

CYTOMORPHOMETRIC AND HISTOMORPHOMETRIC EVIDENCE OF THE DEVELOPMENT OF
CARCINOMA IN SITU OF THE BLADDER IN PATIENTS FOLLOWED FOR LOW-GRADE
PAPILLARY BLADDER CARCINOMA

Mathilde E. Boon (1) and Engelbert C.M. Ooms (2)

(1) Leiden Cytology Laboratory, Groenesteeg 63, Leiden,
The Netherlands.

(2) Westeinde Ziekenhuis, Den Haag, The Netherlands.

ABSTRACT

Carcinoma in situ of the urinary bladder may develop in patients followed after transurethral resection of low-grade papillary bladder carcinoma. In principle, the former lesion has a good prognosis and needs only simple resection while the latter has a bad prognosis and requires aggressive therapy (radiotherapy and removal of the bladder). Therefore it is often of paramount clinical importance to establish this deterioration. In this paper is reported how histomorphometry and cytomorphometry can, in a quantitative way, express this shift.

Keywords: Bladder carcinoma, cytomorphometry, histomorphometry.

INTRODUCTION

Carcinoma in situ of the bladder is by definition a flat lesion with a high-grade atypia: the cancer cells have large nuclei. Lesions with smaller nuclei are classified in our laboratories as dysplasias (Boon et al., 1985). In contrast to low-grade papillary bladder tumors, these lesions have a high tendency to infiltrate, and than one finds high-grade solid growing bladder carcinoma with a poor prognosis. Therefore it is of great importance for the clinician and pathologist to be aware of the fact that carcinoma in situ can develop during follow-up of patients with resected low-grade papillary carcinoma.

In this paper we report our experiences with three of such patients. All three patients were treated with transurethral resection for their low-grade papillary bladder tumors, and were followed with cytology. In all three patients the cytologic report became "positive" during this follow-up (3-5 years after the resection of the papillary tumors). In these patients the first biopsy taken after the positive cytologic follow-up diagnosis was "negative": the sections contained exclusively denuded stroma, and all its surface epithelium had exfoliated. This phenomenon (also called "denuding cystitis") is characteristic for carcinoma in situ. The cancer cells are trapped in the Brunn's nests, and thus they cannot exfoliate in the bladder lumen. In our three cases, only in repeated biopsies, which contained these Brunn's nests filled with cancer cells, the diagnosis carcinoma in situ could be made, and the histomorphometry could be performed on the cancerous epithelium lining these Brunn's nests.

In this paper we report how the cytomorphometry of the cancer cells in the voided urine and the histomorphometry of the cancerous epithelium in the tissue sections express in a quantitative manner the, clinically important, shifts in these three patients followed for low-grade papillary bladder tumors.

HISTOMORPHOMETRY

Of all three patients, histomorphometry was performed on the sections of the resected papillary bladder tumor, and on the cancerous epithelium in the Brunn's nests. For this purpose, nuclei in the superficial layer, and in the deep layer were measured (see Figure 1), and, in addition, cells in the entire cancerous epithelium with giant nuclei were selected for morphometry. For histomorphometry of urothelial neoplasm, 50 cancer cells are selected from the basal layer (deep cells), 50 cancer cells from the superficial layer (B). In addition, 50 cancer cells with giant nuclei are chosen (giant cells) (see for detailed information Ooms et al., 1983). The histomorphometric grading is based on the mean values of the A, the B and the C cells (Ooms et al., 1985).

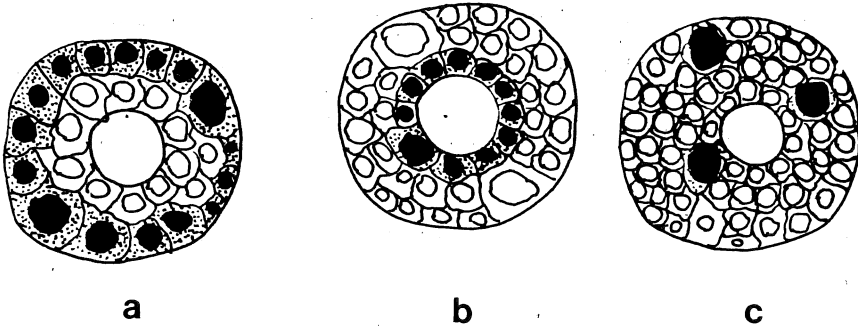


Fig. 1. Histomorphometry of cancerous epithelium lining the Brunn's nests. The measured cells are indicated by their black nuclei. The nuclear area was determined by planimetry, and the malignancy grading was computed with the acquired data.

