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ORIGINAL ARTICLE

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Undifferentiated carcinoma of the prostate with small cell features: immunohistochemical subtyping and reflections on histogenesis

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Abstract To investigate the histogenesis of undifferentiated carcinoma of the prostate with small cell features we analysed the expression of neuroendocrine (NE) markers, the androgen receptor (AR), and prostatic-specific antigen (PSA) in 19 undifferentiated carcinomas of the prostate. The proliferative activity (MIB-1/Ki67) of the tumours was examined, and the clinical data reviewed. The results identified two groups: carcinomas in group 1 were positive for PSA and AR and negative for NE markers. The mean MIB-1 labelling index (LI) was 34.8% and the mean serum PSA value 56.4 ng/ml. Two of the 7 patients died within 12 months after tumour diagnosis. The tumours in group 2 were NE differentiated small cell carcinomas (SCC), which were negative for PSA and AR. The mean MIB-1 LI was 82.6% and the mean serum PSA value 7.1 ng/ml. Seven of the 10 patients died between 2 and 12 months after tumour diagnosis. Positive staining for NE markers in combination with negative staining for PSA and AR and a high MIB-1 LI substantiated the diagnosis of a NE-SCC. We suggest that this tumour has a stem cell origin and does not derive from a dedifferentiated adenocarcinoma or from benign NE cells of the prostatic epithelium. This clear distinction of NE-SCC from NE-negative undifferentiated carcinoma is in accordance with the differing biological behaviour and response to therapy of the two tumour entities.

Key words Prostatic carcinoma · Small cell features · Histogenesis · Neuroendocrine differentiation · Androgen receptor

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Introduction

Undifferentiated carcinoma of the prostate with small cell features has a poor prognosis, because it has usually reached an advanced stage by the time of diagnosis. Survival is measured in months [2, 14, 15, 19, 20]. It seems, however, that prostate carcinomas with small cell features are a heterogeneous group [18] and respond differently to therapy. Subclassification is therefore of therapeutic significance.

Neuroendocrine small cell carcinomas can be expected to behave like their counterparts in the lung, with sensitivity to chemo- and/or radiation therapy. Undifferentiated carcinomas deriving from poorly differentiated adenocarcinomas might be expected to be sensitive to antiandrogen therapy. Therefore, knowledge of the origin of an undifferentiated carcinoma of the prostate with small cell features will give a better understanding of the reasons for the obvious differences in the biological behaviour and response to therapy. The histogenesis of these tumours is unclear. One hypothesis is that they originate from a cell lineage that is different from prostatic epithelium [24], while another favours malignant transformation of normal prostatic NE cells [23]. A third hypothesis is based on dedifferentiation of the typical type of adenocarcinoma [7, 21], and finally an origin from a multipotential prostatic epithelial stem cell [9, 23] has been proposed.

In order to obtain additional information, we performed an immunohistochemical study of 19 cases of undifferentiated carcinoma of the prostate with small cell features. We used the NE markers neuron-specific enolase (NSE), synaptophysin (SNP) and chromogranin A (ChrA), the prostatic marker prostate-specific antigen (PSA), and a marker against the androgen receptor (AR). We analysed the proliferative activity of the tumours with Ki-67/MIB-1. The results were related to clinical data, such as serum PSA values at the time of tumour diagnosis, tumour therapy, death of disease rate, and survival time.

Materials and methods

Among a series of 3503 prostatic carcinomas seen from 1994 to September 1998, we diagnosed 19 cases of undifferentiated carcinoma of the prostate composed of small cells comprising at least 90% of the tumour tissue. Prostatic metastases from small cell carcinomas of other organs, including the urinary bladder and the lungs, were excluded clinically. Core needle biopsy material was available from 11 patients and transurethral resection material from 8 patients. The tissue was fixed in 4% buffered formalin and paraffin embedded. Serial 4-µm sections were cut for routine histology and immunohistochemistry. The following antibodies were used: PSA (Biogenex, Hamburg, Germany,1: 70), AR (Biogenex, Hamburg, Germany, 1:150), ChrA (Camon, Wiesbaden, Germany 1:1), NSE (Camon, Wiesbaden, Germany 1:1), SNP (Biogenex, Hamburg, Germany, 1:100), Ki-67/MIB-1 (Dianova, Hamburg, Germany 1:50).

