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Value of PAX8, PAX2, napsin A, carbonic anhydrase IX, and claudin-4 immunostaining in distinguishing pleural epithelioid mesothelioma from metastatic renal cell carcinoma

Nelson G Ordóñez

Department of Pathology, The University of Texas MD Anderson Cancer Center, Houston, TX, USA

Both mesotheliomas and renal cell carcinomas can present a wide variety of cytomorphologic features and histologic patterns. Because of this, renal cell carcinomas metastatic to the pleura and lung can be confused with mesotheliomas. Recently, a variety of positive carcinoma markers, including kidney-associated markers, have become available. The aim of this study is to investigate the value of some of these markers, specifically PAX8, PAX2, napsin A, carbonic anhydrase IX, and claudin-4, for assisting in distinguishing pleural epithelioid mesotheliomas from metastatic renal cell carcinomas. To do so, a total of 40 pleural epithelioid mesotheliomas and 55 renal cell carcinomas (33 clear cell, 10 papillary, and 12 chromophobe) were investigated. In all, 91% of the renal cell carcinomas expressed claudin-4, 89% PAX8, 60% PAX2, 71% carbonic anhydrase IX, and 29% napsin A. All of the mesotheliomas were positive for carbonic anhydrase IX and were negative for all of the other markers. On the basis of these results, it is concluded that claudin-4 and PAX8 have a higher sensitivity and specificity for assisting in discriminating between pleural epithelioid mesotheliomas and renal cell carcinomas when compared with all of the other positive carcinoma markers that are, at present, recommended to be included in the immunohistochemical panels used in this differential diagnosis. Even though PAX2 and napsin A are highly specific, because of their low sensitivity, they have only a limited value. Carbonic anhydrase IX is not useful.
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Characteristically, mesotheliomas can present a diverse array of cytomorphologic features and grow in a wide variety of histologic patterns. Because of this, they can be easily confused with a number of other malignancies that can involve the serosal membranes. Although the majority of published studies investigating the role of immunohistochemistry in the diagnosis of these tumors have focused primarily on the distinction between pleural epithelioid mesotheliomas and lung adenocarcinomas,[1–7] only a few have looked into discriminating between pleural epithelioid mesotheliomas and renal cell carcinomas.[8–10] Renal cell carcinomas, like mesotheliomas, can also exhibit numerous histologic patterns, some of which can be confused with some morphologic variants of mesothelioma.[11,12] In addition, renal cell carcinomas can also mimic mesotheliomas, both clinically and radiologically, as they frequently metastasize to the lung and pleura, occasionally encase the lung, and sometimes occur in the absence of a known renal tumor or urologic symptoms.[13–17] Furthermore, cases of renal cell carcinomas have been reported in individuals exposed to asbestos and examples of concomitant renal cell carcinoma and mesothelioma have been documented in the literature.[18,19] Finally, distant visceral metastases, which can sometimes occur as a single mass, are not rare in pleural mesotheliomas, and one of the most frequently involved organs is

Correspondence: Dr NG Ordóñez, MD, Department of Pathology, The University of Texas MD Anderson Cancer Center, 1515 Holcombe Boulevard, Houston, TX 77030, USA.
E-mail: nordonez@mdanderson.org
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the kidney.[20] In recent years, a wide variety of immunohistochemical markers, including kidney-associated markers that can assist in the diagnosis and classification of renal epithelial tumors, have become available.[21] Little information exists, however, regarding the expression of these markers in mesotheliomas or on their value in assisting in discriminating these tumors from renal cell carcinomas. The purpose of this study is to determine the practical utility of some of these markers, specifically PAX8, PAX2, napsin A, carbonic anhydrase IX (CA IX), and claudin-4 (CL-4), for assisting in distinguishing between pleural epithelioid mesotheliomas and metastatic renal cell carcinomas and to compare them with other renal cell carcinoma markers that have previously been recommended as being useful in facilitating this differential diagnosis.

Materials and methods

The material used in this study was obtained from the files of the Department of Pathology at The University of Texas MD Anderson Cancer Center. It consisted of 40 pleural epithelioid mesotheliomas and 55 renal cell carcinomas. In all of the mesothelioma cases, the diagnosis was confirmed by the use of histologic and immunohistochemical criteria combined with clinical and radiologic information. Of the 55 renal cell carcinomas, 33 were clear cell (22 metastatic, 11 primary), 10 papillary, and 12 chromophobe renal cell carcinomas. Immunohistochemical studies were performed on 5-μm-thick, formalin-fixed, paraffin-embedded tissue sections using the polymeric biotin-free horseradish peroxidase method on a Leica Microsystems Bond Max Stainer (Bannockburn, IL, USA). The primary antibodies are listed in Table 1. In brief, slides were deparaffinized and hydrated, followed by heat-induced antigen retrieval in which a citrate buffer solution, pH 6.0, was used. Incubation with the primary antibody was followed by development of the immunostaining with 3,3′-diaminobenzidine. The secondary antibody and detection was applied as per the manufacturer's instructions. To evaluate the specificity of the immunoreaction, known positive and negative tissues were used as controls. The immunostaining was graded on a sliding scale of 1+ to 4+ according to the percentage of reactive cells (1+, 1–25%; 2+, 26–50%; 3+, 51–75%; and 4+, >75%).

