CLINICAL AND MORPHOMETRICAL STUDIES ON GRADING OF PROSTATIC CARCINOMA

klinische en morfometrische studies van het graderen van het prostaatcarcinoom

PROMOTIECOMMISSIE

Aan Arina
Aan Maaike
    Robert
    Pieter
    en Johan

CONTENTS

List of abbreviations

ANOVA	analysis of variance
BPH	benign prostatic hyperplasia
CT scan	computer tomography scan
DES	diethylstilbestrol
DF	degree of freedom
PC	prostate cancer
PSA	prostate specific antigen
SV	seminal vesicle
TNM system	Tumor - Node - Metastasis staging system
TUR	transurethral resection
UICC	Union International Contre le Cancer
VACURG	Veterans Administration Cooperative Urological Research Group

TNM-classification for prostate cancer (U.I.C.C. 1978)

T - CATEGORY (primary tumor - clinical examination)

T_0:   No tumor palpable

T_1:   Tumor intracapsular surrounded by palpably normal gland

T_2:   Tumor confined to the gland. Smooth nodule deforming contour, but lateral sulci and seminal vesicles not involved

T_3:   Tumor extending beyond the capsule with or without involvement of the lateral sulci and/or seminal vesicles

T_4:   Tumor fixed or infiltrating neighbouring organs

T_x: The minimum requirements to assess the primary tumor can not be met.

pT - CATEGORY (primary tumor - histopathological examination)

pT_0: No evidence of tumor found on histological examination of specimen

pT_1: Focal (single or multiple) carcinoma

pT_2: Diffuse carcinoma with or without extension to the capsule

pT_3: Carcinoma with invasion beyond the capsule and/or invasion of the seminal vesicles

pT_4: Tumor with invasion of adjacent organs

pT_x: The extent of invasion can not be assessed

N - CATEGORY (lymph nodes - clinical examination)

N_0:   No evidence of regional lymph node involvement.

N_1:   Evidence of involvement of a single ipsilateral regional lymph node.

N_2:   Evidence of involvement of contralateral or bilateral or multiple regional lymph nodes.

N_3:   Evidence of involvement of fixed regional lymph nodes (there is a fixed mass on the pelvic wall with a free space between this and the tumor).

N_4:   Evidence of involvement of juxta-regional lymph nodes.

N_x: The minimum requirements to assess the regional and/or juxta-regional lymph nodes can not be met.

<u>pN - CATEGORY</u> (lymph nodes - histopathological examination)
The pN-categories correspond to the N-categories

<u>M - CATEGORY</u> (distant metastases - clinical examination)
M_0: No evidence of distant metastases.
M_1: Evidence of distant metastases.
M_x: The minimum requirements to assess the presence of distant
metastases can not be met.

<u>pM - CATEGORY</u> (distant metastases - histopathological examination)
The pM-categories correspond to the M-categories

<u>HISTOPATHOLOGICAL GRADING</u>
G_1: high degree of differentiation
G_2: medium degree of differentiation
G_3: low degree of differentiation
G_x: grade can not be assessed

Clinicopathological staging for prostatic carcinoma (Whitmore)

Stage A: Tumor is not suspected on digital rectal examination. Diagnosed
by histopathological investigation of tissue obtained by simple
prostatectomy for presumed benign disease

Stage B: Tumor palpably confined to the prostate

Stage C: Tumor locally extensive beyond the prostatic capsule, but no
evidence of distant spread

Stage D: Metastatic prostatic carcinoma

Chapter 1

Locally confined prostatic cancer - prognostic factors and results of management of 484 patients

1.0 Introduction

In The Netherlands prostatic carcinoma is the third most frequent malignancy in men, after pulmonary and colorectal carcinoma and it is even the second cause of death from cancer in men [1]. In 1984 1,782 men died of prostatic carcinoma, which accounted for 9.2 % of the total number of deaths from cancer in men [2]. In the United States it is even the second most common malignancy in males and it is the third most common cause of death in men older than the age of 55 [3]. There is a slight increase in mortality from prostatic cancer: in 1975/1976 22.8 per 100,000 inhabitants of the Netherlands died of prostatic cancer. In 1984 this figure was 25 per 100,000 [1,2]. One of the causes of the increasing incidence of prostatic carcinoma is the increased life expectancy of the general population during the last decades. Prostatic carcinoma is almost exclusively a disease of elderly men with its peak incidence between 75 and 80 years of age. As in the previous decades many causes of death at a younger age (especially serious infectious diseases) were eliminated, more men can reach an age in which prostatic cancer usually manifests itself.

A large number of investigations on the etiology, histology, biochemistry, therapeutic possibilities and prognosis of prostatic carcinoma has been reported during recent years and many are still being carried out. Some milestones have been reached during the efforts of achieving control of prostatic carcinoma. In 1905 Hugh H. Young [4] gave his first report on radical perineal prostatectomy as a cure for prostate cancer and in 1941 Huggins and coworkers [5, 6] demonstrated the dramatic effects of estrogens as a hormonal therapy for prostatic carcinoma. Both therapies are still used widely all over the world.

Although hormonal therapy, either by estrogens or by orchiectomy, has been shown to be an excellent palliative treatment for prostatic cancer, it is still unknown whether it prolongs life. Until now a curative therapy of metastatic disease has not been found and the only way to cure this tumor is still its eradication during the early stages by radical prostatectomy or radiotherapy. Early diagnosis is not frequently made because of the lack of

symptoms in the early stages of the disease. Many patients present with a tumor in a locally advanced and even metastasized stage. The only ways to recognize prostatic carcinoma in an early stage is by regular rectal examination of the population at risk by means of palpation and/or ultrasound, possibly in combination with the marker substance prostate specific antigen (PSA). Only then it will be possible to offer curative therapy to more patients. This should result in a decrease of mortality.

1.1 Clinical background

The main purpose of this thesis is to correlate long term survival data of patients with localized prostatic carcinoma with histopathological prognostic parameters.

The results reported are all based on the data obtained from reviewing 484 cases of locally confined prostatic cancer and treated by Dr. Elmer Belt of Los Angeles, U.S.A. by means of total perineal prostatectomy between July 1928 and December 1971. A last comprehensive clinical review of the disease and survival status of the patients was carried out by the author in 1985. The series had been subject to several previous publications [7,8,9].

To achieve the main goal of the study the analysis of other than histopathological parameters related to prognosis is essential. Long-term observations and the attitude of the surgeon to use radical prostatectomy in T_3 tumors and the availability of most histological slides offered an opportunity to study long-term survival in relation to prognostic factors such as T-category, pT-category, grade and adjuvant hormonal therapy.

The patients have been followed very carefully during a long period of time. The shortest follow-up time for patients who were still alive at the time of the last review (November 1985) is now more than 12 years. The mean follow up is 23.3 years.

3

1.2 Material and methods

1.2.1 *Patients*

The average age of the 484 patients at the time of operation was 65.4 years, ranging from 42 to 86 (Fig 1).

N
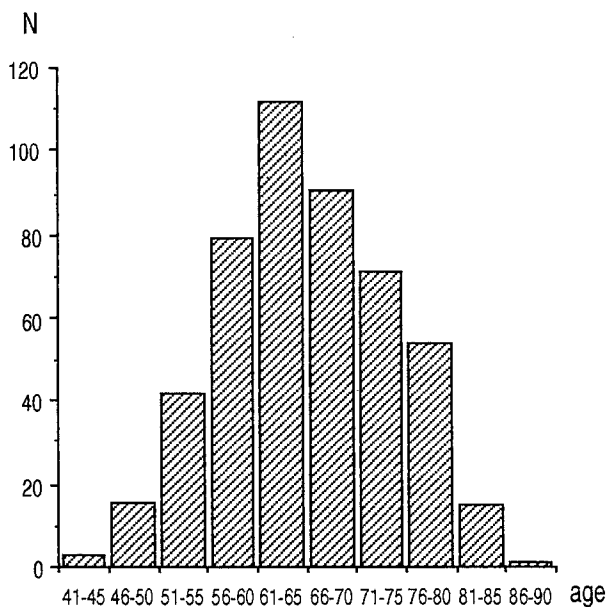
Fig 1 - Age distribution of 484 patients

Unfortunately there is no information about the race of the patients.

1.2.2 *Preoperative examinations*

Neither ultrasound nor CT scan was available in the years these patients were operated. Consequently, local tumor extension was estimated by rectal palpation only. Rectal examination showed a malignant appearing prostate in 259 patients. In 168 patients there was benign prostatic hyperplasia (BPH) on rectal examination, but there was also a suspicion of cancer. In 55 patients (11.4 %) there was absolutely no suspicion of carcinoma. These tumors were incidentally found in the pathological

specimens after prostatectomy for presumed benign hyperplasia. In two patients there were prostatic calculi, but cancer was also suspected on rectal examination. These data are summarized in table 1.

Table 1 - Rectal examination

	No. of patients	%
Definitely carcinoma	259	53.5
BPH, but question of malignancy	168	34.7
BPH with calculi, maybe tumor	1	0.2
Prostatic calculi, maybe tumor	1	0.2
BPH, no suspicion of tumor	55	11.4

The incidence of these incidentally found carcinomas in this series is consistent with the findings of others [10]. However, this consistency is coincidental, as most of the patients in Dr. Elmer Belt's series were referrals from other clinics. These patients with incidental carcinomas have been subject to a separate report [11], which is part of this thesis as chapter 3.

In 372 patients a nodule was palpated in the prostate. Most of the nodules were located in the lateral lobes, whereas only a minority of the nodules (6.8 %) was found in the apex.

1.2.3 Tumor Staging

Staging of the tumors was translated from data in the patient charts to the TNM system of 1978 / 1982 [12] and is presented in table 2.

Table 2 - Tumor Stages (TNM-system)		
T-category	No. of patients	% of total
T_0	55	11.4
T_1	1	
T_2	262	54.1
T_3	152	31.4
T_x	14	2.9
Total	484	

For convenience the one patient with a T_1 tumor was grouped together with the group of patients with a T_2 tumor, resulting in a group of T_{1-2} tumors.

No lymphangiograms and no lymph node biopsies or lymphadenectomies were done in the patients reported in this paper. At the time this series was built up knowledge of early lymph node metastases was poor and it was generally accepted that prostatic carcinoma spread to bone and other distant sites first.

In order to diagnose distant metastases skeletal X-ray photographs were performed and serum phosphatases were estimated. Bone scans were not performed until after 1967.

1.2.4 The Operation

All patients were operated on in the way described by Belt in his paper on the technique of total perineal prostatectomy [13]. As was mentioned before, no lymph node dissection was done.

The amount of operative complications was low. In 463 patients no major complications were encountered. In four patients there was a perforation of the rectum during the dissection. Thus, although Belt used a seemingly dangerous access route beneath the anal sphincter, the rectum was

perforated in only 0.8 %.

In total there were postoperative complications in 164 patients, accounting for a complication rate of 33.8 %. (Table 3)

Postoperative incontinence was regarded as urine loss existing longer than one year in duration. This was the case with 109 patients. Nineteen patients were treated surgically for incontinence. 48 patients needed either a urinary bag or a Cunningham clamp. In 42 patients the incontinence was slight and special measures were not necessary. So in 67 patients there was a significant incontinence after operation. This accounts for 13.8 % of all patients.

1.2.5 Postoperative tissue examination

The slides of 346 radical prostatectomy specimens were available for review. All these slides have been regraded by Dr. Mostofi, using his grading system [14]. The results of grading have been published [15, 16] and are part of this thesis [chapters 4, 5 and 6].

1.2.6 Statistics

The technique of Kaplan and Meier [17] was used to establish survival curves. As many patients in this series died of other causes than prostatic carcinoma the survival curves were corrected for intercurrent and unknown causes of death. In this way the impact of death of cancer is demonstrated more clearly. The Logrank test was applied to evaluate the differences in the survival curves.

1.3   Results

1.3.1 Staging

As may be expected there is a correlation between the clinical stage of the tumor and survival. Table 4 shows that the patients whose tumors are

Table 3 - Postoperative complications in 164 patients

	No.
Perineal fistula	6
Wound infection	18
Wound disruption	8
Hemorrhage	11
Perineal hematoma	1
Acute retention	1
Contracture of the bladder neck	6
Chills, fever	7
Pyelonephritis	1
Oliguria	1
Ureteral obstruction	7
Epididymitis	7
Residual urine	2
Low blood pressure	2
Incontinence	109
Total number of complications	187

clinically judged to have extended beyond the confines of the prostate have a significant worse prognosis than the patients who have localized tumors on rectal examination.

Table 4 - Death from prostate cancer by clinical stage
(Patients with unknown death causes are excluded)

	Total No. of patients	Total No. of deaths	Deaths from prostate cancer		p-value
			No. of deaths	% of dead	
T_0	55	46	11	23.9	
T_{1-2}	263	203	51	25.1	n.s.
T_3	152	122	53	43.4	p < 0.001
T_x	14	12	2	16.7	
Total	484	383	117	30.5	

Total prostatectomy specimens offer an opportunity to determine accurately the true histological extent of the tumor. With the knowledge of the T and pT categories one can study the possible error made at the time of staging by rectal examination. Comparing the pre-operative T-categories with the postoperatively obtained pT-categories it is evident that considerable understaging exists. Table 5 shows that the greatest errors are made by determining the T_0 and T_{1-2} categories. Only 14 out of 55 T_0 tumors proved to be what they should be: focal tumors, limited to a small area of the prostate.

52.4 % of the tumors in category T_{1-2} postoperatively were shown to be more extensive and belong to category pT_3. So in this series there is an understaging of 52.4 %. T_3 tumors were understaged in only 15.8 % of the

		pT$_1$		pT$_2$		pT$_3$		pT$_X$	
		No.	%	No.	%	No.	%	No.	%
T$_0$	55	14	25.5	19	34.5	20	36.4	2	3.6
T$_{1-2}$	263	-	-	123	46.8	138	52.4	2	0.8
T$_3$	152	-	-	24	15.8	125	82.2	3	2.0
T$_X$	14	-	-	6	42.9	5	35.7	3	21.4
Total	484	14		172		288		10	

Table 5 - Comparison T and pT categories

cases. Rectal examination really is most inaccurate in establishing the real extent of the tumor. In the categories T_{1-2} and T_3 there was an agreement between preoperative and postoperative tumor stages in only 51.2 % of the cases (248 out of 484). Unfortunately as the histological extent of the tumor was not re-evaluated, infiltration of the prostatic capsule and its penetration cannot be differentiated. The impact of this staging error on prognosis is seen in Table 6. Only the patients with a pT_1 tumor do significantly better than those with a T_0 tumor.

Table 6 - Death from prostate cancer by clinical and histopathological stage
(Patients with unknown death causes are excluded)

	Total No. of patients	Total No. of deaths	Deaths from prostate cancer		p-value
			No. of deaths	% of dead	
T_0	55	46	11	23.9	$p < 0.05$
pT_1	14	12	1	8.3	
T_{1-2}	263	203	51	25.1	not significant
pT_2	172	124	24	19.4	
T_3	152	122	53	43.4	not significant
pT_3	288	236	91	38.5	
T_x	14	12	2	16.7	not significant
pT_x	10	9	1	11.1	

Figure 2 shows the survival curves for patients whose tumors were preoperatively staged T_2 and T_3.
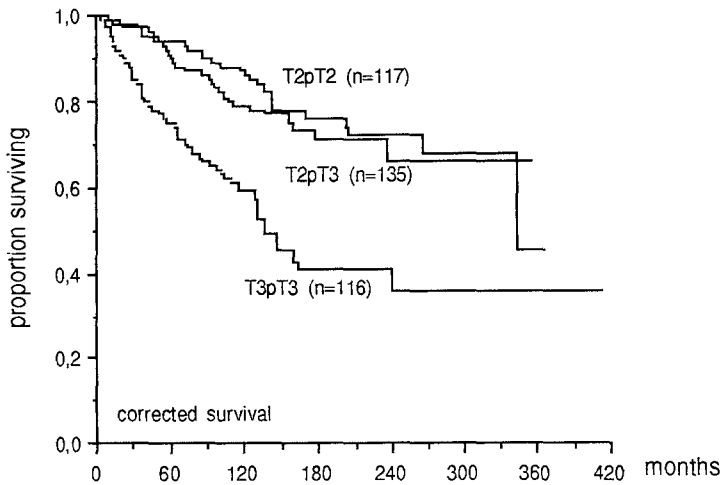
Figure 2 - For intercurrent death corrected survival curves for T2 and T3 tumors

These tumors, who were clinically staged as T_2 or T_3 were postoperatively subdivided in T_2pT_2, T_2pT_3 and T_3pT_3. The patients whose tumors were staged T_3 do significantly worse (p < 0.001) than those whose tumors were T_2. Surprisingly the patients with T_2 tumors, but whose tumors were staged pT_3 postoperatively have a prognosis which is identical to the patients with pT_2 tumors. Apparently, this staging error does not influence prognosis. However, as mentioned before, a differentiation between infiltration of and penetration through the prostatic capsule was not possible retrospectively.