Incubation time was 60 min for all primary markers except PSA (30 min). Before the application of MIB-1 and the antibody against AR, the slides were pretreated in a microwave oven (three times 600 W for 5 min). All immunohistochemical reactions were developed with the avidin–biotin-enhanced immunoperoxidase technique. Tonsillar tissue served as positive control for Ki-67/MIB-1, intestinal tissue for ChrA and NSE, and normal prostatic tissue for PSA and AR. The primary antibody was omitted for negative controls. All immunohistochemical analyses were interpreted by the same observer (B.H.). In each case 1000 cells were counted in two different areas of the small cell component of the tumour for each immunostaining (in an area of highest staining intensity). The nuclear Ki67/MIB-1 and AR labelling index (LI) and the immunostaining of cells for ChrA, NSE, SNP and PSA were expressed as percentages of positive cells.

In 5 patients additional prostatic tumour tissue was obtained by needle biopsy, transurethral resection (TUR), staging pelvic lym-

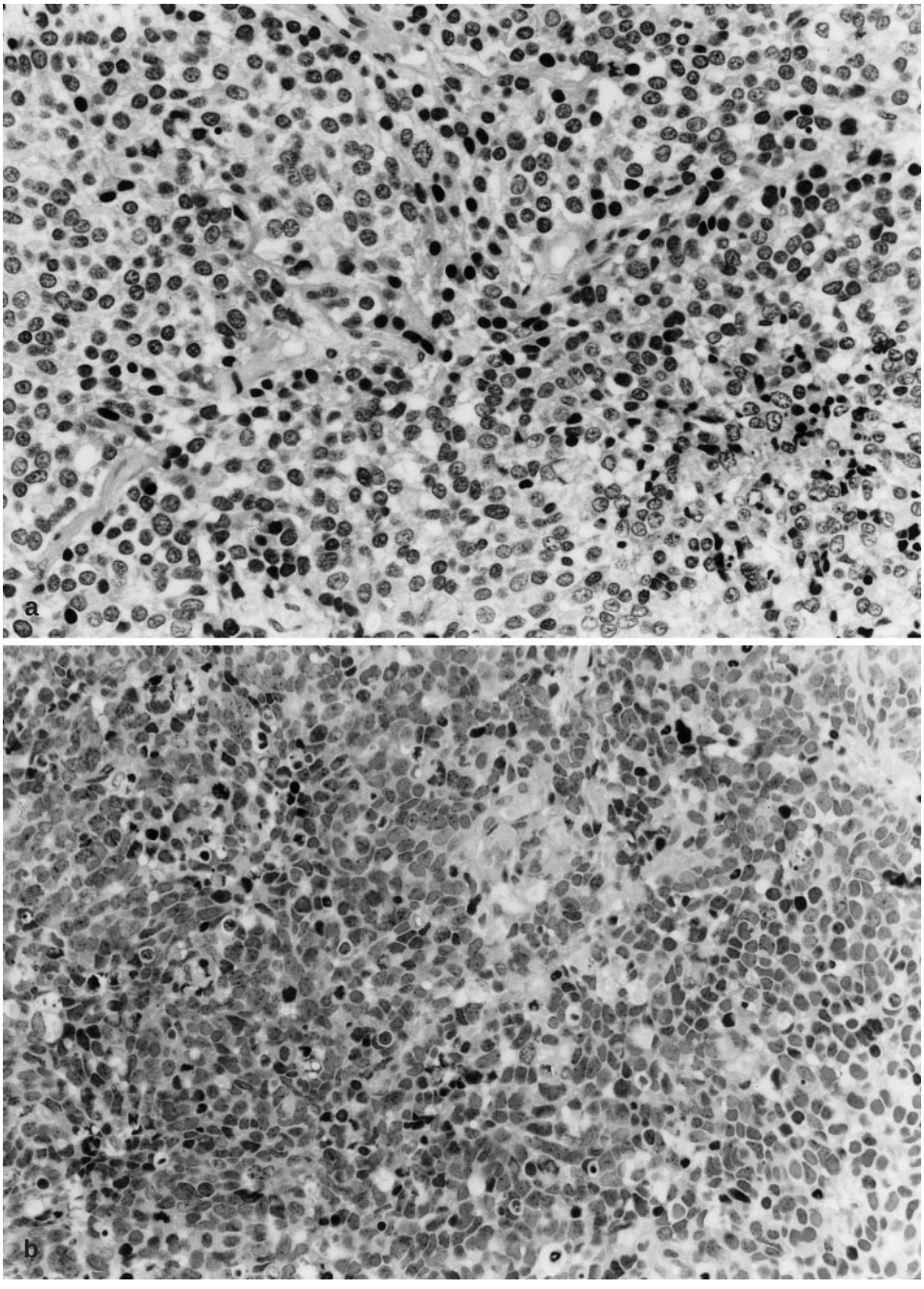

Fig. 1 **a** Undifferentiated adenocarcinoma with small cell features. Tumour cells with nucleoli in some cases, closely packed. No clear glandular differentiation. HE ×400
b NE differentiated small cell carcinoma of the prostate. Small, closely packed tumour cells with hyperchromatic nuclei. No glandular differentiation. HE ×400

phadenectomy or radical prostatectomy prior to or after the diagnosis of undifferentiated carcinoma with small cell features. Evaluation of this material was performed on sections stained with haematoxylin & eosin.

Clinical and follow-up data were obtained from the attending physicians.

Results

On routine haematoxylin & eosin-stained sections the 19 carcinomas showed conspicuous small cell features (Gleason grade 9–10), representing at least 90% of the entire tumour. This made it difficult to decide whether the tumours derived from an usual adenocarcinoma or a typical small cell carcinoma (Fig. 1a, b). Immunohistochemically, however, the tumours could be clearly separated into two groups. Those in group 1 (n=9) were positive for PSA and AR (Fig. 2a) but negative for ChrA, NSE and SNP (Table 1). The mean MIB-1 LI was 34.8%, with a range of 21.7–60.6% (Fig. 3a). The mean age of the patients was 69 (58–85) years. The mean serum PSA level was 56.4 (3.5–146) ng/ml. The material consisted of prostate biopsy specimens in 7 patients and TUR material in 2 patients. The therapy given was radiation and/or androgen deprivation in 3 patients, palliative TUR in 1 patient and radical prostatectomy in 3 patients. In 1 patient no information on therapy was obtainable. In another patient, staging lymphadenectomy revealed

