INTERMEDIATE FILAMENT PROTEINS AS TISSUE SPECIFIC MARKERS IN NORMAL AND MALIGNANT UROLOGICAL TISSUES

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ABSTRACT

Immunocytochemical techniques have become valuable tools in many fields of clinical pathology and medical research. Especially the development of highly specific (monoclonal) antibodies to a large variety of tissue antigens has in recent years led to the establishment of sensitive tissue markers. One of the most promising types of tissue specific markers so far is represented by the intermediate filament proteins. Since the findings of this rapidly expanding field are also being applied in urology, we have reviewed the current data in order to describe the new insights in tumor biology and histogenesis, as well as their application in diagnostic pathology.

TISSUE SPECIFIC DISTRIBUTION OF INTERMEDIATE FILAMENT PROTEINS

Intermediate filaments form a part of the intracellular cytoskeletal matrix present in all mammalian cells. These seven to 11 nm. proteinaceous filament structures occur in cells in addition to microfilaments and microtubules. Figure 1 shows examples of the distribution and pattern of cytoskeletal structures, microtubules and intermediate filaments in cultured human bladder transitional cell carcinoma cell lines.

Five types of intermediate filament proteins (IFP) have been recognized and analyzed by biochemical and immunohistochemical techniques. On the basis of these results the following correlations have been observed: 1) Cells and tissues of mesenchymal origin contain vimentin as their IFP. 2) Muscle tissues contain desmin IFP. 3) Nervous tissues contain the neurofilament proteins. 4) Gial cells contain intermediate filaments of the glial fibrillary acidic protein (GFAP) type. 5) Cytokeratins constitute the intermediate filament system of epithelial cells. Nineteen different cytokeratin polypeptides have been characterized in human epithelial tissues so far. These can be further subdivided into the type I or acidic and type II or basic cytokeratins. These polypeptides are not expressed randomly throughout epithelia, but occur in cell-type specific combinations. The cytokeratin patterns of epithelial tumors are either identical, or at least closely related, to the cytokeratin pattern present in the cell of origin. Several conventional antisera as well as monoclonal antibodies have been raised against IFP, and most of them have been shown to react in a tissue specific manner. Since normally IFP are retained in a cell upon malignant transformation, such antibodies can be exploited in surgical pathology to assist in the final tumor diagnosis. Recent studies using monoclonal antibodies to individual cytokeratin polypeptides have shown that

such antibodies can distinguish between different types of epithelia.

For example, an antibody to cytokeratin 18 mainly recognizes "simple" columnar epithela but not squamous epithelium. On the other hand, a monoclonal antibody directed against the high mol. weight cytokeratins specific for epidermis stains only keratinizing squamous epithelial cells. Furthermore, tumors derived from these different types of epithelium can be distinguished and characterized using these monoclonal anticytokeratin antibodies.

The immunocytochemical evaluation of urological tumors using antisera to several IFP has produced two results. The first is the ability to make a more precise diagnosis in a number of difficult cases. At the level of individual patients' tumors, routine use of a selection of several anti-IFP antisera may lead to a revision or a new interpretation of a diagnosis which seemed highly likely on the basis of routine histology. The second result is the gaining of new insights into the biology and histogenesis of different tumor types. Routine application of multiple anti-IFP antisera to tumors which were thought to be reasonably well-defined on clinical and morphologic grounds provided potentially valuable new insights into the nature of these tumors. Furthermore, applying combinations of monoclonal and polyclonal antibodies to cytokeratins and other IFP's in normal and neoplastic tissues has demonstrated a considerable heterogeneity in IFP expression by tumors, especially those from the urogenital tract. These tissues and tumors will be described in more detail.

NORMAL KIDNEY AND ITS TUMORS

In order to fully understand IFP staining patterns in malignant neoplasms, it is of great importance to examine their normal tissue counterpart(s) or their most likely cells of origin in order to investigate differences occurring during malignant transformation. Furthermore, knowledge of changes occurring in IFP distribution during embryologic differentiation can give important clues about staining patterns recognized in poorly differentiated tumors. Frozen sections from normal monkey, rat and human kidneys (fig. 2 A-D) were examined with

Accepted for publication April 14, 1986.

