d]Comparison between PLND only and PLND with PALND.

[e]All patients with paraaortic LN metastasis have pelvic LN metastasis.

As a normal control, we also investigated normal endometrial tissue blocks from 12 patients undergoing hysterectomy for leiomyoma of uterus: 8 proliferative and 4 secretory phase of the menstrual cycle.

2. Tissue microarray

Tissue microarrays (TMA) were constructed from the core biopsies (diameter 2 mm) of formalin-fixed paraffin-embedded primary EC specimens, using a trephine apparatus (Superbiochips Laboratories, Seoul, Korea). Three core biopsies were taken from each individual specimen. Normal endometrial specimens were also included in each of the array blocks.

3. Immunohistochemistry

After deparaffinization, tissues were rehydrated and subjected to antigen retrieval by immersing in pH 9.0 Tris-EDTA buffer and

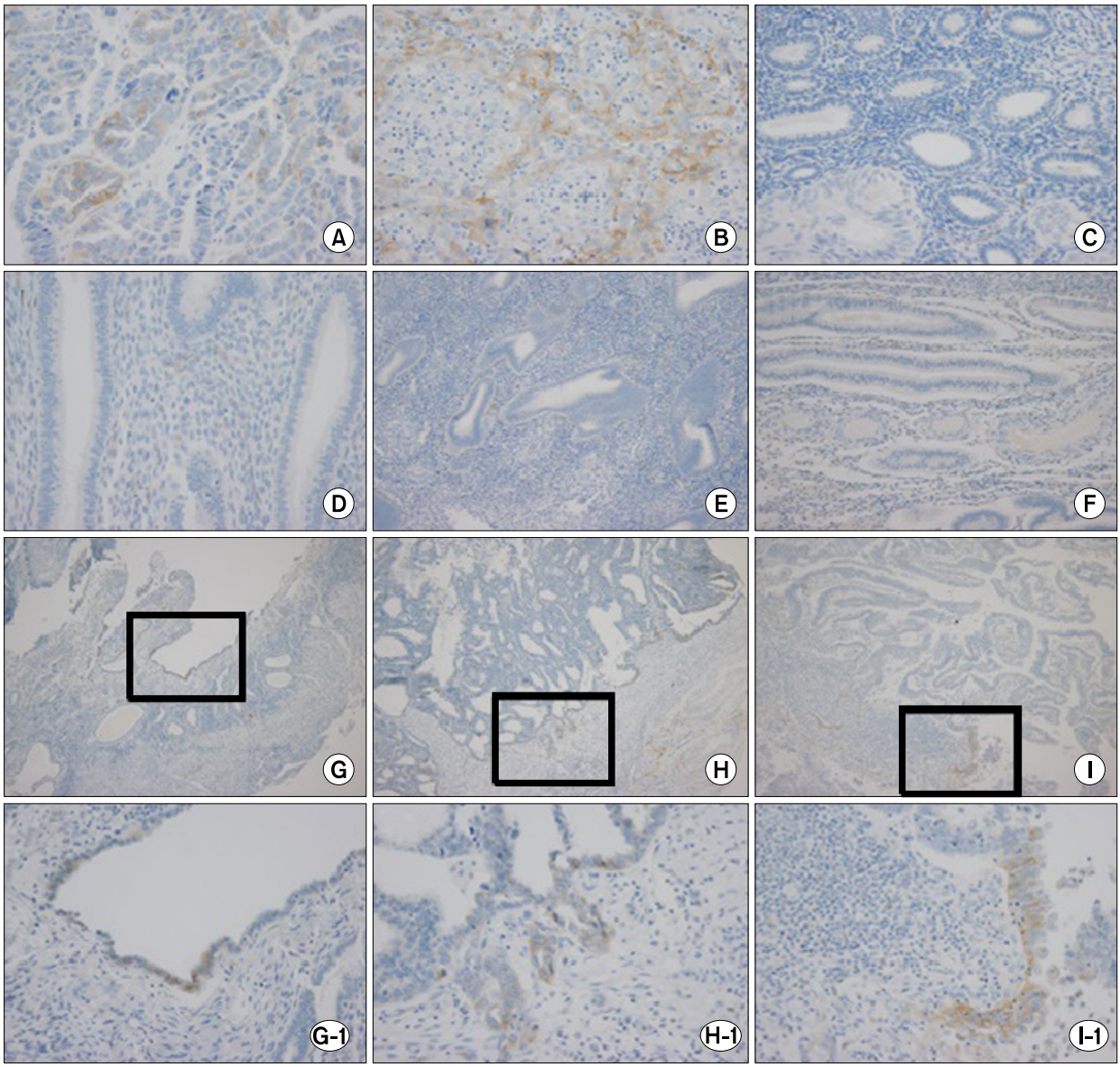

Figure 2. Expression of L1 cell adhesion molecule (L1CAM) in endometrial cancer (EC) tissue. Membranous immunohistochemical staining of EC tissue of L1CAM (brown) with nuclear counterstaining (blue). L1CAM-positive EC (A, endometrioid type; B, clear cell type; ×400). L1CAM-negative EC (C and D; ×400). Negative L1CAM expression in normal proliferative (E) and secretory (F) endometrial tissue (×200). Exclusive staining at the invasive tumor front at low magnification (G, H, and I; ×100). Higher magnification of the boxed areas in (G), (H), and (I) showing L1CAM expression in invading tumor cells into adjacent stroma (G-1, H-1, and I-1; ×400). (A, C, and G, EC tissue of diabetic patients; B, D, E, F, H, and I, EC tissue of non-diabetic patients).

incubating at 100°C for 20 minutes. Endogenous peroxidase activity was blocked with 3% hydrogen peroxide for 6 minutes. After protein blocking for 30 minutes, samples were incubated with a 1:30 dilution of mouse monoclonal antibody (UJ127, Novus Biologicals, Littleton, CO, USA) at 37°C for 32 minutes. After incubating with biotinylated anti-goat IgG for 30 minutes, samples were treated with ABC reagent for another 30 minutes. After incubating for 2 minutes, using a 3,3'-diaminobenzidine substrate kit, samples were counterstained with hematoxylin. Samples were then incubated in a bluing reagent for 4 minutes. Signals were detected using an Ultra view polymer (Ventana, Tucson, AZ, USA).

