Annals of Diagnostic Pathology 42 (2019) 7–11

Contents lists available at ScienceDirect

Annals of Diagnostic Pathology

journal homepage: www.elsevier.com/locate/anndiagpath

Original Contribution

Large nested urothelial carcinoma: A clinicopathological study of 22 cases on transurethral resection materials☆

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ARTICLE INFO

Keywords:
Large nested variant
Urothelial carcinoma
Bladder cancer
Histology
Cytology

ABSTRACT

Large nested urothelial carcinoma (LNUC) is a recently recognized variant of urothelial carcinoma (UC), which is characterized with inverted growth pattern and bland cytology. Diagnosis can be extremely challenging, especially in transurethral resection (TUR) materials.

Haematoxylin-eosin stained slides of TUR materials with UC, submitted to our department between 2008 and 2017 were re-examined. Twenty-two LNUC cases were found.

LNUC frequency was 0.7%. Mean age was 69.9, 82% were male. Mean tumor diameter was 4.9 cm. Non-invasive UC was present in all cases; low-grade in 6, high-grade in 1, low and high-grade in 15. Five and 16 cases were pT1 and pT2, respectively, no invasion was detected in one case. In addition to medium-large nests, small nests and conventional UC areas was present in 2 and 3 cases, respectively. Stroma-tumor interface was irregular in 19 cases and 3 cases had invasive nests with rounded contours. Fibrous stromal reaction and/or stromal lymphoid infiltration were present in 21 cases. Budding, described as small nests in stromal interface of medium-large nests was found in 16 cases. Follow-up was available for 18 cases with an average of 59 months. Four cases had metastasis, 1 patient died of disease.

LNUC causes diagnostic difficulty, especially in TUR materials. Large size of tumor, irregularity of nests, presence of stromal reaction and budding can be clues for correct diagnosis.

1. Introduction

The morphological spectrum of urothelial carcinoma (UC) shows great diversity with different lines of differentiation and many variants, some of which are recently described [1]. Some of these UC variants have important impact on prognosis and treatment and some of them challenge the pathological diagnosis by its deceptive morphology [2-7]. Recognition of these variants by pathologists is important for evaluation of invasion and therefore staging. One of these challenging variants is 'large nested UC', which can be mistaken for a benign neoplasm or noninvasive UC with endophytic growth pattern because of its pattern of invasion and bland cytological features [6].

The importance of endophytic growth pattern in urothelial carcinoma has been recognized for a long time. However, the nomenclature 'Large nested variant UC' was first used by Cox and Epstein in 2011 [6]. This entity was accepted as a variant of urothelial carcinoma in the 2016 edition of WHO Urogenital Tumor Classification [1]. Cox and Epstein describe this variant as papillary urothelial carcinoma with an inverted growth pattern, characterized by large rounded nests of carcinoma in the lamina propria/muscularis propria with the same cytology as the surface papillary component [6]. These carcinomas share features of both the nested pattern of urothelial carcinoma and papillary urothelial carcinomas with an inverted growth pattern. According to WHO 2016 classification, nested (including large nested) urothelial carcinoma is a cytologically bland variant of invasive urothelial carcinoma. With its bland cytological features and special pattern of invasion that is different than the classical pattern of invasion in urothelial carcinoma, this variant requires utmost attention, especially in transurethral resection (TUR) materials, due to its capacity for presenting in an advanced stage and metastasizing [6,8].

In this study, we aim to describe the clinical and histopathological features of large nested variant urothelial carcinoma in bladder TUR

Abbreviations: UC, urothelial carcinoma; LNUC, large nested urothelial carcinoma; TUR, transurethral resection; WHO, World Health Organization

☆ The authors have disclosed that we have no significant relationships with, or financial interest in, any commercial companies pertaining to this article.

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https://doi.org/10.1016/j.anndiagpath.2019.06.005

Table 1
Clinical features, pathological stages and follow-up information of cases.