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Supported by grants from the Netherlands Cancer Foundation.
cytokeratin antibodies that parietal cells of Bowman’s capsule represent a continuation of the proximal tubular cells. The mesangium and endothelium of the glomerulus are stained with the polyclonal antibody to vimentin (fig. 2 D). The parietal epithelium seems to show variable staining with vimentin antibodies and also we occasionally observed faint vimentin positivity in some of the distal convoluted tubules. This co-expression of cytokeratin and vimentin deserves further investigation. In this context it is interesting to note that Holthoefer et al.15,16 have observed a transient co-expression of vimentin and cytokeratin in collecting ducts of fetal kidneys. Connective tissue fibroblasts, endothelium, and smooth muscle cells in blood vessels are also positive for vimentin. In our experiments, desmin was only found in the smooth muscle cells of the blood vessels.

Bachman et al.17 have shown co-existence of vimentin and desmin in glomerular and extra-glomerular mesenchymal cells, which is in agreement with the notion that these cells are derived from a specific subset of vascular smooth muscle cells.18

RENAI CELL TUMOR (GRAWITZ TUMOR)

Renal cell carcinoma (Grawitz tumor) is a malignant tumor of kidney cells. Since its initial description, the origin and nature of this tumor has been controversial.21 However, in recent years it has become clear that it is in fact a primary tumor probably arising from proximal tubular epithelial cells. It has been considered an adenocarcinoma because of its light and electron microscopic features, especially the presence of intercellular bridges and the formation of ducts and gland-like structures. However, the clinical behavior of this tumor was never totally supportive of the interpretation that it is a simple adenocarcinoma. Two features of the clinical behavior of this tumor which suggest a more complicated interpretation are its marked tendency to early blood vessel invasion and lung metastasis as well as a noteworthy resistance to chemotherapy, both features of sarcomas. Routine use of anti-IFP antibodies in evaluating renal cell tumors of adults has shown that these tumors usually contain both cytokeratins and vimentin. Individual cells with both IFP types are routinely demonstrable in renal cell tumors.15,22 We have found that most of the tumors examined (37 out of 40) express vimentin in virtually all cells. Cytokeratins are expressed to a lesser extent in the tumors. In this respect double immunofluorescence labelling demonstrated that some of the vimentin-containing cells co-expressed cytokeratin (fig. 2 E, F), while others did not. However, occasionally tumor cells were found to contain cytokeratin but not vimentin. Some lymph node metastases were found to express only vimentin in cytokeratin. In summary we find higher percentage of tumors co-expressing vimentin and cytokeratin than described by Holthoefer et al.19 who found simultaneous expression of both types of intermediate filaments in only about half of their samples. On the basis of these findings, we have proposed that renal cell tumors may better be considered carcinomas than pure adenocarcinomas.23 This interpretation of the biology of renal cell tumors fits well with both the clinical course and the immunopathologic findings. Using the polypeptide specific monoclonal antibodies directed against subtypes of cytokeratins we found cytokeratin 18 but not cytokeratin 7 in most tumors examined. These data confirm results described by Achsttaetter et al.24 who could demonstrate cytokeratins 8, 18 and 19 in kidney tumors using biochemical techniques. The absence of cytokeratin 7 both in normal proximal tubules and in renal cell tumors supports the assumption that such malignancies may be derived from proximal tubular epithelium.

NEPHROBLASTOMA (WILMS’ TUMOR)

Unlike its adult counterpart, the nephroblastoma’s origin as a renal tumor has been recognized from the first descriptions of this tumor, since the three cell types which comprise the

