The utility of Alpha-methyl CoA Racemase (P504S) Expression as a marker of Renal Cell Carcinomas

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L'utilité de l'expression de l'alpha methyl Coa racemase (P504S) dans le diagnostic des carcinomes à cellules rénales

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RÉSUMÉ

Prérequis : Les tumeurs rénales sont hétérogènes et nombreuses. Les données morphologiques et immunohistochimiques sont parfois insuffisantes pour faire le diagnostic d'où la nécessité de nouveaux marqueurs comme la P504S.
Buts: Les auteurs aspirent à démontrer le rôle de l'alpha methyl Coa racemase (P504S) dans le diagnostic des carcinomes à cellules rénales.
Methodes : Les auteurs rapportent une étude rétrospective de 62 patients traités pour des tumeurs rénales diagnostiquées entre janvier 1994 et novembre 2005. L'immuno-positivité a été évaluée grâce à des méthodes qualitatives. La positivité a été définie par un marquage cytoplasmique granuleux.
Résultats: Les 62 tumeurs rénales renfermaient 22 carcinomes papillaires, 18 carcinomes à cellules claires, 12 carcinomes chromophobes et 10 oncocytomes. L'expression de l'alpha méthyl coa racémase a été observée dans tous les carcinomes tubulo-papillaires et 22% des carcinomes à cellules claires. Tous les carcinomes chromophobes n'exprimaient pas la P504 et un seul oncocytome était positif.
Conclusion: Cette étude montre la forte sensibilité de la P504 dans le diagnostic du carcinome tubulo-papillaire du rein sans pour autant négliger le fait qu'une faible positivité peut être observée dans les autres types histologiques.

Mots-clés
alpha-methyl CoA racemase expression, tumeur rénale, spécificité

SUMMARY

Background: Renal cell tumours are numerous and heterogeneous. Because of their clinicopathological heterogeneity, their accurate diagnosis may be challenging. In case of an equivocal diagnosis, immunohistochemistry may be a useful mean of diagnosis. Recently, alpha-methyl CoA racemase has been identified as a useful marker in kidney cancers.
Aims: Our objectives are to highlight the role of alpha-methyl CoA racemase (AMACR) as a diagnostic marker in papillary renal carcinoma and to assess its utility in the other tumour types.
Methods : A retrospective review was performed on 62 patients who were treated for renal tumours between January 1994 and November 2005. Immunoreactivity was evaluated with a qualitative manner. Positive AMACR staining was defined as a coarse dense cytoplasmic granularity.
Results: The 62 renal tumours were diagnosed as papillary tumours in 22 cases, clear cell tumours in 18 cases, chromophobe carcinoma in 12 cases and oncocytoma in 10 cases among the 22 cases of papillary tumours, all the cases (100%) showed cytoplasmic immunoreactivity staining. 4 cases between the 18 clear cell carcinomas (22%) showed positivity with AMACR. The 12 cases of chromophobe carcinoma didn't express AMACR by immunohistochemistry. Only one case between the oncocytomas (1%) expressed AMACR.
Conclusion: This study confirms the high sensitivity of AMACR for papillary renal cell carcinomas but we must keep in mind that weak focal AMACR staining could be present in other renal cell carcinomas.

Key-words
alpha-methyl CoA racemase expression, renal tumour, specificity

Renal tumours account for 3% of all cancers. Among these tumours, renal cell cancers represent 90% of all malignancies that occur in adults in both sexes (1). They are subdivided into clear cell carcinoma, multilocular renal cell carcinoma, papillary renal cell carcinoma, chromophobe renal cell carcinoma, carcinoma of the collecting ducts of Bellini, renal medullary carcinoma, mucinous tubular and spindle cell carcinoma, unclassified renal cell carcinoma, papillary adenoma and oncocytoma according to the World Health Organization International histological classification of kidney tumours (1). Because of their clinicopathological heterogeneity, their accurate diagnosis may be challenging. In case of an equivocal diagnosis, immunohistochemistry may be a useful mean of diagnosis. Recently, alpha-methyl CoA racemase has been identified as a useful marker in kidney cancers.

Our aims are to highlight the role of alpha-methyl CoA racemase (AMACR) as a diagnostic marker in papillary renal carcinoma, to assess its utility in the other tumour types and to justify its relative efficiency in cases of metastatic renal tumours.