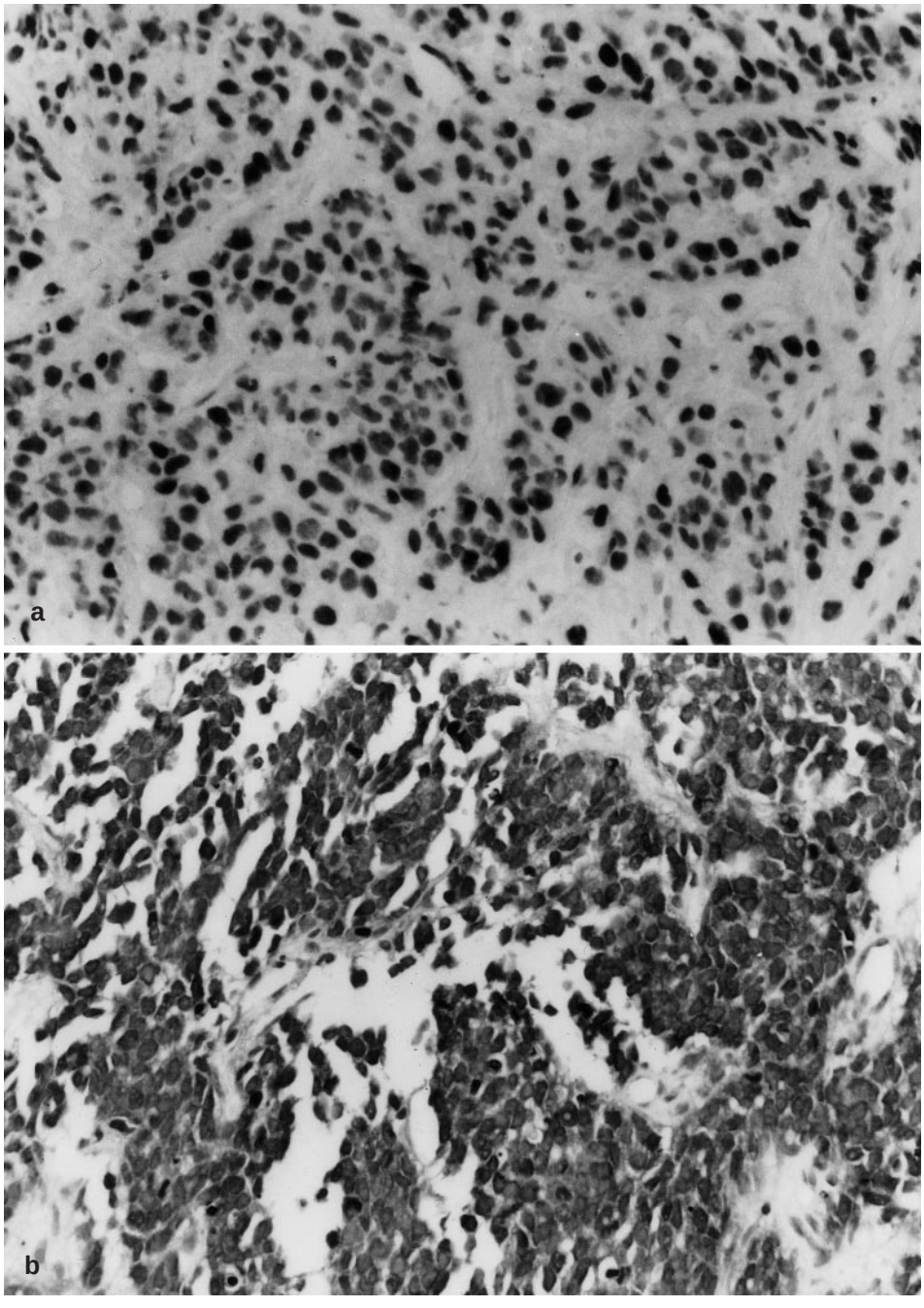

Fig. 2 **a** Positive reaction to androgen receptor of undifferentiated adenocarcinoma of the prostate with small cell features. ABC technique, ×400 **b** NE differentiated small cell carcinoma. Labelling with synaptophysin. ABC technique, ×400

Table 1 Neuroendocrine (*NE*) differentiated small cell carcinoma of the prostate versus undifferentiated adenocarcinoma with small cell features. Comparison of mean labelling index of different markers

	PSA %	AR %	NSE %	SNP %	ChrA %
Undifferentiated adenocarcinoma with small cell features (*n*=9)	61.8 (21.7–98.9)	90.3 (81.7 – 94.7)	0	0	0
NE-small cell cancer (*n*=10)	0	0	82.6 (37.6 – 97.6)	60.6 (12.7 – 81.7)	24.7 (0.1 – 81.7[a])