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**Fig. 1.** Immunofluorescence micrographs of human bladder carcinoma cells in culture showing cytoskeletal structures present in these cells. A, microtubules; B, cytokeratin intermediate filaments; C, vimentin intermediate filaments. (A, B, C × 550).
tumor are easily recognized as malignant counterparts of the cells and structures forming the kidney during embryological development. Recent studies of nephroblastoma using antibodies to IFPs (Fig. 2 G, H) have shown that, similar to its adult counterpart, nephroblastoma contains both vimentin and cytokeratin, with co-expression of both IFPs in some of the tumor cells. Immunofluorescence microscopy studies revealed expression of cytokeratins and vimentin in blastema cells, while we have found co-localization of these two type of IFP also in the tubular structures of nephroblastoma (Fig. 2 G, H). In undifferentiated nephroblastomas with absence of tubule formation, the blastema cells were only vimentin positive and cytokeratin negative. We have found that part of the tubules are positive using monoclonal antibodies to cyto-
keratins 7 and 18, which is in accordance with the suggestion of Yeger et al. that Wilms’ tumor tubules more closely resemble distal than proximal convoluted tubules of adult kidneys. Vogel et al. have described that rhomboid tumors of the kidney contain both vimentin and cytokeratin. Occasionally we have observed desmin positive cells in nephroblastomas in the myoid component. From this point of view it is therefore interesting to note the almost perfect separation of patients with one of the two types of kidney tumor. The peak incidence of nephroblastoma is in the preschool years with very few well-documented cases occurring after the age of 20. Conversely, well-documented cases of pure Grawitz tumor before the age of 20 are quite unusual.

The demonstration that both childhood and adult primary renal tumors have an important biological property in common, namely expression of both vimentin and cytokeratin IFP, has focused attention on abnormalities of these tumors heretofore difficult to explain. Both tumors in addition to carcinomatous components frequently contain elements which histologically are thin fibrous stroma and similar to the presence of areas of adult type (Grawitz) renal tumor in otherwise typical nephroblastomas of young patients has been reported in the U.S. National Wilms’ Tumor Study. Finally, nephroblastoma shows two variants, clear cell sarcoma of the kidney and malignant rhomboid tumor of the kidney. These are both relatively more aggressive neoplasms than the usual nephroblastoma. In addition clear cell sarcomas, which have been shown to contain vimentin IFP, have a marked propensity for bone metastases. Thus, prompted by the immunopathological study of IFP expression in both childhood and adult renal cell tumors, a unifying concept of these tumors emerges. Both Wilms’ and Grawitz tumor may be considered carcinomas, since they express epithelial as well as mesenchymal properties. Both in children and adults an unbalanced proliferation of elements differentiated more along epithelial lines or more along mesenchymal lines leads to the bewildering variety of tissue types seen in these tumors. Finally, this interpretation of the biology of renal cell tumors correlates well with the clinical behavior of these tumors as mentioned above.

RETAIL TUMOR MODEL SYSTEMS

Frozen sections of xenografts of human renal cell tumors in nude mice (fig. 2 I, J) and a rat renal cell tumor (fig. 2 K, L) could also be shown to contain cytokeratins and vimentin. In double labeling studies it was seen that vimentin positive cells were also positive for cytokeratin. This may give support to the suggestions that these experimental tumors may be used as proper renal cell tumor model systems for further biological and chemosensitivity studies.

NORMAL AND NEOPLASTIC UROTHELIUM

The normal mucosal lining of the urinary tract between the renal pelvis and the urethra, the transitional epithelium, is a thin multilayered cell layer. The cells composing the deeper layers show little morphologic or functional differentiation until they reach the luminal surface. Here the amount of cytoplasm increases significantly and the cells assume the shape of umbrella cells, responsible for maintaining the water-tightness of the urinary tract. In normal adult and embryotational transitional cells only IFPs of the cytokeratin type have been detected. Using a polyclonal rabbit antiserum to skin keratins or the broadly cross-reacting monoclonal cytokeratin antibody (RCK102), all layers of the bladder epithelium were stained (fig. 3 A, C). Biochemical and immunochemical studies using polyclonal and monoclonal antibodies have demonstrated the presence of cytokeratins 7, 8, 18 and 19 with small amounts of cytokeratins 4 and 5 in this type of epithelium. Interestingly, several authors have found that certain monoclonal antibodies specific for cytokeratin 18 stain only a subset of transitional epithelial cells, the umbrella cells (fig. 3 E, D). However, other monoclonal antibodies specific for this cytokeratin subtype recognize all cell layers of the urothelium. A possible explanation for this phenomenon would be the selective unmasking of certain cytokeratin 18 epitopes as a result of urothelial cell differentiation.