4. Evaluation of the staining

Staining was evaluated simultaneously by a specialized pathologist and a gynecologic oncologist who were unaware of the patients' clinical features. Any discrepancy was resolved by a third observer. The slides were evaluated for the distribution of staining in the tissue (cytoplasmic or stromal), the percentage of positively stained tumor cells (low, 1-10%; moderate, 11-30%; high, 31-100%), and the intensity of staining (grade 0-3). Cases were defined positive for L1CAM expression when > 5% tumor cells showed > grade 1 staining intensity. Negative cases had to show definitely no L1CAM immunoreactivity in any field.

5. Statistical analysis

Continuous variables were compared using the student t-test, and categorical variables were compared using the chi-square test or Fisher's exact test, as appropriate. Progression-free survival (PFS) was calculated and compared using the Kaplan-Meier method with a log rank test. A 2-sided P-value < 0.05 indicated statistical significance. The statistical analyses were performed using SPSS for Windows version 19.0 (SPSS, Chicago, IL, USA).

RESULTS

1. L1 cell adhesion molecule expression

Of the 68 patients, 13 (19.1%) were positive for L1CAM immunostaining in the cytoplasm of tumor cells. L1CAM was stained with membranous pattern and was often localized in some area rather than diffusely stained (Fig. 2). Interestingly, we observed that L1CAM expression was often localized, exclusively, at the leading front of tumors irrespective of DM status. All of the 12 normal endometrial tissue samples failed to show any evidence for the expression of L1CAM in the proliferative or secretory phase. There was no difference of L1CAM expression, according to the presence of DM.