Results

The immunohistochemical results are summarized in Table 2.

PAX8

Of the 55 renal cell carcinomas, 49 (89%) demonstrated PAX8 nuclear positivity, including 31 (94%) of 33 clear cell, 10 (100%) of 10 papillary, and 9 (75%) of 12 chromophobe (Figures 1a–c). The reaction was strong and diffuse (3+ or 4+) in the 31 clear cell, 8 of the papillary, and 4 of the chromophobe renal cell carcinomas, and focal (1+ or 2+) in the remaining cases. All of the mesotheliomas were PAX8 negative.

PAX2

In all, 33 (60%) of the 55 renal cell carcinomas exhibited PAX2 positivity. In total, 23 of the positive cases were clear cell, 6 papillary, and 4 chromophobe. The staining in these cases was strong (3+ or 4+) in 7 of the clear cell, 3 of the papillary, and 2 of the chromophobe renal cell carcinomas (Figure 1d), whereas in the remaining cases, it was focal (1+ or 2+). None of the mesotheliomas expressed PAX2.

Napsin A

Only 8 (24%) of the 33 clear cell and 7 (70%) of the 10 papillary renal cell carcinomas, but none of the

Table 2 Immunohistochemical results in renal cell carcinoma

	n	*PAX8 + (%)*	*PAX2 + (%)*	*Napsin A + (%)*	*CA IX + (%)*	*Claudin-4 + (%)*
Clear cell	33	31 (94)	23 (70)	8 (24)	32 (97)	28 (85)
Papillary	10	10 (100)	6 (60)	7 (70)	7 (70)	10 (100)
Chromophobe	12	9 (75)	4 (33)	0 (0)	0 (0)	12 (100)
Total	55	49 (89)	33 (60)	15 (28)	39 (71)	50 (91)

Table 1 Antibodies used in this study

Marker	*Source*	*Type*	*Dilution*	*Antigen retrieval*
Carbonic anhydrase IX	Novocastra (Buffalo Grove, IL, USA)	TH22 MAb	1:100	Yes (citrate)
Claudin-4	Invitrogen (Camarillo, CA, USA)	3E2C1 MAb	1:250	Yes (citrate)
Napsin A	Novocastra	IP64 MAb	1:300	Yes (citrate)
PAX2	Invitrogen	Z-RX2 PAb	1:25	Yes (citrate)
PAX8	ProteinTech Group (Chicago, IL, USA)	PAb (rabbit)	1:100	Yes (citrate)

Abbreviations: MAb, monoclonal antibody; PAb, polyclonal antibody.