In addition to these findings it is of importance to note that Summerhayes and Chen have shown in mouse bladder epithelium the presence of a 62 KD cytokeratin which occurred exclusively in the basal epithelial cell layer, but seemed to be absent in the intermediate and superficial layers.

TRANSITIONAL CELL CARCINOMA

The simplest neoplastic growth in transitional epithelium, the flat carcinoma in situ, is a replacement of normal transitional cells by cells with features of malignant cells with increased nuclear/cytoplasmic ratio, irregular nuclear and chromatin features. Development of non-invasive papillary transitional cell tumors is characterized by the formation of papillary fronds projecting into the lumen of the urinary tract. These fronds have a thin connective tissue stalk containing blood vessels and are lined on the outside by more or less abnormal transitional epithelium. These non-invasive papillary transitional cell tumors are graded according to both histologic and cytologic criteria. The best differentiated papillary tumors (Grade 0; papilloma) show a lining epithelium on the papillary surface which is composed of cells and tissue structures identical to normal transitional epithelium.

Increasing dedifferentiation of these papillary tumors (Grades 1 to 3) is marked both by increasing thickness of the epithelium caused by increased numbers of cells and by increasing cellular atypia. Thus grade 3 papillary transitional cell carcinomas show a marked increase in the number of cell layers in the non-lining epithelium and most of these cells have nuclear and cytoplasmic features of carcinoma cells. The third form of neoplastic growth in transitional epithelium is frankly invasive carcinoma. However these carcinomas may be composed of cells with more or less malignant features and are therefore also graded 1 to 3, grade 3 being the most malignant appearing cell type. As a consequence of the complex relationship of cytologic and histologic dedifferentiation in transitional cell neoplasia, grading of the lesions provides relatively less insight into the expected tumor behavior than grading of many other carcinomas. This is especially true at the level of the individual patient’s prognosis.

The study of IFP expression in neoplastic transitional epithelium has provided new insights into the nature and progression of carcinogenesis in this tissue. Cytokeratins 7, 8, 18, and 19 can readily be detected in these tumors by gel-electrophoretic and immunocytochemical techniques. In addition to these, cytokeratins 5, 13 and 17 were found in some tumors. Although all cell layers of neoplastic transitional epithelium express cytokeratins as recognized by several cytokeratin antibodies (fig. 3 A, G), application of one of the monoclonal antibodies to cytokeratins 18 (RCK553) shows an interesting series of changes associated with neoplastic progression. In non-invasive papillary transitional cell carcinomas, the tumors which appear cytologically and histologically well differentiated (Grades 0 and 1) show a pattern of cytokeratin 18 expression similar to that of normal urothelium with only the most superficial cell layer reacting with the cytokeratin 18 antibody (fig. 3 F). Somewhat less differentiated tumors with an increased cellular atypia (Grade 2) begin to show single cells and small clusters of cells deeper in the epithelium which express this specific cytokeratin 18 epitope. The least differentiated most atypical non-invasive tumors (Grade 3) show cytokeratin 18 positive cells throughout the whole thickness of the epithelium (fig. 3 H). In a lymph node metastasis all transitional epithelial tumor
cells showed reactivity with this cytokeratin 18 antibody. From these findings it appears that both during urothelial cell differentiation and during its neoplastic progression similar alterations in the epitope availability of the cytokeratin 18 protein occur.

These findings are especially striking since transitional cell neoplasia seems to consist of morphologically less differentiated cells, analogous to those comprising the basal and parabasal cell layers of normal transitional epithelium, rather than the more differentiated umbrella cells of the epithelial surface. This reactivity pattern may be useful for a better classification of transitional cell carcinomas. In some cases of bladder carcinoma we have observed small numbers of cells reactive with a monoclonal antibody specific for keratinizing stratified epithelial tissues (fig. 3 f). This suggests the presence of small areas with squamous cell differentiation or single cells keratinizing within these tumors.