Case no	Age/sex	Tumor diameter	Stage	Clinical findings	Treatment	Follow-up	Status
1	78/F	1,5	T1	Hematuria	TURB	*Re*-TURB, cystectomy (pT2b)	No evidence of disease
2	79/M	1	T1	Hematuria	TURB	Re-TURB	DOC
3	59/M	5	T2	Routine examination	TURB	Cystoprostatectomy (pT3bN1)	Alive with disease at 74 months
4	58/M	1,5	T2	Hematuria	TURB	Cystoprostatectomy (pT2b)	Alive with disease at 48 months
5	83/M	4,5	T2	Hematuria	TURB	Cystoprostatectomy (pT2b)	DOC
6	62/M	2	T2	Routine examination	TURB	Cystoprostatectomy (pT3bN1)	NA
7	37/M	4	T2	Hematuria	TURB	Cystoprostatectomy (pT3a)	Bone metastasis, Dead of disease
8	68/M	3	T2	Hematuria	TURB	NA	NA
9	66/F	9	T1	Hematuria, anemia	TURB	*Re*-TUR	No evidence of disease
10	64/M	8	T2	Hematuria	TURB	Cystoprostatectomy (pT0)	No evidence of disease
11	68/M	4,5	T2	Hematuria	TURB	Cystoprostatectomy (pT0)	No evidence of disease
12	77/M	3	T2	Hematuria, dysuria	TURB	NA	NA
13	61/M	9	T2	Hematuria	TURB	NA	NA
14	79/M	6	Ta	Hematuria	TURB	Chemotherapy	Lung metastasis-metastasectomy, Alive with disease at 20 months
15	77/M	7	T2	Hematuria	TURB	Cystoprostatectomy (pT3b)	No evidence of disease
16	73/M	5	T2	Hematuria	TURB	Cystoprostatectomy (pT3b)	DOC
17	79/F	4	T2	Hematuria, dysuria	TURB	Cystectomy (pT3b)	Alive with disease at 62 months
18	84/M	9	T1	Hematuria	TURB	Re-TUR, cystoprostatectomy (pT2b)	DOC
19	65/M	5	T2	Hemoptysis	TURB	Chemotherapy	Presented with multiple lung metastases, Alive with disease at 10 months
20	70/M	4	T2	Hematuria	TURB	NA	No evidence of disease
21	73/M	6	T2	Hematuria, weight loss	TURB	Cystoprostatectomy (pT3b)	Lung metastasis, Alive with disease at 26 months
22	78/F	6	T2	Hematuria	TURB	Cystoprostatectomy (pT3b)	DOC

F, female; M, male; NA, data not available; DOC, Dead of other causes; TUR, Transurethral resection.