2. Analysis of clinicopathologic characteristics

At 50.5 months of the mean follow-up (range 2-113 months), a total of three recurrences, 2 diabetic and 1 non-diabetic patients, were observed without disease related mortality. Of the 68 cases in total, 60 (88.2%) were endometrioid histological type and 8 (11.8%) were non-endometrioid histological type: 2 serous, 2 clear cell, and 2 mucinous type in diabetic group and each one serous and clear cell type in non-diabetic group. Due to the matching process, there was no significant difference of the risk factors for recurrence between the diabetic and non-diabetic groups. Four (11.8%), two in each group did not undergo lymphadenectomy because they seemed to have little chance to have LN metastasis: grade 1 endometrioid type EC without myometrial invasion. There was no significant difference of the performance of paraaortic LN dissection between the diabetic and non-diabetic groups (35.3% vs. 20.6%, $P = 0.177$). Positive rate of L1CAM expression in diabetic EC patients was also similar to that in the non-diabetic EC patients (14.7% vs. 23.5%, $P = 0.355$) (Table 1).

Table 2. L1CAM expression and risk factors for recurrence for all enrolled patients (N = 68)

	L1CAM expression		*P*
	Positive	Negative	
FIGO stage, n (%)			**0.014**
Early	2 (3.7)	52 (96.3)	
Advanced	4 (28.6)	10 (71.4)	
Histological type			0.543
Endometrioid	5 (8.3)	55 (91.7)	
Non-endometrioid	1 (12.5)	7 (87.5)	
Histological grade			0.394
I	2 (5.3)	36 (94.7)	
II, III	4 (13.3)	26 (86.7)	
Myometrial invasion, n (%)			0.543
≤ 1/2	5 (8.3)	55 (91.7)	
> 1/2	1 (12.5)	7 (87.5)	
LVSI, n (%)			1.000
Positive	1 (9.1)	10 (90.9)	
Negative	4 (7.3)	51 (92.7)	
LN metastasis, n (%)			0.159
Positive	2 (25.0)	6 (75.0)	
Negative	4 (7.1)	52 (92.9)	
PALN metastasis, n (%)			0.530
Positive	1 (33.3)	2 (66.7)	
Negative	3 (18.8)	13 (81.3)	

FIGO, International Federations of Gynecology and Obstetrics; LN, lymph node; LVSI, lymphovascular space invasion; PALN, paraaortic lymph node.

3. L1 cell adhesion molecule expression and lymph node metastasis

Of the 64 patients, who underwent lymphadenectomy, 8 (12.5%) had LN metastasis: 4 diabetic and 4 non-diabetic EC patients. Of the 19 who underwent paraaortic lymphadenectomy, 3 (15.8%) had paraaortic LN metastasis; all of them had diabetes. Tumor recurred more in patients with positive L1CAM expression than those with negative L1CAM expression for all enrolled patients (33.3% vs. 1.6%, $P = 0.019$) (data not shown). However, we failed to find any significant association between L1CAM expression and LN metastasis for the entire subjects (Table 2).

Only for the diabetic EC patients (n = 34), patients with pelvic

Table 3. L1CAM expression and risk factors for recurrence according to the status of diabetes

	L1CAM expression of diabetic group (N = 34)		P	L1CAM expression of non-diabetic group (N =34)		*P*
	Positive	Negative		Positive	Negative	
FIGO stage, n (%)			**0.006**			0.511
Early	0	27 (100.0)		2 (7.4)	25 (92.6)	
Advanced	3 (42.9)	4 (57.1)		1 (14.3)	6 (85.7)	
Histological type			1.000			0.171
Endometrioid	3 (10.7)	25 (89.3)		2 (6.3)	30 (93.8)	
Non-endometrioid	0	6 (100.0)		1 (50.0)	1 (50.0)	
Histological grade			1.000			0.544
I	1 (5.9)	16 (94.1)		1 (4.8)	20 (95.2)	
II, III	2 (11.8)	15 (88.2)		2 (15.4)	11 (84.6)	
Myometrial invasion, n (%)			0.322			1.000
≤ 1/2	2 (6.7)	28 (93.3)		3 (10.0)	27 (90.0)	
> 1/2	1 (25.0)	3 (75.0)		0	4 (100.0)	
LVSI, n (%)			0.464			1.000
Positive	1 (16.7)	5 (83.3)		0	5 (100.0)	
Negative	2 (7.4)	25 (92.6)		2 (7.1)	26 (92.9)	
Pelvic LN metastasis, n (%)			**0.035**			1.000
Positive	2 (50.0)	2 (50.0)		0	4 (100.0)	
Negative	1 (3.6)	27 (96.4)		3 (10.7)	25 (89.3)	
PALN metastasis, n (%)			1.000			
Positive	1 (33.3)	2 (66.7)				
Negative	2 (22.2)	7 (77.8)				

FIGO, International Federations of Gynecology and Obstetrics; LN, lymph node; LVSI, lymphovascular space invasion; PALN, paraaortic lymph node.

