Immunohistochemical Evaluation of GATA3 Expression in Tumors and Normal Tissues

A Useful Immunomarker for Breast and Urothelial Carcinomas

Haiyan Liu, MD, Jianhui Shi, MD, PhD, Myra L. Wilkerson, MD, and Fan Lin, MD, PhD

Key Words: GATA3; Breast carcinoma; Urothelial carcinoma; Normal tissue; Cytology

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CME/SAM

Upon completion of this activity you will be able to:
- apply GATA3 antibody when working on a tumor of unknown origin in both surgical and fine-needle aspiration specimens.
- define effective immunostaining panels in distinction of breast carcinoma from endometrial carcinoma and from ovarian serous carcinoma.
- describe the pattern of GATA3 antibody reactivity in estrogen receptor–positive breast carcinomas.

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Questions appear on p 158. Exam is located at www.ascp.org/ajcpcme.

Abstract

GATA3 expression has been reported in urothelial and breast carcinomas; however, the published data on GATA3 expression in tumors from other organs are limited. Immunohistochemical evaluation of GATA3 expression in 1,110 carcinomas and 310 cases of normal tissue using tissue microarray sections, 48 breast and bladder biopsy specimens, and 53 breast fine-needle aspiration biopsy specimens was performed. Sixty-two of 72 urothelial carcinomas (86%) and 138 of 147 breast carcinomas (94%) tested positive for GATA3. All other cases, except for 2 of 96 endometrial carcinomas, tested negative for GATA3. On fine-needle aspiration biopsy samples, 88% of primary breast carcinomas and 82% of metastatic breast carcinomas tested positive for GATA3. Our study revealed that GATA3 is a sensitive and specific marker for the diagnosis of breast and urothelial carcinomas. When working on a tumor of unknown origin, GATA3 should be routinely included in the initial screening panel if either a breast or urothelial primary tumor is suspected.

GATA3 (GATA binding protein 3 to DNA sequence [A/T]GATA[A/G]) is 1 of 6 members of a zinc finger transcription factor family, and it plays an important role in promoting and directing cell proliferation, development, and differentiation in many tissues and cell types,[1,2] including luminal glandular epithelial cells of the mammary gland,[3-5] T lymphocytes,[6,7] thymocytes,[8,9] adipose tissue,[10] kidney,[11] sympathetic nervous system,[12] and hair follicles of the skin.[13] Together with S100P it has recently been reported to be a useful immunohistochemical marker for the detection of urothelial carcinoma and ovarian Brenner tumors.[14-16] Overexpression of GATA3 has been reported in breast carcinomas by complementary DNA microarray analysis.[17] Low GATA3 expression has also been suggested to correlate with poor prognosis in breast cancer[3,18-20]; however, the published data on GATA3 expression in tumors from other organs and normal tissues are limited. In this study, we investigated the expression of GATA3 in a large series of carcinomas from various organs using a single immunostaining system (DAKO, Carpinteria, CA).

Materials and Methods

Construction of Tissue Microarray Blocks

The study was approved by the institutional review board at Geisinger Medical Center. A total of 1,110 tumors and 310 cases of normal tissues from various organs were identified from the archives of the Department of Laboratory Medicine

at Geisinger Medical Center from 2000 to 2010. Seventy-two cases of urothelial carcinoma were high-grade urothelial carcinoma with muscularis propria invasion. The number of tumor cases for each specific entity is summarized in **Table 1**. Multiple tissue microarray (TMA) blocks with 2 punches of 0.75 mm or 1.0 mm each for each case were constructed as previously described.[21,22]

Immunohistochemical Stains on Surgical Specimens

Immunohistochemical evaluation of the expression of GATA3 (catalogue No. GATA3 [HG3-31]:sc-268; Santa Cruz Biotech, Santa Cruz, CA) in 1,110 carcinomas and 310 cases of normal tissue from various organs using TMA sections, 38 breast core biopsy specimens (9 ductal carcinoma in situ [DCIS], 10 fibroadenoma, 9 ductal hyperplasia, and 10 normal breast tissue), and 10 benign bladder mucosa biopsy specimens was performed. The staining was performed on the DAKO staining system (1:25 dilution, EDTA antigen retrieval, and 45-minute incubation for primary antibody) based on the previously published protocol.[23,24] The staining intensity for the tumors and normal or nonneoplastic breast tissues was graded as weak or strong. Only nuclear staining for GATA3 was regarded as positive. The distribution was recorded as negative (<5% of tumor cells stained), 1+ (5%-25%), 2+ (26%-50%), 3+ (51%-75%), or 4+ (>75%). Two surgical pathologists (F.L. and H.L.) independently evaluated the immunostained slides.

Table 1 Summary of GATA3 Immunostaining Results on 1,110 Tumors

Tumor	No. (%) of GATA3+ Cases
Seminoma (n = 30)	0
Embryonal carcinoma (n = 24)	0
Yolk sac tumor (n = 12)	0
Lung neuroendocrine carcinoma (n = 61)	0
Lung adenocarcinoma (n = 61)	0
Lung SCC (n = 49)	0
Papillary thyroid carcinoma (n = 47)	0
Follicular thyroid carcinoma (n = 37)	0
Medullary thyroid carcinoma (n = 10)	0
Clear cell RCC (n = 82)	0
Papillary RCC (n = 20)	0
Colonic adenocarcinoma (n = 43)	0
Esophageal adenocarcinoma (n = 30)	0
Gastric adenocarcinoma (n = 21)	0
Pancreatic adenocarcinoma (n = 50)	0
Urothelial carcinoma (n = 72)	62 (86)
Prostatic adenocarcinoma (n = 136)	0
Cholangiocarcinoma (n = 11)	0
Breast ductal carcinoma (n = 99)	90 (91)
Breast lobular carcinoma (n = 48)	48 (100)
Endocervical adenocarcinoma (n = 17)	0
Endometrial carcinoma (n = 96)	2 (2)
Ovarian serous carcinoma (n = 56)	0
Hepatocellular carcinoma (n = 18)	0
Pancreatic endocrine neoplasm (n = 15)	0
Skin melanoma (n = 100)	0

RCC, renal cell carcinoma; SCC, small cell carcinoma.