Three cell types were selected for measurement: deep cells (A), superficial cells (B), and giant cells scattered in the cancerous epithelium (C). The three drawings are made from different sections of the same Brunn's nests lined with cancer cells, and illustrate the principles on which the histomorphometric grading according to Ooms et al. is based.

The mean values of the three cell types, and the resulting computer gradings are presented in Table 1.

Table 1.

Histomorphometrical Results and Computer Grading.

	papillary tumor		carcinoma in situ	
Patient I	deep:	32	deep:	64*
	superficial:	28	superficial:	56
	giant:	52	giant:	127
	Low grade		High grade	
Patient II	deep:	38	deep:	69
	superficial:	37	superficial:	48
	giant:	69	giant:	122
	Low grade		High grade	
Patient III	deep:	42	deep:	61
	superficial:	36	superficial:	41
	giant:	61	giant:	117
	Low grade		High grade	

* values μm^2

CYTOMORPHOMETRY

The cytomorphometry was performed on wet-fixed Papanicolaou-stained slides made from the sediments of freshly voided urine (Ooms et al., 1982). In this context it is important to stress that if the cytopreparatory technique used differs from ours, or when the cells are harvested from urine that is not processed immediately, the morphometric values can differ significantly from ours. The problem with measuring papillary cell groupings is that of often occurring nuclear overlapping. In each smear, 50 urothelial cells were measured. Except for the exclusion of cells with degenerative features, cells were selected at random.

In the samples prior to the resection of the papillary tumors, the cancerous cells were predominately in dense cell groupings, whereas in the later positive follow-up specimen the cancer cells were predominantly single.

The cytomorphometric findings are tabulated in Table 2. As in the histologic specimen the nuclear sizes of the cells (exfoliated form) the papillary tumors were much smaller than those of carcinoma in situ.

Table 2.

Cytomorphometry, nuclear values of wet-fixed Papanicolaou stained cells. (values μm^2)

	Papillary tumor		carcinoma in situ	
	mean	SD	mean	SD
Patient I	72	26	107	42
Patient II	58	18	130	56
Patient III	42	20	140	49

DISCUSSIONS AND CONCLUSIONS

Histomorphometric grading is far more reproducible than visual grading (Ooms et al., 1985). If this method is applied on cases of carcinoma in situ, special care has to be taken to measure intact epithelium. Therefore it is important to search for cancerous epithelium in the Brunn's nests. The cancerous surface epithelium, if present at all, is often partly desquamated; When this incomplete epithelium is used for morphometry, the histomorphometrical grading might be too low (Ooms et al., 1983).

In some cases, the blocks had to be sectioned further in order to obtain enough cancer cells in Brunn's nests for measurements. All three cases contained several Brunn's nests filled with cancer cells which could be used for quantitation.

The three cases presented herewith demonstrate that a shift in (cytomorphometric) pattern in patients followed after resection of low-grade bladder carcinoma can be an indication that a high-grade carcinoma developed. If the positive cytologic follow-up results are not directly confirmed in the biopsies, new biopsies must be taken from areas in which Brunn's nests can be anticipated, that is: protruding areas with a yellowish colour (comedo-like). The histomorphometric grading of cancerous epithelium in the Brunn's nests can be used to confirm the higher grade of the cancer.

Computer grading in cytomorphometry on voided urine shows a larger intraobserver and interobserver variability (unpublished data M.E. Boon and E.C.M. Ooms), but cytomorphometry is a very powerful method to show shifts in exfoliated cell populations.

Using quantitative findings it is easier to convince the clinician that the prognosis of the patient has changed in a negative way, and that more aggressive therapy is indicated.

REFERENCES

- Boon ME, Blomjous CEM, Zwartendijk RJ, Heinhuis RJ, Ooms ECM. Carcinoma in situ of the urinary bladder: clinical presentation, cytologic pattern and stromal changes. *Acta Cytologica*, 1985, IN PRESS.

- Ooms ECM, Blok ARP, Veldhuizen RW. The reproducibility of a quantitative grading system of bladder tumours. *Histopathology* 1985; 9: 501-509.
- Ooms ECM, Kurver PJH, Boon ME. Morphometric analysis of urothelial cells in voided urine of patients with low grade and high grade bladder tumours. *J Clin Pathol* 1982; 35: 1063-1065.
- Ooms ECM, Kurver PHJ, Veldhuizen RW, Alons CL, Boon ME. Morphometric grading of bladder tumors in comparison with histologic grading by pathologists. *Human Pathology* 1983; 14: 144-150.