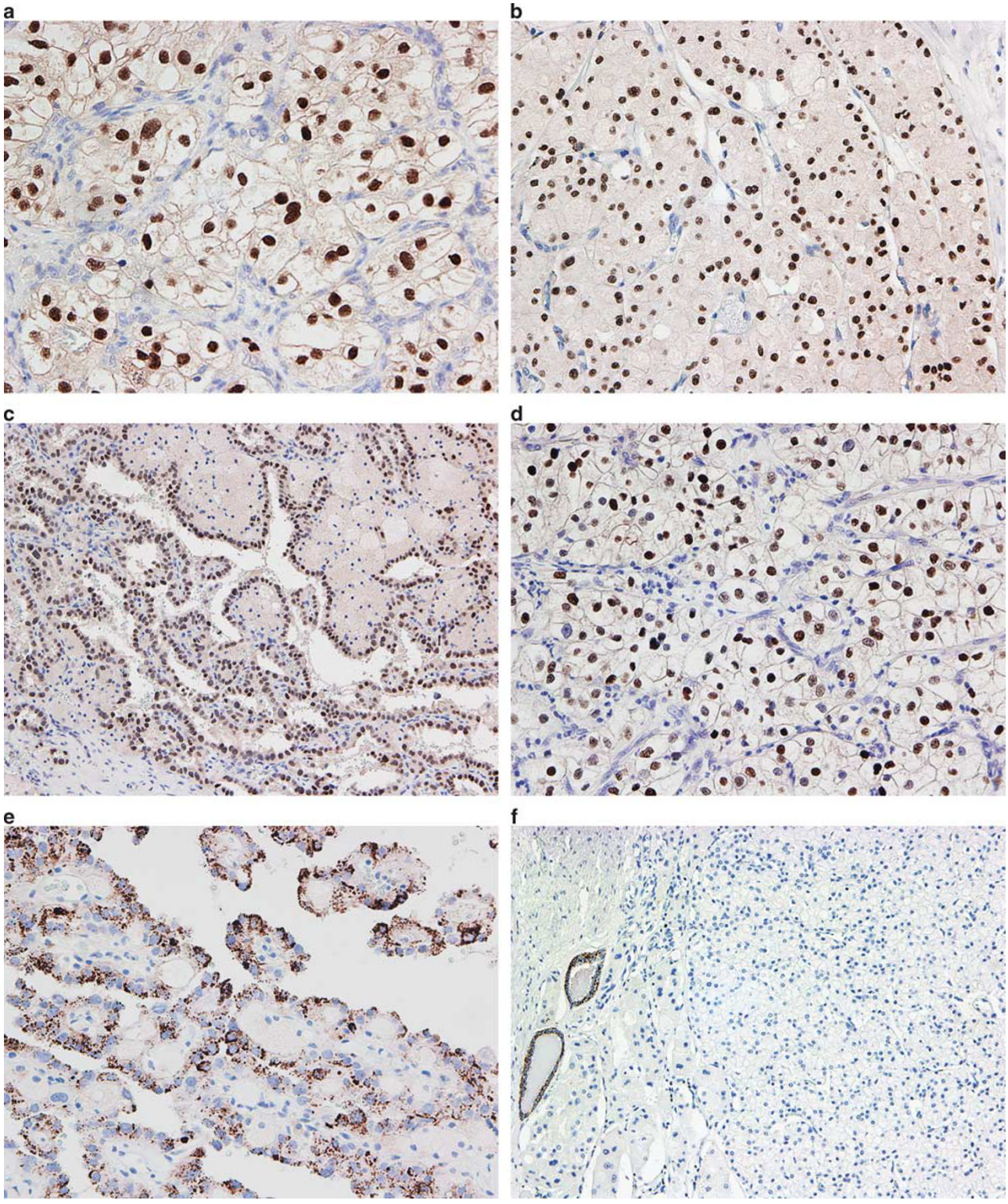

Figure 1 (**a**) Clear cell carcinoma exhibiting strong nuclear positivity for PAX8. (**b**) Chromophobe renal cell carcinoma displaying nuclear positivity for PAX8. (**c**) Papillary renal cell carcinoma showing nuclear PAX8 reactivity. (**d**) Clear cell renal cell carcinoma exhibiting PAX2 nuclear positivity. (**e**) Papillary renal cell carcinoma showing cytoplasmic positivity for napsin A. (**f**) Chromophobe carcinoma demonstrating a lack of napsin A expression in the neoplastic cells. Strong positivity for this marker is seen in two atrophic non-neoplastic renal tubules (left).

12 chromophobe renal cell carcinomas, were napsin A positive (Figures 1e and f). The staining in these cases was granular and cytoplasmic, and it was strong (3+ or 4+) in 4 of the 8 clear cell and in all 7 of the positive papillary renal cell carcinomas. In the remaining 4 clear cell renal cell carcinomas, the

reactivity was focal (1+ or 2+). No immunoreactivity was seen in any of the mesotheliomas.

Carbonic Anhydrase IX

CA IX expression was demonstrated in 32 (97%) of 33 clear cell and 7 (70%) of 10 papillary, but in none of the 12 chromophobe renal cell carcinomas. The staining in these cases occurred along the cell membrane and was strong (3+ or 4+) in the 32 clear cell and in 3 of the papillary renal cell carcinomas (Figure 2a). All 40 of the epithelioid mesotheliomas expressed CA IX (Figure 2b). In 28 of the cases, the staining was strong and diffuse (3+ or 4+), whereas it was focal (1+ or 2+) in the remaining cases.

Claudin-4

In all, 51 (91%) of the 55 renal cell carcinomas exhibited CL-4 expression, including 28 (85%) of 33 clear cell, 10 of 10 papillary, and 12 of 12 chromophobe renal cell carcinomas. The staining in these cases occurred along the cell membrane in a continuous or punctuated pattern, and it was strong and diffuse (3+ or 4+) in the majority of clear cell (20 of the cases), and in all of the papillary and chromophobe renal cell carcinomas (Figures 2c–f). None of the mesotheliomas showed CL-4 positivity.