Immunohistochemical Stains on Fine-Needle Aspiration Biopsy Specimens on Cell Blocks

On the basis of the study results, immunohistochemical evaluation of the expression of GATA3 was further performed on 53 cases of fine-needle aspiration biopsy (FNAB) specimens of the breast on cell block sections. The preparation of the cell block was as previously described.[25] The immunostaining protocol was as described for surgical specimens.[23,24]

The 53 cases were divided into 3 groups. Group 1 included 17 cases of breast ductal carcinoma with adequate cellularity (12 cases were well-differentiated or moderately differentiated ductal carcinoma; the remaining 5 were poorly differentiated ductal carcinoma). Group 2 included 22 cases of metastatic breast ductal carcinoma in axillary lymph nodes (12 cases with low to intermediate nuclear grade and 10 with high nuclear grade). Group 3 included 14 cases of benign breast proliferative lesion without atypia (9 cases with apocrine metaplasia). Only carcinoma cases with adequate cellularity in the cell block preparation were included in this study. Adequate cellularity was tentatively defined as at least 3 groups of atypical epithelial cells (>10 cells in each group) and single atypical cells. For benign breast lesions, a minimum of 3 groups of ductal cells and apocrine cells in the cell block was considered adequate. Most cell blocks contained more than 5 groups of atypical epithelial cells or benign ductal cells. The staining intensity (weak or strong) and distribution (negative [<5% of tumor cells stained], 1+ [5%-25%], 2+ [26%-50%], 3+ [51%-75%], or 4+ [>75%]) were recorded. Two surgical pathologists (F.L. and H.L.) independently evaluated the immunostained slides. Follow-up surgical biopsy or resection confirmed the diagnosis for each malignant case.

Results

GATA3 Expression on Tumors on TMA Sections

The positive staining results and the total number of cases for each entity are summarized in Table 1. Sixty-two of 72 urothelial carcinomas (86%), 90 of 99 ductal carcinomas (91%) and 48 of 48 lobular carcinomas (100%) tested positive for GATA3. Diffuse (3+ or 4+) and strong nuclear staining was noted in 65% of urothelial carcinomas, 84% of ductal carcinomas, and 77% of lobular carcinomas. Two of 96 endometrioid-type endometrial carcinoma cases tested positive for GATA3. All others cases in this study tested negative for GATA3. Representative photomicrographs of GATA3 in breast carcinoma are shown in **Image 1** and in urothelial carcinoma in **Image 2**. The detailed staining results for breast carcinoma and urothelial carcinoma are summarized in **Table 2**.

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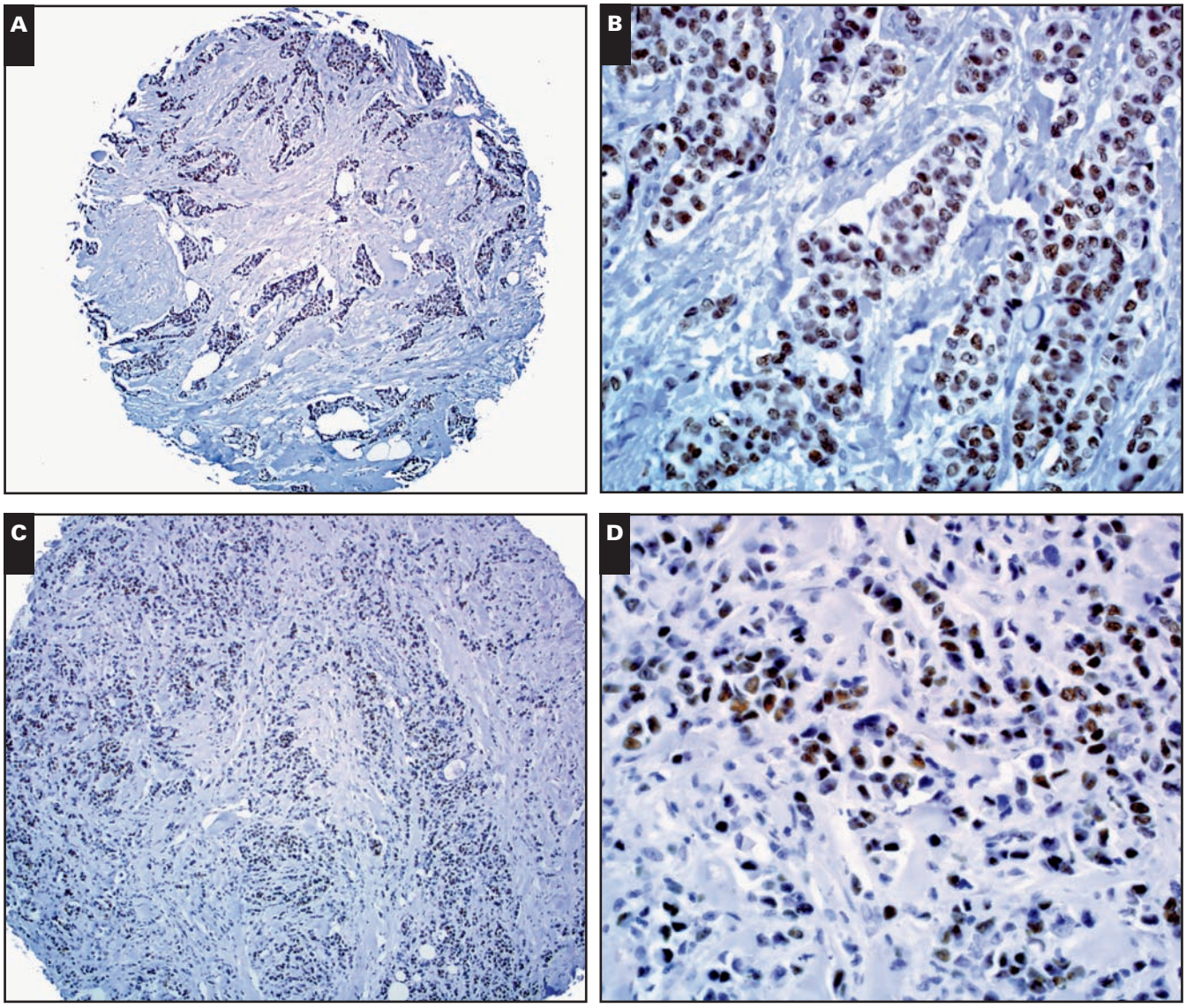