1.3.2 Seminal vesicle invasion and extraprostatic tumor growth

Apart from studying the impact of the clinical and histopathological stages on survival it was also examined whether invasion of the seminal vesicles by tumor without further signs of extraprostatic tumor growth carried a better prognosis than patients with tumor growth beyond the

12

confines of the prostate. There were 51 patients whose slides showed invasion of the seminal vesicle as the only sign of tumorgrowth beyond the confines of the prostate. Survival of these patients was compared with that of the patients who had clear perforation of the prostatic capsule, but no visible invasion of the seminal vesicles. Figure 3 shows the corrected survival curves for these two groups.
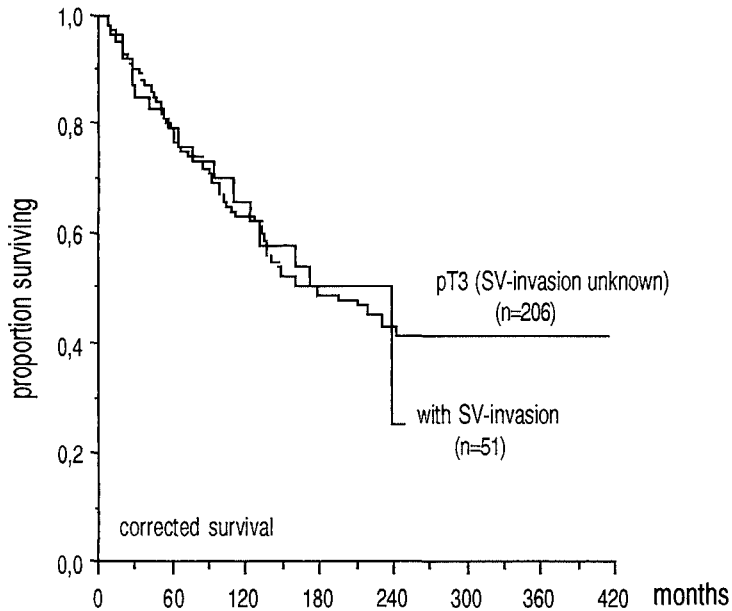
Figure 3 - Survival curves for patients whose tumors show seminal vesicle invasion only and for patients with tumorgrowth in the periprostatic tissues

It is evident that seminal vesicle invasion alone does not carry a better prognosis than tumors with extraprostatic growth with or without seminal vesicle invasion and these tumors should really be included in the group of pT_3 tumors.

1.3.3 *Grading*

It is well-known that the grade of malignancy of the tumor correlates with the prognosis. Table 7 shows the impact of grade and stage on the death rates. As can be seen the high grade tumors show the largest percentages of deaths from carcinoma. It is also shown that the impact of the T category on prognosis is far less than the impact of grade. This difference diminishes when grade is compared with pT categories (Table 8).

Grading is extensively dealt with in the next chapters of this thesis.

1.3.4 *Adjuvant hormonal therapy*

For various reasons a group of 188 patients received estrogens and/or castration as an adjuvant therapy during a certain period after operation. 34 of them again received estrogen treatment when local recurrence or metastases were discovered. 46 patients received estrogen therapy only when local recurrence or metastases were discovered. As can be seen in figures 4 and 5 there is no significant difference in survival (both in the low and in the high stage tumors) whether the patients received adjuvant hormonal therapy or not.
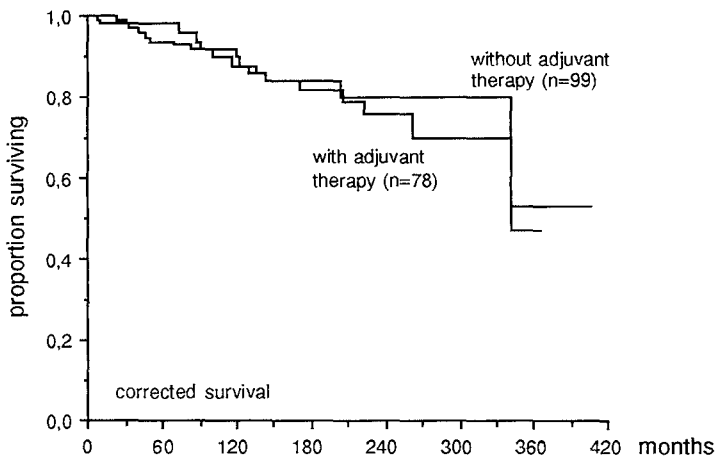
Figure 4 - Survival curves for patients with tumors Stage pT1-2
and with or without having received adjuvant hormonal treatment

14

Table 7 - Death from prostate cancer by clinical stage and grade

(Patients with unknown death causes are excluded)

Grade	T_0 Total No. of deaths	Deaths from PC No. of deaths	% of dead	T_{1-2} Total No. of deaths	Deaths from PC No. of deaths	% of dead	T_3 Total No. of deaths	Deaths from PC No. of deaths	% of dead
G_1	16	1	6.2	26	3	11.5	13	4	30.8
G_2	26	8	30.8	140	32	22.9	76	29	38.2
G_3	4	2	50.0	37	16	43.2	33	20	60.6
Total	46	11	23.9	203	51	19.4	122	53	43.4

Table 8   -   Death from prostate cancer by histopathological stage and grade

(Patients with unknown death causes are excluded)

| Grade	T_0 Total No. of deaths	T_0 Deaths from PC No. of deaths	T_0 Deaths from PC % of dead	T_{1-2} Total No. of deaths	T_{1-2} Deaths from PC No. of deaths	T_{1-2} Deaths from PC % of dead	T_3 Total No. of deaths	T_3 Deaths from PC No. of deaths	T_3 Deaths from PC % of dead
G_1	8	0	-	30	4	13.3	15	4	26.7
G_2	4	1	25.0	80	16	20.0	158	52	32.9
G_3	0	-	-	14	4	28.6	63	35	55.6
Total	12	1	8.3	124	24	19.4	236	91	38.5
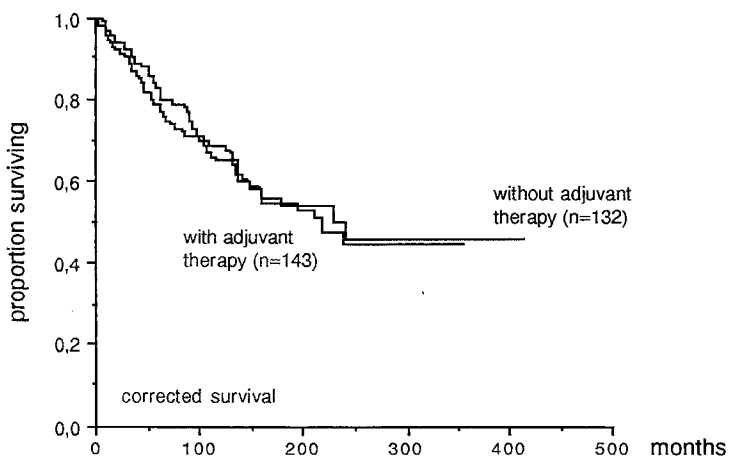
Figure 5 - Survival curves for patients with tumors Stage pT3
and with or without having received adjuvant hormonal treatment

1.4   Discussion

It is a well-known fact that the local extent (T-category) of prostatic carcinoma at the time of diagnosis is one of the factors that determine the prognosis of the disease and many authors stressed the importance of an accurate staging system [3,18, 19]. Catalona and Stein [20] found a 40 % understaging in 96 patients with stage A or B disease. They found early treatment failures in 5 patients and each of them were clinically understaged. Furthermore they found a higher predictive value of staging and grading combined than of each of them separately. Elder and associates [21] found that in the clinical stage B2 the prognosis of those tumors that showed extraprostatic extension on histological examination was poorer than for the patients with a B2 tumor confined to the prostate. In the present study histological tumor understaging ($T_{1-2}pT_{1-2}$ versus $T_{1-2}pT_3$) did not have a negative influence on prognosis. Tumors, clinically thought to be confined to the prostate but pathologically extended at least

17

into the capsule behaved as if they were really confined to the gland. Those patients who were thought to have T_3 tumors and in whom the local extension was confirmed survived significantly shorter (fig 2). Unfortunately, a differentiation between infiltration and penetration of the capsule was not made in this series of patients. A similar observation was made by Bosch and associates who found a significant difference in progression of disease between patients with a tumor staged T_3pT_3, and those with a tumor staged $T_{0-2}pT_3$ [22]. Another significant difference in survival was detected for clinical and pathological stage T_0 tumors. So, in localized prostatic cancer clinical stage is a prognostic factor of only limited clinical value, mainly because of the difficulties to accurately determine tumor extension prior to treatment. However, one must realize that no lymphadenectomies were done in this series of patients.

Grade as a prognostic factor is the main issue of this thesis. In a previous study on this material it was shown that especially differentiation and nuclear variation in size and shape show a good correlation with survival and death due to prostate cancer [15, 16]. In this study the prognostic importance of grade seems to be of greater value than of clinical stage and as important as pathological stage.

The impact of adjuvant hormonal therapy on prognosis in this study is questionable. Both in the group of patients with stage pT_{1-2} tumors and in the group of patients with tumor growth beyond the confines of the prostate (pT_3) there is no beneficial effect of adjuvant hormonal treatment on survival. There are hardly any papers that report on the long-term effect of adjuvant hormonal treatment. In the first VACURG study [23] in the stages I and II radical prostatectomy plus DES was compared with radical prostatectomy plus placebo. The study showed no significant advantage of hormonal treatment. On the contrary: the study demonstrated clearly the cardiovascular hazards of DES in doses of 5 mg daily. Zincke and associates [24] reported on 101 patients with pathological stage C or D1 disease. 42 of these patients received adjuvant hormonal treatment, being castration, estrogens or both. They found that for the 12 out of 49 patients

with stage C disease, who received castration, there was no improvement in total survival or survival free of progression as compared with the 37 patients who did not receive this form of adjuvant treatment. On the contrary, there was a statistically significant longer progression free survival of the 30 patients with stage D1 disease who received an adjuvant castration as compared with 22 patients who had no orchiectomy. Overall survival also in the D1 group was not influenced.

From the present study it may be concluded that tumor stage is an important parameter in the estimation of the prognosis of the patient with prostate cancer. Clinical understaging does have an important impact on the prognosis. However, microscopic extension through the prostatic capsule does not worsen the prognosis in a significant way. Tumor grade has an equally prognostic importance.

1.5 References

1. Central Bureau of Statistics. Kanker: Morbiditeit en Mortaliteit 1975 / 1976. Staatsuitgeverij, 's-Gravenhage, 1979

2. Central Bureau of Statistics. Compendium of health statistics of the Netherlands 1986. Staatsuitgeverij, 's-Gravenhage, 1986

3. Catalona WJ, Scott WW: Carcinoma of the prostate. In: Walsh, Gittes, Perlmutter, Stamey (eds): "Campbell's Urology", Ed 5. Philadelphia, WB Saunders Co, 1463-1534, 1986

4. Young HH: The early diagnosis and radical cure of carcinoma of the prostate. Being a study of 40 cases and presentation of a radical operation which was carried out in 4 cases. Johns Hopkins Hosp Bull 16:315-321, 1905.

5. Huggins C, Hodges CV: Studies on prostatic cancer. I. The effect of castration, of estrogen and of androgen injection on serum phosphatases in metastatic carcinoma of the prostate. Cancer Res 1:293-297, 1941

6. Huggins C, Stevens RE, Hodges CV: Studies on prostatic cancer. II. The effect of castration on advanced carcinoma of the prostate gland. Arch Surg 43:209-223, 1941

7. Turner RD, Belt E: A study of 229 consecutive cases of total perineal prostatectomy for cancer of the prostate. J Urol 77: 62-77, 1957

8. Belt E, Schroeder FH: Total perineal prostatectomy for carcinoma of the prostate. J Urol 107: 91-96, 1972

9. Schroeder FH, Belt E: Carcinoma of the prostate: A study of 213 patients with stage C tumors treated by total perineal prostatectomy. J Urol. 114: 257-260, 1975

10. Battaglia S, Barbolino G, Botticelli AR: Early (Stage A) prostatic cancer. Virchows Arch Pathol Anat 382: 245-259, 1979

11. Schröder FH, Blom, JHM, Hop WCJ, Mostofi FK: Incidental carcinoma of the prostate treated by total prostatectomy. The prognostic impact of microscopic tumor extension and grade. World. J. Urol. 1: 15-23, 1983

12. Spiessl B, Scheibe O, Wagner G, (ed.): U.I.C.C. TNM-atlas. Illustrated guide to the classification of malignant tumours. Springer-Verlag, New York, 1982

13. Belt E, Ebert CE, Surber AC: A new anatomic approach in perineal prostatectomy. J Urol 41:482-497, 1939

14. Mostofi FK: Grading of prostatic carcinoma. Cancer Chemoth Rep Part 1, 59:111-117, 1975

15. Schröder FH, Blom JHM, Hop WCJ, Mostofi FK: Grading of Prostatic Cancer (I) - An Analysis of the Prognostic Significance of Single Characteristics. The Prostate 6:81-100, 1985

16. Schröder FH, Blom JHM, Hop WCJ, Mostofi FK: Grading of Prostatic Cancer (II) - The Prognostic Significance of the Presence of Multiple Architectural Patterns. The Prostate 6:403-415, 1985

17. Kaplan EL, Meier P: Nonparametric estimation from incomplete observations. Jr Am Stat Ass 53:457-481, 1958

18. Whitmore WF: Natural history and staging of prostate cancer. Urol Clin N Am 11:205-220, 1984

19. Spirnak JP, Resnick MI: Clinical staging of prostatic cancer: new modalities. Urol Clin N Am 11:221-235, 1984

20. Catalona WJ, Stein AJ: Staging errors in clinically localized prostatic cancer. J Urol 127:452-456, 1982

21. Elder JS, Jewett HJ, Walsh PC: Radical perineal prostatectomy for clinical stage B2 carcinoma of the prostate. J Urol 127: 704-706, 1982

22. Bosch RJLH, Kurth KH, Schroeder FH: Surgical treatment of locally advanced (T3) prostatic carcinoma: Early results.J Urol 138: 816-822, 1987

23. Byar DP: VACURG studies on prostatic cancer and its treatment. In: Tannenbaum M (ed): "Urologic Pathology: The Prostate", Philadelphia, Lea & Febiger, 241-267, 1977

24. Zincke H, Utz DC, Taylor WF: Bilateral pelvic lymphadenectomy and radical prostatectomy for clinical stage C prostatic cancer: role of adjuvant treatment for residual cancer and in disease progression. J Urol 135: 1199-1205, 1986

Chapter 2

Clinical studies on grading of prostatic carcinoma Historical review

2.1 Broders' system

Since Broder's publications in the 1920's [1, 2] many attempts have been made to correlate the histological findings of tumors with the course of the disease.

Realizing that the percentage of undifferentiated cells in a tumor determines the malignant potential, Broders proposed a grading system based upon the proportion of differentiated and undifferentiated cells in the tumor. When about 75 % of a tumor consisted of differentiated epithelium and about 25 % was undifferentiated, the tumor was graded 1. If the amounts of differentiated and undifferentiated epithelium were about equal, the tumor was graded 2. In grade 3 tumors the undifferentiated epithelium formed about 75 % and the differentiated about 25 % of the growth. In grade 4 tumors there was no tendency of the cells to differentiate. Broders graded and followed 537 cases of epithelioma of the lip, finding a very close correlation between the tumor grade and the prognosis of the patients after treatment (table 1)

Table 1 - Correlation between grade and
death rate in epithelioma of the lip [1]

Grade	Dead from epithelioma
1	0 %
2	54.9 %
3	84.2 %
4	100 %

Kahler [3] was the first who applied Broders' system to prostate cancer. He examined 189 post-mortem specimens. Ninety-nine of these 189 tumors were not confined to one lobe of the prostate. In these 99 patients distant metastases were found in 33 % of those in grade 1, in 50 % of those in grade 2, in 84 % of those in grade 3 and in 100 % of those in grade 4. These figures showed that it was justified to apply Broders'

classification also to adenocarcinomas of the prostate.

Greene and Simon [4] studied 83 cases of occult carcinoma of the prostate (incidentally found after prostatectomy for presumed benign disease). The tumors were graded 1 in 71 cases, grade 2 in eleven cases and grade 3 in the remaining case. The system of grading was the Broders system. Five year survival in their series was 70.7 % and ten year survival was 39.4 % with conservative therapy (mostly hormonal). They suggested a correlation between the good survival rates in their patients and the fact that almost all tumors were of low grade. In fact they did not show the correlation between grade and survival.

Broders was not the first to realize that tumors did not all behave equally aggressive. Virchow, cited by Evans and associates [5], had a rather clear conception of the fact that certain groups of cancers varied in degree of malignancy. In his well known lectures in the 1850's he clearly stated: *"Cancer is not malignant because it contains heterologous cells, nor cancroid benignant because its cells are homologous - they are both malignant and their malignity only differs in degree."*

Hansemann [6], in 1893, was the first to suggest the practicability of recognizing grades of clinical malignancy based on histological degrees of abnormality. He used the term **anaplasia**, (now used as practically synonymous with "undifferentiation"), as descriptive of the essential quality of cancers. Later, in 1902, he again published a report on actual attempts at grading cancer, showing statistically that it is applicable in routine practice. His work attracted little attention, and it was not until 1920 that Broders published his first, above mentioned, paper on the grading of epithelioma of the lip.

2.2 Muir's classification

Muir [7] studied 48 cases of prostatic carcinoma and divided the tumors into three groups, based on the tendency of the primary growth to

reproduce the prostatic tubules. Contrary to Broders he also added some cytological parameters:

Group I (high degree of differentiation) included those cases in which the prominent feature of the growth was tubule formation. The tubules were smaller than those in the normal prostate and were sometimes branched. The cells were regular and there were no mitoses.

In **group II** the tubules were less frequent and not so well formed. There were masses of spheroidal cells which were less regular and might show mitotic figures.