BLADDER TUMOR MODEL SYSTEMS

In the examination of rat bladders bearing tumors induced by butyl-(4-hydroxybutyl)-nitrosamine (BBN) by routine histology and immunohistochemical staining of intermediate filament types smaller lesions were stained similar to human urothelial dysplasia (fig. 3 J, K). Progression of the lesions demonstrated large exophytic papillomas with extensive endo-
phytic epithelial growth into abundant stroma. These lesions showed increasing predominance of squamous over transitional elements. Immunohistochemical findings confirmed these results and also demonstrated that morphologically indifferent cells, even in early lesions, express cytokeratin 10, characteristic of keratinizing squamous epithelium (fig. 3L). These results demonstrate that BBN-induced bladder tumors show marked quantitative and qualitative differences from the most common, purely transitional, human bladder carcinomas. Sumerhayes and Chen 26 concluded from a similar study using an antibody to a 50 KD cytokeratin polypeptide that BBN-induced hyperplastic bladder lesions in mice result primarily from the proliferation of cells in the basal compartment. They also found that this antibody, which in normal mouse bladder only stained the basal cell layer, reacted homogeneously with all squamous cell components, whereas the transitional cell component showed mainly reaction in the basal cells of the epithelial islands. Hasegawa and Cohen 27 have also suggested changes in cytokeratin expression in proliferative rat bladder lesions. All in all, these studies strongly indicate that the cytokeratin composition and/or distribution in the bladder urothelium changes following induction of carcinogenesis.

NORMAL AND NEOPLASTIC PROSTATE TISSUE

In normal and hyperplastic prostate tissue, a clear distinction can be made between the epithelial and the stromal cell component with the help of cytokeratin and vimentin antibodies (compare fig. 4A and E), although occasionally some co-expression of both types of IFP is found in the columnar epithelium (fig. 4E). With a rabbit antiserum to human skin keratins a strong reaction is seen in the basal cells (fig. 4A) with a weak staining of the columnar epithelium. Similar observations have been made by Achttetster et al. 18 when using a guinea pig antiserum to epidermal keratins. The monoclonal cytokeratin antibody RCK102, which stains virtually all epithelia, reacts equally strongly with both basal and columnar cells (fig. 4B). Brawer et al. 24 have described a monoclonal cytokeratin antibody (recognizing several keratins from human stratum corneum) which in prostate specifically recognized basal cells of the glandular ducts. 35 A similar staining pattern found with the monoclonal cytokeratin antibody RCK103 is shown in fig. 4C. On the other hand monoclonal antibodies to cytokeratin 18 can be shown to react only with normal and hyperplastic columnar (secretory) epithelial cells (fig. 4D). 18 It can therefore be concluded that cytokeratin expression differs among the epithelial cell populations of the human prostate.

PROSTATE TUMORS

When prostate tumors of several degrees of malignancy (well-differentiated adenocarcinoma to anaplastic prostate carcinoma) were tested with different monoclonal cytokeratin antibodies, virtually all tumor cells were stained with the broadly cross-reacting cytokeratin antibody RCK102. The rabbit antiserum to skin keratin, however, gave only a weak to negative reaction in the tumor cells (fig. 4F, H). Similarly, the monoclonal antibody RCK103 recognizing only basal cells in normal and hyperplastic prostate also showed a negative or occasionally weak reaction in the tumor cells. On the contrary, the monoclonal antibody to cytokeratin 18, which reacted strongly with the "normal" columnar epithelial cells, also gave a strong positive reactivity pattern in virtually all tumor cells (fig. 4G, I).

Therefore we, in accordance with Brawer et al. 24 conclude that the differential staining pattern of the (monoclonal) cytokeratin antibodies available so far may assist in distinguishing hyperplastic from neoplastic prostate epithelium as well as in the recognition of basal cell hyperplasia, transitional cell metaplasia and premalignant changes. We can conclude from our data and those of Brawer et al. 24 that prostatic intratubular neoplasia may arise directly from the columnar (secretory) epithelial cells or that the cytokeratin profile changes during malignant transformation of the prostate basal cell layer.