Image 1 Positive nuclear staining for GATA3 in breast ductal carcinoma at low (**A**, ×40) and high (**B**, ×400) magnification and positive staining for GATA3 in breast lobular carcinoma at low (**C**, ×40) and high (**D**, ×400) magnification.

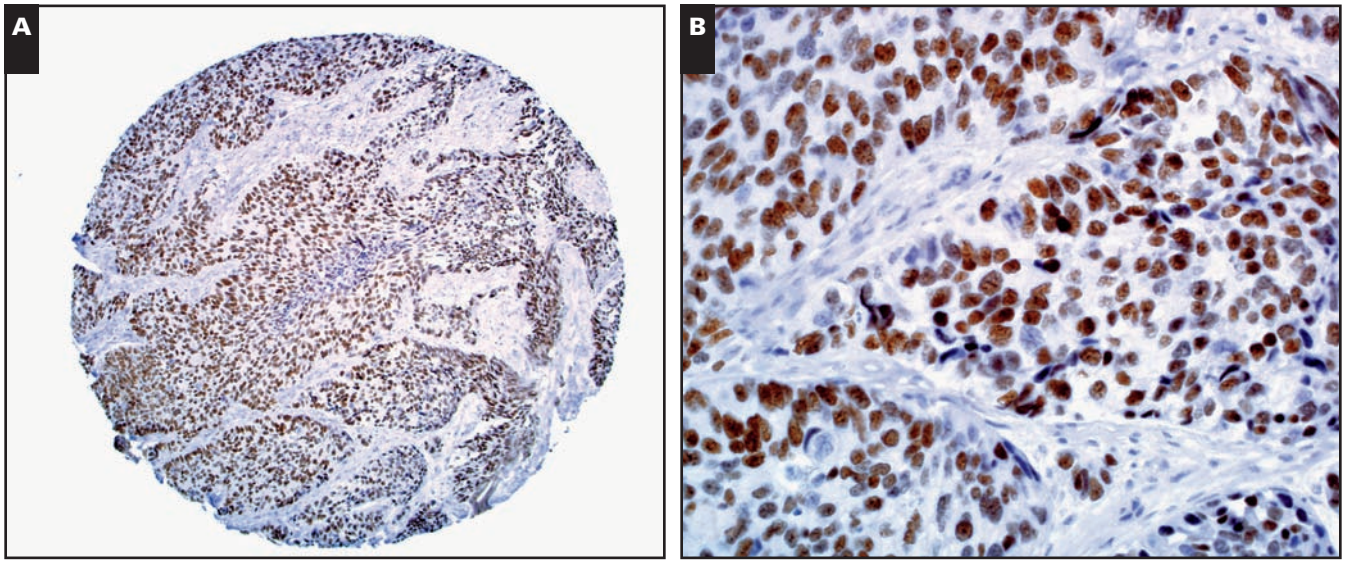

Image 2 Positive staining for GATA3 in urothelial carcinoma at low (**A**, ×40) and high (**B**, ×400) magnification.

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Table 2 GATA3 Immunostaining Results on Breast and Urothelial Carcinomas

Diagnosis	Negative	1+	2+	3+	4+	Total No. (%) of Positive Cases
DCA grade I (n = 25)	0	0	0	5	20	25 (100)
DCA grade II (n = 46)	5	0	2	5	34	41 (89)
DCA grade III (n = 28)	4	0	5, W	5	14	24 (86)
CLCA (n = 48)	0	0	0	11	37	48 (100)
UC (n = 72)	10	7, W	10	8	37	62 (86)

CLCA, classic lobular carcinoma; DCA, ductal carcinoma; UC, invasive urothelial carcinoma; W, weak nuclear staining.