Discussion

Because an absolutely sensitive and specific marker for either mesothelioma or renal cell carcinoma has not yet been identified, establishing the differential diagnosis between these two malignancies largely depends on the use of immunohistochemical panels composed of positive mesothelioma markers (ie, those that are frequently expressed in mesotheliomas, but not in carcinomas) and positive carcinoma markers (ie, those that are frequently expressed in carcinomas, but not in mesotheliomas). To my knowledge, only three studies have been published comparing the value of a relatively large number of immunohistochemical markers for assisting in discriminating between mesotheliomas and metastatic renal cell carcinomas.[8–10] The first of these studies was conducted by Attanoos *et al*[8] in 1995, who evaluated the diagnostic utility of CD15 (Leu-M1), Ber-EP4, Tamm–Horsfall protein, and thrombomodulin for assisting in the differential diagnosis between renal cell carcinomas and mesotheliomas.[8] The conclusion of that investigation was that only thrombomodulin and CD15 were useful in discriminating between renal cell carcinomas and epithelioid mesotheliomas; however, these two markers had no value in distinguishing between sarcomatoid mesotheliomas and sarcomatoid renal cell carcinomas.[8] The second study was conducted by Osborn *et al*[9] in 2002, who investigated the value of calretinin, keratin 5/6, thrombomodulin, CEA, Ber-EP4, and BCA225 immunostaining in discriminating metastatic renal cell carcinomas from mesotheliomas. Based on the results of that study, the authors concluded that calretinin, keratin 5/6, and Ber-EP4 were the most useful markers for assisting in this differential diagnosis. In the third study, which was by this author in 2004, the expression of a large number of positive mesothelioma markers (calretinin, mesothelin, keratin 5/6, WT1, thrombomodulin, and N-cadherin) and positive carcinoma markers (Ber-EP4, MOC-31, CD15, BG8 (Lewis^y), TAG-72 (B72.3), CEA, renal cell carcinoma marker (RCC Ma) and CD10) was investigated.[10] The conclusion of that investigation was that calretinin, mesothelin, and keratin 5/6 were the best positive mesothelioma markers, and CD15, MOC-31, and RCC Ma were the best positive carcinoma markers for discriminating between epithelioid mesotheliomas and renal cell carcinomas.

In recent years, a relatively large number of markers whose expression is considered to be somewhat restricted to the kidney and renal cell carcinomas have become available. These renal-associated markers include PAX8, PAX2, napsin A, CA IX, RCC Ma, and CD10. Very few studies have been published on the expression of these markers in mesotheliomas and on their value in assisting in distinguishing these tumors from renal cell carcinomas.

PAX8 is a transcription factor that, during embryogenesis, plays an important role in the development of several organs, including the kidney, thyroid, certain areas of the nervous system, and organs derived from the mesonephric (Wolffian) duct and those related to the Müllerian duct.[22,23] The percentage of PAX8 positivity reported in various types of renal cell carcinomas has ranged from 91–98% for clear cell, 71–100% for papillary, and 57–88% for chromophobe.[24–26] It has also been reported to be expressed in the vast majority (83–100%) of metastatic clear cell renal cell carcinomas, regardless of their degree of differentiation[24,26–29] and in 100%[24,25] of the metastatic papillary renal cell carcinomas investigated. These results are comparable with those of the present investigation in which 10 (91%) of 11 primary and 21 (95%) of 22 metastatic clear cell, 10 (100%) of 10 papillary, and 9 (75%) of 12 chromophobe renal cell carcinomas were found to express PAX8. Only one study has been published on PAX8 expression in epithelioid mesotheliomas.[30] In that investigation, PAX8 positivity was demonstrated in 2 (9%) of 23 peritoneal epithelioid mesotheliomas, but none was found in any of the 24 pleural epithelioid mesotheliomas included in the study. In the present investigation, none of the 40 pleural epithelioid mesotheliomas that were stained for PAX8 were positive. This finding indicates that, because PAX8 is usually absent in mesotheliomas, this marker could be very useful for assisting in