PROSTATE TUMOR MODEL SYSTEMS

Prostate tumor model systems include in vitro systems, spontaneously occurring tumors, spontaneously transplantable tumors and chemically or hormonally inducible tumors and xenografts. One of the model systems that has been investigated elaborately is the Dunning rat prostate cancer model system. This system comprises a spectrum of different types of prostatic tumors with varying degrees of malignancy or biological behavior. One of the most malignant and highly metastatic sublines within this system is the MATLyLu tumor. 30 Immunohistochemical investigation of frozen sections from MATLyLu subcutaneous tumors as well as from lymph-node metastases showed a slightly positive reaction with the polyclonal cytokeratin antibodies and a moderate to strong reaction to the vimentin antibodies. This pattern contrasts with the strong reaction to the monoclonal antibody to cytokeratin 18 seen in this tumor line. Co-expression of vimentin and cytokeratin IFP was seen in the same cells using double label immunofluorescence (fig. 4J, K). Similar observations were made in tissue cultures of this tumor (fig. 4L, M). These data suggest that the MATLyLu tumor may originate from the columnar glandular epithelial cell layer in rat prostate.

NORMAL AND NEOPLASTIC TESTIS TISSUE

When frozen or paraffin sections of normal human testis (fig. 5A-F) are incubated with the antisera to cytokeratin, a strong positive reaction is only seen in the epithelial cells lining the ductus efferens and the rete testis (fig. 5E). No other cell types are positive for cytokeratin (fig. 5A), except for one case where we have found some cytokeratin positive cells in the seminiferous tubules (fig. 5B). This case, however, represented testis tissue in which spermatogenesis was lacking because of the presence of a tumor. Similar observations have been described by Miettinen et al. 40 and by Damjanov et al. 41 for intratubular atypical ("carcinoma in situ") cells in embryonal carcinoma-bearing testis. From the use of cytokeratin-polyepitide-specific monoclonal antibodies described above and twodimensional gel electrophoresis it can be concluded that the cells lining the rete testis contain cytokeratins 7, 8, 18 and 19 while the epithelium of the ductus efferens in addition contains cytokeratin 5. According to Achttetster et al. 18 these cells, ductus deferens and seminal vesicles also contain cytokeratins 5, 7, 8, 18, 19 and may contain cytokeratin 17.

The antibodies to vimentin do not stain the epithelial cells of the ductus efferens. Strikingly, however, a considerable
Fig. 5. Immunofluorescence micrographs of normal and malignant testis tissues. A–F, normal human testis shows strong positive reaction with cytokeratin antibodies in epithelial cells lining ductus efferentes and rete testis (F) and except for occasionally positive cells in some seminiferous tubules (B) no other cells are cytokeratin positive (A). Vimentin positive reaction is found in Sertoli and Leydig cells (C). Myoid cells surrounding seminiferous tubules were strongly positive with antibodies to desmin (D). Co-expression of cytokeratin (E) and vimentin (F) can be seen in epithelial cells of rete testis (E, F) and classic and anaplastic seminomas are positive for vimentin (H) and negative for cytokeratin (G) although cytokeratin-positive cells can occasionally be seen in such tumors. I, non-seminomatous testis tumors such as embryonal cell carcinomas show positive reaction with cytokeratin antibodies in tumor cells but not with the vimentin antibodies, which stain only stromal components. K, in L, in teratocarcinomas differentiation can be made between glandular and squamous components using monoclonal antibody to cytokeratin 18 (L) in combination with polyclonal keratin antiserum (K). A–L: x 150.
percentage of the epithelial cells of the rete testis were also positive for vimentin (fig. 5 F). Double-labeling experiments clearly proved this co-expression of cytokeratin and vimentin in rete testis-lining epithelial cells (compare fig. 5 E and 5 F). Also in the cytokeratin-positive cells present in the seminiferous tubules described above, co-expression of cytokeratins and vimentin was seen (not shown). Furthermore, a positive reaction with the antivimentin antibody is found in Sertoli cells and in Leydig cells (fig. 5 C). The Leydig cells, however, showed a variable reaction, with some cells negative for vimentin and others containing perinuclear clusters of this antigen. Fibroblasts and blood vessels were also stained with the antivimentin antibody. We could not find any staining reaction with any of the antibodies in spermatozoa. Ito et al., however, could show that human spermatozoa have a highly specialized cytoskeletal organization with vimentin forming band-like structures in the sperm head.

