PAX8 is expressed in the majority of renal epithelial neoplasms: an immunohistochemical study of 223 cases using a mouse monoclonal antibody

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ABSTRACT

Aims PAX8 is a cell lineage-specific transcription factor which plays a crucial role in the organogenesis of the kidney, thyroid gland and Müllerian duct. A previous study showed that PAX8 is a specific and sensitive marker for both renal and ovarian carcinomas. The purpose of this study is to investigate PAX8 expression using a new monoclonal PAX8 antibody in a larger number of renal epithelial neoplasms including clear cell renal cell carcinoma, papillary renal cell carcinoma, chromophobe renal cell carcinoma and renal oncocytoma.

Methods PAX8 immunohistochemical staining was performed on tissue microarrays containing 84 cases of clear cell renal cell carcinoma, 66 cases of chromophobe renal cell carcinoma, 57 cases of papillary renal cell carcinoma and 16 cases of renal oncocytoma.

Results PAX8 expression was detected in 93% (78/84) of cases of clear cell renal cell carcinoma, 80% (53/66) of cases of chromophobe renal cell carcinoma, 95% (54/57) of cases of papillary renal cell carcinoma and 94% (15/16) of cases of renal oncocytoma.

Conclusions PAX8 is expressed in the majority of renal epithelial neoplasms including renal cell carcinomas and oncocytomas and the monoclonal PAX8 antibody is more sensitive than polyclonal antibody to detect chromophobe renal cell carcinoma. These results showed that PAX8 is a valuable marker for nephric neoplasms.

INTRODUCTION

PAX8 is a cell lineage-specific transcription factor which plays a crucial role in the organogenesis of the kidney, thyroid gland and Müllerian duct. Our previous study using a rabbit polyclonal antibody anti-PAX8 in a wide variety of normal and neoplastic tissues showed that PAX8 is a sensitive and specific marker for both renal and ovarian carcinomas.¹ While there are several studies including ours on renal cell carcinomas using polyclonal PAX8 antibody immunohistochemistry,^{1–3} the information for a new monoclonal PAX8 antibody in renal epithelial neoplasms is very limited. Therefore, using a new monoclonal PAX8 antibody, this study aims to investigate PAX8 expression in a larger number (223 cases) of renal epithelial neoplasms including 84 cases of clear cell renal cell carcinoma, 57 cases of papillary renal cell carcinoma, 66 cases of chromophobe renal cell carcinoma and 16 cases of renal oncocytoma.

MATERIALS AND METHODS

Construction of tissue microarrays

Tissue microarrays, containing 223 cases of human renal epithelial neoplasms including 84 cases of clear cell renal cell carcinoma, 66 cases of chromophobe renal cell carcinoma, 57 cases of papillary renal cell carcinoma and 16 cases of renal oncocytoma, were constructed as previously described^{4,5} at the Institute for Pathology, University of Erlangen, Germany. All tumours were reviewed by an experienced uropathologist and diagnosis of the histological type according to the 2004 WHO classification⁶ was confirmed. Sections of 5 µm thickness were cut from tissue microarrays and were stained with H&E to confirm the presence of the expected tumour histology within each tissue core. Additional sections were cut for immunohistochemistry.

Immunohistochemistry

Tissue sections from the tissue microarray were deparaffinized, rehydrated using graded alcohols and washed with tris buffered saline. Antigen retrieval for PAX8 was performed by heating sections at 125°C in a pressure cooker for 20 min. After endogenous peroxidase activity was quenched and non-specific binding was blocked, tissue sections were incubated with mouse monoclonal antibody PAX8 (BC12, Biocare Medical, Concord, California, USA) for 30 min. Sections were then rinsed and incubated in a polymer HRP-detection system for 30 min and visualised with diaminobenzidine (DAB) chromogen. Slides were briefly counterstained in a modified Mayer haematoxylin. A normal renal tissue served as positive control. Negative control was performed by replacing the PAX8 antibody with normal serum. Only nuclear staining was considered positive.

RESULTS

The results of PAX8 immunohistochemical staining are summarised in table 1 and figure 1. Overall, 90% (200/223) of all renal epithelial neoplasms stained positive for PAX8. Among renal cell carcinomas, PAX8 expression was detected in 93% (78/84) of cases of clear cell renal cell carcinoma, in 80% (53/66) of cases of chromophobe renal cell carcinomas and in 95% (54/57) of cases of papillary renal cell carcinoma. PAX8 expression was detected in 94% (15/16) of cases of renal oncocytoma.