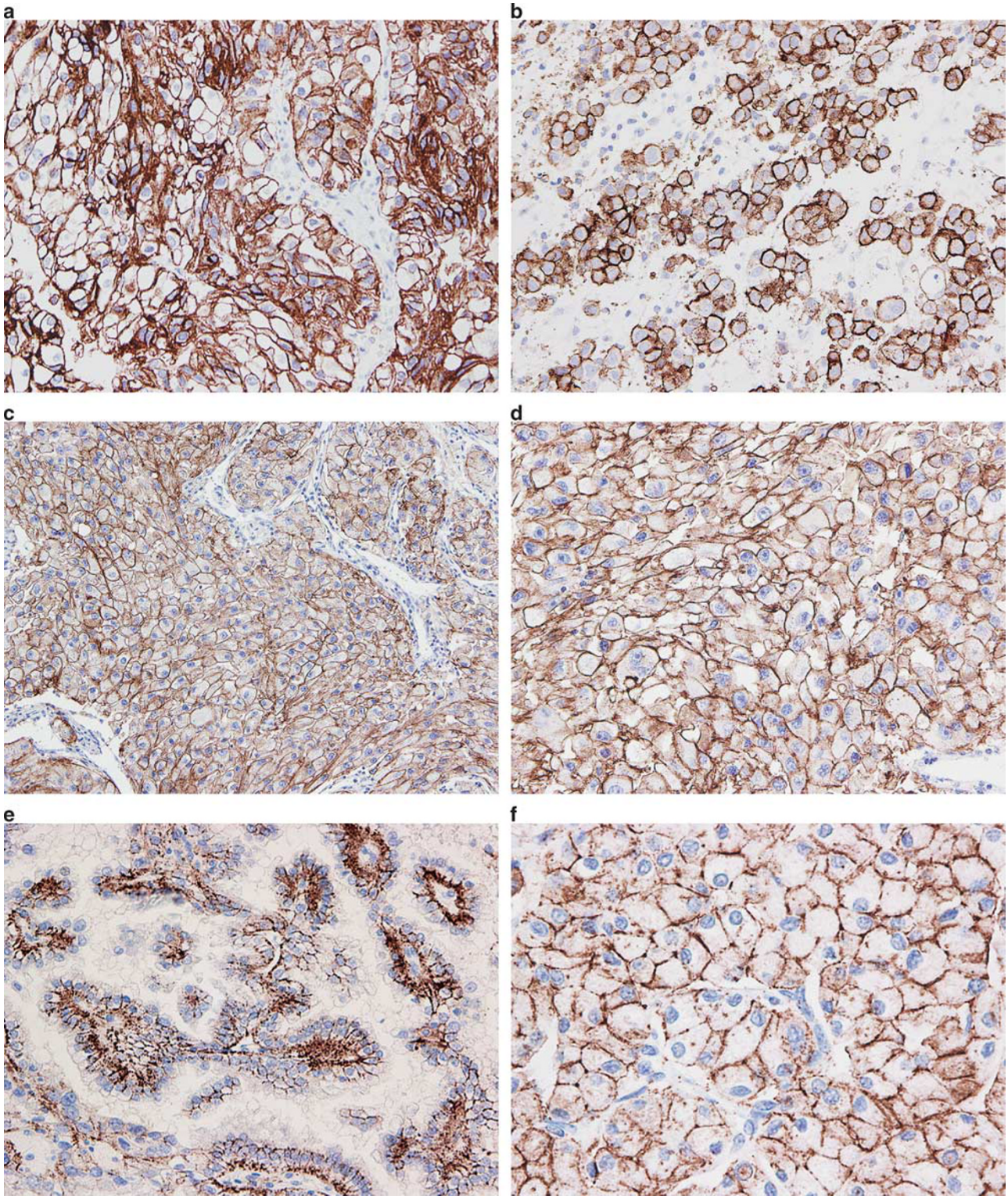

Figure 2 (**a**) Clear cell renal cell carcinoma displaying strong membranous staining for carbonic anhydrase IX. (**b**) Epithelioid mesothelioma reacting for carbonic anhydrase IX. (**c**) Clear cell renal cell carcinoma exhibiting diffuse positivity for claudin-4. (**d**) Higher magnification showing continuous membranous staining for claudin-4. (**e**) Papillary renal cell carcinoma reacting for claudin-4. (**f**) Higher magnification of a chromophobe renal cell carcinoma demonstrating a punctated staining for claudin-4 along the cell membrane.

distinguishing these tumors from metastatic renal cell carcinomas in those instances in which the differential diagnosis is difficult on routine light microscopy. It should be mentioned, however, that even though PAX8 is regarded as a renal cell carcinoma-associated marker, it is not specific for

this type of tumor as it can also be expressed in other carcinomas, particularly those originating in the thyroid, ovary (non-mucinous carcinomas), endometrium, and thymus.[31]