In **group III** (undifferentiated tumor) there was little or no tendency for tubule formation. The masses of cells which determined the histological picture resembled closely round-celled sarcoma. Variation in the cells and mitotic figures were more common in this group.

Certain cases easily fell in group I and group III, while group II merely contained "border" cases. With this classification Muir was able to correlate the presence of metastases at autopsy with the histological picture of the tumor in 24 out of his 48 patients (table 2).

Table 2 - Muir's classification. Correlation between histological group and prognosis

Group	Cases	Metastases present in	Average duration of life in months
I	4	1	18
II	14	12	14
III	6	6	8

As can be seen from this table: in group I only 25 % of the patients showed metastases, while this was the case in 85 % of group II patients and in all of the group III patients. Muir already found at that time that grading of prostate cancer could be difficult because of the fact that variations may occur in the same growth.

2.3 Evans' system

In 1942, Evans and co-workers [5] published a paper on the study of 100 consecutive clinical cases of carcinoma of the prostate. Until that time grading of prostate cancer, contrary to other types of malignancy, was far from being a routine. An explanation for this may have been the complexity of the histological features in carcinoma of the prostate, but also the fact that in those days, cure of prostate cancer was almost impossible, in contrast to certain more accessible tumors. In an attempt to create a system, sufficient for proper grading of prostate cancer they selected eight main aspects of the tumor's structure as possible valid criteria for grading:

1. acinar structure
2. cell structure
3. density of cytoplasm
4. nuclear characteristics
5. presence of nucleoli
6. mitotic figures
7. fibrosis
8. inflammation

Although Muir [7] had used some cytological parameters, it is remarkable that in Evans' grading system, more than 20 years after Broders, the first attempt is made to include cytological parameters into grading of prostate cancer, and that it was for the first time attempted to apply the hetero-geneity of prostate cancer in grading.

Evans and co-workers graded each of their criteria on a scale of four divisions. Grade 1 represented the closest relationship to normal, while grade 4 represented the extreme degree of abnormality. They also took into account the fact that prostatic carcinoma has a tendency to vary in appearance in different parts in the same tumor. Three of the above-mentioned structural features (acinous structure, cell structure and nuclear size) were also recorded by the proportion of each grade present in a

given cancer. So, each of these eight criteria was graded separately and the grade of each was correlated with the clinical course of the disease in the patient. Evans and co-workers found that of these eight criteria only three (acinar structure, cell structure and nuclear characteristics) showed a good correlation with the clinical course and they created a grade using only these three parameters. Correlation between grade and survival time and incidence of clinical metastases are shown in Tables 3 and 4 respectively.

Table 3 - Correlation between Grade and survival (Evans et al, [5])

Grade	No. of cases	Average survival
I and II	20	5.4 years
III	62	4.4 years
IV	18	2.2 years

Table 4 - Correlation between grade and incidence of metastases (Evans et al, [5])

Grade	No. of cases	Cases with metastasis
I and II	20	4
III	62	18
IV	18	8

The other five parameters showed no correlation with survival and/or incidence of metastases. The comparisons of histological and clinical data were based on the predominant type of histological structure in each

28

specimen. It was remarkable that in their study comparisons with grades, based only on the most malignant elements present in each tumor showed no significant correlations.

2.4 The Mayo Clinic system

In 1969, Utz and Farrow [8] discussed their Mayo Clinic grading system. This system was developed because it was felt that not only histological, but also cytological characteristics of a prostate cancer had to be dealt with. The Mayo Clinic system consisted of four grades:

Grade 1: A well demarcated tumor with minimal invasion. Abnormally small and closely packed acini. Low, cuboidal cells with enlarged nuclei and prominent nucleoli. Infrequent mitoses.

Grade 2: Acini smaller, less regular and more flattened and elongated. Cells become progressively smaller, the nuclei become darker and more irregular.

Grade 3: Progression of grade 2. Utz and Farrow did not describe separately the features of grade 3.

Grade 4: Complete loss of recognizeable acinar pattern. The tumor is markedly invasive and composed of solid masses of cells with very irregular, dark nuclei with little apparent cytoplasm. Mitotic figures are numerous.

This grading system is comparable to that of Evans et al. [5] in that it also takes into account both the histological and the cytological characteristics of the tumor.

In 1956 Pool and Thompson [9] already presented a study on 1,534 patients and found a good correlation between tumor grade (Mayo Clinics system) and survival (Table 5):

Table 5 - Correlation between tumor grade and survival
(Pool and Thompson, [9])

Grade	Patients		5-yrs or more survival	
	Total	Traced	No.	%
1	293	284	169	59.5
2	486	478	163	34.1
3	463	457	74	16.2
4	252	250	14	5.6

Rous and Mallouh [10] used The Mayo Clinic system in an effort to assess the relationship between histological grading of the tumor, clinical findings and the incidence of metastases. They reviewed the records of 66 patients with prostatic cancer and the histological slides of the biopsy and/or surgical material were regraded. They found that in grade 1 tumors 6 out of 15 patients (40 %) had demonstrable metastases at the time of diagnosis. Out of 31 patients with grade 2 tumors, 9 (29 %) had metastases at the time of diagnosis. Out of 17 patients with grade 3 carcinoma, 3 (18 %) had metastases, whereas of 3 patients with grade 4 carcinoma, 2 (67 %) had demonstrable metastases at the time of diagnosis and died shortly thereafter. They noted a high number of patients with low grade disease developing metastases, but did not find a clear correlation between grade and the incidence of metastases.

2.5 Auerbach's classification

In 1958 Shelley and co-workers [11] presented a new system of classification of prostatic tumors, based on differentiation and on changes in histological pattern. Their arguments to propose a new grading system was that in Broders' system only the percentage of malignant cells and not the histological picture presented by a tumor were indicative for a certain grade. They divided the prostatic carcinomas into four classes (Auerbach's

classification):

Class I: Small, morphologically uniform cells with moderately abundant, faintly granular cytoplasm. The cells are arranged in regular glandular patterns around small alveolar spaces. Orientation is well maintained. Mitotic figures are not common.

Class II: Less regular and less uniform arrangement of the neoplastic cells, which are larger than normal and in some tumors arranged in large sheets with narrow fenestrations.Larger glandular structures. Irregular and imperfect luminal spaces with scanty or absent content. Increased ratio of the size of the nucleus to the amount of cytoplasm. More prominent nuclear irregularity and hyperchromatism. More numerous, but not abundant mitotic figures than in Class I tumors. More evident invasion into surrounding glandular tissue and extension into the capsular structures.

Class III: Increase in epithelial cellularity over Class II tumors. Cells usually grow in large, irregular sheets, interrupted by narrow and poorly defined lumina, sometimes a tendency toward a papillary disposition is apparent. Cells are large and show marked variation in size. Relatively scanty and granular cytoplasm. Disproportionally large and hyperchromatic nuclei, often with a vesicular appearance. More numerous mitotic figures than in Class II. Invasion of surrounding structures is evident. Atrophied adjacent tissues.

Class IV: No recognizable glandular pattern; cells grow in solid sheets or cords, which are sometimes very narrow and appear to ramify through the prostatic stroma almost in single file. Very large tumor cells with moderately abundant, granular cytoplasm. Frequent multinucleated tumor cells. Disproportionally large, often acidophilic nuclei with marked hyperchromatism and often with nucleoli. Numerous mitotic figures. Increased tendency to permeate lymphatic spaces and to grow into preexisting prostatic ducts.

With this Auerbach classification it was possible to distinguish four groups of patients with clearly different survival rates (Table 6):

Table 6 - Auerbach's classification. Correlation between grade and survival [11]

Class	No. patients	Dead with carcinoma No.	%	Average survival
I	27	0	0	3.0 years
II	35	8	20	2.78 years
III	19	12	47	1.9 years
IV	2	2	100	0.3 years

Wiederanders and co-workers [12], using the classification proposed by Shelley et al. [11] in 171 cases of prostatic carcinoma also found a good correlation of this classification with survival. For patients with Class I tumors there was a 5-year survival of 76.5 %; Class II patients showed a 5-year survival of 28.5 % ; Class III patients had a 5-year survival of 8.7 % and none of the patients with Class IV tumors survived for five years.

2.6 Gleason's system

In 1966 Gleason [13, 14] proposed a system based on a completely different concept. Realizing that both stage and grade of disease were important prognostic factors he developed a system in which glandular differentiation was combined with the stage of the disease. He recognized five different tumor patterns (figure 1):

Pattern 1: Very well-differentiated. The tumor consists of single, separate, round to oval glands, which are quite uniform in size. They grow abnormally in closely packed, roughly rounded masses with definitely limited edges relative to the uninvolved tissue.

Pattern 2: Well-differentiated. The tumor consists of single, separate, round to oval glands, which are similar in size and shape but vary more than those in pattern 1. They also show more stromal spacing between the

32

glands (up to one gland diameter average). The tumor masses are less well circumscribed and not as definitely rounded as those in pattern 1.
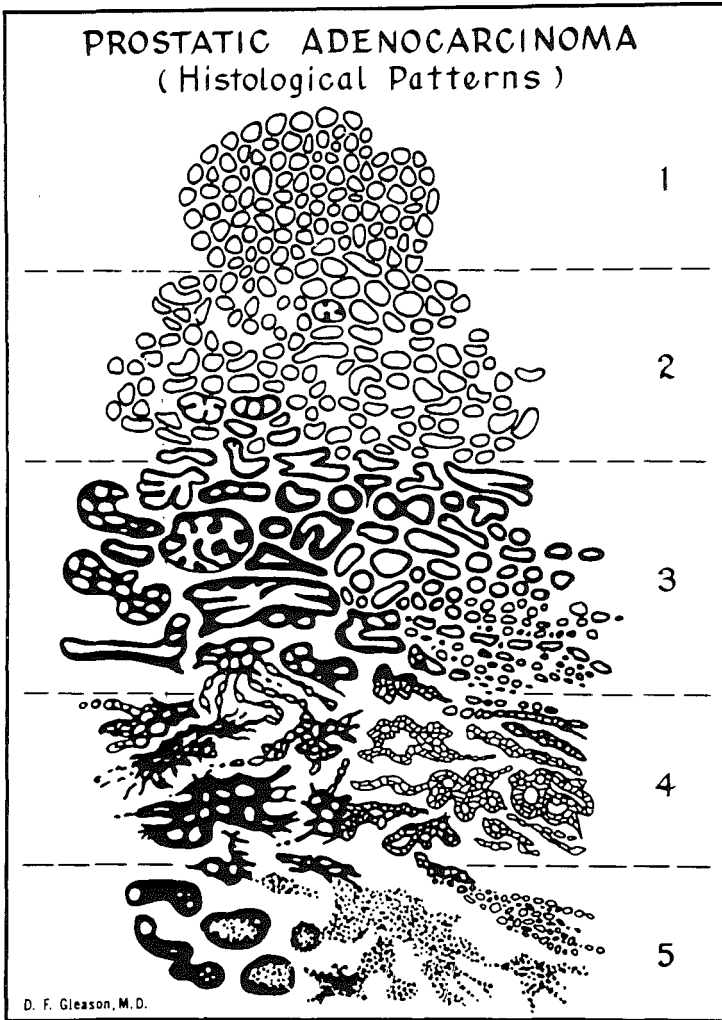

Fig 1. Simplified drawing of histological patterns, emphasizing degree of glandular differentiation and relation to stroma (with permission)

Pattern 3: <u>Moderately differentiated</u>. This pattern includes two distinctive appearances. One is an extension of patterns 1 and 2, with single, separate glands which may be much smaller or much larger, or have about the same size as those in patterns 1 and 2. The individual

glands are more irregular in size and shape than in patterns 1 and 2, many being more elongated or angular. They may be closely packed together but are much more commonly quite widely separated by stroma.The areas of tumor are usually quite irregular in outline, without a clearly definable boundary. Larger glands may have some papillary infoldings or a thick epithelium containing additional tiny glandular lumina. These latter appearances provide the transition to the second distinctive appearance in pattern 3, namely the occurrence of sharply circumscribed masses of papillary or cribriform tumor, or both. These vary in size and may be quite large, but the essential feature is the smooth and usually rounded edge around all of the sharply circumscribed masses of tumor. Any or all of these patterns may appear in one case. There may be tiny glands, large irregular glands and sharply circumscribed papillary and cribriform masses, all included under the designation of pattern 3.

Pattern 4: Poorly differentiated. The tumor consists of irregular masses of fused glands. That is, the glands are not single and separate but coalesce and branch. The fusion may be so extreme that the appearance is that of solid masses of epithelium containing multiple glandular lumina lined by poorly oriented layers of polygonal cells. Some cells may have two surfaces facing separate rounded gland spaces. The multiple glandular lumina are usually of small or medium size. In contrast to the sharply circumscribed and smoothly rounded masses in pattern 3, the pattern 4 tumors grow in very raggedly outlined masses, appearing to infiltrate the stroma very aggressively. Also included are essentially similar tumors composed of large cells with very pale cytoplasm, sometimes resembling the clear cell adenocarcinoma of the renal cortex or "hypernephroma".

Pattern 5: Very poorly differentiated. The tumor shows minimal glandular differentiation and consists of raggedly infiltrating masses of epithelial cells with only a few poorly formed glandular lumina or signet ring cells to confirm that it is an adenocarcinoma. A second rare pattern is also included in pattern 5. This consists of sharply circumscribed broad

cords and masses of compactly arranged epithelial cells with only occasional poorly formed tiny glandular lumina,sometimes with central necrosis of the masses like the "comedocarcinoma" of the breast.

The pattern most extensive in area is called the "primary" pattern and the less extensive pattern the "secondary" pattern. The numbers determined for the grades of the primary and secondary pattern are added together for each case, achieving a scaling effect of averaging. The sum of the two patterns is called the "pattern score" The pattern scores showed a very good correlation with mortality rates (table 7, page 36) [14].

Gleason also found that the mortality rate for Stage I tumors was about equal to the mortality rate for histological pattern 1. The same was found for Stage II and pattern 2 and for stage III and pattern 3. Stage IV showed a mortality rate about equal to that of pattern 5 tumors and was designated a weighting value of 5. By adding Stage numbers to pattern score numbers, Gleason created a combined histological grading and clinical staging score, ranging from 3 to 15. Thus, a score 3 means that there is a low stage, low grade disease and score 15 indicates high stage, high grade disease. There was a very good correlation of this combined scoring with the mortality rates (table 8, page 37) [14].

This system was shown to be so easy to apply and to correlate so well with prognosis, that it soon found wide acceptance [15, 16, 17]. It still is used in many centers especially in the U.S.A.

Corriere and co-workers [18] reviewed the records of 525 patients with the pathologically proven diagnosis of prostatic carcinoma presenting at the Hospital of the University of Pennsylvania from 1955 through 1964. All the tumors were regraded. They divided the tumors into five histological grades according to Gleason's definition. Only the histological grading was used and not combined with clinical staging. As table 9 shows, these authors also found a good correlation between survival and the histological type of the tumor.

Table 7   -   Gleason grading system. Correlation between pattern score and tumor mortality rate [14]

Histol. pattern score	No. patients	Total yrs followup	No. dead	No. dead-CA	Dead / total yrs.	Dead-CA / total yrs.
2	14	46.3	7	0	0.151	0
3	84	381.0	34	3	0.089	0.008
4	68	272.4	31	3	0.114	0.011
5	558	2423.8	279	37	0.115	0.015
6	1240	4057.7	726	179	0.179	0.044
7	256	718.5	158	49	0.220	0.068
8	537	1527.6	384	199	0.251	0.130
9	61	137.3	44	27	0.320	0.197
10	93	165.8	70	41	0.422	0.247
totals	2911	9730.4	1733	538	0.178	0.055

Table 8 - Gleason grading system. Combined histological grading and clinical staging [14]

Histol. pattern score	No. patients	Total yrs followup	No. dead	No. dead-CA	Dead / total yrs.	Dead-CA / total yrs.
3	5	12.1	0	0	0.000	0.000
4	36	156.4	11	0	0.070	0.000
5	37	136.9	11	0	0.080	0.000
6	156	623.8	55	2	0.088	0.003
7	180	730.6	75	5	0.103	0.007
8	438	1865.5	219	24	0.117	0.013
9	662	2407.4	390	46	0.162	0.019
10	229	781.3	129	32	0.165	0.041
11	586	1692.7	410	181	0.242	0.107
12	134	314.8	91	39	0.289	0.124
13	348	818.3	270	160	0.330	0.196
14	35	82.0	26	21	0.317	0.256
15	65	108.6	46	28	0.424	0.258
totals	2911	9730.4	1733	538	0.178	0.055

Table 9 - Survival by histological grade (Corriere et al. [18])

Survival	histological grade (alive / total)				
	1	2	3	4	5
3-year	8/11	42/49	90/145	11/24	4/12
%	72.8	85.8	62.0	45.8	33.3
5-year	5/10	21/39	49/134	7/24	2/11
%	50.0	54.0	36.6	29.2	18.2

Kramer and associates [19] proposed to use Gleason's system as a predictor of lymph node metastases. In their series of 144 patients who underwent a staging pelvic lymphadenectomy none of the 31 patients with a Gleason score 2 to 4 had lymph node metastases, whereas 27 of the 29 patients with a Gleason score 8 or more had nodal metastases. With the Gleason system they furthermore were able to predict response to chemotherapy in patients with stage D prostate cancer [20]. Olsson [21] doubted the reliability of the Gleason grading system in predicting lymph node involvement. He observed patients having lymph node metastases whereas their tumors were graded Gleason 2-4 and he reported some patients whose tumors were graded score 8-10 and not having lymph node involvement. Olsson advised against using the Gleason score as a replacement for lymph node dissection.