[a] Range of labelling index

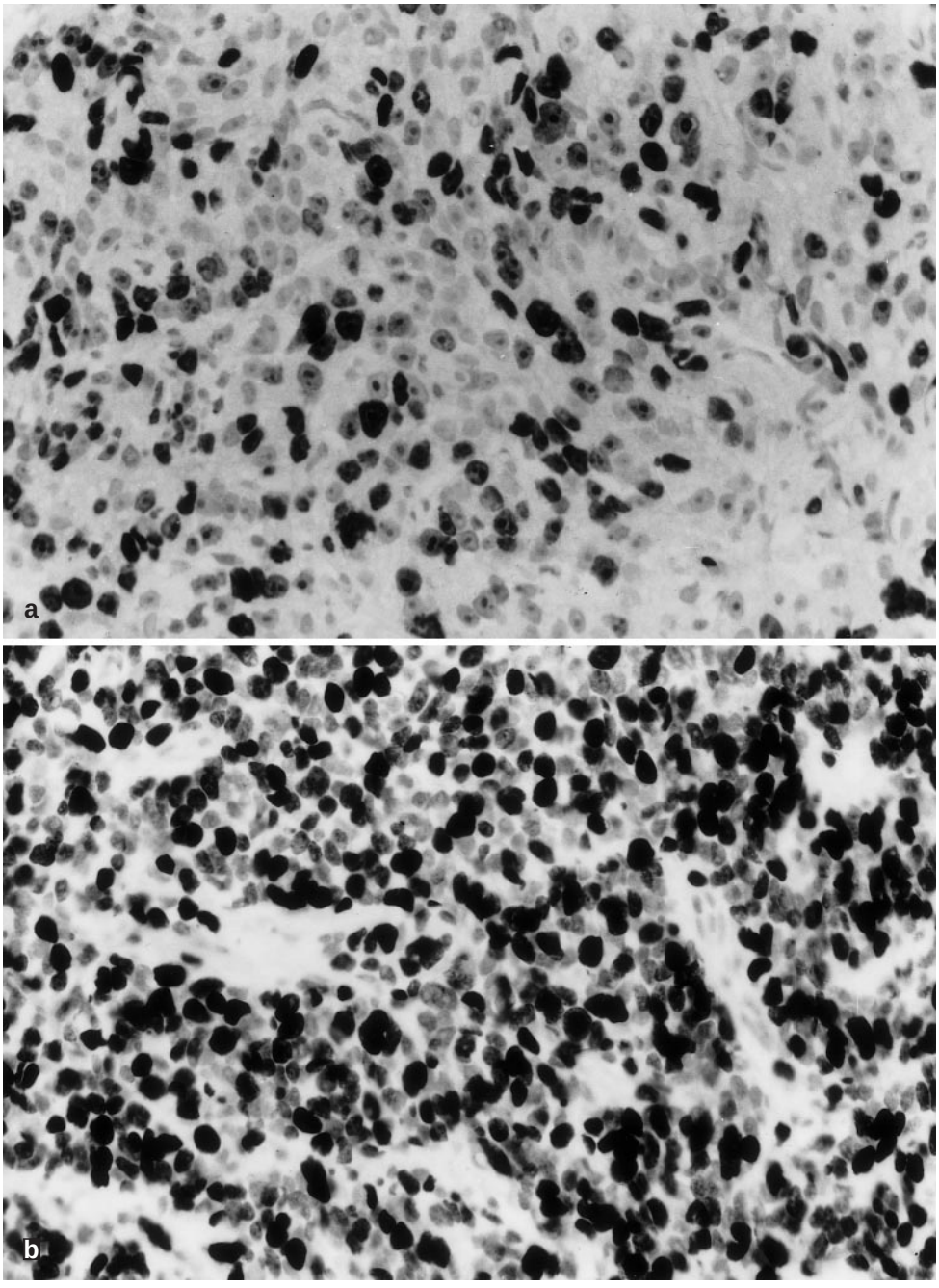

Fig. 3 **a** Undifferentiated adenocarcinoma with small cell features. MIB-1 labelling of tumour cells is clearly lower than in NE differentiated small cell carcinoma (**b**). ABC technique, ×560 **b** NE differentiated small cell carcinoma with very high MIB-1 labelling index. ABC technique, ×560

lymph node metastases; therefore radical prostatectomy was not performed.

The radical prostatectomy specimens revealed stage pT3pN0Mx and Gleason grade 9 tumours in 2 patients, and a stage pT4pN0Mx, Gleason grade 9 tumour in 1 patient. Dedifferentiation of glandular carcinoma was found in 2 patients with previous biopsies demonstrating Gleason grade 6 (3+3) and 7 (3+4) tumours. Biopsies and TUR material of group 1 patients did not reveal preneoplastic lesions. The radical prostatectomy specimens

Table 2 Summary of differences in clinical and immunohistochemical characteristics in undifferentiated adenocarcinoma with small cell features and NE differentiated pure small cell carcinoma (*AR* androgen receptor)

	Undifferentiated adenocarcinoma with small cell features (*n*=9)	NE differentiated pure small cell carcinoma (*n*=10)
Age (years)	69 (range 58–85)	64.6 (range 59–78)
Initial blood serum PSA (ng/ml)	56.4 (range 3.5–146)	7.1 (range 0.94–23.6)
MIB-1 LI (%)	34.8 (21.7–60.6)	82.6 (61.5–96.3)
NE marker	Negative	Positive
AR	Positive	Negative
Dead of disease (%)	2/7 (22.6)	7/10 (70)
Time to death (months)	12	7.7 (range 2–12)

revealed foci of high-grade prostatic intraepithelial neoplasia (PIN) without NE differentiation in all specimens.