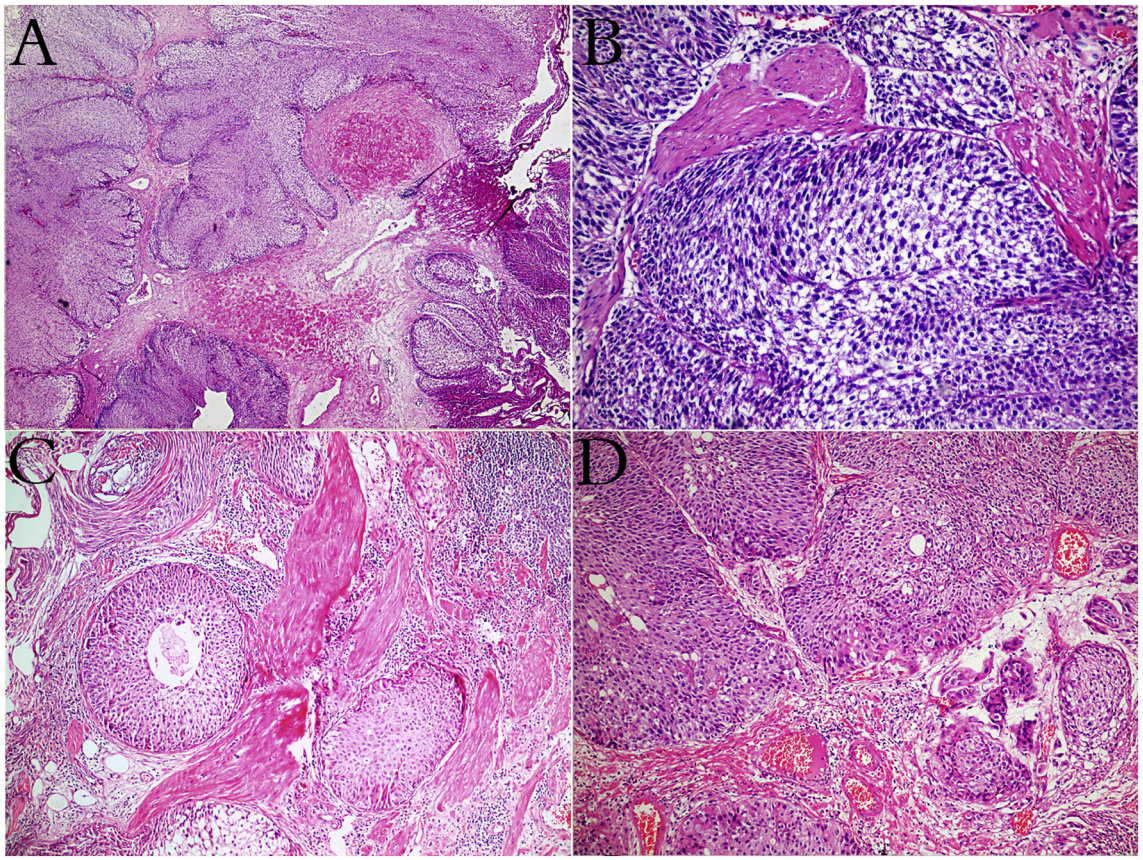

Fig. 1. Large nested urothelial carcinoma. (A) Endophytic non-invasive growth pattern (Case 11). (B) Invasive nests are well-demarcated with monotonous bland appearing cytology and do not show stromal reaction, tumor-stroma interface irregularity or stromal lymphoid infiltration (Case 11). (C) Invasive large nests and stromal lymphoid infiltration are present between muscle bundles (Case 10). (D) Conventional UC areas and medium sized nests are present among large invasive nests (Case 9).