Mills and Fowler [22], using Gleason's grading system found that only in 51% of the cases the Gleason score of the biopsy specimen was identical to that of the radical prostatectomy specimen. The reason for this was that in small biopsies not always a sample representative for the whole tumor was obtained. Their advice was to take repeated biopsies when only limited amounts of tumor with a low Gleason score were present in the biopsy. The problem of undergrading is not typical for the Gleason system.

Every grading system is hampered by the fact that on small biopsies conclusions concerning the whole tumor have to be made. This problem was also stressed by Ackermann and Müller [23], by Kastendieck [24], and by Garnett and co-workers [25]. Not everyone was able to demonstrate the advantages of the Gleason grading system. Guinan and associates [26] compared the accuracy of this system with Whitmore's staging system [27] and with Broders' grading system [1 ,2] in 111 patients undergoing radical surgery. A classification was considered correct if a patient with a Gleason score 2-5, a Whitmore score A-B, or a Broders I-II was free of disease or if a patient with disease had a Gleason score 6-10, a Whitmore score C-D, or a Broders score II-IV. Of the three systems, the Gleason system was shown to be the least accurate, with 59 % of the cases being classified accurately. The Whitmore staging system scored better with 67% accuracy, while Broders' system was the most accurate with 76% of the cases being scored accurately.

2.7 Mobley and Frank's grading system

Mobley and Frank [28], in a paper on the influence of grade on survival and on serum acid phosphatase in metastatic carcinoma of the prostate, used a grading system where Grade 1 lesions were characterized by residual acinar formations and minimal nuclear changes of the columnar epithelium. Grade 2 lesions showed acinar formation, but had loss of nuclear uniformity. Grade 3 lesions showed an anaplastic carcinoma with loss of acinar formation and with undifferentiated cells. Their system did not differ very much from Muir's system [7] and seemed to be a simplification of the system of Evans and co-workers [5]. With this system they reviewed 96 cases of metastatic prostatic carcinoma and regraded the histological specimens. They could demonstrate a positive correlation between grade and survival and between grade and serum acid phosphatase levels. When they regarded the survival from the time of diagnosis of metastases they found that 1 and 2 years after diagnosis of metastasis the patients with low grade tumors survived longer than the patients with moderate or high

grade disease. Over a 5-year period the patients with grade 1 tumors did as poorly as those with grade 2 or grade 3 tumors.

Byar and Mostofi [29] in co-operation with the VACURG studied several prognostic histological parameters in step-sections of 208 radical prostatectomy specimens. They found that capsular and/or seminal vesical invasion, whether detected clinically or pathologically, had an unfavorable prognosis. They also tried to grade the tumors, but their attempts were quite frustrating. The investigator was not able even to reproduce his own results, unless a specific focus on the slide was marked and the examiner went back to that particular focus. Although they did not discuss this observation in their paper (their main interest was the extent of the tumor), this was the first indication that intra-observer and inter-observer variation is a major limitation in widespread use of any grading system.

2.8 Mostofi's system

In 1975 Mostofi [30, 31] analysed the problem of grading prostatic carcinoma and he was impressed by the fact that in all grading systems the terms "differentiation" and "anaplasia" had not been clearly defined. He reserved the term "differentiation" for a tendency of a tumor to form glands and their characteristics as compared to normal prostatic glands. The term "anaplasia" was reserved for nuclear characteristics such as variations from normal in size, shape, staining, chromatin distribution, and mitotic activity. So, here the term "anaplasia" has a different meaning than in Hansemann's original description [6].

With this conception Mostofi defines a tumor as being differentiated if it forms glands, which may be large, intermediate or small. The glands may even be fused, have a cribriform pattern or have a papillary configuration. In contrast, he defines tumors that grow in rows, columns or sheets as undifferentiated.

Furthermore he defines a tumor as having slight anaplasia when the nuclei show slight variations from normal. A tumor with moderate

40

anaplasia shows moderate variation as compared to normal nuclei and in markedly anaplastic tumors there is marked variation of nuclear size and shape.

Combining differentiation with the degree of anaplasia results in different grades:

Grade I: well-differentiated glands with nuclei that show slight nuclear anaplasia.

Grade II: gland formation but the nuclei show moderate nuclear anaplasia.

Grade III: glands with marked nuclear anaplasia or tumors that are undifferentiated (not forming glands).

Although in his original paper Mostofi did not support his grading system with any clinical results, he had the impression that this system showed a good correlation with the prognosis. In 1977 Harada and associates [32] showed that Mostofi's grading system indeed showed a good correlation with the prognosis of the disease. They furthermore and again stressed the fact that each grading system had the disadvantage of being most accurate in the hands of the one who developed the system, but this accuracy decreased when the system was used by other pathologists. They compared the Mostofi system with the Gleason system on the same cases and they found agreement between their grading with the Mostofi system and the grading performed by Gleason himself in only 38 % of the cases. However, when they graded the tumors themselves repeatedly using the Gleason system they found agreement between their first and their second reading in 71 % of the cases. So, the intra-observer variability was much less than the inter-observer variability. Also others have confirmed the value of Mostofi's grading system [33,34].

In 1976 Epstein and Fatti [35] correlated morphological features in 146 cases of prostatic carcinoma with a 5-year survival. They used McNeal's system of typing of prostatic tumors [36], which in fact is no grading system.

Besides typing of prostatic carcinomas they also graded the tumors. Their grading system resembled very much that of Mostofi in that they regarded both histological differentiation and cytological parameters. Their cytological parameters included nuclear pleomorphism, amount of cytoplasm, nucleoli, cell borders, lymphocytic inflammatory reaction, and vascular and perineural invasion. When they correlated a five-year survival with McNeal's classification they found a 40.2 % survival for the medullary-alveolar type, a 20.8 % survival for the tubular-scirrhous type and a 43.7 % survival for the mixed type. The difference between the three groups was significant at the 5 % level. However, they could not find a significant correlation between the histological differentiation and a five-year survival. Nuclear pleomorphism on the other hand, showed a significant correlation at the 1 % level with survival (table 10).

Table 10 -   Epstein and Fatti, [35]

5-year survival correlated with pleomorphism

	Untraced	Total traced	Alive 5 years	% survival
Class I	8	32	16	50
Class II	8	53	21	39.6
Class III	5	61	12	19.7

Cell borders, whether distinct or indistinct, showed a significant correlation with a 5-years survival at the 1 % level. The group of 75 patients, whose tumors showed distinct cell borders had a five-year survival of 45.3 %, whereas the group of 71 patients whose tumors did not show distinct cell borders had a five-year survival of only 21.1 %. Also the presence or absence of lymphocytic infiltrations showed a significant correlation with survival. The presence of such infiltrates showed a better prognosis: 47.7 % five-year survival against 27.5 % when lymphocytes were absent. It did not make any difference whether these infiltrates were

focal, diffuse or were both. These parameters had never been studied before. The amount of cytoplasm, the presence or absence of nucleoli, vascular invasion and perineural invasion did not show any significant correlation with the prognosis. Interaction of the various parameters showed that the prognostic effect of McNeal's classification was due to an unequal distribution of parameters. In the medullary-alveolar type tumors there were distinct cell borders in 49 %, whereas this was the case in 30 % of the tubular-scirrhous tumors. When corrected for this unequal distribution, McNeal's typing did not show any prognostic significance any more. The same was seen for nuclear pleomorphism: once corrected for the presence or absence of distinct cell borders this parameter did not show any prognostic importance in Epstein and Fatti's patients. The conclusion of Epstein and Fatti was that only cell borders and lymphocytic infiltration was of prognostic importance. They suggested that the presence of lymphocytic infiltration was a sign of cell-mediated immune response to the tumor. As presence or absence of distinct cell borders was prognostically important in their study, Epstein and Fatti tried to assess the intra-observer variability. Four different pathologists regraded 20 of the tumors and they found an inter-observer correlation between 80 and 90%.

Gibbons and co-workers [37], reporting on behalf of the National Prostatic Cancer Project (NPCP) tried to correlate grade of the tumor with the response to chemotherapy. They studied 125 patients with metastatic prostatic carcinoma, not (or no longer) responding to hormonal therapy. These patients were randomized to receive either 5-fluorouracil, cyclophosphamide or any other treatment (non-chemotherapeutic). They restudied all histological slides (biopsies and other slides available) and graded them without indicating which grading system they used. They did however indicate their criteria for grading and from that it can be seen that their system resembles somewhat the Mayo Clinic system [8] and the Auerbach classification, proposed by Shelley et al [11]. Gibbons and co-workers found response to therapy in both chemotherapeutic treatment

groups, regardless of grade of tumor. However, progression of disease was significantly less rapid in cyclophosphamide treated patients with poorly differentiated or anaplastic carcinomas than in the other two treatment groups with this grade of tumor. In this way there was some prognostic importance of grading, but favorable responders could not always be identified in advance on the basis of grade alone.

2.9 Hohbach and Dhom's classification

In 1979 Böcking and associates [38, 39], using Hohbach and Dhom's classification [40], which is a system resembling very much the above depicted systems of the Mayo Clinic and Auerbach's system, created a combined grade by adding values for nuclear anaplasia (according to Mostofi) to values for each of the histological patterns in their classification. As Hohbach and Dhom used four classes of differentiation (well-differentiated, poorly differentiated, cribriform and solid-anaplastic carcinoma) Böcking and co-workers numbered these classes 0 to 3. Accordingly nuclear anaplasia was graded from 0 to 2. Adding the grade of histological differentiation to the grade of nuclear anaplasia resulted in the combined grades 0 - 5 of prostatic carcinoma. In 1980 they proposed to simplify the system by combining the combined grades 0 and 1 to a new grade I, the grades 2 and 3 to a new grade II and the grades 4 and 5 to a new grade III. With this system they were able to identify groups of patients with significantly different survival rates.

2.10 Gaeta's system

Gaeta and co-workers [41] presented a new grading system in which the architectural arrangement of glandular structures and the nuclear characteristics were evaluated separately. Each was graded one to four (Table 11)

Table 11 - histological grades of prostatic cancer (Gaeta et al, [41])

Grade	Glands	Cells
I	Well defined, small, medium or large, separated by scant stroma	Uniform and normal size, nucleoli may be present but inconspicous
II	Medium and small, moderate amount of stroma	Slight pleomorphism, nucleoli prominent
III	Small acini, frequent loss of glandular organization, cribriform and scirrhous patterns	Pronounced pleomorphism, vesicular nuclei, acidophilic nucleoli
IV	Round expansile masses of tumor cells, no formation of glands	Small or large, uniform or pleomorphic with significant mitotic activity (> 3 per high power field)

Based on these criteria 169 cases of prostatic carcinoma were reviewed and assigned to the highest category according to the glandular pattern or to the nuclear features. Their findings are given in table 12:

Table 12 - Correlation between grades and mortality rates - Gaeta et al, [41]

Grade	Cases No.	Cases (%)	Total yrs.	deaths	Cancer deaths No.	Cancer deaths (%)	Mortality index
I	6	3.5	27.3	2	1	16	0.036
II	43	25.5	161.5	25	16	64	0.099
III	83	49.2	332.4	67	60	89	0.180
IV	37	21.8	98.0	36	34	94	0.346

As can be seen the mortality index increases with increasing grade. The difference between this system and Mostofi's system is that in Mostofi's

system nuclear characteristics are not evaluated separately but together with the architectural pattern, combining these two to an "overall" grade. It remains however rather subjective what weight each parameter should have in Mostofi's system. In the Gaeta-system this problem does not play an important role. Both glandular pattern and nuclear characteristics are graded and the highest number for grade is the overall grade of the tumor. The only problem not solved by this system is the problem of multiple tumor patterns. Gaeta and co-workers take into account the predominant pattern. An occasional focus of any given pattern of glands or nuclei did not alter the assigned grade of the remainder of the tumor if that remainder made up approximately 90 % of the tumor. It was not indicated how the system deals with a small amount of tumor of poor differentiation.

2.11 The M.D. Anderson system

In 1982 Brawn and co-workers [42] presented their M.D. Anderson system of grading. The poor reproducibility of the previous grading systems and the problem of determining which of the histological features of the tumor were important in assigning a grade was the reason for searching for a new system. They developed a new system attempting to improve both the reproducibility and the simplicity of grading prostatic adenocarcinoma. They created 4 grades, using the percentage of the tumor that is able to form glands. In grade 1 75-100% of the tumor formed glands, in grade 2 this percentage was 50-75 (including $\geq$ 50% cribriform-papillary pattern), grade 3 showed 25-50% gland formation, while in grade 4 only 25% or less of the tumor was able to form glands. In fact their grading system looked very much a combination of the Broders, the Mostofi and the Gleason systems. It is remarkable that cytological characteristics are completely absent in this system. With this system Brawn and co-workers were able to identify patients with significantly different prognoses. Out of 84 patients with M.D.Anderson (MDA) grade 1 only 12 (14%) died from prostate cancer, whereas 35 out of 75 patients (46.6%) with MDA grade 2-3 tumors, and 18 out of 23 patients (78.2%) with

MDA grade 4 tumors died from prostate cancer. They found however, that there was virtually no difference between grade 2 and grade 3. They therefore proposed to combine these two grades, leaving a modified three grade system. The advantage of their system indeed was that it was a simple, low-power microscopic method of grading. The authors did however not evaluate the reproducibility of the system, whereas reproducibility was one of their reasons to create their new system.

2.12 Cytological grading

Since the introduction of transrectal fine needle aspiration by Franzén and co-workers [43] an increasing interest has grown for this means of diagnosis. The advantages were obvious: the procedure could easily be performed without any form of anaesthesia and more samples than with the thick needle biopsy could be taken without much harm to the patient . A disadvantage is that the technique requires great skill in obtaining good material for proper diagnosis [44].

In 1971 Esposti [45] reported on the cytological grading of 469 prostate cancers. All patients had at least a 5-year follow-up. He used three grades and could identify a good correlation between cytological grade and crude 3- and 5-year survival. Also Faul and associates [46] found a good correlation between cytological estimated tumor grade and prognosis.

Voeth and co-workers [47] attempted to correlate cytological grading with histological grading. They found a 46.1% correlation in 92 histologically and cytologically proven prostate cancers. In 39.1% the difference was one grade. In more than 60% the cytologic grading was reproducible. They explained their rather poor results by mentioning the possibility that by a core biopsy only a small fragment of the tumor could be taken. This small sample might be not representative for the whole tumor. Fine needle aspiration allowed the investigator to obtain a more generous sample from the prostate, with a higher chance of obtaining adequate material for proper grading.

2.13 Comparing systems

The NPCP and the American Cancer Society conducted a workshop in an attempt to verify current systems of classification. In 1979 a report on this workshop was published [48]. Four major grading systems were compared: the Gleason system [13, 14], the Mostofi system [30, 31], the Mayo Clinic system [8] and the new Gaeta system [41]. It was concluded that all grading systems had to deal with some limitations:

1.   the amount of tissue available for grading
2.   the objective definability of grading criteria
3.   the degree of reproducibility of interpretation
4.   the simplicity
5.   the predictive value of the system relative to the biologic potential of the tumor

The general recommendations after the workshop were that histological grading of prostate cancer should be employed routinely, that the system of Gleason should be employed at least in conjunction with any other system, that nuclear and cytologic characteristics should be considered in prospective studies to further the discriminative capabilities of the Gleason system, and that data should be accumulated on correlations between histological grade and the natural history of the tumor, or the response to various forms of treatment.

In 1981 Cantrell and co-workers [49] studied the influence of extent of tumor and grade on the prognosis in patients with Stage A cancer. They referred to their own grading system (Hopkins system [50]) which was rather vaguely described. Comparison of their system with that of Gleason showed that out of the 14 patients with either Hopkins grade 1 or Gleason score 2 to 4, none had progression of the disease. Out of the 12 patients whose tumors showed progression, 8/12 were graded Hopkins grade 2 and 4/12 Hopkins grade 3. Out of these 12 patients, 5/12 were scored as Gleason score 5 or 6 and 58 % were scored as more than 6. So, in the low grade tumors there was uniformity in the two systems, whereas in the higher grade tumors the Gleason system had a tendency to score the

tumors higher than the Hopkins system. However, for stage A tumors this difference is not really important. More importantly stage A comprised the low grade tumors and there was complete uniformity between the Hopkins and the Gleason systems.

In 1982 Albertsen [51] reviewed the most important grading systems and concluded that three histological features were significant for the prognosis of prostate cancer. These were tumor volume, glandular differentiation and nuclear anaplasia. In several studies in the literature he pointed out the problems with objectivity and reproducibility, which made most of the grading systems less suitable for daily urological practice. As has already been stated before most grading systems work best in the hands of the people who developed them [32].

In this thesis the weight of each of the parameters in Mostofi's grading system [30] is evaluated in chapters 3, 4, 5 and 6. The conclusion is reached that only glandular differentiation, nuclear pleomorphism and amount of tumor seen in the slide are important parameters in relation to the prognosis of the disease. The presence of mitoses is also important, but the vast majority of prostatic carcinomas contain no or very few mitoses. The application of Mostofi's grading system in a large series of 346 cases of prostatic carcinoma resulted in the proposal of a simplification of the Mostofi grading system.