Group 2 tumours (*n*=10) were positive for ChrA, NSE and SNP (Fig. 2b), but negative for PSA and the AR. They had a very high MIB-1 LI, the mean being 82.6% and the range 61.5–96.3% (Fig. 3b). The mean age of these patients was 64.6 (59–78) years. The mean serum PSA level of these patients was 7.1 (0.94–23.6) ng/ml. None of the patients had endocrine symptoms. In 5 of the patients the diagnosis was confirmed on needle biopsies, in another 4 patients TURP was performed with diagnostic and palliative intention. In 1 patient TURP was performed because of suspected local recurrence of a previously diagnosed NE-SCC of the prostate, which was treated with antiandrogen and radiation therapy. In 2 patients tumour therapy consisted of androgen deprivation. In 1 patient radical prostatectomy was performed. In 6 patients no therapy was initiated. None of the patients received chemotherapy. Preneoplastic lesions could not be identified; specifically, we found no foci of PIN.

Follow-up data were available from 17 of the 19 patients. The mean follow-up time was 19.2 months (2–46 months). Nine of the patients died (52.9%) after 2–12 months (mean 8.7 months). In the group of patients with PSA-positive carcinomas follow-up data were available in 7 patients; 2 died within 12 months. In the group of patients with NE differentiated pure small cell carcinoma 70.0% died (7/10) with a mean survival time of 7.7 months (2–12 months). A summary of the results is given in Table 2.

Discussion

Undifferentiated carcinomas of the prostate with small cell features are very rare [14]. In our series of prostatic carcinomas, spanning a period of 4.5 years, they account for 0.5% of all cases. Two groups are distinguishable, which differ clearly in their clinical and immunohistochemical characteristics. Group 1 tumours represent dedifferentiated adenocarcinomas of the prostate, reflecting a terminal aggressive change of these neoplasms [21, 22]. This process of dedifferentiation of the carcinomas was demonstrable in 2 of our patients, in whom previous prostate biopsies had shown the usual type of adenocarcinoma. Group 2 tumours represent NE-SCC of the prostate. Their highly aggressive character [14, 19], clearly exceeding that of usual prostatic adenocarcinoma, is reemphasized by our data.

The histogenesis of NE SCC of the prostate is still controversial. There are four competing theories that are most commonly discussed, which are summarized in the introduction: these postulate variously that the tumours derive from NE cells of the diffuse NE system or from normal prostate NE cells that undergo malignant transformation, that this entity is an undifferentiated variant of the usual adenocarcinoma [7, 21] or that it arises directly from a putative stem cell of the prostatic epithelium [9, 23].

Pearse postulated that NE tumours of the different organs are derived from the APUD cell system [16], which is now called the diffuse NE system (DNS). The assumed neural crest origin of most of the cells is not confirmed by embryological studies, which indicate an endodermal origin [10, 11]. The final differentiation of the cells is believed to occur under the influence of a specific site or organ, such as the lungs, the prostate or the urinary bladder [17]. Therefore, NE cells of the prostate are integrated into the stem cell concept [5], and a second cell lineage need not be implicated. Furthermore, it has been shown that NE cells in the normal prostatic epithelium represent postmitotic cells [6], which makes it unlikely that these cells suddenly start to proliferate and become a highly aggressive small cell carcinoma (Fig. 4). Based on the observation that SCC was detected during the course of a previously diagnosed adenocarcinoma it has been postulated that SCC represents a final step of dedifferentiation of the usual type of adenocarcinoma [21]. In studies on prostatic tumour cell lines it was shown that NE differentiation of the tumour cells can be induced by raising cyclic adenosine monophosphate activity [3]. Coexpression of PSA supports the prostatic epithelium origin of these NE cells [1]. Nevertheless, the cells seem to represent postmitotic cells [3, 6]. Therefore it is unlikely that they will suddenly start to proliferate and become a highly aggressive small cell carcinoma (Fig. 4). Schron et al. [21] reported on three cases of small cell carcinoma of the prostate detected during the course of treatment of a typical prostatic adenocarcinoma. Absence of prostate-specific acid phosphatase staining (PSAP) in the small cell component was interpreted as loss during the process of dedifferentiation and taken as confirmation of the assumption that SCC derives from the usual adenocarcinoma. However, the authors did not differentiate between the two groups of