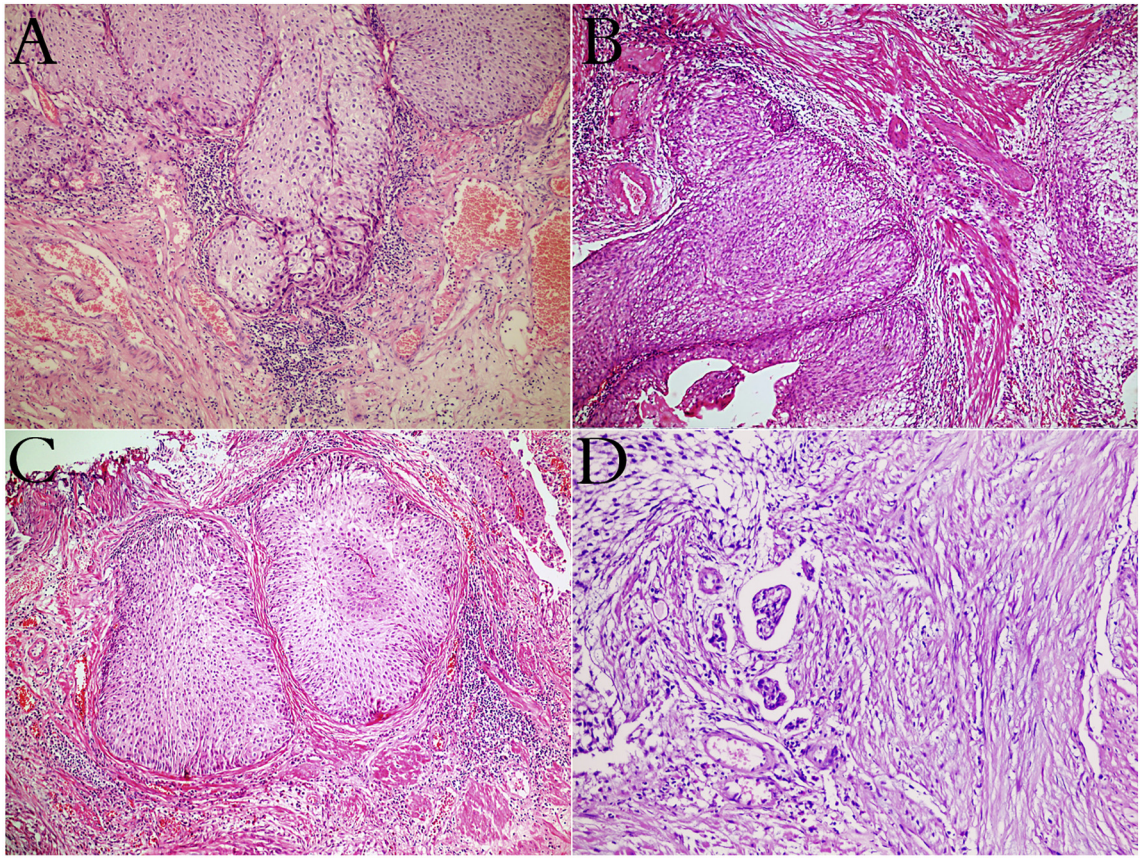

Fig. 2. Large nested urothelial carcinoma. (A) Budding, tumor-stroma interface irregularity and marked lymphoid infiltration in pre-invasive large nested urothelial carcinoma areas (Case 5). (B) Pushing invasion of large nests with budding, tumor-stroma interface irregularity, marked lymphoid infiltration and stromal reaction (Case 9). (C) Large invasive urothelial nests between muscle bundles with bland cytological features (Case 6). (D) Note the presence of lymphovascular invasion (Case 7 and Case 10).

materials and to find out the possible clinicopathological clues for accurate diagnosis of this variant, which is especially challenging in TUR materials.

2. Materials and methods

Electronical database was searched for TUR materials diagnosed as urothelial carcinoma between 2008 and 2017. A total of 1980 TUR materials were retrieved from the archive and reviewed for the purpose of variant stratification. All Haematoxylin-eosin stained slides of the cases with large nested variant were re-examined. Also, one recut for each paraffin block of the cases was prepared and examined. The clinical data of the patients, including age and medical history, were obtained from the patient charts.

All cases with large nested pattern were evaluated according to the clinical and histological parameters, which were age, sex, tumor size, surface component of urothelial carcinoma (presence and grade), extent of invasion, conventional pattern of invasion, stromal reaction (fibrous reaction and/or stromal lymphoid infiltration), angiolymphatic invasion and necrosis. Follow-up and disease status was assessed, where available.

3. Results

3.1. Clinical

There were 22 UC cases with large nested pattern of invasion among 1980 TUR materials. Eight of the cases were retrieved from the consultation files of one of the authors (KB). The frequency of large nested variant was 0.7% (14/1972). The mean age was 69.9 (range: 37–84) and 82% of the patients (18:4) were male. Mean tumor diameter was 4.9 cm. The clinical information on cases is summarized in Table 1.