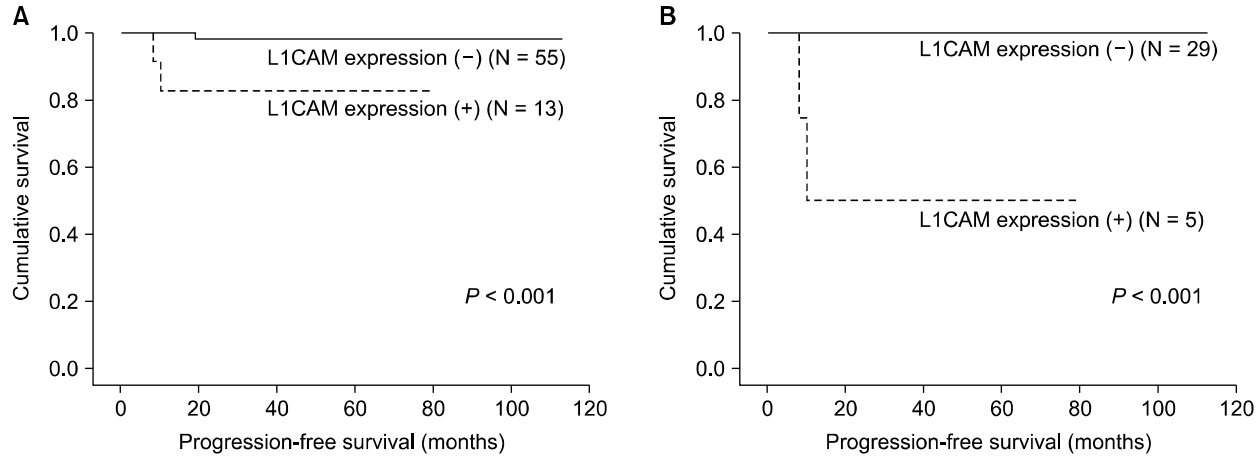

Figure 3. Progression-free survival according to L1 cell adhesion molecule (L1CAM) expression in endometrial cancer (EC). Kaplan-Meier survival curves for all enrolled patients (n = 68) (A) and diabetic EC patients (n = 34) (B).

LN metastasis showed L1CAM expression more than those without LN metastasis (50.0% vs. 3.6%, $P = 0.035$) (Table 3). Nevertheless, there was no significant association between paraaortic LN metastasis and L1CAM expression.

4. L1 cell adhesion molecule expression and other risk factors for recurrence

Advanced stage (FIGO stage II, III) tumor was the only risk factor for recurrence that showed a significant association with L1CAM expression in EC ($P = 0.014$). Moreover, in a subgroup analysis with the diabetic EC patients, the significant relationship between advanced stage and L1CAM expression was also observed ($P = 0.006$). However, no significant association was found between L1CAM expression in diabetic EC patients and other known risk factors for recurrence, such as paraaortic LN metastasis, high grade tumor and deep myometrial invasion.

5. L1 cell adhesion molecule expression and progression-free survival

Kaplan-Meier method with log-rank test revealed that there was no significant difference of PFS between the diabetic and non-diabetic groups (data not shown). However, EC with L1CAM expression recurred significantly earlier than that without L1CAM expression ($P = 0.017$) (Fig. 3A). In a subgroup analysis of diabetic patients, PFS was also significantly shorter in EC patients with L1CAM expression than those without L1CAM expression ($P < 0.001$) (Fig. 3B).