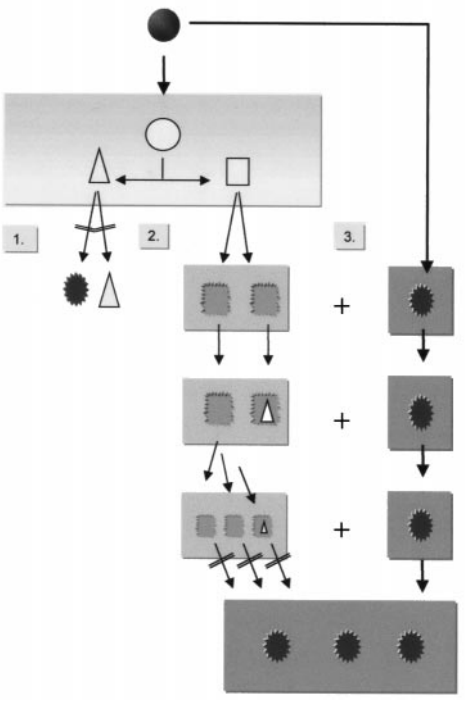

Fig. 4 Different concepts of the origin of SCC of the prostate (for detail see "Discussion"). The first theory that NE SCC (✹) develops from normal NE cells (△) seems unlikely, because normal NE cells are postmitotic. The second theory is based on the assumption that adenocarcinoma (▩) deriving from secretory epithelium (□) further dedifferentiates into carcinoma with small cell pattern (▩). Though transformation of tumour cells into cells with NE differentiation (▲) is possible, further dedifferentiation of these NE cells into NE SCC seems unlikely, because these cells have been proven to be postmitotic. The third concept postulates that NE SCC derives de novo from a putative stem cell (●) as an early step in dedifferentiation. Combinations with typical adenocarcinoma or dedifferentiated adenocarcinoma with small cell features are possible, but adenocarcinoma and NE SCC still represent independent tumor cell lineages. ○ Basal cell of normal prostatic epithelium

SCC discussed in our paper. As staining for NE differentiation was negative in 2 cases, at least these cases may have represented undifferentiated adenocarcinomas with a small cell pattern, and not pure NE SCC. Analysis of the proliferative activity would have provided additional information but was not performed.

In our opinion a direct stem cell origin for NE SCC seems to be the most convincing (Fig. 4). According to this theory, NE SCC represents an independent tumour cell lineage that is distinct from the usual prostatic adenocarcinomas and their variants at the stem cell level. This is supported by the fact that immunostaining of NE SCC for PSA and AR is negative and that the MIB-1 LI is extremely high, clearly exceeding that of even dedifferentiated adenocarcinomas. All this is in agreement with its very aggressive biological behaviour and the failure of the hormonal therapy normally given in typical adenocarcinomas. Consequently, it is obviously necessary to apply therapeutic regimens that take account of the biological characteristics of this tumour entity. Following experience in treatment of small cell carcinoma of the lung, chemotherapy, perhaps combined with radiation therapy, has been adopted with some success [8, 12]. New therapeutic approaches directed at neuroendocrine differentiation, as applied in endocrine tumours of other organs, might be of additional benefit [13]. None of our patients with NE small cell carcinoma received therapy specifically directed against NE tumours. If treated at all, their tumours were treated as typical adenocarcinomas. In conclusion, subtyping of undifferentiated prostatic carcinoma with small cell features is of clinical importance. This indicates that the pathologist has to recognize NE small cell carcinomas and has to indicate this diagnosis explicitly to the urologist. The urologist, on the other hand, has to be aware of the tumour biology, which differs from that of the typical adenocarcinoma with or without focal NE differentiation. This requires recognition of the NE character of this tumour entity when therapy is planned.