3.2. Histology

Non-invasive UC component was present in all cases; low-grade in 10, high-grade in 1, low and high-grade in 15 cases. 5 and 16 cases were pT1 and pT2, respectively. In one case (case no: 14), no invasion was detected despite serial recuts. However, no muscularis propria was identified either. In the follow-up, this patient had lung metastasis. All cases had invasive component composing of medium-large nests (Figs. 1B–C, 2C). Seventeen cases were pure LNUC (77%). Two cases had focal small invasive nests (nested variant UC); the percentage of accompanying nested variant was 5% and 20% in these cases. Three other cases had areas of conventional invasive UC, percentages ranging between 5% and 50% (Fig. 1D). Stromal-tumor interface was irregular in 19 cases, whereas 3 cases had invasive nests with rounded contours. Fibrous stromal reaction and stromal lymphoid infiltration were present at varying proportions in 19 and 21 cases, respectively. We described “budding” as small nests in stromal interface of medium large nests and this was a remarkable feature in 16 of our cases (Fig. 2A and B). Among these 16 cases, 12 were pure LNUC, 3 had accompanying conventional UC and 1 had accompanying nested variant UC. The small nests in stromal interface were not entirely separate from the larger ones. They were clinging to the medium-large nests that composed the tumor. This feature mostly accompanied tumor-stroma interface irregularity and stromal lymphoid infiltration (Fig. 2A and B). Angiolymphatic invasion and necrosis were detected in 2 and 5 cases, respectively (Fig. 2D).