DISCUSSION

This study demonstrated that L1CAM expression was significantly associated with pelvic LN metastasis only in EC patients with DM. L1CAM expression in EC was found to be greater in the advanced stage tumor than in the early stage tumor, irrespective of diabetic condition of the patients. However, L1CAM expression was not associated with the histological type and grade of the tumor, which was consistent with the findings of the previous study.[8] EC patients with L1CAM expression showed a significant shorter PFS than those without L1CAM expression for the diabetic EC patients, as well as all the enrolled patients.

Many studies have shown that transforming growth factor-β, an well-known epithelial mesenchymal transition (EMT) inducer, is a key contributor to diabetic nephropathy.[15-17] Moreover, metformin, a widely prescribed biguanide derivative for the treatment of type 2 DM, has been shown to be able to inhibit EMT pathway in various cancer cells.[18,19] These findings suggest that DM could negatively affect the prognosis of EC through facilitating EMT. EMT is known as an initial step of morphogenic changes of polarized epithelial tumor cells to motile mesenchymal cells, allowing them to invade the basement membrane, enter the vessels and disseminate to secondary organs, during the metastatic progression.[20] We hypothesized that L1CAM expression might play a crucial role in LN metastasis in diabetic patients with EC via EMT. In this study, we observed the distinctive pattern of L1CAM expression, which is often localized exclusively at the leading front of tumors. This finding is consistent with previous observations in colon and ovarian cancer, as well as EC.[6,20,21] An immunohistohcemical study with EC tissue demonstrated that L1CAM was strongly stained at the leading front of the tumor, in contrast to the completely negative staining of the rest of the tumor; whereas, E-cadherin was stained in the opposite way, negative staining of the cells of the leading edge and positive staining of the bulk of the tumor.[20] Another immunohistochemical study of colon cancer tissue also showed that L1CAM expression correlated with high levels of nuclear β-catenin in cells was exclusively localized at the invasive front of the tumor tissue, suggesting that L1CAM might promote the invasiveness of colorectal cancer cells.[6] It suggests that L1CAM expression may be associated with EMT in the progression of these tumors.

In addition, recent studies have shown that the treatment of EC cells with transforming growth factor-β augments L1CAM expression and downregulation of E-Cadherin and this process is blocked by knockdown of Slug that is known to be an essential factor during EMT.[20,22] Slug and β-catenin are involved in the regulation of L1CAM transcription in the EC cell lines,[23] which suggests that L1CAM might be regulated in a fashion similar to that of EMT.

The expression of L1CAM has been shown to be associated with the poor prognosis in EC.[8] We found a significant shorter PFS in EC with L1CAM expression than in EC without L1CAM expression, irrespective of diabetic condition of the patients. By contrast, the association of L1CAM expression with pelvic LN metastasis was observed, only in diabetic patients with EC. It suggests that L1CAM might have important roles in LN metastasis in diabetic EC patients possibly via EMT. This is the first study demonstrating the clinical association between L1CAM expression and LN metastasis in diabetic EC patients, despite the lower expression rate of L1CAM than that of the previous studies (19.1% vs. 28%[8] to 29%[20]). The possible explanation of this finding could include younger age (56.7 years vs. 66.6 years[20] to 68.1 years[8]), more low grade tumor (55.8% vs. 39.0%[20]), and less non-

endometrioid type (11.8% vs. 20.0%[8]). Moreover, racial difference of the subjects between this study and previous ones might account for this finding.

Our study has a few limitations. First, this is a retrospective study with a small sample size. However, the possibility of the selection bias and lower statistical power seemed to be minimal because of the matching comparison design. Second, TMA has been subject to the criticism of representativeness. We used three core sections per case in order to keep the representativeness of TMA. Through this multi-core technique, many studies indicated the excellent quality and good reliability of TMA method.[24,25] Third, unfortunately, we have no disease-related mortality. Therefore, we could not perform the overall survival analysis, according to L1CAM expression in our study population, although there was a report of shorter overall survival of EC patients with L1CAM-positive tumors than EC patients with L1CAM-negative tumors.[8]