PAX2 is another transcription factor that, similar to PAX8, is involved in the development of the central nervous system, kidney, and organs related to the Müllerian system.[32,33] PAX2 has been reported to be expressed in all types of renal cell carcinomas, except urothelial cell carcinomas.[34–36] The percentage of PAX2 positivity reported in various types of renal cell carcinomas has ranged from 50 to 93% for clear cell,[34–41] 18 to 100% for papillary,[34,35,37,38,40–42] and 6 to 83% for chromophobe.[35–38,40,41] It has also been reported to be frequently expressed in metastatic clear cell carcinomas (49–85%).[28,29,36,43,44] These results are comparable with those of the present investigation in which 7 (64%) of 11 primary and 16 (73%) of 22 metastatic clear cell, 6 (60%) of 10 papillary, and 4 (33%) of 12 chromophobe renal cell carcinomas were found to be PAX2 positive. The sensitivity of PAX2, when compared with that of PAX8, is lower as 60% and 89% of all renal cell carcinomas were positive for these markers, respectively (Table 2). Because all of the PAX2-positive cases were also positive for PAX8 and the staining for the latter marker was often stronger, immunostaining for both markers does not appear to be necessary. To my knowledge, only one study has been published on PAX2 expression in mesotheliomas.[45] In that study, PAX2 positivity was demonstrated in 2 (12%) of 17 peritoneal mesotheliomas in women, whereas all 37 peritoneal mesotheliomas in men were negative for this marker. That PAX2 was negative in all of the 40 pleural epithelioid mesotheliomas in the present investigation is an indication that this marker is usually absent in these tumors. It should be mentioned that, similar to PAX8, PAX2 is also considered to be a renal cell carcinoma-associated marker, but as in the case of PAX8, it is not specific for this type of tumor as it can be expressed in some neoplasms of the female genital tract, particularly non-mucinous carcinoma of the ovary and endometrial adenocarcinomas.[46]

Napsin A is an aspartic proteinase that is predominantly expressed in the lung and kidney.[47] In the kidney, it is expressed in the proximal tubules, where it is thought to function as a lysosomal proteinase in protein catabolism.[48] Recent investigations have shown that in tumors, napsin A expression is largely restricted to lung adenocarcinomas and renal cell carcinomas, especially the clear cell and papillary subtypes. The percentage of napsin A positivity reported in clear cell and papillary renal cell carcinomas has ranged from 17% to 43% and from 75% to 80%, respectively.[49–52] Only 1 (3%) of 38 chromophobe carcinomas investigated in two published studies was reported to be napsin A positive.[50,52] Mesotheliomas have been consistently negative for this marker.[50,53–55] That only 8 (24%) of 33 clear cell and 7 (70%) of 10 papillary renal cell carcinomas, but none of the mesotheliomas, in this study were found to express napsin A is an indication that, even though this marker is highly specific in distinguishing these two subtypes of renal cell carcinomas from mesotheliomas, its sensitivity is low, as only a relatively small percentage of clear cell (24%) and papillary (70%) renal cell carcinomas were napsin A positive.

CA IX is a transmembrane, zinc-containing metalloenzyme that catalyzes the reversible hydration of carbon dioxide into carbonic acid.[56] CA IX is thought to play a role in the adaptation of tumors to hypoxic conditions by regulating the intracellular and extracellular pH.[57,58] It is also believed to play a role in the control of cell proliferation, cell transformation, and tumor cell progression.[58] Because CA IX has been reported to be frequently expressed in renal cell carcinomas, especially the clear cell type, it is, at present, considered to be a diagnostic marker for clear cell renal cell carcinomas.[59–64] The percentage of CA IX reported in various types of renal cell carcinomas has ranged from 50 to 100% for clear cell,[35,41,62–68] 23 to 100% for papillary,[35,41,62,64,66] and 0 to 31% for chromophobe.[35,41,62,64,66] It has also been reported to be frequently expressed in metastatic clear cell renal cell carcinomas (100%).[69] CA IX, however, is not a specific marker for renal cell carcinomas as it has also been reported to be expressed in other malignancies, including carcinomas of the lung, breast, stomach, and colon.[70] Since, in this study, 32 (97%) of 33 clear cell and 7 (70%) of 10 papillary renal cell carcinomas, as well as all 40 epithelioid mesotheliomas, that were investigated were found to express CA IX, immunostaining for this marker has no utility for assisting in distinguishing between epithelioid mesotheliomas and metastatic renal cell carcinomas.

Renal cell carcinoma marker is the term used to designate a 200 kDa glycoprotein that is present along the brush border of the pars convoluta and pars recta segments of the proximal tubule, but it is absent in most other normal tissues.[71] Although RCC Ma expression has been reported in all types of renal cell carcinomas, it is more frequently expressed in clear cell (~40–85%)[10,71–77] and papillary (~50–95%)[40,42,72,74,76–79] renal cell carcinomas. This marker has also been reported in a large percentage of metastatic renal cell carcinomas (~40–85%).[43,68,71,75,76,79] It should be emphasized, however, that RCC Ma is not absolutely specific for these tumors as it can be expressed in other malignancies, including embryonal carcinomas,[74,80] adenocarcinomas of the breast,[71,74,76] prostate and colon,[76] adrenal cortical carcinomas,[76] and mesotheliomas.[10,75] Only two studies have been published on RCC Ma expression in mesotheliomas.[10,75] The first was by this author who reported focal RCC Ma positivity in

3 (8%) of 40 epithelioid mesotheliomas and concluded that this marker should be included in the panel used to distinguish these tumors from metastatic renal cell carcinomas.[10] The second was by Butnor *et al*,[75] who reported RCC Ma expression in 39 (26%) of 145 mesotheliomas. Because the staining was often focal, these authors concluded that strong, diffuse staining for RCC Ma, together with a similar reaction for CD10, would support the diagnosis of metastatic renal cell carcinoma over mesothelioma.