MATERIALS AND METHODS

A retrospective review of the pathological databases of the Charles Nicolle Hospital was performed on 62 patients who were treated for renal tumours between January 1994 and November 2005.

Immunohistochemical staining was performed on 5-um sections cut from formalin-fixed, paraffin-embedded tissue blocks. Heat-mediated antigen retrieval was performed in pH 6.0 citrate in water both for 30 minutes. Immunostaining was performed on a Dako autostainer using a peroxidase-labeled polymer-based detection system (Envision plus) and diaminobenzidine as a chromogen. Rabit monoclonal prediluted anti-AMACR antibody was used and incubated. Appropriate negative and positive controls were used.

Immunoreactivity was evaluated in a qualitative manner that assessed the staining intensity. Positive AMACR staining was defined as a coarse dense cytoplasmic granularity (Fig 1). We divided the staining intensity into three categories: negative, weak and strong.

RESULTS

Histological results

The 62 renal tumours were diagnosed as papillary tumours in 22 cases (Fig 2), clear cell tumours in 18 cases, chromophobe carcinoma in 12 cases and oncocytoma in 10 cases according to the current 2004 World Health Organization classification. These results are listed in table n°1. In all cases, normal kidney tissue adjacent to tumours was present attesting the validity of the reaction.

Figure 1 : Positive AMACR staining defined as a coarse dense cytoplasmic granularity (HEX400).

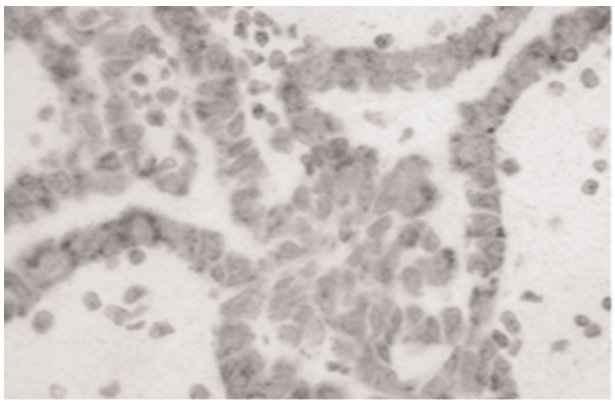

Figure 2 : Papillary renal cell carcinoma (HE X 250). Inset: Intense positive staining for AMACR (HE X 250).

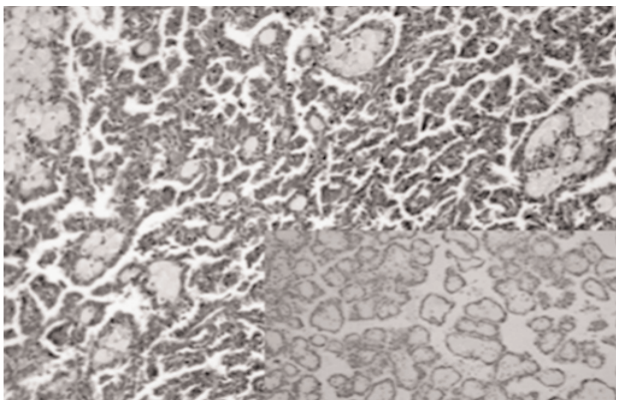

Tableau 1 : Histological subtypes

Histologic subtypes	Number of cases
Clear cell carcinomas	18
Papillary carcinomas	22
Chromophobe carcinomas	12
Oncocytomas	10

AMACR in papillary renal cell carcinoma

Among the 22 cases of papillary tumours, all the cases (100%) showed cytoplasmic immunoreactivity staining even in type 1 and type 2.

AMACR in clear cell carcinoma

Four cases between the 18 clear cell carcinomas (22%) showed positivity with AMACR.

AMACR in chromophobe carcinoma

The 12 cases of chromophobe carcinoma didn't express AMACR by immunohistochemistry.

AMACR in oncocytoma

Only one case between the oncocytomas (1%) expressed AMACR by immunohistochemistry.

All these results are listed in table n° 2.