LN metastasis is one of the most important prognostic determinants in EC, and enormous efforts are still being put into identifying patients at low or high risk for LN metastasis pre- and intra-operatively.[26,27] There is, however, no fully reliable method to assess an individual EC patient's risk category, either pre- or intra-operatively. The sensitivity and specificity of computed tomography or magnetic resonance image for the detection of LN metastasis in EC were reported to be 27-66% and 73-99%, respectively.[28] Previous reports revealed the sensitivity, specificity, and accuracy of positron emission tomography/CT were 41-77%, 56-100%, and 76-89%, respectively.[28-30]

Our results suggest that L1CAM expression constitutes a new biomarker for EC of diabetic patients associated with LN metastasis and poor prognosis. Fogel et al.[8] reported that preoperative L1CAM immunohistochemical staining of endometrial curettage samples could identify patients with aggressive tumor, and consequently, at high risk for recurrence. Our study results confirmed the prognostic value of L1CAM in patients with EC, especially those who are at risk of LN metastasis due to DM. Through further studies with a larger number of samples, preoperative LN metastasis prediction model could be developed, based on L1CAM expression in the diabetic patients with EC. Further study with a large population is warranted.

ACKNOWLEDGMENTS

This work was supported by Seoul National University Hospital [grant number 0420100810].

CONFLICTS OF INTEREST

We declare that there is no potential conflict of interest to disclose.

REFERENCES

1. Hill EK, Dizon DS. Medical therapy of endometrial cancer: current status and promising novel treatments. Drugs 2012;72:705-13.
2. Fujimoto T, Nanjyo H, Nakamura A, Yokoyama Y, Takano T, Shoji T, et al. Para-aortic lymphadenectomy may improve disease-related survival in patients with multipositive pelvic lymph node stage IIIc endometrial cancer. Gynecol Oncol 2007;107:253-9.
3. Pecorelli S. Revised FIGO staging for carcinoma of the vulva, cervix, and endometrium. Int J Gynaecol Obstet 2009;105:103-4.
4. Weiderpass E, Persson I, Adami HO, Magnusson C, Lindgren A, Baron JA. Body size in different periods of life, diabetes mellitus, hypertension, and risk of postmenopausal endometrial cancer (Sweden). Cancer Causes Control 2000;11:185-92.
5. Steiner E, Schmidt M, Weikel W, Koelbl H. Influence of diabetes mellitus and nodal distribution in endometrial cancer and correlation to clinico-pathological prognostic factors. Eur J Gynaecol Oncol 2006;27:477-80.
6. Raveh S, Gavert N, Ben-Ze'ev A. L1 cell adhesion molecule (L1CAM) in invasive tumors. Cancer Lett 2009;282:137-45.
7. Brummendorf T, Kenwrick S, Rathjen FG. Neural cell recognition molecule L1: from cell biology to human hereditary brain malformations. Curr Opin Neurobiol 1998;8:87-97.
8. Fogel M, Gutwein P, Mechtersheimer S, Riedle S, Stoeck A, Smirnov A, et al. L1 expression as a predictor of progression and survival in patients with uterine and ovarian carcinomas. Lancet 2003;362:869-75.
9. Zecchini S, Bianchi M, Colombo N, Fasani R, Goisis G, Casadio C, et al. The differential role of L1 in ovarian carcinoma and normal ovarian surface epithelium. Cancer Res 2008;68:1110-8.
10. Boo YJ, Park JM, Kim J, Chae YS, Min BW, Um JW, et al. L1 expression as a marker for poor prognosis, tumor progression, and short survival in patients with colorectal cancer. Ann Surg Oncol 2007;14:1703-11.
11. Fogel M, Huszar M, Altevogt P, Ben-Arie A. L1 (CD171) as a novel biomarker for ovarian and endometrial carcinomas. Expert Rev Mol Diagn 2004;4:455-62.
12. Jiang H, Guan G, Zhang R, Liu G, Cheng J, Hou X, et al. Identification of urinary soluble E-cadherin as a novel biomarker for diabetic nephropathy. Diabetes Metab Res Rev 2009;25:232-41.
13. Gavert N, Conacci-Sorrell M, Gast D, Schneider A, Altevogt P, Brabletz T, et al. L1, a novel target of beta-catenin signaling, transforms cells and is expressed at the invasive front of colon cancers. J Cell Biol 2005;168:633-42.
14. Sengupta U, Ukil S, Dimitrova N, Agrawal S. Expression-based network biology identifies alteration in key regulatory pathways of type 2 diabetes and associated risk/complications. PLoS One 2009;4:e8100.
15. Hills CE, Squires PE. TGF-beta1-induced epithelial-to-mesenchy-