GATA3 Expression on Breast and Bladder Biopsy Specimens

In addition to the invasive ductal and lobular carcinomas of the breast being tested on TMA section, GATA3 expression in DCIS, ductal hyperplasia, fibroadenoma, and normal breast tissue was also performed, and the results showed nuclear positivity in 100%, 78%, 100%, and 50%, respectively. Only 50% of normal breast tissues tested positive for GATA3, and the staining was generally focal (1+-2+) and weak. In the ductal hyperplasia group, 2 cases of apocrine metaplasia tested negative for GATA3. Similarly, 2 cases of apocrine DCIS had only focal (1+) and weak nuclear staining for GATA3. All 10 urothelial mucosa biopsy specimens tested positive for GATA3. Representative photomicrographs of GATA3 expression in DCIS, ductal hyperplasia, fibroadenoma, and normal breast tissue are shown in **Image 3**. The results are summarized in **Table 3**.

GATA3 Expression on Normal Tissues

Of the 310 cases of normal tissues tested, 20 (100%) of 20 cases of distal renal tubules tested positive for GATA3. Proximal renal tubules, glomeruli, and interstitium tested negative. All other normal tissues (lung, parotid gland, stomach, duodenum, small intestines, colon, pancreas, liver, thyroid, prostate, seminal vesicles, ovary, and endometrium) tested in the study were negative for GATA3 (normal breast and bladder tissues were not included here). The results are given in Table 3.

GATA3 Expression on FNAB Specimens of the Breast on Cell Blocks

Strong and diffuse staining (3+ or 4+) for GATA3 was seen in 88% of group 1, 82% of group 2, and 36% of group 3. Two cases in group 1 (primary breast ductal carcinoma) tested negative for GATA3. One was triple-negative (estrogen receptor [ER], progesterone receptor [PR], and Her2/neu negative), high-grade ductal carcinoma, and the other was high-grade ductal carcinoma with apocrine features. Four cases in group 2 tested negative for GATA3; 2 were ductal carcinoma with high nuclear grade, and the other 2 were ductal carcinoma with intermediate nuclear grade. All 9 cases of apocrine metaplasia in group 3 tested negative for GATA3. The results are summarized in **Table 4**. Representative photomicrographs of the expression of GATA3 on cell block sections with H&E stain are shown in **Image 4**.

Discussion

The primary focus of the current study is to determine whether GATA3 is an organ-specific biomarker, its potential utility in a tumor of unknown origin on surgical and FNAB specimens, and its expression in normal tissues. This study demonstrates that GATA3 expression is primarily limited to breast carcinoma and urothelial carcinoma and only rarely seen in tumors from other organs, such as endometrial adenocarcinoma (endometrioid type), which confirms the previous report by Higgins et al[15] and broadens the potential diagnostic utility of GATA3, especially for a tumor of unknown origin. In the study by Higgins and coworkers, all 4 cases of breast carcinoma tested positive for GATA3. Our current study with a large series of cases demonstrates a similar finding, with GATA3 expression in 100% of lobular carcinomas and 91% of ductal carcinomas (combined grade I, II, and III ductal carcinomas). The expression of GATA3 was observed in 100% of grade I ductal carcinomas and was reduced to 86% in grade III ductal carcinomas, which shows the downregulation of GATA3 expression with progression of tumor grade. Furthermore, GATA3 expression appears to be even lower in luminal B subtype breast carcinoma. In our study with a limited number of cases (F. Lin, MD, PhD, unpublished data, October 2011), only 4 of 8 cases of luminal B subtype breast carcinoma tested focally positive for GATA3. All 8 cases were triple-negative breast carcinomas (ER, PR, and Her2/neu negative).

It has been well documented that there is a good correlation between ER and PR status and GATA3 expression (ie, a strong ER positivity with a high level of GATA3 expression).[3,18,19] Our current data on ER, PR, and Her2/neu are incomplete; therefore, a complete correlation study of the expression of GATA3 and ER, PR, and Her2/neu status cannot be performed at this time. However, preliminary results indicated that (1) all grade I ductal carcinomas tested positive for ER and GATA3 as expected, (2) 8 cases of grade