Tableau 2 : p504 S immunoreactivity in renal tumours in our study

Histologic subtypes	Percentage (%)
Papillary carcinomas	100 (22/22)
Clear cell carcinomas	22 (4/18)
Chromophobe carcinomas	0 (0/12)
Oncocytomas	1 (1/10)

DISCUSSION

Renal tumours are diverse and heterogeneous. This fact makes their diagnosis challenging in some cases. Cytogenetic analysis and gene expression microarray analyses tried to assess the profile of the major renal cell carcinomas including papillary, clear cell and chromophobe sub-types (2, 3, 4, 5). Papillary carcinomas are characterized by a gain of 2 or more chromosomes, 3q, 7, 8, 12, 16, 17 or 20 (4, 6, 7). However, all these analyses seem to be difficult to realize as a routine technique. For these reasons, many authors tried to find a new reliable marker for renal carcinomas and they recently identified AMACR. The gene for AMACR is located in chromosome 5 and encodes 382 amino acid protein that play a key role in the , oxidation of branched chain fatty acids and the bile acid intermediates dihydroxycholestanoic acid and trihydroxycholestanoic acid. Mutations in the AMACR gene, associated with decreased enzyme activity, have been implicated in the development of adult onset sensory motor neuropathy. This is thought to be a consequence of sustained rises in plasma branched chain fatty acids (8). AMACR expression was initially observed in prostatic carcinomas. This overexpression has been proved to be not only unique to prostate cancer. Many authors proved its expression in normal tissue like the liver, the kidney, the gallbladder, the nervous system, the colon, the kidney, the breast, the pancreas and blood elements (9, 10). High expression of AMACR mRNA was also found in prostate, liver, kidney, colon, ovarian, breast, lung carcinomas in addition to melanoma and lymphoma (9, 10, 11). All these findings prove that AMACR isn't a specific marker of renal tumours and it can't be used in secondary localizations. AMACR positivity should only be interpreted when the primitive localization is known. Despite this lack of specificity to identify secondary renal tumours, this marker seems to be of interest in order to differentiate mixed types tumours or unclassified ones. Our results showed that the expression of AMACR was observed in 100% of papillary renal cell carcinomas. These results are identical to those reported by yang et al and Molinié and coworkers (12, 13). Concerning chromophobe carcinoma and oncocytoma, we found 0% of expression in the former and 1% in the later. These results are lower than those reported in the literature. In fact, Molinié et al reported positivity in 16% of chromophobe carcinoma and 11% in oncocytoma. Tretiakova et al reported in their study that 15% of oncocytoma and 0% of chromophobe carcinoma expressed AMACR (13, 14). Lin et al demonstrated an overexpression of AMACR in 29% of chromophobe carcinomas and 25% of oncocytomas (15). These results make the separation between chromophobe carcinomas and oncocytomas, based on the expression of AMACR difficult. Chen et al studied the expression of AMACR in 63 cases of renal carcinomas and they found a highest expression in papillary renal cell carcinomas. They could only separate some, but not all, oncocytomas from chromophobe carcinomas (16). Clear cell carcinomas expressed AMACR in 22% of the cases in our study. These results are quite similar to those reported by Molinié et al and Tretiakova et al who reported respectively 16% and 25% of positivity in clear cell carcinomas (13, 14). Lin et al have reported higher rates estimated to 68, 6%. All these results that are represented in table n°3 highlight the fact that the expression of AMACR is highest in papillary carcinomas but it could be noticed in other types of renal tumours. Molinié et al reported the expression of this marker in mucinous, tubular and spindle cell tumours. These authors support the point of view that mucinous tubular and spindle cell carcinomas and papillary renal cell carcinomas are related (13). The purpose of all the studies made on AMACR was to assess its ability as a diagnostic marker in order to provide the effective treatment. Langner et al reported also the efficiency of this marker as an additional prognostic indicator in upper urinary tract urothelial cancer. In fact, they found that its expression was correlated with advanced tumour stage and grade (17).

CONCLUSION

This study confirms the high sensitivity of AMACR for papillary renal cell carcinomas but we must keep in mind that weak focal AMACR staining could be present in case of clear cell, chromophobe carcinomas and oncocytomas. More studies are necessary in order to give value to this marker and to prove its possible utility as a prognostic indicator.

Tableau 3 : p504 positivity in renal tumours: a comparison between our results and those in the literature

	Papillary	Clear cell	Oncocytoma	Chromophobe
Tretiakova et al	100	25	15	0
Lin et al	100	68.6	25	29
Molinié et al	100	16	11	16
Present study	100	22	1	0

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