Table 1 Summary of PAX8 immunostaining in renal epithelial neoplasms

	n	Negative (%)	Positive (%)
Clear cell renal cell carcinoma	84	6 (7)	78 (93)
Chromophobe renal cell carcinoma	66	13 (20)	53 (80)
Papillary renal cell carcinoma	57	3 (5)	54 (95)
Renal oncocytoma	16	1 (6)	15 (94)
Total	223	23 (10)	200 (90)

DISCUSSION

In this study, using a monoclonal PAX8 antibody, we found that 90% (200/223) of renal epithelial neoplasms were positive for PAX8. Among renal cell carcinomas, papillary renal cell carcinomas had the highest percentage of positivity for PAX8, followed by clear cell renal cell carcinomas and chromophobe renal cell carcinomas. Most renal oncocytomas were positive for PAX8. These findings confirm earlier studies by us and other groups using a polyclonal PAX8 antibody and further show that PAX8 is a very useful immunomarker for renal epithelial neoplasm.^{1–3}

PAX8 is one of the nine members of the paired box (PAX) family of cell lineage transcription factors, which play crucial roles during fetal development and neoplastic growth.^{3,7–9} PAX8 is a nephric lineage nuclear transcription factor which plays an

important role in renal organogenesis.^{3,10–14} PAX8 may also be involved in maintaining the differentiated features and specialised functions of renal epithelial cells.^{15,16} Our previous study, using a rabbit polyclonal PAX8 antibody, found PAX8 positivity in 91% (86/94) of clear cell renal cell carcinomas, 100% (14/14) of papillary renal cell carcinomas and 57% (4/7) of chromophobe renal cell carcinomas.¹ The current study with a mouse monoclonal antibody showed that 93% of clear cell renal cell carcinomas, 95% of papillary renal cell carcinomas and 80% of chromophobe renal cell carcinomas were positive for PAX8. These findings are similar to those from other groups and our previous study using polyclonal PAX8 antibody with the exception of a lower positive rate for chromophobe renal cell carcinoma (57% vs 80%). The lower positive rate in our previous study may be due to the limited number (only seven cases) of chromophobe renal cell carcinomas. Our current study, which consisted of a larger number of cases, further confirmed our previous findings that chromophobe renal cell carcinoma has the lowest PAX8 positive rate among the common renal cancers.¹ These findings are consistent with those reported by Laury *et al*.²

Using a polyclonal PAX8 antibody, Laury *et al* found that 93% (115/123) of clear cell renal cell carcinomas, 76% (19/25) of papillary renal cell carcinomas, 80% (4/5) of chromophobe renal cell carcinomas and 81% (13/16) of renal oncocytomas were

Figure 1 Immunohistochemical staining of PAX8 in renal epithelial neoplasms. Positivity for PAX8 was seen in clear cell renal cell carcinoma (A), papillary renal cell carcinoma (B), chromophobe renal cell carcinoma (C) and renal oncocytoma (D) (magnification 200×).

Original article

Take-home messages

- ▶ PAX8 is a cell lineage-specific transcription factor which plays a crucial role in the organogenesis of the kidney, thyroid gland and Müllerian duct.
- ▶ PAX8 is expressed in the majority of renal epithelial neoplasms.
- ▶ Chromophobe renal cell carcinoma has a lower PAX8 positive rate among the common renal tumour types.

positive for PAX8.² Similarly, using a polyclonal PAX8 antibody, Tong *et al* detected PAX8 positivity in 98% (58/59) of clear cell renal cell carcinomas, 90% (19/21) of papillary renal cell carcinomas, 82% (9/11) of chromophobe renal cell carcinomas and 95% (21/22) of renal oncocytomas.³ Our findings are very similar to those reported by Laury *et al* and Tong *et al*, with the exception of a slightly higher positive rate (95%) in papillary renal cell carcinomas in our current study of 223 cases of renal tumours.^{2 3}

Tissue microarray immunohistochemistry is a very useful screening tool to identify promising biomarkers.^{4 5} However, its limitation should be realised as the positive rate in the current study may be underestimated when dealing with heterogeneous expressions, such as PAX8. Previous studies by our group and other groups have shown that the concordance between tissue microarray and whole section is over 90% as representative areas based on whole section reviews were preselected to construct the tissue microarray. Our results in the current study are similar to those reported by other groups.^{1–3}

In summary, our results in the 223 cases of renal epithelial neoplasms showed that PAX8 is expressed in a high percentage (90%) of renal neoplasms including both malignant renal tumours (renal cell carcinomas, 80–95%) and benign renal tumours (oncocytomas, 94%), which in turn confirm that PAX8 is a valuable marker for nephric tumours.

Competing interests None.

Provenance and peer review Not commissioned; externally peer reviewed.

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