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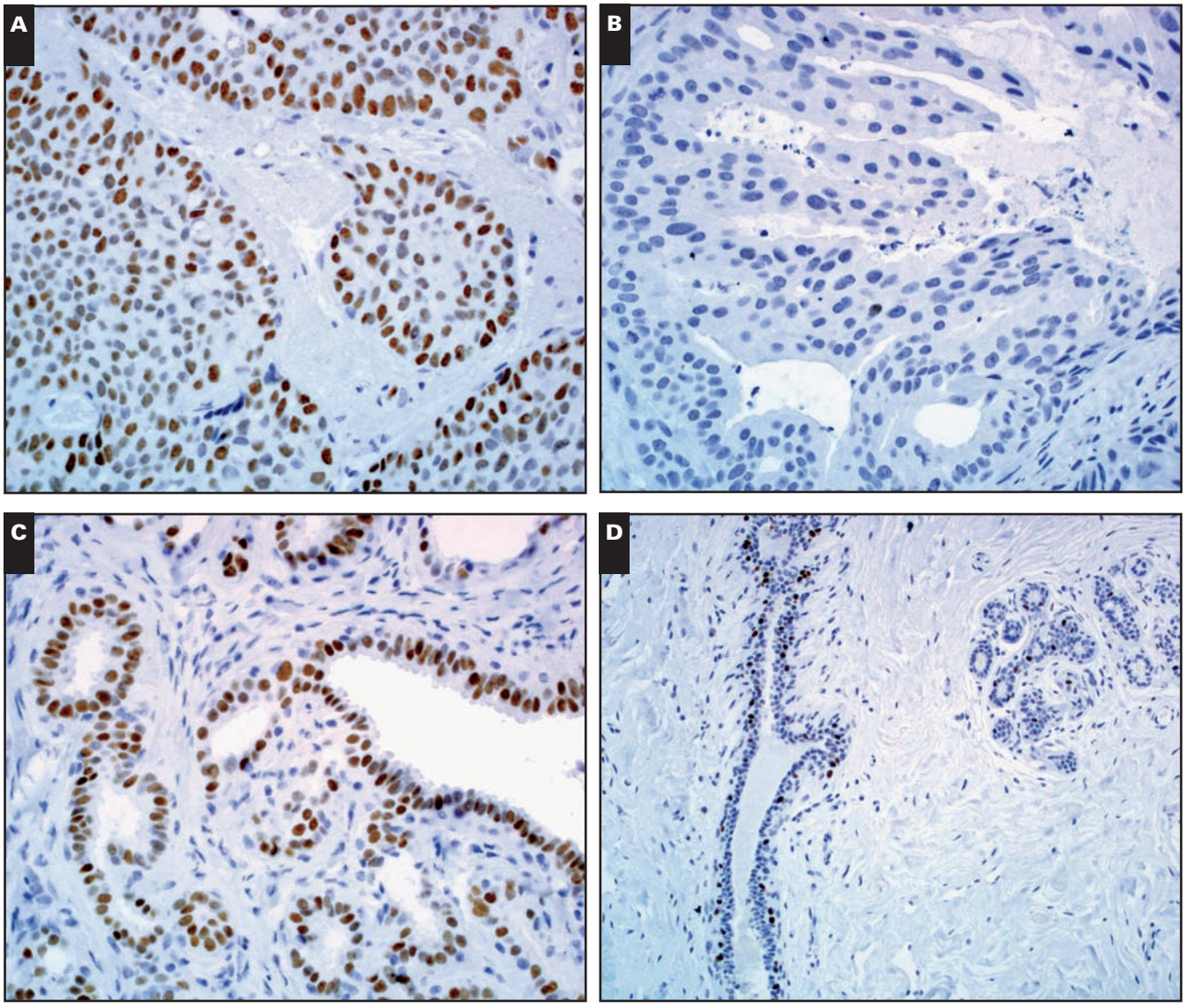

Image 3 GATA3 expression in breast core biopsy specimens with diffuse and strong staining in ductal carcinoma in situ (DCIS) (**A**, ×400), only focal and weak staining in apocrine DCIS (**B**, ×400), strong staining in ductal hyperplasia (**C**, ×400), and focal staining in normal breast ducts and lobules (**D**, ×200).

Table 3 GATA3 Immunostaining Results on DCIS, DH, FA, NBT, and BUM

GATA3	Negative	1+	2+	3+	4+	Total No. (%) of Positive Cases
DCIS (n = 9)	0	2	0	3	4	9 (100)
DH (n = 9)	2	0	0	3	4	7 (78)
FA (n = 10)	0	1	4	4	1	10 (100)
NBT (n = 10)	5	3	2	0	0	5 (50)
BUM (n = 10)	0	0	3	4	2	10 (100)

BUM, benign urothelial mucosa; DCIS, ductal carcinoma in situ; DH, ductal hyperplasia; FA, fibroadenoma; NT, normal breast tissue.

II ductal carcinoma tested negative for ER but expressed GATA3 with an overall sensitivity of 89% vs 73% for ER, (3) 8 cases of grade III ductal carcinoma tested negative for ER but positive for GATA3, and (4) 3 cases of nuclear grade II lobular carcinoma tested negative for ER, and 4 cases of nuclear grade III lobular carcinoma were only weakly positive for ER, whereas all lobular carcinoma cases tested positive for GATA3. Subtypes of breast carcinoma, such as colloid, medullary, apocrine, and pleomorphic lobular, were not tested in this study.

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The sensitivity of GATA3 expression in urothelial carcinoma is higher in the current study (86% vs 67%) than in the study by Higgins et al.[15] The explanation for the discrepancy between these 2 studies can be difficult for the following reasons. All 72 cases in our study are invasive, high-grade urothelial carcinoma because we are more interested in knowing when a urothelial carcinoma presents as an unknown primary tumor; in contrast, in the study by Higgins and coworkers,[15] 208 cases were invasive, and 113 cases were noninvasive. We have routinely used several antigen retrieval methods to test a new antibody, with multiple different dilutions for the primary antibody as previously described.[26] These methods include

Table 4 GATA3 Immunostaining Results on Fine-Needle Aspiration Biopsy Specimens

GATA3	Negative	1+	2+	3+	4+	Total No. (%) of Positive Cases
BDCA (n = 17)	2	2	3	7	3	15 (88)
MDCA (n = 22)	4	4	4	5	5	18 (82)
Benign breast lesions (n = 14)	9	1	2	2	0	5 (36)

BDCA, breast ductal carcinoma; MDCA, metastatic breast ductal carcinoma.