CD10 is a cell surface metalloendopeptidase that was originally named common acute lymphoblastic leukemia antigen as it was first identified in acute lymphoblastic leukemia.[81] Subsequent investigations demonstrated CD10 expression in a variety of non-hematopoietic tumors, including endometrial stromal sarcomas,[82] hepatocellular carcinomas,[83] and renal cell carcinomas.[72,84] Because of its frequent and somewhat restricted expression in the latter tumors, CD10 is often regarded as a renal cell carcinoma-associated marker, even though it is not specific for these tumors. CD10 is expressed in all subtypes of renal cell carcinomas, including clear cell (~80–100%),[10,72,77,78,84–86] papillary (~65–100%),[41,72,77,78,84–87] and chromophobe (~40–65%).[10,77,78,85,88] Only a few studies have been published on the expression of CD10 in mesotheliomas.[10,75,78] The percentage of CD10 positivity reported in these tumors has ranged from 18 to 54% of the cases investigated.[10,75,78] In a previous study by this author, 39 (81%) of 48 renal cell carcinomas and 19 (47%) of 40 epithelioid mesotheliomas were found to express CD10.[10] The conclusion of that investigation was that CD10 immunostaining has no utility is discriminating between epithelioid mesotheliomas and metastatic renal cell carcinomas.

CL-4 is a transmembrane protein located within the tight junctions that is widely expressed in most epithelial cells, including those of the kidney, lung, breast, prostate, thyroid, thymus and bladder, but not in hepatocytes or mesothelial cells.[89] Because CL-4 is frequently expressed in a wide variety of carcinomas, including adenocarcinomas of the lung, breast, ovary, and kidney, as well as in most squamous and transitional cell carcinomas, but it is often absent in epithelioid mesotheliomas,[89,90] it can be regarded as a broad-spectrum positive carcinoma marker.[91] In 2006, Soini *et al*[90] were the first to investigate CL-4 expression in mesotheliomas and to examine its potential utility for assisting in the differential diagnosis of these tumors. In that study, 7 (29%) of 24 epithelioid mesotheliomas and none of the 7 biphasic mesotheliomas investigated were CL-4 positive, whereas all 23 (100%) metastatic adenocarcinomas expressed this marker. A subsequent study published in 2007 by Facchetti *et al*,[89] using the same anti-CL-4 antibody, reported expression in 248 (88%) of 278 primary carcinomas of various sites and 57 (98%) of 58 serosal metastases, whereas all 60 epithelioid, 11 biphasic, and 9 sarcomatoid mesotheliomas were negative. Only a few studies have been published on CL-4 expression in renal cell carcinomas.[89,92] In one of these investigations, CL-4 expression was demonstrated in 7 (87.5%) of 8 clear cell, 6 (100%) of 6 papillary, and 7 (100%) of 7 chromophobe renal cell carcinomas.[89] These results are comparable with those obtained in this study in which 28 (85%) of 33 clear cell, 10 (100%) of 10 papillary, and 12 (100%) of 12 chromophobe renal cell carcinomas were CL-4 positive. CL-4 has a higher sensitivity for discriminating epithelioid mesotheliomas from metastatic renal cell carcinomas when compared with other broad-spectrum carcinoma markers. In a previous study by this author in which several of the latter markers, including Ber-EP4, MOC-31, CD15, BG8, TAG-72, and CEA, were evaluated, it was concluded that CD15 and MOC-31 were the best markers for distinguishing between epithelioid mesotheliomas and renal cell carcinomas.[10] The sensitivity of these two markers for renal cell carcinoma was rather low, however, as only 50% and 42% of these tumors, respectively, were found to be positive for these markers.

At present, a relatively large number of positive mesothelioma markers that can assist in the differential diagnosis between epithelioid mesotheliomas and renal cell carcinomas are currently available. These include calretinin, keratin 5/6, WT1, mesothelin, and thrombomodulin.