2.14   Objectivity in grading

Despite the many attempts to make grading easier and more accurate and reproducible, there is to date no uniform grading system that combines these qualities. Application of all grading systems is rather subjective and works better in the hands of the ones who developed them than in other, even experienced hands. It is therefore mandatory to put objectivity in grading. This can be achieved by trying to objectivate the most important parameters of a certain grading system. Chapters 7 and 8 of this thesis deal with the objectivation of nuclear anaplasia in Mostofi's grading system.

2.15 References

1. Broders AC: Squamous-cell epithelioma of the lip. A study of five hundred and thirty-seven cases. JAMA 74: 656-664, 1920

2. Broders AC: Carcinoma grading and practical application. Arch Path 2:376-381, 1926

3. Kahler JE (1939) Carcinoma of the prostate gland: a pathologic study. J Urol 41: 557-574, 1939

4. Greene LF, Simon HB: Occult carcinoma of the prostate. Clinical and therapeutic study of eighty-three cases. JAMA 158: 1494-1498, 1955

5. Evans N, Barnes RW, Brown AF: Carcinoma of the prostate: correlation between the histologic observations and the clinical course. Arch Path 34: 473-483, 1942

6. Hansemann DP (1893), cited by Haagensen CD: Am J Cancer 19: 285, 1933

7. Muir EG: Carcinoma of the prostate (Hunterian lecture). Lancet I: 667-672, 1934

8. Utz DC, Farrow GM (1969) Pathologic differentiation and prognosis of prostatic carcinoma. JAMA 209: 1701-1705, 1969

9. Pool TL, Thompson GJ: Conservative treatment of carcinoma of the prostate. JAMA 160: 833-837, 1956

10. Rous SN, Mallouh C: Prostatic carcinoma: relationship between histologic grade and incidence of early metastases. J Urol 108: 905-907, 1972

11. Shelley HS, Auerbach SH, Classen KL, Marks CH, Wiederanders RE: Carcinoma of the prostate. A new system of classification. Arch Surg 77: 751-756, 1958

12. Wiederanders RE, Stuber RV, Mota C, O'Connell D, Haslam GJ: Prognostic value of grading prostatic carcinoma. J. Urol. 89, 881-888, 1963

13. Gleason DF: Classification of prostatic carcinomas. Cancer

Chemother Rep 50: 125-128, 1966

14. Gleason DF and the VACURG: Histologic grading and clinical staging of prostatic carcinoma.In: Tannenbaum M (ed.) Urologic Pathology: The Prostate, Lea & Febiger, Philadelphia, pp 171-197, 1977

15. Sogani PC, Israel A, Liebermann PH,Lesser ML, Whitmore WF: Gleason grading of prostate cancer: a predictor of survival. Urology 25: 223-227, 1985

16. Guileyardo JM, Sarma DP, Johnson WD, Akazaki K, Correa P: Incidental prostatic carcinoma: tumor extent versus histologic grade. Urology 20: 40-42, 1982

17. Barzell W, Bean MA, Hilaris BS, Whitmore WF: Prostatic adenocarcinoma: relationship of grade and local extent to the pattern of metastases. J. Urol. 118: 278-282, 1977

18. Corriere JN, Cornog JL, Murphy JJ: Prognosis in patients with carcinoma of the prostate. Cancer 25: 911-918, 1970

19. Kramer SA, Spahr J, Brendler CB, Glenn JF, Paulson DF: Experience with Gleason's histopathologic grading in prostatic cancer. J Urol 124: 223-225, 1980

20. Kramer SA, Tatum AC, Walker A, Hinshaw W, Cox E, Paulson DF: Histologic grading as a predictor of response to chemotherapy in patients with prostatic adenocarcinoma. Urology 18: 377-379, 1981

21. Olsson CA: Prostate cancer. Does the Gleason score accurately predict regional lymph node involvement? Urol. Corresp. Club. 40-41, 1982

22. Mills SE, Fowler Jr JE: Gleason histologic grading of prostatic carcinoma. Correlations between biopsy and prostatectomy specimens. Cancer 57: 346-349, 1986

23. Ackermann R, Müller H-A: Der Wert der perinealen Stanzbiopsie zur Beurteilung des histologischen Differenzierungsgrades des Prostata-Carcinoms. Verhandl. Ber. der Deutsch. Gesellsch. für Urol. 32: 79-81, 1980

24. Kastendieck H: Morphologie des Prostatacarcinoms in Stanzbiopsien und totalen Prostatektomien. Untersuchungen zur Frage der Relevanz bioptischer Befundaussagen. Pathologe 2: 31-43, 1980

25. Garnett, JE, Oyasu R, Grayhack JT: The accuracy of diagnostic biopsy specimens in predicting tumor grades by Gleason's classification of radical prostatectomy specimens. J Urol 131: 690-693, 1984

26. Guinan P, Talluri K, Nagubadi S, Sharifi R, Ray V, Shaw M: Evaluation of Gleason classification system in prostate cancer. Urology 21:458-460, 1983

27. Whitmore WF Jr: The natural history of prostatic cancer. Cancer 32: 1104-1112, 1973

28. Mobley TL, Frank IN: Influence of tumor grade on survival and on serum acid phosphatase levels in metastatic carcinoma of the prostate. J Urol 99: 321-323, 1968

29. Byar DP, Mostofi FK and the VACURG: Carcinoma of the prostate: prognostic evaluation of certain pathologic features in 208 radical prostatectomies. Cancer 30: 5-13, 1972

30. Mostofi FK: Grading of prostatic carcinoma. Cancer Chemoth Rep part 1, 59: 111-117, 1975

31. Mostofi FK: Problems of grading carcinoma of prostate. Semin in Oncol 3: 161-169, 1976

32. Harada M, Mostofi FK, Corle DK, Byar DP Trump BF: Preliminary studies of histologic prognosis in cancer of the prostate. Cancer Treatm Rep 61: 223- 225, 1977

33. Bandhauer K: Primary grading of prostatic carcinoma and clinical course. Proc. Symp. on Prostatic Carcinoma, 40-55, 1978

34. De las Morenas A, Siroky MB, Merriam J, Stilmant M: Prostatic adenocarcinoma: reproducibility and correlation with clinical stages of four grading systems. Hum Pathol 19: 595-597, 1988

35. Epstein NA, Fatti, LP: Prostatic carcinoma. Some morphological features affecting prognosis. Cancer 37: 2455-2465, 1977

36. McNeal JE: Morphogenesis of prostatic carcinoma. Cancer 18: 1659-1666, 1965

37. Gibbons RP, Scott WW, Johnson DE, Prout GR, Schmidt JD, Murphy GP:Prostatic carcinoma. Relationship between primary tumor, histologic grade, and response to chemotherapy. Urology 8: 222-226, 1976

38. Böcking A, Sinagowitz E, Thon W, Nattenmüller E, Wagner J, Sandritter W: Histologisches Grading des Prostatakarzinoms. Med Welt 30: 275-279, 1979

39. Böcking A, Sinagowitz E: Histologic grading of prostatic carcinoma. Path Res Pract 168: 115-125, 1980

40. Hohbach M, Dhom G: Aktuelle Probleme des Prostatakarzinoms. Münch Med Wchschr 114: 45-54, 1972

41. Gaeta JF, Asirwatham JE, Miller G, Murphy GP: Histologic grading of primary prostatic cancer: a new approach to an old problem. J Urol 123: 689-693, 1980

42. Brawn PN, Ayala AG, Von Eschenbach AC, Hussey DH, Johnson DE: Histologic grading study of prostate adenocarcinoma. The development of a new system and comparison with other methods - A preliminary study. Cancer 49: 525-532, 1982

43. Franzén S, Giertz G, Zajicek J: Cytological diagnosis of prostatic tumours by transrectal aspiration biopsy: A preliminary report. Brit J Urol 32: 193-196, 1960

44. Wajsman Z, Chu TM: Detection and diagnosis of prostatic cancer. In: Murphy GP (ed.). Prostatic Cancer. PSG Publishing Company, Inc. Littleton, Massachisetts, 1979, pp 111-128

45. Esposti PL: Cytologic malignancy grading of prostatic carcinoma by transrectal aspiration biopsy. Scand J Urol Nephrol 5: 199-209, 1971

46. Faul P, Schmiedt E, Kern R: Die prognostische Bedeutung des zytologischen Differenzierungsgrades beim östrogenbehandelter Prostatacarcinom. Urologe A 17: 377-381, 1978

47. Voeth Ch, Droese M, Steuer G: Erfahrungen mit dem zytologischen Grading beim Prostatakarzinom. Urologe A 17: 367-370, 1978

48. Murphy GP, Whitmore WF: A report of the workshops on the current status of the histologic grading of prostate cancer. Cancer 44: 1490-1494, 1979

49. Cantrell BB, DeKlerk DP, Eggleston JC, Boitnott JK, Walsh PC: Pathological factors that influence prognosis in stage A prostatic cancer: the influence of extent versus grade. J Urol 125: 516-520, 1981

50. Jewett HJ, Bridge RW, Gray GF, Shelly WM: The palpable nodule of prostatic cancer. Results 15 years after radical excision. JAMA 203: 403, 1968

51. Albertsen P: Histologic grading and the practicing urologist. Prostate 3: 333-338, 1982

Chapter 3

Incidental carcinoma of the prostate treated by total prostatectomy

The prognostic impact of microscopic tumor extension and grade

F.H. Schröder, J.H.M. Blom, W.C.J. Hop, and F.K. Mostofi

Incidental Carcinoma of the Prostate Treated by Total Prostatectomy

The Prognostic Impact of Microscopic Tumor Extension and Grade

F. H. Schroeder[1], J. H. M. Blom[1], W. C. J. Hop[2] and F. K. Mostofi[3]

[1] Department of Urology and
[2] Institute of Biostatistics, Erasmus University, P.O. Box 1738, NL-3000 DR Rotterdam, The Netherlands
[3] Armed Forces Institute of Pathology, Washington DC, USA

Summary. A retrospective study of 55 patients with incidental prostatic carcinoma with long term follow-up is presented. All patients were treated with total perineal prostatectomy, 43 received some form of endocrine treatment after the initial diagnosis was made. In order to contribute to the establishment of low and high risk groups which do not or do require agressive treatment, a careful histological analysis of the 39 patients was carried out on whom total prostatectomy slides with tumor were available. The amount of tumor, grade and parameters commonly used to establish grading were determined and correlated with corrected survival. The findings indicate that a small amount of tumor, grade 1, the presence of small, intermediate or large glands (but not cribriform and/or solid tumor) and the presence of slight but not moderate or marked variation in size and shape of the nucleus are strong predictors of not dying from prostatic carcinoma. There is agreement with the literature, where similar groups of patients not further treated after the initial diagnosis had been established showed a comparably low number of progressions. It is concluded that small, well differentiated prostatic carcinomas (category $T_0pT_1N_xM_0G_1$, stage A_1) do not require an aggressive diagnostic work-up or further treatment. A group of 11 patients (27%) showed more extensive but well differentiated tumors. Only two of these patients died of prostatic carcinoma. The natural history of this entity is not sufficiently known to make definite treatment decisions. Staging, radical prostatectomy, radiotherapy or deferred treatment may be indicated. Grade 3 carcinoma or the presence of cribriform and/or solid tumor were strong predictors of progression and death from prostatic carcinoma. Seven of 14 patients with these characteristics died of their disease. It is concluded that at least the experience in this series does not show radical prostatectomy to be an optimal treatment in this group of patients. Smaller rates of progression are however described in the literature. Endocrine management does not seem to have any beneficial effect, reports on radiotherapy are scarce. The optimal treatment for this group of patients with a high risk of dying from their tumor and a significantly shortened overall survival is not known.

This communication deals with prostatic carcinoma found in prostatic specimens removed for treatment of BPH which were not suspected on rectal examination (Incidental carcinoma).

Many authors of recent articles on this subject conclude from their findings, that within this tumor category a group of patients can be identified who do not require treatment because the tendency of their tumors to progress and to kill is negligibly small or even absent [2, 3, 6, 7, 8, 11, 13, 22, 23, 27, 32]. In reviewing these articles there seems to be agreement that grade of differentiation and the microscopically defined extension of the tumor within the resected specimen are the most important determinants of tumor free survival or the occurrence of progression. The purpose of this communication is to contribute to the further definition of criteria allowing to predict a benign or malignant course of individual patients.

According to international agreement the TNM classification will be used to express the characteristics of incidental (T_0) carcinoma [33]. This system translates easily into the A, B, C, D classification originally suggested by Whitmore [35], which is commonly used in the United States of America. The TNM system eliminates the confusing necessity to use the same letters for the clinical and histological classification of a tumor. The division of "stage A" prostatic carcinoma in stage A_1 and A_2 on the basis of histological extension can for example easily be expressed by categorizing such tumors as T_0pT_1, respectively T_0pT_{2-3}. Increasing knowledge of the prognostic significance of morphological properties of incidental prostatic carcinoma can and should be used to improve the TNM system. Table 1 attempts to correlate the American classification of incidental prostatic carcinoma with the TNM system.

Table 1. Comparison of the American and TNM staging systems for incidental carcinoma of the prostate

American staging system	TNM system	Description of tumor characteristics. Incidental carcinoma
A$_1$	T$_0$pT$_1$N$_0$M$_0$G$_1$	*American system:* Focal, well differentiated $\leqq$3 chips [5], only one lobe involved [9], focal, well differentiated [17], < 5% surface [8] *TNM:* Focal (single or multiple) carcinoma (pT$_1$)
A$_2$	T$_0$pT$_{1-3}$N$_0$M$_0$G$_{1-3}$	*American system:* > 3 chips, poorly differentiated [5], > 5 chips, poorly differentiated [20] multifocal or diffuse [9], > 50% of specimen or poorly differentiated [17], > 5% surface [8] *TNM:* Histological extension defined by pT categories, differentiation by G categories pT$_2$: Diffuse carcinoma with or without extension to the capsule pT$_3$: Carcinoma with invasion beyond the capsule and/or invasion of the seminal vesicles
A$_f$		Several authors propose a separate "focal" classification [9, 32]

The incidence of clinically unsuspected prostatic carcinoma found in operative prostatic specimens varies greatly in the literature. The subject has recently been reviewed [1, 32]. Routine examination (which again varies institutionally) has produced an incidence of 6–14% of incidental carcinoma in patients undergoing surgery for benign disease (6.0%, 8%, 10%, 10.3%, 14% for references [2, 8, 11, 24, 15, 29]). Autopsy studies have shown a much higher incidence of occult or latent prostatic carcinoma. The incidence increases with age and varies with the number of sections taken [19]. The incidence of incidental carcinoma not palpable on rectal examination should in addition be adversely influenced by the fact that the prostatic capsule and some adjacent tissue where most prostatic carcinomas originate remains in situ. Denton [14] and Battaglia [1] have reported incidences of 6 vs. 15% and 41% vs. 86% with different step section techniques. Even if one considers that about 50% of Battaglia's cases were probably atypical hyperplasia it is evident that the incidence figures obtained by "routine pathological examination" do not reflect the number of tumors nor the volume of tumor which is in fact present in patients with BPH. On the background of this information all considerations of tumor volume for prognostic classification of this disease should be looked at with great caution and must be related to the tissue preparation techniques and resection techniques used at any given institution.

For this reason grade may be the more important determinant in daily clinical use and efforts have been taken in this study to apply individually the parameters used in grading prostatic carcinoma [31] to a selected material taking advantage of the availability of radical prostatectomy specimens and long term follow-up. Because of surgical and in many instances endocrine treatment being applied it is not possible to study the natural history with this material. Cancer deaths are treatment escapes. However, data resulting from untreated patient populations (except for the initial surgery) are available for comparison [11, 24, 7, 8].

It is not the intention of this publication to completely review the subject of incidental prostatic carcinoma.

Materials and Methods

Patients

Of 484 patients treated by the late Dr. Elmer Belt by means of total perineal prostatectomy, 55 were diagnosed incidentally at the time of prostatic surgery for benign disease.

The age at diagnosis varied from 44 to 85 years with an average of 64.6 years. The average age of 484 patients was 65.5 years. None of the tumors was suspected on previous rectal examination. Pre-total prostatectomy histological slides were destroyed in one of the Los Angeles hospitals. In 8 radical prostatectomy specimens no tumor was found and in 3 sets of slides grading was impossible because of estrogen induced morphological changes. Three sets of slides were lost. Of the remaining 40 patients the histological slides revealed evaluable prostatic cancer, however of one of these patients the cause of death was unknown and he therefore was excluded from most of the evaluations.

The 39 patients included into the histological study were operated upon between 1948 and 1970. No evaluation of the lymph node status was done. Bone scans were done from 1967 on. Serum acid phosphatase was normal in all patients initially.

At the time of the last evaluation of their clinical status in September 1982, 9 of 39 patients were alive, 30 were dead. Two of those being recorded as "alive" were lost to follow-up after 8 and 16 years. Only two patients of those being alive have evidence of carcinoma. All of the ones that are still alive were followed for 15 years and have therefore completed their desired period of follow-up.

Eleven of 55 patients (20%) and 8 of 39 patients (21%) died of prostatic carcinoma. A patient was considered dead of cancer if metastases were documented prior to his death. Other causes of death were cardiovascular (20 patients), other cancer (4), other causes (4) respiratory (2), unknown cause (1). 43 of 55 patients received some form of endocrine treatment.

Pathological Evaluation

All available slides were re-graded by one of us (FKM). An overall grade was determined on a scale of 1 to 3 and the individual architectural and cytological criteria used for grading were evaluated individually and correlated with the rate of cancer deaths (corrected survival). Table 2 shows the individual criteria used for grading.