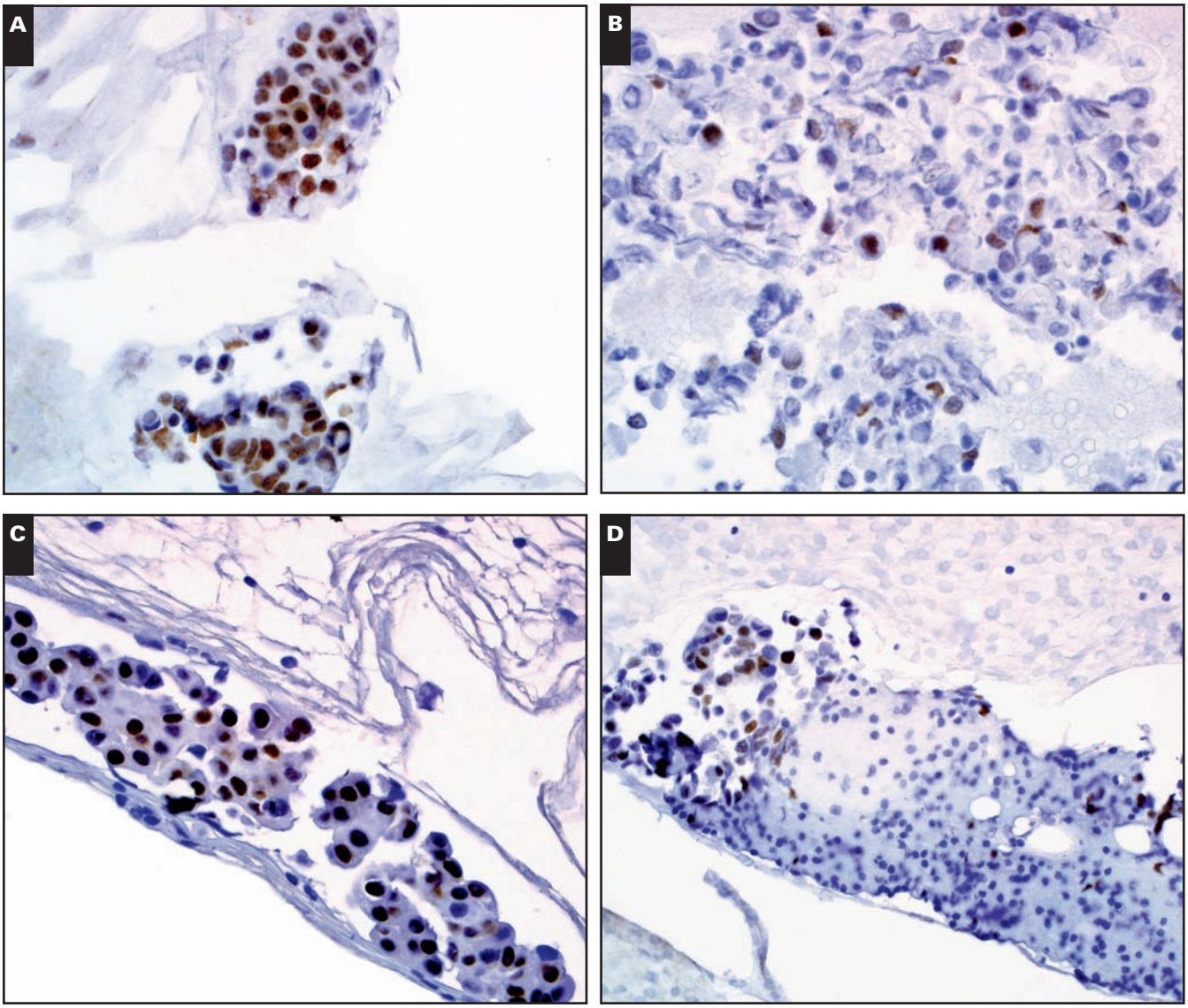

Image 4 GATA3 expression on breast fine-needle aspiration biopsy specimens on cell blocks with strong and diffuse staining in ductal carcinoma (**A**, ×600), focal staining in ductal carcinoma (**B**, ×600), diffuse and strong staining in metastatic breast carcinoma (**C**, ×600), and focal staining in metastatic breast carcinoma (**D**, ×600).

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EDTA, Tris buffer, high pH, citrate buffer, proteinase K, and no retrieval. For GATA3, we noticed that the antigen retrieval method and primary antibody dilution have a significant effect on the staining intensity and distribution. Following the comparison of different antigen retrieval methods, 3 different dilutions for GATA3 (1:25, 1:50, and 1:100), and different incubation times for a primary antibody (30 minutes, 45 minutes, and 60 minutes), we concluded that the optimal working condition for GATA3 with the DAKO system is EDTA antigen retrieval, 1:25 dilution, and 45 minutes of incubation time. In the study by Higgins et al,[15] the primary antibody was the same one (same clone from the same vendor) used in this study; however, the dilution was 1:50, and the antigen retrieval and incubation time were not specified in the article. Perhaps the higher sensitivity in our study is attributable to the higher primary antibody concentration, better antigen retrieval method, and longer incubation time. No major technical issues were encountered in our study. Staining background was generally absent, and the stain was easily reproducible.

The expression of GATA3 was observed in 100% of DCIS, 50% of normal breast tissue, and 78% of ductal hyperplasias. Therefore, GATA3 has a limited value in the distinction of breast carcinoma from benign breast tissue or ductal hyperplasia. Similarly, because GATA3 was expressed in normal urothelium,[15] it has little utility in differentiating urothelial carcinoma from benign or reactive urothelium. GATA3 expression was also observed in normal renal distal tubules but absent in proximal renal tubules, glomeruli, and stromal cells; however, the papillary renal cell carcinomas and clear cell renal cell carcinomas examined in this study tested negative for GATA3.