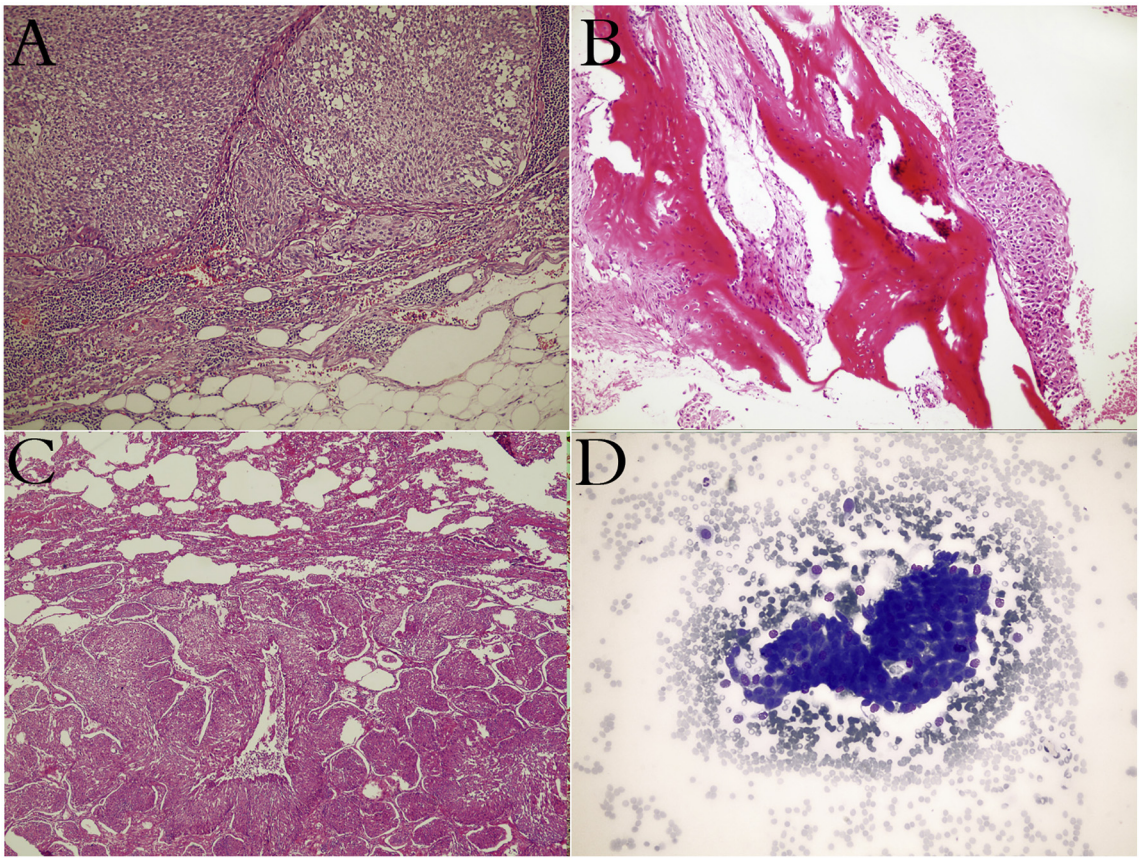

Fig. 3. Large nested urothelial carcinoma. (A) Lymph node metastasis with large nested pattern (Case 6), (B) Bone metastasis (Case 7), (C) Lung metastasis (Case 14). (D) Urothelial cells with bland cytologic features in lung aspiration biopsia (Case 14).

Table 2
Histological features of cases.

Pattern of invasion	
Medium-large nests	22/22
Small invasive nests	2/22
Conventional invasive UC	3/22
Stroma-tumor interface	
Irregular	19/22
Rounded	3/22
Fibrous stromal reaction	19/22
Stromal lymphoid infiltration	21/22
Surface precursor component	
Low grade UC	6/22
High grade UC	1/22
Low and high grade UC	15/22
Stage	
Lamina propria invasion	5/22
Muscularis propria invasion	16/22
No invasion detected*	1/22
Budding	16/22
Necrosis	5/22
Angiolymphatic invasion	2/22

In one case, no invasion was detected despite serial recuts.

Cytologically, the urothelial cells within the nests had bland morphologic features in all of the cases, with uniform, vesicular nuclei and small indistinct nucleoli (Figs. 1A–D, 2A–D). Similar morphological appearance with medium-large nest structures and bland cytology were also seen in the biopsies from the metastatic sites of the patients (Fig. 3A–D). Histological features of the cases are summarized in Table 2.

3.3. Prognosis

Follow-up was available for 18 cases with an average interval of 59 months (10 to 98 months). In 3 cases, lung metastasis was present. In fact, one of these 3 patients (case no: 19) presented with haemoptysis and metastatic lung mass was detected before bladder tumor. In other patient with lung metastasis (case no: 14), no invasion was detected in the TUR materials despite serial recuts and the case was reported as pTa. But as mentioned in the previous section, this biopsy did not include muscularis propria. Other patient with lung metastasis (case no: 21) had previous TUR material diagnosed as pT2. One case (case no: 7) had bone metastasis, detected 10 years after the initial diagnosis of pT2 urothelial carcinoma. It could be argued that a metastasis this late could be related to a new primary tumor. However, we were informed by the clinicians that the patient was searched thoroughly and no new tumor could be detected. Among all cases, only this patient with bone metastasis died of disease, and this patient died 56 months after being diagnosed with metastasis.

4. Discussion

Large nested urothelial carcinoma is among the newest variants of urothelial carcinoma accepted in WHO 2016 and described as urothelial carcinoma with an inverted growth pattern, characterized by large rounded nests of carcinoma in the lamina propria/muscularis propria with the same cytology as the surface component [1,6]. Other than its special invasion pattern, it is reported to have bland cytological features. However, despite this bland cytology, it has been shown that presentation at advanced stage and metastasis are not uncommon in large nested urothelial carcinoma cases [6,8]. Due to its inverted growth pattern, recognizing this variant in TUR materials may be

challenging, especially when the biopsy does not include muscularis propria.