Myoid cells surrounding the seminiferous tubules were strongly positive with the antibodies to desmin (fig. 5 D). Some of these myoid cells could be shown to co-express desmin and vimentin as demonstrated in the double-label indirect immunofluorescence technique. The desmin antibodies did not stain any other cell types than muscle cells. Neurofilament antibodies stained only nerve cells.

SEMINOMAS

Classic seminomas, anaplastic seminomas in the testis and metastases of anaplastic seminomas are positive for vimentin. This staining reaction is, however, usually observed in a variable number of the tumor cells (fig. 5 H). Identical results are obtained with monoclonal and polyclonal vimentin antibodies. When incubated with polyclonal or monoclonal antibodies to cytokeratins almost all cases are negative (fig. 5 G), although in some of these cases few cytokeratin-positive cells scattered through the tumor tissue sections were seen. In a lymph node metastasis which was morphologically a pure anaplastic seminoma, a considerable fraction of cytokeratin positive cells was detected. This correlated well with the finding that the primary tumor in the testis was a mixed yolk-sac tumor/ seminoma.

Interestingly, in one of the cases classified as seminoma that we examined which had elevated levels of the serum tumor markers alpha-fetoprotein and beta HCG, it was obvious that some of the tumor cells were positive for cytokeratin. This immunohistochemical finding therefore confirmed the clinical diagnosis of a non-seminoma testis.

NON-SEMINOMATOUS TESTIS TUMORS

Non-seminomatous testis tumors, including primary and metastatic embryonal carcinomas, primary and metastatic endodermal sinus tumor, choriocarcinomas and teratocarcinomas react with cytokeratin antibodies but not with vimentin antisera. Only stromal components of the tumor were positive for vimentin. Double-label immunofluorescence microscopy shows that the vimentin and cytokeratin reaction pattern are mutually exclusive in these tumors (fig. 5 I, J). Different types of epithelial differentiation in teratocarcinomas can be identified using the different monoclonal cytokeratin antibodies. In, for example, with a monoclonal antibody to cytokeratin 18 (RIGE5) a distinction can be made between the glandular and the squamous component of the tumor (fig. 5 K, L). Haugen and Taylor and Trojanowski and Hickey have shown the presence of neural IF (neurofilaments and GAPAP) in teratomas. Detection of these IF in such testis tumors can be useful in their diagnosis, subclassification and investigation of their histogenesis and potentials of differentiation.

Based on these studies we conclude that intermediate filament typing of testis tumors provides a useful adjunct to routine histology and determination of serum markers in determining therapy and prognosis, especially in cases of anaplastic tumors.

TESTIS TUMOR MODEL SYSTEMS

Extensive studies have appeared with respect to teratocarcinoma model systems. The primary focus of these studies has been differentiation rather than oncology. Thus little information is available on the suitability of these model systems for studies of therapy and prognosis.

CONCLUSIONS

From the foregoing it may have become clear that the study of IFP expression in normal and neoplastic tissues of the urogenital tract is of exceptional interest, especially since it has produced important new insights in both fundamental cell biological and clinical aspects of this system. We have summarized current knowledge on this subject and shown that antibodies to IFP are important immunohistochemical reagents in the study of histogenesis and differentiation of urogenital tract tissues. Moreover, these antibodies are useful tools in tumor grading and tumor classification.

Acknowledgments. We wish to thank Mr. O. Moesker, Mrs. A. Huizmans, Mr. G. Schaart, Mr. B. Hendriks and Mr. P. Peelen for excellent technical assistance and all members of the Dept. of Urology and Pathology for providing the tumor material and for their help in tumor diagnosis.

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TISSUE SPECIFIC MARKERS IN UROLOGICAL CANCERS