The expression of GATA3 on FNAB specimens of the breast on cell blocks was slightly lower than that of surgical specimens. The sensitivities for ductal carcinoma and metastatic ductal carcinoma were 88% and 82%, respectively. For FNAB of benign breast lesions, the expression of GATA3 was seen in 36% of cases. All 9 cases with apocrine metaplasia tested negative for GATA3. Even though the staining signal was weaker in benign breast lesions than in primary and metastatic carcinomas, the utility of using GATA3 to distinguish breast carcinoma from benign breast lesion or normal breast tissue is limited.

When working on a tumor of unknown origin with an immunostaining profile of CK7+/CK20−/ER+, the primary sites being considered should include the breast, ovary, and uterus, especially breast ductal and lobular carcinomas, ovarian serous carcinoma, and endometrial adenocarcinoma. PAX8 is a recently described nuclear marker, which is usually expressed in müllerian tumors, renal cell carcinomas, and thyroid carcinomas of follicular cell origin but negative in breast carcinomas.[27-30] Our unpublished data demonstrated that PAX8 was positive in 58 of 58 cases of endometrial adenocarcinoma (endometrioid type) and 41 of 41 cases of ovarian serous carcinoma and negative in 98 of 98 breast ductal carcinomas. WT-1 was also a sensitive positive nuclear marker for ovarian serous carcinoma. Our unpublished data revealed that 88% of ovarian serous carcinomas (36 of 41 cases) tested positive for WT-1, but 0 of 98 breast carcinomas tested positive. Therefore, a panel of immunostaining markers, including PAX8 and GATA3, should be considered when differentiating a breast primary tumor from an endometrial primary tumor. Similarly, a panel of immunomarkers, including GATA3, PAX8, and WT-1, should be considered when the differential diagnosis includes breast carcinoma and ovarian serous carcinoma.

Urothelial carcinoma frequently presents as a tumor of unknown origin, and there are no highly sensitive and specific markers for it. Nearly all urothelial carcinomas are positive for p63 and CK7, and most are also positive for high-molecular-weight cytokeratin (CK903 and CK5/6). Approximately 50% of cases are also positive for CK20. Uroplakin III has been described as a specific marker for urothelial carcinoma; however, the sensitivity is relatively low (approximately 50%).[31,32] Our experience showed a poor reproducibility of uroplakin III. In addition, S100P and thrombomodulin are relatively sensitive markers,[14,15,31] but the specificity is low. Both p63 and high-molecular-weight cytokeratin are positive in squamous cell carcinomas. To our knowledge, GATA3 is the most sensitive and specific marker for urothelial carcinomas.

In our limited experience, GATA3 has been proven to be a useful marker for tumors of unknown origin. Recently, we encountered 2 cases of urothelial carcinoma with metastasis to the rectum. Both cases tested positive for GATA3. One case was only focally positive for CK7 and p63 with squamous differentiation, and the other was positive for CK20 and p63. Many high-grade urothelial carcinomas may show squamous differentiation. Because squamous cell carcinoma and urothelial carcinoma are positive for high-molecular-weight cytokeratin and p63, GATA3 can be important in distinguishing between these 2 entities. We also had a case of breast carcinoma with pulmonary hilar metastasis. The tumor tested negative for ER, PR, GCDFP-15, and mammaglobin but positive for GATA3, which confirmed the clinical suspicion of a breast primary tumor.

In summary, on the basis of our current study, we recommend that GATA3 be routinely included in a panel of immunomarkers on surgical and FNAB specimens when working with a tumor of unknown origin, especially when either a breast carcinoma or urothelial carcinoma is being considered.

From Geisinger Medical Center, Danville, PA.

Address reprint requests to Dr Lin: Department of Laboratory Medicine, MC 01-31, Geisinger Medical Center, 100 N Academy Ave, Danville, PA 17822; Flin1@geisinger.edu.