Here in this study, we present a series of 22 cases of large nested variant UC. Only TUR materials were included in the study for the purpose of finding out any morphological clues for accurate diagnosis of this variant in TUR materials. To the best of our knowledge, this study is the largest on LNUC in TUR materials until now. In the study by Cox et al. 18 out of 23 cases were TUR materials and Comperat et al. had only 10 TUR materials out of 36 cases [6,8]. In their series, Cox et al. described 23 cases of UC with a deceptively benign pattern of invasion characterized by large round-to-irregularly shaped nests of bland urothelial cells, often present within the muscularis propria and named it as large nested variant of UC [6]. They emphasized that the presence of nests of urothelial cells in muscularis propria, regardless of the cytology is diagnostic of UC. They also reported that variable size and shape, irregular contours and infiltrative haphazard pattern of the nests may be suggestive of large nested variant UC. Later, Comperat et al. aimed to collect more data on the clinical behavior and immunophenotype of this variant and studied on 36 cases [8]. Their inclusion criteria were large invasive nests with round contour, minimal stromal reaction and low grade cytological features. Similar to Cox et al., Comperat et al. also emphasized the importance of irregular distribution of the nests with variable intervening stroma in the diagnosis of large nested UC. However, it is not always possible to see these findings in TUR materials. We focused on finding more morphological clues regarding the nature of these large nests, that may lead us to the correct diagnosis of large nested UC. In our study, irregularity of stromal-tumor interface was seen in 19 cases, only 3 cases had invasive nests with rounded contours. Fibrous stromal reaction and/or stromal lymphoid infiltration were present at varying proportions in all but one case. “Budding”, which we describe as small nests in stromal interface of medium large nests, was a remarkable feature in 16 cases, 12 of which were pure LNUCs.

This study is the largest single center study on LNUCs up-to-date and the frequency of this variant was found to be 0.7%, which means LNUC is relatively rare among other variants of UC. The mean age in this study was 69.9, which is slightly higher than other studies regarding this variant (63.7 in Cox et al. [6] and 66.7 in Comperat et al. [8]). A male preponderance of 4.5:1 was detected in this study, which is consistent with the literature. In the recent studies on this variant by Cox et al. and Comperat et al., information on tumor size was not provided. In 1997, Amin et al. concluded that the endophytic pattern was not related to tumor size, because even tumors with 1 cm diameter exhibited this growth pattern in their study. In our study, we also had cases with small diameters, however; the mean diameter of the cases was 4.9 cm. This may be a result of these tumors presenting at an advanced stage. In large tumors with endophytic growth pattern, utmost caution should be exercised during histopathological evaluation in order not to miss invasion.

When the difficulties of evaluating a TUR material, such as tangential sectioning, crush artifact and thermal injury, are added to the bland cytological features and special pattern of invasion of this tumor, the correct diagnosis of invasive carcinoma can be extremely challenging. We found out that one of our cases with lung metastasis, which was evaluated with serial recuts and diagnosed as pT2 UC in our hospital, was previously diagnosed as pTa UC in a different center. In the TUR material of the other case with lung metastasis in our series, invasion was not detected although all biopsy material was examined with recuts. But it is important to note that this biopsy did not include muscularis propria. Thus, invasion can be missed in the cases which are not well-sampled.

Metastatic spread and tumor related death have been reported in LNUC in the previous studies [6-8]. A remarkable outcome of our study was that only one patient died of disease, even though capacity for local invasion and metastasis was high. This makes us think that low grade histological features of LNUCs may provide better prognosis than conventional UCs.

LNUC is a tumor with low grade cytological features but high capacity for muscularis propria invasion and metastasis. This rare variant of UC should always be kept in mind, especially when dealing with TUR materials, in order to avoid misdiagnosis resulting from the bland cytological features and inverted growth pattern of this tumor. More caution should be exercised in this kind of cases if the diameter of the lesion is large, and histological findings supporting invasion such as ‘irregularity of tumor-stroma interface’, ‘fibrous stromal reaction and/or stromal lymphoid infiltration’ and ‘budding’ should be searched tenaciously. Furthermore, in cases that raise suspicion for LNUC during histopathological evaluation, re-TUR can be suggested if the current biopsy does not include muscularis propria.

Author contributions

Conception and design of the study: EH, KB.
Acquisition and analysis of data: EH, KB.
Drafting the manuscript or figures: EH, KB.

Declaration of Competing Interest

We declare that we have no conflict of interest.

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