Calretinin is one of the positive mesothelioma markers that, because it is commonly expressed in epithelioid mesotheliomas, but not in carcinomas, is usually recommended as one of the primary markers in the various immunohistochemical panels used in the diagnosis of these tumors.[91] In my experience, as well as that of others, calretinin expression can be demonstrated in nearly all epithelioid mesotheliomas when polyclonal antibodies against recombinant human calretinin are used.[6,7,93–95] In a combined review of four published studies on calretinin expression in renal cell carcinomas, 5 (2%) of 204 clear cell, 7 (10%) of 77 papillary, and 3 (6%) of 50 chromophobe renal cell carcinomas were reported to express calretinin.[10,96–98] That none of the clear cell, papillary, or chromophobe renal cell carcinomas studied in a previous investigation by this author were calretinin positive indicates that this marker is uncommonly expressed in these types of tumors.[10]

Keratin 5/6 is another marker that, similar to calretinin, is expressed in nearly all epithelioid mesotheliomas.[7,99] In my experience, as well as that of other investigators, however, this marker has been consistently negative in all clear cell, papillary, and chromophobe renal cell carcinomas investigated.[10,85,100,101]

Depending on the antibody used, WT1 expression has been reported in 43–100% of epithelioid

mesotheliomas.[4,7,95,102,103] When the 6F-H2 monoclonal antibody, which reacts with the N-terminal of the WT1 protein, is used, positivity can be demonstrated in over 90% of epithelioid mesotheliomas. Only a few studies with a relatively small number of cases have been published on WT1 expression in renal cell carcinomas.[10,102,104,105] In the largest of these studies, which was by this author, WT1 positivity was demonstrated in only 1 (4%) of 24 clear cell, but in none of 8 papillary or 12 chromophobe, renal cell carcinomas included in that investigation.[10]

Mesothelin is a marker that has been reported to be commonly expressed in epithelioid mesotheliomas (~90–100%).[10,106–109] Only a few studies have been published on the expression of this marker in renal cell carcinomas. The first of these was by Frierson *et al*,[110] who reported mesothelin positivity in 1 (3%) of 33 renal cell carcinomas.[110] In a subsequent study, however, I was unable to demonstrate the expression for this marker in any of 44 renal cell carcinomas (24 clear, 8 papillary, 12 chromophobe).[10] Because all of the 40 epithelioid mesotheliomas included in that investigation exhibited strong mesothelin positivity, it was concluded that this was a highly sensitive and specific marker for discriminating between epithelioid mesotheliomas and renal cell carcinomas.

Thrombomodulin was the first positive mesothelioma marker that became generally accepted as being useful in assisting in the differential diagnosis of epithelioid mesothelioma. The percentage of thrombomodulin positivity reported in these tumors ranged from ~50 to 100% of the cases.[3,7,10,93,95,111–113] Several studies have also investigated the expression of this marker in renal cell carcinomas.[8–10,114] One of these was in 2002 by Osborn *et al*,[9] who reported thrombomodulin expression in 13 (33%) of 40 renal cell carcinomas and concluded that this marker has no utility in distinguishing between these tumors and epithelioid mesotheliomas. This is in contrast to two previous studies by other investigators in which all 28 renal cell carcinomas included in those investigations were negative for this marker.[8,114] In a more recent study by this author, only 1 (2%) of 44 renal cell carcinomas was thrombomodulin positive.[10]

The results of the present investigation indicate that, because of their sensitivity and specificity, CL-4 and PAX8 should be considered to be the best positive carcinoma markers. In renal cell carcinomas, positivity for CL-4 and PAX8 was demonstrated in 91% and 89% of the cases, respectively, while all of the mesotheliomas were negative for these markers. Because their sensitivity for renal cell carcinomas is lower, PAX2 and napsin A have limited value for assisting in distinguishing these tumors from epithelioid mesotheliomas. As CA IX is commonly expressed in both renal cell carcinomas and epithelioid mesotheliomas, it has no value in discriminating between these malignancies. Finally, since the International Mesothelioma Interest Group[115] recommends an immunohistochemical panel composed of two positive carcinoma markers and two positive mesothelioma markers for distinguishing between epithelioid mesotheliomas and metastatic carcinomas to the serosal membranes, based on the findings of this study, as well as the results obtained in previous investigations by this author, CL-4 and PAX8 combined with keratin 5/6 and calretinin (or mesothelin) will usually make it possible to distinguish epithelioid mesotheliomas from metastatic renal cell carcinomas. In addition, if the differential diagnosis between pleural mesothelioma and metastatic renal cell carcinoma includes other tumors with features resembling these two types of neoplasms, such as solid adenocarcinomas and squamous cell carcinomas of the lung with clear cell morphology, and papillary serous carcinomas of the ovary, some other markers, such as thyroid transcription factor 1, p63, and estrogen receptor, which are commonly expressed in adenocarcinomas of the lung, squamous cell carcinomas, and papillary serous carcinomas, respectively, but not in epithelioid mesotheliomas or renal cell carcinomas, could be added to the immunohistochemical panel.[10,116–118]

Disclosure/conflict of interest

The authors declare no conflict of interest.

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