57

Table 2. Histological and cytological parameters used for grading (Mostofi [28])

A. Histological
Pleomorphism (number of tumor formations)
Glands
    small, intermediate, large
    cribriform and solid tumor
Amount of tumor
Amount of stroma

B. Cytological
Size of cell
Cytoplasm (clear or granular)
Nucleus:
    size
    pleomorphism
    vacuoles
    mitoses
    nucleoli

Fig. 1. Uncorrected survival rates and expected survival of 470 patients with prostatic cancer treated by radical prostatectomy split in categories T_0, T_{1-2} and T_3

Table 3. Uncorrected 5, 10, 15 and 20 year survival data by T-categories

T-category	Number of patients	Uncorrected survival (%)			
		5 years	10 years	15 years	20 years
T_0	55	76	62	34	22
T_{1-2}	263	72	50	30	18
T_3	152	62	32	18	10

The amount of tumor seen in the slides was recorded as being small, intermediate or much. A small amount of tumor was defined as not more than 3 foci or 2 microscopic fields at magnification $100 \times$.

In the case of presence of several architectural formations within one specimen, the worst formation was evaluated. Re-grading was done in a blind fashion. The pathologist was not aware of the clinical information. The methods of pathological evaluation have been described at greater extent elsewhere [31].

Statistics

The technique of Kaplan and Meier [26] was used to establish survival curves. For evaluation of the histological properties and their impact on prognosis it seemed more adequate to compare the number of cancer deaths in the subgroups. Therefore for intercurrent deaths corrected survival curves are shown in most instances rather than overall survival. For this reason the data in the plots do not agree with the overall survival data (Table 3, Fig. 1). Progression was not studied as a separate parameter because most patients have completed 15 years of follow-up or have died. The number of patients being alive with tumor is small [15]. In this situation and with the definition of cancer deaths used time to progression and progression rate would add little information.

The Logrank test was applied to evaluate the differences of survival curves.

As mentioned in the introduction the main purpose of the study is to more clearly identify and define parameters which allow to predict a benign or malignant course of the disease. With all precautions of interpretation the proportions of patients presenting with a given parameter who die from prostatic cancer or do not die from prostatic carcinoma can be established. These proportions will in a collective where very few patients are still at risk represent an approximate measure of the probability of dying from prostatic carcinoma or not.

Example. Of the 26 patients in whose slides moderate or large amounts of tumor were found, 9 died of prostatic carcinoma. The proportion dying of carcinoma of those having moderate or large amounts of tumor present is $9:26 = 0.35$. The probability of dying of carcinoma of the prostate if more than small amounts of tumor are found in this study was 35%.

Results

The prognostic impact of the parameters under study can be evaluated by their ability to predict tumor free survival on one side and death from prostatic carcinoma

on the other. One should, however, in looking at the data not forget that the patients have been treated. Tumor free survivors may be "cures". Patients dying from prostatic carcinoma have escaped treatment, their survival time may have been influenced.

Figure 1 compares the uncorrected survival rates of T_0, T_{1-2} and T_3 tumors from the whole series to the theoretically expected survival of this group of patients if they had not had prostatic carcinoma. It can be seen that there is a significant difference for the three different degrees of local tumor extension studied. The survival of the T_0 group is not identical with the expected survival for 10 years. Uncorrected survival figures are indicated in Table 3.

Only those histological criteria which were earlier shown to have significant prognostic influence will be discussed in detail. In a previous study such criteria were identified as being tumor architecture (glands, differentiation), small amount of tumor (not intermediate or large), nuclear pleomorphism (anaplasia, variation in size and shape of the nucleus), presence or absence of mitoses and grade [28].

Architectural Parameters

Glands. Figure 2 shows the influence of "glands" (differentiation) on for intercurrent death corrected survival. If only large, intermediate or small glands, but no cribriform or solid tumor was present, a subgroup of 26 patients was identified of which only two died of prostatic carcinoma. Seven patients who had solid or cribri-
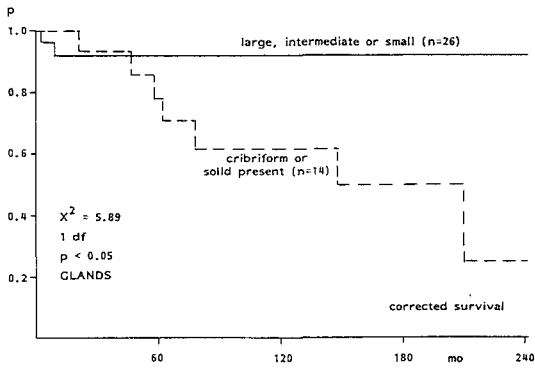
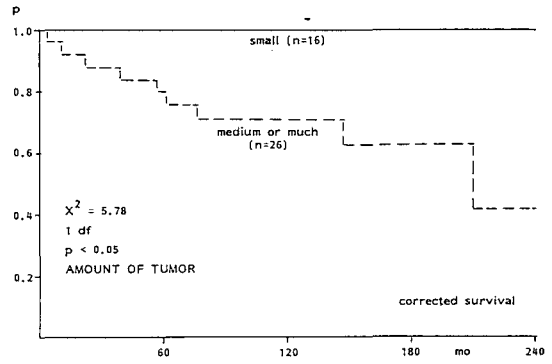Fig. 2. Glandular differentiation and corrected survival in 40 patients with incidental prostatic carcinoma

Fig. 3. Amount of tumor and corrected survival in 40 patients with incidental prostatic carcinoma

Table 4. Death from prostatic carcinoma by amount of tumor and glandular differentiation in 40 patients with incidental prostatic carcinoma

Amount of tumor	Glands, small, intermediate, large		Cribriform or solid tumor		Total
	Number of patients	Dead from carcinoma	Number of patients	Dead from carcinoma	
Small	15	0	0	0	15
Intermediate	2	0	4	2	6
Much	9	2	10	5	19
Total	26	2	14	7	40

form tumor in their specimen eventually died of prostatic carcinoma. The difference is significant. It is remarkable that most cancer deaths occur within 5 years (6 of 11) but that a significant proportion of patients dies of this disease after 5, 10 and even 15 years (5 of 11).

The proportion of patients not dying from prostatic carcinoma of those who had large, intermediate or small glands (but not cribriform and/or solid tumor) was 24 of $26 = 0.92$. The probability of not dying from prostatic carcinoma in this group was therefore 92%. It is unfortunately not possible to state how much the natural history and how much an effect of treatment is reflected in these data.

The proportion of patients dying from carcinoma of the prostate if cribriform and/or solid tumor is present is $7:14 = 0.50$. The probability that a patient with cribriform and/or solid tumor present died from carcinoma of the prostate was 50% in this patient material. This means that radical prostatectomy may have saved half of these patients. Patients escaping treatment do in some way reflect the natural history of a disease. Survivors may reflect the natural history rather than an effect of treatment. It remains unclear how far endocrine treatment was the reason for the prolonged course in some of these patients.

Amount of tumor. It is not unexpected that the "amount of tumor" found in the radical prostatectomy specimens has impact on corrected survival (Fig. 3). Of 16 patients

with small amounts of tumor none died of prostatic carcinoma. All 9 cancer deaths occurred in the group of 26 patients with more than a small amount of tumor (medium or much). The difference is significant. Further differentiation in "medium" or "much" tumor produced no significant spread of the corrected survival curves (data not shown).

The chance of not dying from prostatic carcinoma if only a small amount of tumor was found in the specimen of the radical prostatectomy was 100%. If more than a small amount of tumor was present (medium or much), the proportion of patients dying from carcinoma was 9 of 26 or 0.35. The chance of dying from carcinoma was therefore 35%.

Table 4 correlates the findings obtained with the parameters "differentiation" and "amount of tumor". It can be seen that in none of the patients with a small amount of tumor cribriform and/or solid formations were found. It is of interest that there are 11 patients with an intermediate or large amount of tumor with no cribriform or solid formations present. Only two of those died of carcinoma.

Unfortunately, we do again not know how much the natural history and how much radical prostatectomy contributes to these figures. Data available from the literature, which will be discussed later, show that in the groups of small, well differentiated tumors (stage A_1, category $T_0pT_1N_0M_0G_1$) identified from TUR or enucleation specimens rarely a patient will die of car-

Fig. 4. Size of cells and corrected survival in 39 patients with incidental prostatic carcinoma

Fig. 5. Nuclear size and corrected survival in 39 patients with incidental prostatic carcinoma
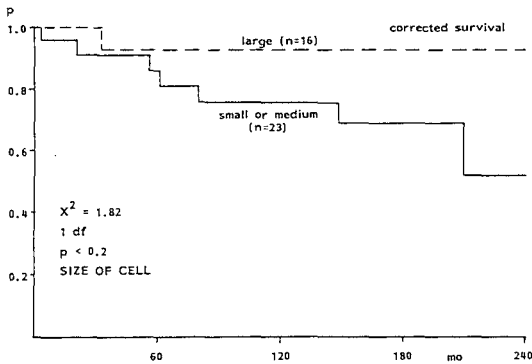

Fig. 6. Nuclear pleomorphism and corrected survival in 39 patients with incidental prostatic carcinoma

cinoma, even if no further treatment is applied [7, 8, 11, 24]. How many of the patients with larger amounts of tumor have been "cured" by radical prostatectomy is an open question. The patients with cribriform and/or solid tumor were about equally distributed within the groups where moderate or large amounts of tumor were seen.

The other architectural parameters studied, the number of formations (pleomorphism) and the amount of stroma present did not have any impact on corrected survival, but showed an almost equal distribution of cancer death for the criteria analysed. For this reason the data are not shown.

Cytological Parameters

These parameters were studied in histological slides at high magnification. It is at variance with the study of the whole material of 484 patients that none of the cytological parameters produced a significant spread of the corrected survival curves [31]. Still, because they indicate trends, some examples will be shown.

Size of Cell. Corrected survival curves are shown in Fig. 4. The parameter "large cells" identifies a group of 16 patients of whom 2 died of prostatic carcinoma. The chance of not dying from the tumor if large cells (but not small and medium size cells) are present is 94%. Of the 23 patients with small or intermediate size cells 7 died of carcinoma. The chance of dying from the tumor in this group is 30%. The difference between the two survival curves is not significant.

Nuclear Size. The corrected survival curves are shown in Fig. 5. The difference is not significant. In the group of 22 patients with small or intermediate size nuclei, 2 patients died of prostatic cancer. The probability of not dying from the disease if large nuclei are not present is 91%. Of the 17 patients with large nuclei, 6 died of prostatic cancer. The probability of dying from the disease is 35%.
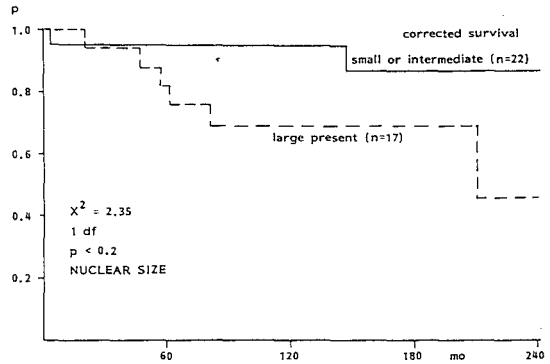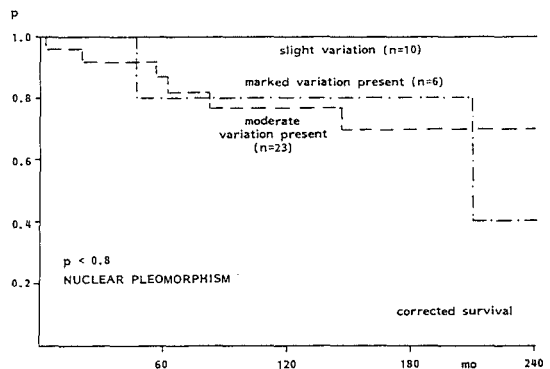
Nuclear Pleomorphism. In the previous study of the whole patient material this parameter separated the patients in 3 groups with a significantly different uncorrected and corrected survival. The corrected survival curves of 39 patients with T0 carcinomas are shown in Fig. 6. "Slight variation" was found in 10 patients of whom none died of carcinoma of the prostate. There was however no significant difference from the two other groups which produce almost coinciding curves. The chance of not dying from prostatic carcinoma within the group of 10 patients with slight variation was 100%. The chance of dying from carcinoma with moderate or marked variation was 28%.

Mitoses were present in only 8 of the 39 specimens. The results are shown in Fig. 7. Patients with mitoses in their tumor had a slightly greater chance of dying of prostatic carcinoma. The difference is not significant.

The other parameters: presence or absence of prominent nucleoli, clear vs. granular cytoplasm, presence or absence of nuclear vacuoles did produce almost coinciding corrected survival curves and are therefore not shown.
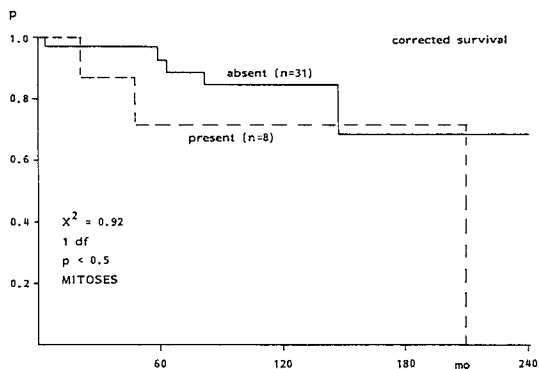
60

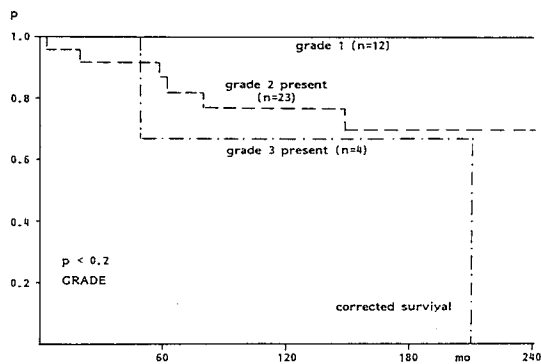**Fig. 7.** Mitoses and corrected survival in 39 patients with incidental prostatic carcinoma

Fig. 8. Grade and corrected survival in 39 patients with incidental prostatic carcinoma

Table 5. Indicators of good and poor prognosis in 40 patients with incidental carcinoma of the prostate treated by total perineal prostatectomy

Indicators for not dying of prostatic carcinoma			Indicators for dying of prostatic carcinoma		
Indicator	Number of patients at risk	Chance of not dying of carcinoma	Indicator	Number of patients at risk	Chance of dying of carcinoma
Small amount of tumor	15	100%	Grade 3	4	50%
Grade 1	12	100%	Cribriform or solid tumor	14	50%
Nuclear variation, slight	10	100%	Nuclear size, large	17	35%
Size of cell, large	16	94%	Moderate or large amounts of tumor	26	35%
Glands, large, medium or small	26	92%	Size of cell, small or intermediate	23	30%
Size of nucleus, small or intermediate	22	91%			
Grade 2	23	74%			

Grade. The parameter "grade" computed by the pathologist did not produce significantly different corrected survival curves. These are shown in Fig. 8. For 12 patients with grade 1 tumors the chance of not dying from prostatic cancer was 100%. Two of 4 patients with grade 3 tumor died of their tumor. Unfortunately there is a large group of 23 patients in the middle of which 6 died of prostatic cancer. For patients in this group the chance of dying from the disease was 26%, the chance of not dying from carcinoma 74% respectively.

In summarizing the results it can be said that a number of strong indicators of a benign and malignant course of the disease has been identified within this group of 39 patients. Table 5 puts these in an order of sequence.

Finally it should be mentioned that 18 of the 39 and 21 of the 55 patients had penetration of the fibrous capsule by tumor and belonged to the category $T_0pT_3N_0M_0G_{1-3}$.

Discussion

Death Rate from Prostatic Cancer

In this series of 39 patients the rate of death from carcinoma of the prostate is 20.5%. In comparison to similar data in the literature, this seems to be relatively high. Byar, in reporting on the VACURG focal study and study I [7] found progression in 3 of 61 patients, of which 31 were treated by radical prostatectomy (5.0%). Sheldon [32] and Battaglia [1] in reviewing the literature, found an incidence of progression varying from 4.76% to 33.33% and of cancer deaths varying from 1.9% to 50.0% for various series of patients. There is agreement that tumor size as determined by histological examination and grade are the main determinants.

The high progression rate in our series could be due to a selective accumulation of large and poorly differentiated tumors. Sixteen of 42 (38%) were considered

61

small and 26 intermediate or large (65%) with 17 (46%) penetrating the capsule, 26 of our patients had tumor forming glands (65%) 14 had cribriform or solid tumors with an obviously poor prognosis (35%). Figures published by Dhom [15], who found in 141 T_0 patients 41% well differentiated and in 30% small amounts of tumor and data reviewed by Sheldon [32] suggest however, that our findings are well within the range of the data reported in the literature. Another possible determinant for the incidence of cancer deaths is the time of follow-up. In this series only 6 of 11 patients died of cancer within the first five years and 4 died later than 10 years after the diagnosis was made.

Predictive Value of Results

In spite of a small number of patients significant differences in corrected survival have been found for some of the parameters analysed. Some of these show indeed a strong association with a benign or malignant course of the disease in this study. The fact that all patients were treated by radical prostatectomy and that most patients received some kind of endocrine treatment limits the general applicability of these observations.