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References

1. Burch JB. Regulation of GATA gene expression during vertebrate development. *Semin Cell Dev Biol*. 2005;16:71-81.
2. Zheng R, Blobel GA. GATA transcription factors and cancer. *Genes Cancer*. 2010;1:1178-1188.
3. Yoon NK, Maresh EL, Shen D, et al. Higher levels of GATA3 predict better survival in women with breast cancer. *Hum Pathol*. 2010;41:1794-1801.
4. Kouros-Mehr H, Slorach EM, Sternlicht MD, et al. GATA-3 maintains the differentiation of the luminal cell fate in the mammary gland. *Cell*. 2006;127:1041-1055
5. Asselin-Labat ML, Sutherland KD, Barker H, et al. Gata-3 is an essential regulator of mammary-gland morphogenesis and luminal-cell differentiation. *Nat Cell Biol*. 2007;9:201-209.
6. Ting CN, Olson MC, Barton KP, et al. Transcription factor GATA-3 is required for development of the T-cell lineage. *Nature*. 1996;384:474-478.
7. Naito T, Tanaka H, Naoe Y, et al. Transcriptional control of T-cell development. *Int Immunol*. 2011;23:661-668.
8. Pai SY, Truitt ML, Ting CN, et al. Critical roles for transcription factor GATA-3 in thymocyte development. *Immunity*. 2003;19:863-875.
9. Hendriks RW, Nawijn MC, Engel JD, et al. Expression of the transcription factor GATA-3 is required for the development of the earliest T cell progenitors and correlates with stages of cellular proliferation in the thymus. *Eur J Immunol*. 1999;29:1912-1918.
10. Tong Q, Dalgin G, Xu H, et al. Function of GATA transcription factors in preadipocyte-adipocyte transition. *Science*. 2000;290:134-138.
11. Grote D, Souabni A, Busslinger M, et al. Pax 2/8-regulated Gata 3 expression is necessary for morphogenesis and guidance of the nephric duct in the developing kidney. *Development*. 2006;133:53-61.
12. Tsarovina K, Pattyn A, Stubbusch J, et al. Essential role of Gata transcription factors in sympathetic neuron development. *Development*. 2004;131:4775-4786.
13. Kaufman CK, Zhou P, Pasolli HA, et al. GATA-3: an unexpected regulator of cell lineage determination in skin. *Genes Dev*. 2003;17:2108-2122.
14. Esheba GE, Longacre TA, Atkins KA, et al. Expression of the urothelial differentiation markers GATA3 and placental S100 (S100P) in female genital tract transitional cell proliferations. *Am J Surg Pathol*. 2009;33:347-353.
15. Higgins JP, Kaygusuz G, Wang L, et al. Placental S100 (S100P) and GATA3: markers for transitional epithelium and urothelial carcinoma discovered by complementary DNA microarray. *Am J Surg Pathol*. 2007;31:673-680.
16. Raspollini MR, Sardi I, Giunti L, et al. Plasmacytoid urothelial carcinoma of the urinary bladder: clinicopathologic, immunohistochemical, ultrastructural, and molecular analysis of a case series. *Hum Pathol*. 2011;42:1149-1158.
17. Davidson B, Stavnes HT, Holth A, et al. Gene expression signatures differentiate ovarian/peritoneal serous carcinoma from breast carcinoma in effusions. *J Cell Mol Med*. 2011;15:535-544.
18. Hoch RV, Thompson DA, Baker RJ, et al. GATA-3 is expressed in association with estrogen receptor in breast cancer. *Int J Cancer*. 1999;84:122-128.
19. Ciocca V, Daskalakis C, Ciocca RM, et al. The significance of GATA3 expression in breast cancer: a 10-year follow-up study. *Hum Pathol*. 2009;40:489-495.
20. Jacquemier J, Charafe-Jauffret E, Monville F, et al. Association of GATA3, P53, Ki67 status and vascular peritumoral invasion are strongly prognostic in luminal breast cancer. *Breast Cancer Res*. 2009;11:R23.
21. Lin F, Zhang PL, Yang XJ, et al. Human kidney injury molecule-1 (hKIM-1): a useful immunohistochemical marker for diagnosing renal cell carcinoma and ovarian clear cell carcinoma. *Am J Surg Pathol*. 2007;31:371-381.
22. Wilkerson ML, Powell E. Tissue microarray. In: Lin F, Prichard J, Liu H, et al, eds. *Handbook of Practical Immunohistochemistry: Frequently Asked Questions*. New York, NY: Springer; 2011:45-54.
23. Lin F, Shi J, Liu H, et al. Diagnostic utility of S100P and von Hippel-Lindau gene product (pVHL) in pancreatic adenocarcinoma—with implication of their roles in early tumorigenesis. *Am J Surg Pathol*. 2008;32:78-91.
24. Lin F, Shi J, Liu H, et al. Immunohistochemical detection of the von Hippel-Lindau gene product (pVHL) in human tissues and tumors: a diagnostic marker for metastatic renal cell carcinoma and clear cell carcinoma of the ovary and uterus. *Am J Clin Pathol*. 2008;129:592-605.
25. Liu H, Shi J, Wilkerson M, et al. Immunohistochemical detection of p16INKa in liquid-based cytology specimens on cell block sections. *Cancer*. 2007;111:74-82.
26. Lin F, Shi J. Technique and troubleshooting of antibody testing. In: Lin F, Prichard J, Liu H, et al, eds. *Handbook of Practical Immunohistochemistry: Frequently Asked Questions*. New York, NY: Springer; 2011:17-22.
27. Nonaka D, Chiriboga L, Soslow RA. Expression of pax8 as a useful marker in distinguishing ovarian carcinomas from mammary carcinomas. *Am J Surg Pathol*. 2008;32:1566-1571.
28. Lacroix L, Mian C, Barrier T, et al. PAX8 and peroxisome proliferator-activated receptor gamma 1 gene expression status in benign and malignant thyroid tissues. *Eur J Endocrinol*. 2004;151:367-374.
29. Laury AR, Hornick JL, Perets R, et al. PAX8 reliably distinguishes ovarian serous tumors from malignant mesothelioma. *Am J Surg Pathol*. 2010;34:627-635.
30. Hu Y, Hartmann A, Stoehr C, et al. PAX8 is expressed in the majority of renal epithelial neoplasms: an immunohistochemical study of 223 cases using a mouse monoclonal antibody. *J Clin Pathol*. 2012;65:254-256.
31. Parker DC, Folpe AL, Bell J, et al. Potential utility of uroplakin III, thrombomodulin, high molecular weight cytokeratin, and cytokeratin 20 in noninvasive, invasive, and metastatic urothelial (transitional cell) carcinomas. *Am J Surg Pathol*. 2003;27:1-10.
32. Kaufmann O, Volmerig J, Dietel M. Uroplakin III is a highly specific and moderately sensitive immunohistochemical marker for primary and metastatic urothelial carcinomas. *Am J Clin Pathol*. 2000;113:683-687.

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