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References

1. Aprikian AG, Cordon-Cardo C, Fair WR, Reuter VE (1993) Characterization of neuroendocrine differentiation in human benign prostate and prostatic adenocarcinoma. Cancer 71:3952–3965
2. Aygun C (1997) Small cell carcinoma of the prostate: a case report and review of the literature. Md Med J 46: 353–356
3. Bang Y, Pirnia F, Fang W, Kang WK, Sartor O, Whitesell L, Ha MJ, Tsokos M, Sheahan MD, Nguyen P (1994) Terminal neuroendocrine differentiation of human prostate carcinoma cells in response to increased intracellular cyclic AMP. Proc Natl Acad Sci USA 91:5330–5334
4. Bonkhoff H, Remberger K (1996) Differentiation pathways and histogenetic aspects of normal and abnormal prostatic growth: a stem cell model. Prostate 28:98–106
5. Bonkhoff H, Remberger K (1998) Morphogenetic concepts of normal and abnormal growth in the human prostate. Virchows Arch 433:195–202
6. Bonkhoff H, Stein U, Remberger K (1995) Endocrine-paracrine cell types in the prostate and prostatic adenocarcinomas are postmitotic cells. Hum Pathol 26:167–170
7. Bostwick DG (1997) Neoplasms of the prostate. In: Bostwick DG, Eble JN (eds) Urologic surgical pathology. Mosby, St Louis, p 375
8. Debras B, Chautard D, Delva R, Pabot du Chatelard P, Guyetant S, Soret JY (1994) Small cell carcinoma of the prostate. Complete remission after chemoradiotherapy: apropos of a case. Prog Urol 4:569–571
9. Leader M, Kay E, Walsh CB (1998) Soft tissue neoplasms and other unusual tumors of prostate, including uncommon carcinomas. In: Foster CS, Bostwick DG (eds) Pathology of the prostate. (Major problems in pathology, vol 34) Saunders, Philadelphia, pp 364–383
10. LeDouarin N (1982) The neural crest. Cambridge University Press, Cambridge
11. LeDouarin N, Teillet MA (1973) The migration of neural crest cells to the wall of the digestive tract in the avian embryo. J Embryol Exp Morphol 30:31–48
12. Moore SR, Reinberg Y, Zhang G (1992) Small cell carcinoma of the prostate: effectiveness of hormonal versus chemotherapy. Urology 39:411–416
13. Oberg K (1998) Advances in chemotherapy and biotherapy of endocrine tumors. Curr Opin Oncol 10:58–65

14. Oesterling JE, Hauzeur CG, Farrow GM (1992) Small cell anaplastic carcinoma of the prostate: a clinical, pathological and immunohistological study of 27 patients. J Urol 147:804–807
15. Okada H, Gotoh A, Ogawa T, Arakawa S, Ohbayashi C, Kamidono S (1996) Two cases of small cell carcinoma of the prostate. Scand J Urol Nephrol 30:503–508
16. Pearse AGE (1969) The cytochemistry and ultrastructure of polypeptide hormone producing cells (the APUD series) and the embryologic, physiologic and pathologic implications of the concept. J Histochem Cytochem 17:303–313
17. Pearse AG, Takor T (1979) Embryology of the diffuse neuroendocrine system and its relationship to the common peptides. Fed Proc 38:2288–2294
18. Ro KY, Tetu B, Ayala AG (1987) Small cell carcinoma of the prostate. II. Immunohistochemical and electron microscopic studies of 18 cases. Cancer 59:977–982
19. Rubenstein JH, Katin MJ, Mangano MM, Dauphin J, Salenius SA, Dosoretz DE, Blitzer PH (1997) Small cell anaplastic carcinoma of the prostate: seven new cases, review of the literature, and discussion of a therapeutic strategy. Am J Clin Oncol 20:376–380
20. Sano K, Miyai K, Yoshida S (1997) Small cell carcinoma of the prostate: a case report. Int J Urol 4:321–323
21. Schron DS, Gipson T, Mendelsohn G (1984) The histogenesis of small cell carcinoma of the prostate. An immunohistochemical study. Cancer 53:2478–2480
22. Valle J, von Boguslawsky K, Stenborg M, Andersson LC (1996) Progression from adenocarcinoma to small cell carcinoma of the prostate with normalization of prostate-specific antigen (PSA) levels. Scand J Urol Nephrol 30:509–512
23. Weaver MG, Abdul-Karim FW, Srigley JR (1992) Paneth cell-like change and small cell carcinoma of the prostate. Two divergent forms of prostatic neuroendocrine differentiation. Am J Surg Pathol 16:1013–1016
24. Wenk RE, Bhagavan BW, Levy R, Miller D, Weisburger WR (1977) Ectopic ACTH, prostatic oat cell carcinoma and marked hypernatremia. Cancer 40:773–778