The decision to exclude a patient with a presumably non-progressing tumor from treatment requires solid knowledge of the natural history of a given tumor without treatment. In the patient material presented herein, there is no way of knowing how many of the patients who did not die of cancer of the prostate would have died of this disease if radical prostatectomy had not been carried out. Still, if in any given group no patient or a very low percentage of patients died of prostatic cancer (escaped treatment) this would indicate, that the risk of dying from such a tumor is not very high. Careful comparison of the groups identified with the experience of others who have not further treated similar patients helps to interpret these results.

If on the other hand large proportions of patients die of prostatic carcinoma in subgroups identified by use of prognostically important parameters, it is safe to state, that in these groups the treatment applied may not be very effective.

Indicators for no Progression

Three parameters identify groups of 10–16 patients of whom none died of the disease under study (small amount of tumor, grade 1 and slight nuclear variation). Many of these coincide within the three groups in the sense that most of the small tumors are well differentiated. However the largest number of those not showing progression is identified by the parameter "small amount of tumor".

This finding is much in agreement with the literature. Cantrell [8] has recently found that only 2% of 48 patients who had involvement of less than 5% of

their resected and examined BPH material involved by tumor showed progression. This parameter was a stronger predictor of progression than grade and other estimates of tumor volume like the diameter of the largest focus, the number of foci, the number of involved blocks or the percentage of involved blocks. These patients received no treatment after the initial diagnosis. It was also demonstrated that all of these small tumors, which amounted to 60% of the whole group, were well differentiated. Considering the information related by a number of other authors who present series of patients not receiving further treatment after the initial diagnosis has been made [7, 11, 24], it seems safe to conclude that such patients do not require treatment.

The question remains however, how safely this group can be identified. Suggestions for defining T_0pT_1 tumors (stage A_1) have recently been summarized by Sheldon [32]. They include: focal, less than 3 chips, only one lobe involved; focal, well differentiated; focal, well differentiated, less than 5 chips; less than 3 foci. The TNM booklet [33] states: focal (single or multiple) carcinoma.

All these definitions may or may not be suitable. It is necessary to remember that the incidence and the tumor volume of T_0 carcinoma depend on the number of sections taken, on the resection technique and on the localization of most carcinomas close to the prostatic capsule. Variation of preparative techniques could easily introduce a factor of 2–3 of variation of incidence and tumor volume.

Blackard [3] reported that 87.5% of 24 patients who underwent radical prostatectomy for T_0 carcinoma had residual tumor. Similar figures published by Heaney [24] and Lehman [27] are 50% and 60%. For these reasons it will be very difficult to establish a generally accepted definition of the tumor volume that should be used as a cut off point for not considering treatment. Cantrell has indicated the preparative technique used at the Johns Hopkins Hospital. It may be suitable to use this technique and a cut off point of 5% as suggested in his paper. It may, however, be easier and just as effective to define the incidental carcinoma not requiring further treatment as a well differentiated, small tumor with emphasis on differentiation *and* size. In the series of patients presented in this paper no patient with a small or intermediate amount of tumor who did not have cribriform or solid formations died of tumor.

The best separation of patients with respect to death of carcinoma is achieved in this series by the parameters "small, intermediate or large glands" present (but no cribriform and/or solid tumor) and "cribriform and/or solid tumor present" on the other side. It is surprising that this parameter produces a sharper separation than "grade" and "small amount of tumor". The chance of not dying from carcinoma with "small, intermediate or large glands" (but not cribriform or solid

tumors present) was 92% for 26 involved patients. Seven of the remaining 14 who had cribriform and/or solid tumors died of carcinoma. The first group contains the 15 patients with small amounts of tumor and 9 patients with larger tumor volumes, only one of whom died of prostatic carcinoma. It has been pointed out by Sheldon [32] that this group may represent a separate entity ($T_0pT_{2-3}G_1$). This group of patients may have benefitted from radical prostatectomy. The natural history of larger, well-differentiated tumors is unknown.

Indicators for Progression

The prognosis of poorly differentiated tumors is poor in spite of treatment if these are defined as showing cribriform and/or solid growth patterns. It must however be considered that 18 of these patients had capsular penetration and belong to the pT_3 category. They are examples for the understaging known to occur in the T_0 category [17].

Radical perineal prostatectomy may cure half of these patients. Probably lymph node staging can identify the group of potentially curable patients with poorly differentiated tumors as suggested by the data of Donohue [17]. The results presented cast some doubt on the effectiveness of radical prostatectomy in patients with T_0G_3 tumors.

At variance with previous results is the fact that one of the cytological parameters produced a significant spread of the corrected survival curves. Small numbers probably play a role. The future will show, whether determination of nuclear pleomorphism (variation in size and shape of the nucleus) by morphometrical techniques will be of major value for better identification of risk factors for this tumor as has recently been shown for other groups of patients [4, 16].

Indications for Treatment

It was mentioned earlier that the uncorrected survival of the 55 patients with T_0 prostatic carcinoma was identical with their theoretical life expectancy during 10 years (Fig. 1). This was also observed by several other authors [2, 7, 8, 11, 23, 24]. However, in several series a significant impact of poorly differentiated and/or large T_0 carcinomas on uncorrected survival was found [2, 23, 24].

As discussed above, a subgroup of small and well differentiated $T_0pT_1G_1$ carcinomas is identifyable which is very unlikely to progress and kill the patient. The chance for death from carcinoma in untreated populations has been demonstrated to be smaller than 2% in at least 5 publications [8, 11, 24, 32, 34]. Also, lymph node dissection seems to be unnecessary because the available experience recently summarized by Sheldon [32] and the data of Catalona [10] which have shown no lymph node involvement in A_1 patients. Also, we would not advocate a second TUR because empirically the findings established by one resection seem to be sufficient for decision making.

Within the material presented in this publication a group of 11 patients (27%) with intermediate or large amounts of tumor and absence of cribriform and/or solid formations is found of which only two escaped treatment (category $T_0pT_{2-3}N_0M_0G_1$). Such a group has also been identified by others [8, 32] who have not treated these patients. The rate of progression was low. However, the natural history of these selected tumors is insufficiently known for definite conclusions. These patients could benefit from radical prostatectomy or radiotherapy. Lymph node staging may help to further classify these patients, but it also seems to be justified to delay invasive staging and treatment and to follow the patient with 3-monthly rectal examination or ultrasound measurements of the prostate.

Patients with moderately or poorly differentiated T_0 prostatic carcinoma have been shown to have a high chance to progress and to die from their tumor. Disease related death rates were found to be 14–50% [32]. In this series 7 of 14 patients with cribriform and/or solid carcinomas died of their tumor. In the TNM system these tumors can be classified as $T_0pT_{2-3}N_xM_0G_{2-3}$. Considering the high risk of progression and death from carcinoma as well as the significant impact on overall survival one certainly wants to treat these patients. Unfortunately it is doubtful which form of treatment is most effective. In this series a considerable number of long term survivors was observed.

Radical prostatectomy was used by several authors and lead to the report of long term results [3, 21, 27, 30]. High complication rates were described by most. Reliable information on rates of progression for the large, not well differentiated tumors which are under discussion are rare. All studies suffer from the unavailability of control groups. In the only prospective, randomized studies reported [7], the numbers of patients are too small to detect differences in progression rates which could be significant for treatment decisions. Still, it is remarkable that in the VACURG "focal study" and in "study I" no significant differences in progression were found for the radical prostatectomy and for the placebo groups. Progression occurred significantly more frequent in poorly differentiated tumors (Gleason grade 6–10, 12 of 60 patients, 20%). From our own material we must conclude that 50% of these patients escaped radical prostatectomy. The relatively small proportion of long term survivors however, may or may not justify surgical staging and radical prostatectomy in these cases.

The effect of endocrine treatment cannot be evaluated in the present series and was not shown by others to have any beneficial effect [6, 11, 24, 27]. Adverse palliative effects were described by Byar and Heaney. No evidence was found that radiotherapy is effective in these patients.

References

1. Battaglia S, Barbolino G, Botticelli AR (1979) Early (stage A) prostatic cancer. Virchows Arch [Pathol Anat] 382:245–259
2. Bauer WC, McGavran MH, Carlin MR (1960) Unsuspected carcinoma of the prostate in suprapubic prostatectomy specimens. Cancer 13:370–378
3. Blackard CE, Mellinger GT, Gleason DF (1971) Treatment of stage I carcinoma of the prostate: a preliminary report. J Urol 106:729–733
4. Blom JHM, ten Kate FWJ, Schroeder FH, van der Heul RO (1982) Nuclear pleomorphism as a parameter in grading prostatic carcinoma. A morphometrical study. Presented at the 77th Annual Meeting of the AUA, Kansas City 1982
5. Boxer RJ (1977) Adenocarcinoma of the prostate gland. Urol Surv 27:75–79
6. Byar DP, Veterans Administration Cooperative Urological Research Group (1972) Survival of patients with incidentally found microscopic cancer of the prostate: results of a clinical trial of conservative treatment. J Urol 108:908–913
7. Byar DP, Corle DK, Veterans Administration Cooperative Urological Research Group (1981) VACURG randomized trial of radical prostatectomy for stages I and II prostate cancer. Urology [Suppl] 17:7–11
8. Cantrell BB, de Klerk DP, Eggleston JC, Boitnott JK, Walsh PC (1981) Pathologic factors that influence prognosis in stage A prostate cancer: the influence of extent versus grade. J Urol 125:516–520
9. Catalona WJ, Scott WW (1978) Carcinoma of the prostate: a review. J Urol 119:1–4
10. Catalona WJ, Stein AJ (1982) Staging errors in clinically localized prostatic cancer. J Urol 127:452–456 (1982)
11. Correa RJ Jr, Anderson RG, Gibbons RP, Mason JT (1974) Latent carcinoma of the prostate – why the controversy? J Urol 111:644–646
12. Corriere JN Jr, Cornog, JL, Murphy JJ (1970) Prognosis in patients with carcinoma of the prostate. Cancer 25:911–918
13. deVere White R, Paulson DF, Glenn JF (1977) The clinical spectrum of prostate cancer. J Urol 117:323–327
14. Denton SE, Choy SH, Valk WI (1965) Occult prostatic carcinoma diagnosed by the step-section technique of the surgical specimen. J Urol 93:296–298
15. Dhom G, Hautumm B (1975) Die Morphologie des klinischen Stadiums 0 des Prostatacarcinoms (incidental carcinoma). Urologe A 14:105–111
16. Diamond DA, Berry SJ, Umbricht C, Jewett HJ, Coffey DS (1982) Computerized image analysis of nuclear shape as a prognostic factor for prostatic cancer. Prostate 3:321–332
17. Donohue RE, Pfister RR, Weigel JW, Stonington OG (1977) Pelvic lymphadenectomy in stage A prostatic cancer. Urology 9:273–275
18. Ekman H (1959) Late results of prostatectomy for benign prostatic hyperplasia. A clinical study based on 370 cases. Acta Chir Scand [Suppl] 250:1–141
19. Franks LM (1954) Latent carcinoma of the prostate. J Pathol Bacteriol 68:603–616
20. Golimbu M, Schinella R, Morales P, Kurusu S (1978) Differences on pathological characteristics and prognosis of clinical A_2 prostatic cancer from A_1 and B disease. J Urol 119:618–622
21. Goodwin WE (1952) Radical prostatectomy after previous prostatic surgery: technical problems encountered in the treatment of occult prostatic carcinoma. JAMA 148:799–803
22. Greene LF, Simon HB (1955) Occult carcinoma of the prostate. Clinical and therapeutic study of eighty-three cases. JAMA 158:1494–1498
23. Hanash KA, Utz DC, Cook EN, Taylor WF, Titus JL (1972) Carcinoma of the prostate: a 15-year follow-up. J Urol 107:450–453
24. Heaney JA, Chang HC, Daly JJ, Prout GR (1977) Prognosis of clinically undiagnosed prostatic carcinoma and the influence of endocrine therapy. J Urol 118:283–287 (1977)
25. Jewett HJ (1975) The present status of radical prostatectomy for stages A and B prostatic cancer. Urol Clin North Am 2:105
26. Kaplan EL, Meier P (1958) Nonparametric estimations from incomplete observations. J Am Stat Assoc 53:457–458
27. Lehman TH, Kirchheim D, Braun E, Moore R (1968) An evaluation of radical prostatectomy for incidentally diagnosed carcinoma of the prostate. J Urol 99:646–650
28. Mostofi FK (1975) Grading of prostatic carcinoma. Cancer Chemother Rep (Part I) 59:111–117
29. Mostofi FK (1976) Problems of grading carcinoma of the prostate. Semin Oncol 3:161–169
30. Nichols RT, Barry JM, Hodges CV (1977) The morbidity of radical prostatectomy for multifocal stage I prostatic adenocarcinoma. J Urol 117:83–84
31. Schroeder FH, Blom JHM, Hop WCJ, Mostofi FK (1983) Mostofi grading of prostatic cancer – a study of 346 patients. Prostate (accepted for publication)
32. Sheldon CA, Williams RD, Fraley EE (1980) Incidental carcinoma of the prostate: a review of the literature and critical reappraisal of classification. J Urol 124:626–631
33. TNM – classification of malignant tumors, 3rd edn (1978) Harmer MH (ed) UICC – International Union against Cancer, Geneva
34. Varkarakis M, Castro JE, Azzopardi JG (1950) Prognosis of stage I carcinoma of the prostate. Proc R Soc Med 63:1077–1081
35. Whitmore WF (1956) Hormone therapy in prostatic cancer. Am J Med 21:697–713
36. Whitmore WR Jr (1973) The natural history of prostatic cancer. Cancer 32:1104–1112

Chapter 4

Grading of prostatic cancer (I) An analysis of the prognostic significance of single characteristics

F.H. Schröder, J.H.M. Blom, W.C.J. Hop, and F.K. Mostofi

Grading of Prostatic Cancer (I): An Analysis of the Prognostic Significance of Single Characteristics

Fritz H. Schroeder, Jan H.M. Blom, Wim C.J. Hop, and F.K. Mostofi

Department of Urology (F.H.S., J.H.M.B.), the Department of Biostatistics (W.C.J.H.), Erasmus University, Rotterdam, The Netherlands and the Armed Forces Institute of Pathology (F.K.M.), Washington, DC

This paper contains the first part of an attempt to quantitate the impact on prognosis of various parameters used in grading of prostatic cancer. Out of 346 patients of Elmer Belts series, 113 were identified whose tumors showed homogeneity with respect to single characteristics of a total of 12 parameters applied for grading in Mostofi's system. By this procedure it was possible to eliminate the possible influence on prognosis of the presence of several tumor formations within the same tumor. By using overall and intercurrent death corrected survival as end points, the impact of each of the 12 parameters on prognosis was studied. Only the architecture of the tumor (the parameter "glands"), variation in size and shape of the nucleus (anaplasia), and grade significantly influenced overall survival. In addition, corrected survival was significantly dependent on the amount of tumor seen and on the presence of mitoses.

Subsequently, an attempt was made to replace "grade" by single parameters which had been shown previously to be of prognostic significance. It turned out that this was not possible. Grade is largely dependent on architecture and nuclear pleomorphism, but neither one of these parameters alone can reproduce "grade." Multivariate analysis was next used to further determine the prognostic weight of the individual parameter, and, if possible, to construct a new, more efficient grading system. These results will be reported separately [8]. It is unknown at the present time what the impact of several architectural formations within the same tumor on prognosis may be. The number of different formations found ranges from 1 to 4 in this material; 668 different formations belonging to 346 tumors were graded. The results of this analysis will be reported in part two of this series of papers [7].

Key words: prostatic cancer, grading, single tumor formations, correlation survival and grading

INTRODUCTION

The morphological picture of the cell and the nucleus as well as the architectural pattern of prostatic cancer are related to the prognosis of patients bearing this tumor. The computation of such prognostic parameters has been called "grading of malignancy." Grading of prostatic cancer has become common clinical practice. It serves as a prognostic indicator and in some instances as the basis of therapeutic decisions. Many different systems for grading of prostatic cancer have been proposed and correlation to prognosis has been established [1–5].

At present, there is no generally accepted system which is simple enough to be reproduced easily, and it is still unclear which system provides the most reliable prediction of prognosis. Furthermore, it remains uncertain which role fine-needle aspiration cytology may play and how grading obtainable in such specimens compares to grading obtainable from histological biopsy samples.

One of the difficulties of grading prostatic carcinoma lies in the fact that usually these tumors are pleomorphic and consist of two or more architectural formations, each with a different grade. It is still unknown which formation determines prognosis, the predominant pattern or the most malignant one, which may be very small.

The purpose of the work presented in this paper was to study some of these clinically relevant questions in a large series of patients with a long-term follow up. Mostofi's system was used because it is based on the evaluation of multiple architectural and cytological parameters and because it takes into account the presence of multiple formations within one tumor. In fact, this system includes most of the parameters used in other grading systems and its comprehensive evaluation should therefore also contribute to their understanding.

The goals of this study are as follows: 1. An evaluation of the prognostic impact of each of the parameters proposed by Mostofi [6]. The system was slightly simplified, as shown in Table I. 2. To obtain an answer to the question of whether "grading" can be replaced by the analysis of single parameters. 3. To establish the prognostic significance of the presence of single vs multiple histological patterns. Does the most malignant pattern determine the outcome; does the presence of well-differentiated tumor improve prognosis? The results of this study are presented in the second paper

TABLE I. Architecture and Cytology of Prostate Carcinoma

Tumor architecture		Cells and nucleus	
Parameters	Characteristics	Parameters	Characteristics
Pleomorphism (number of formations)	One formation Two formations Three formations Four formations	Size of the nucleus	Small Intermediate Large
Glands	Small glands Intermediate glands Large glands Cribriform glands No glands	Variation in size and shape of the nucleus (nuclear anaplasia) Nuclear vacuoles	Slight Moderate Marked Present Absent
Amount of tumor	Small Medium Much	Prominent nucleoli	Present Absent
Amount of stroma	Normal Increased Decreased	Mitoses Size of the cells	Present Absent Small Medium Large
Grade	Grade 1, grade 2, grade 3	Cytoplasm	Clear Granular None

of this series [7]. 4. Finally, an attempt will be made by taking advantage of the technique of multivariate analysis to determine the degree of independence of the different parameters from each other and their relative prognostic importance. This information will be used to construct a system of prognostic factors which can be used as a new grading system. The results will be presented in the third paper of this series [8].