mal transition and therapeutic intervention in diabetic nephropathy. Am J Nephrol 2010;31:68-74.
16. Holian J, Qi W, Kelly DJ, Zhang Y, Mreich E, Pollock CA, et al. Role of Kruppel-like factor 6 in transforming growth factor-beta1-induced epithelial-mesenchymal transition of proximal tubule cells. Am J Physiol Renal Physiol 2008;295:F1388-96.
17. Hills CE, Al-Rasheed N, Al-Rasheed N, Willars GB, Brunskill NJ. C-peptide reverses TGF-beta1-induced changes in renal proximal tubular cells: implications for treatment of diabetic nephropathy. Am J Physiol Renal Physiol 2009;296:F614-21.
18. Barrière G, Tartary M, Rigaud M. Metformin: a rising star to fight the epithelial mesenchymal transition in oncology. Anticancer Agents Med Chem 2012;13:333-40.
19. Del Barco S, Vazquez-Martin A, Cufi S, Oliveras-Ferraros C, Bosch-Barrera J, Joven J, et al. Metformin: multi-faceted protection against cancer. Oncotarget 2011;2:896-917.
20. Huszar M, Pfeifer M, Schirmer U, Kiefel H, Konecny GE, Ben-Arie A, et al. Up-regulation of L1CAM is linked to loss of hormone receptors and E-cadherin in aggressive subtypes of endometrial carcinomas. J Pathol 2010;220:551-61.
21. Gast D, Riedle S, Riedle S, Schabath H, Schlich S, Schneider A, et al. L1 augments cell migration and tumor growth but not beta3 integrin expression in ovarian carcinomas. Int J Cancer 2005;115:658-65.
22. Geismann C, Morscheck M, Koch D, Bergmann F, Ungefroren H, Arlt A, et al. Up-regulation of L1CAM in pancreatic duct cells is transforming growth factor beta1- and slug-dependent: role in malignant transformation of pancreatic cancer. Cancer Res 2009;69:4517-26.
23. Pfeifer M, Schirmer U, Geismann C, Schäfer H, Sebens S, Altevogt P. L1CAM expression in endometrial carcinomas is regulated by usage of two different promoter regions. BMC Mol Biol 2010;11:64.
24. Jawhar NM. Tissue microarray: a rapidly evolving diagnostic and research tool. Ann Saudi Med 2009;29:123-7.
25. Shergill IS, Shergill NK, Arya M, Patel HR. Tissue microarrays: a current medical research tool. Curr Med Res Opin 2004;20:707-12.
26. Kang S, Kang WD, Chung HH, Jeong DH, Seo SS, Lee JM, et al. Preoperative identification of a low-risk group for lymph node metastasis in endometrial cancer: a Korean gynecologic oncology group study. J Clin Oncol 2012;30:1329-34.
27. Suh DH, Kim K, Kim JW. Major clinical research advances in gynecologic cancer in 2011. J Gynecol Oncol 2012;23:53-64.
28. Kitajima K, Yamasaki E, Kaji Y, Murakami K, Sugimura K. Comparison of DWI and PET/CT in evaluation of lymph node metastasis in uterine cancer. World J Radiol 2012;4:207-14.
29. Park JY, Kim EN, Kim DY, Suh DS, Kim JH, Kim YM, et al. Comparison of the validity of magnetic resonance imaging and positron emission tomography/computed tomography in the preoperative evaluation of patients with uterine corpus cancer. Gynecol Oncol 2008;108:486-92.
30. Chung HH, Park NH, Kim JW, Song YS, Chung JK, Kang SB. Role of integrated PET-CT in pelvic lymph node staging of cervical cancer before radical hysterectomy. Gynecol Obstet Invest 2009;67:61-6.