Obviously, the questions indicated under 1 and 2 can best be answered by considering tumors which are homogeneous with respect to individual parameters shown in Table I. The subjects indicated under 3 and 4 must be dealt with by also considering patients with multiple architectural formations and multiple characteristics of the various parameters present in the same tumor.

The end point of this study, the time of the death of the patient, is influenced by multiple factors. Some have been identified as being significant: age at the time of diagnosis, other disease and local tumor extension. Other factors that may be of significance are treatment by radical prostatectomy, adjuvant treatment by endocrine measures, the time of diagnosis, and preoperative treatment. An analysis of these parameters is reported elsewhere [9]. Their impact has been reduced in this paper by considering cancer death (survival corrected for intercurrent death) and overall survival. By studying the patients who obviously have not been cured, a glimpse of the natural history of this disease should be possible.

For the understanding of this work it is essential to define categories (parameters) and subcategories (characteristics) for classification of the morphological findings. This was done in Table I.

MATERIALS AND METHODS

The series of patients on which this study is based has been subject to several reports [10–13]. At the time of the last review of the clinical data in May 1980, data from 484 patients were collected. They were all treated by total perineal prostatectomy for prostatic carcinoma of the categories T_0, T_1, T_2, $T_3N_XM_0$. The first operation was carried out in October 1938 and the last one in November 1971. The series represents the life-long experience of the late Dr. Elmer Belt of Los Angeles.

Unfortunately, the histological slides of the total prostatectomy specimen of only 346 of the 484 patients were available for review. These were reviewed by one of the authors (F.K.M.) according to the evaluation sheet shown as Table I. The microscopic analysis was carried out without knowledge of clinical information. The slides were evaluated for architectural characteristics at low microscopic power, and subsequently cells and nuclei were looked at with high-power magnification.

In the slides of 346 patients a total of 668 histological patterns were found. The number of architectural formations per tumor cell varied from one to four. The relevant information is summarized in Table II. For every one of these different patterns a separate evaluation sheet was completed.

All clinical and pathological information was stored on data carriers for computer analysis. Survival data were calculated using the technique of Kaplan and Meier [14]. Survival was studied for each of the morphological parameters listed in Table I. Since death from other disease was found to be very frequent, survival curves were also corrected for intercurrent death. Such survival curves allow study of the impact of prostatic cancer on survival more specifically (cancer death). The possible effect

TABLE II. The Number of Tumor Formations and
Their Distribution in 346 Patients With Prostatic
Carcinoma

No. of tumor formations	No. of patients	%	No. of formations
1	113	32.6	113
2	152	44.0	304
3	73	21.1	219
4	8	2.3	32
Total	346	100	668

of treatment cannot be recognized in overall survival or in the corrected data since no control group is available.

To solve the problem of the impact of multiple architectural and multiple cytological characteristics within one tumor on the prognosis of the patient, the tumors with single characteristics were analysed separately. There were 113 patients with only one tumor formation, 209 patients with only one size of nuclei (small, medium, or large), and 232 patients with only granular or clear cytoplasm. These tumors were used to identify the prognostic significance of the individual parameters and the order of sequence of the prognostic weight of the different characteristics. The chi-square test was applied to evaluate the significance of differences in the survival curves obtained.

Deceased patients with documented recurrent or metastatic carcinoma of the prostate present at their last evaluations were considered as cancer deaths.

RESULTS
Tumor Architecture

The survival curves calculated by the use of the parameter "glands" are shown in Figure 1A and 1B. Figure 1A, the crude overall survival according to the individual characteristics of the parameter "glands," indicates two groups of prognostically different patients: those with small and intermediate glands and those with cribriform and solid tumors. Patients with large glands do well at 5 years. The subsequent decrease in survival can be identified as being due to intercurrent death when the same patients are traced in Figure 1B (bottom). Still, overall, the differences are significant (p $\leq$ 0.002). The tendency toward two prognostic groups is confirmed when the data are corrected for intercurrent death. The number of cancer deaths is equally low in patients with small, intermediate, or large glands; 20 patients with cribriform and three patients with solid tumors did significantly less well (p $\leq$ 0.000).

The number of patients within each characteristic differs slightly between top and bottom Figure 1B. This is due to the fact that within the whole series of 484 patients, the cause of death is unknown in 23. Patients with an unknown cause of death are omitted from all evaluations of survival corrected for intercurrent death.

In Figure 2, the parameter "amount of tumor" is analysed. No significant differences are found between the crude survival curves. After correction for intercurrent death, it becomes evident that within the group "small amount of tumor"
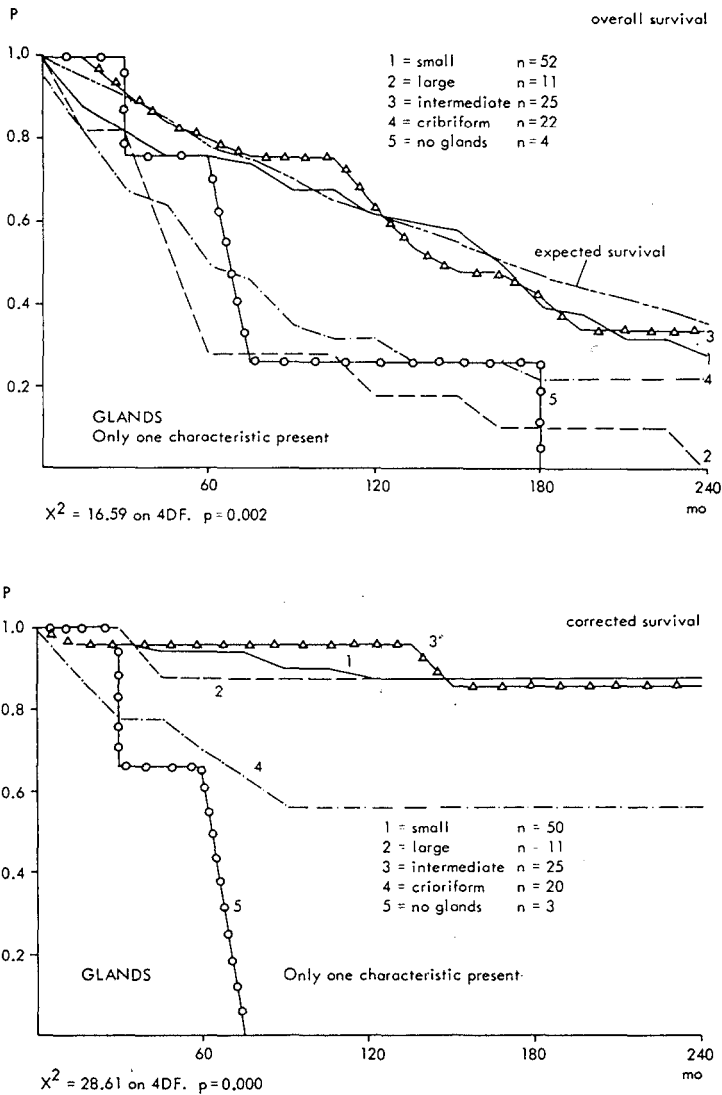
69

Fig. 1. Top: Overall survival by tumor architecture (glands). Only one characteristic is considered. The overall difference is significant. Bottom: Corrected survival of the same group of patients. Two prognostically different groups are evident.

(n = 54), only two (4%) patients died of prostatic carcinoma, as compared to much larger proportions of 30 and 40% in the other subgroups. The overall differences are significant (p $\leq$ 0.003). No significant differences are found for normal and decreased amount of stroma (Fig. 3).

Both parameters, ie, amount of tumor and amount of stroma, were evaluated in routine sections; sections of the whole prostates were not available. Variation within the whole tumor can not therefore be taken into account. Thus, absolute measurements
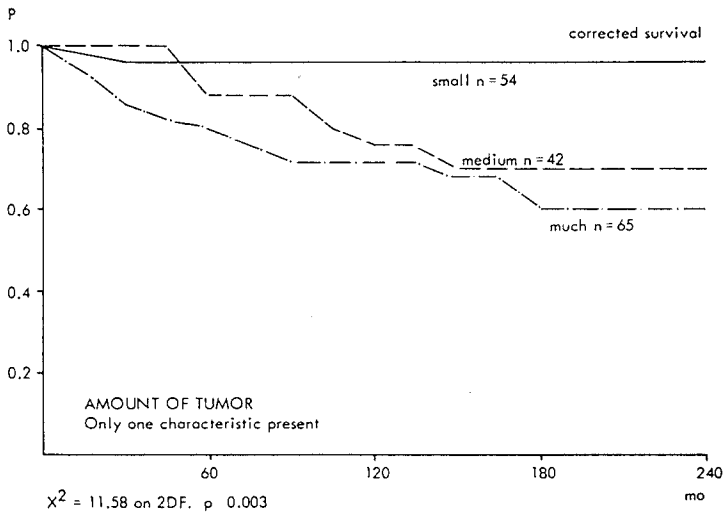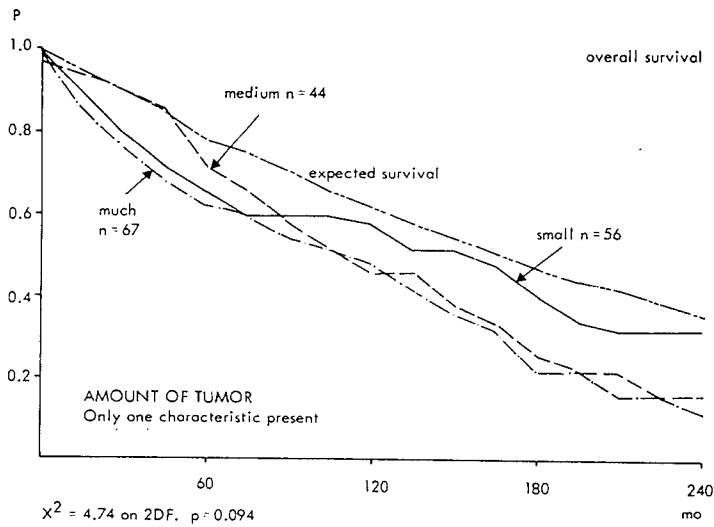
Fig. 2. Top: Overall survival by amount of tumor. Tumors with single characteristics are considered. There is no significant difference. Bottom: Corrected survival of the same group of patients. Again, there is no significant difference. Only the group of 54 patients with small amounts of tumor has a significantly better prognosis than the rest.

or estimates which are representative for the whole tumor are not possible in this study.

The cytological parameters and characteristics listed in Table I address the nucleus and the cytoplasm. The parameter "nuclear size" reflects the nuclear-cytoplasmic relation. Figure 4 shows the results obtained. More patients with large nuclei
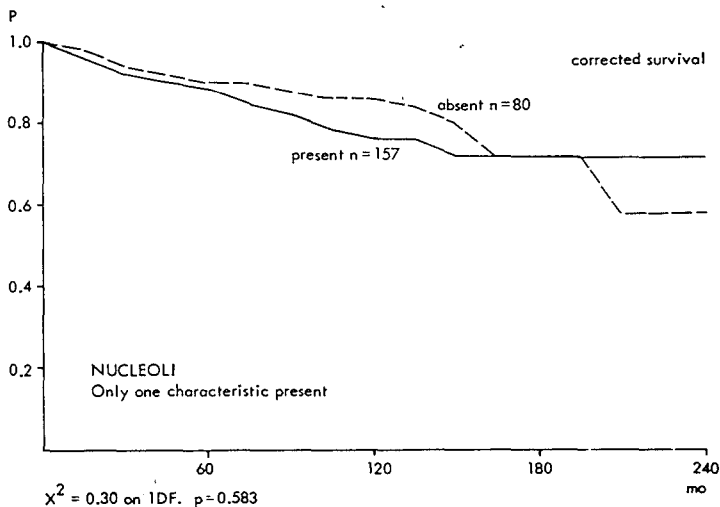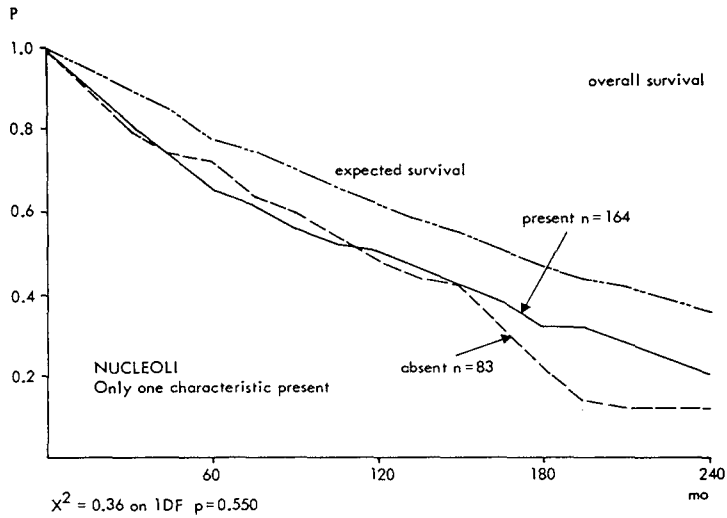
Fig. 3. Top: Overall survival by amount of stroma. No significant difference betweeen normal and decreased is observed. Bottom: Corrected survival: same parameters, no significant difference.

die of prostatic cancer, and the corrected curve for intermediate nuclei runs intermediate. However, the differences are not significant.

Nuclear variation in size and shape, ie, nuclear pleomorphism, is evaluated in Figure 5. Overall survival data and corrected survival differ significantly within the slight, moderate, and marked variation groups (p $\leq$ 0.017 and p $\leq$ 0.000). Within the group of 56 patients with slight variation, no patient died of carcinoma within the first 5-year interval. After 10 years, four patients, after 15 years, six patients, and after 20 years, eight patients, or 20% succumbed to their malignancy. Patients with

72
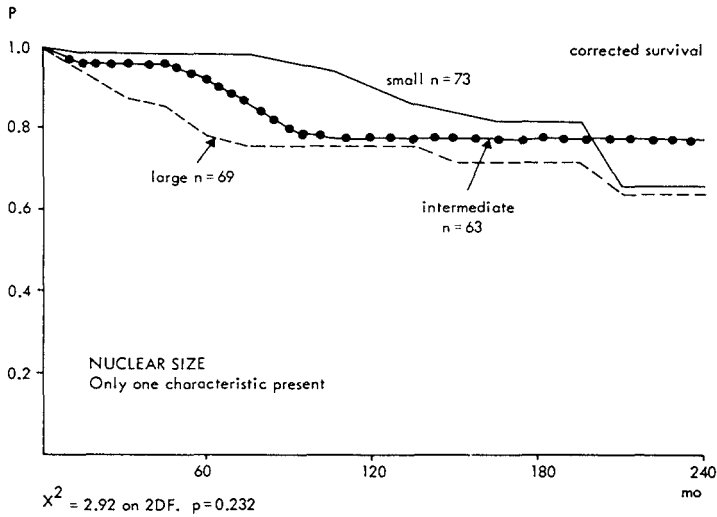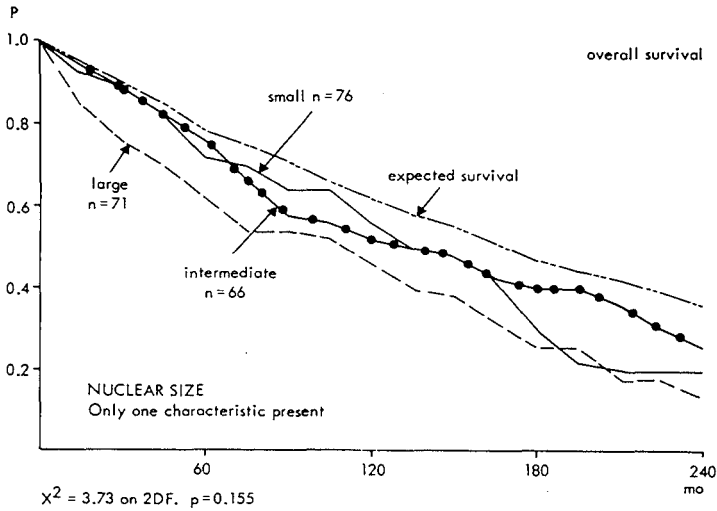
Fig. 4. Top: Overall survival by nuclear size, only one characteristic is present. Bottom: Corrected survival: same patients, same parameter, no significant difference.

marked variation in size and shape had a much greater chance, about 50%, of dying of their disease within the first 5 years, but some cancer deaths in this group also occurred 15 to 20 years after the diagnosis was made.

The presence and absence of nuclear vacuoles (Fig. 6) and the presence and absence of nucleoli (Fig. 7) have no bearing on prognosis when overall and intercurrent death corrected survival is considered.

The presence and absence of mitoses and its effect on survival is analysed in Figure 8. The number of tumors with mitoses is very small. But if mitoses are
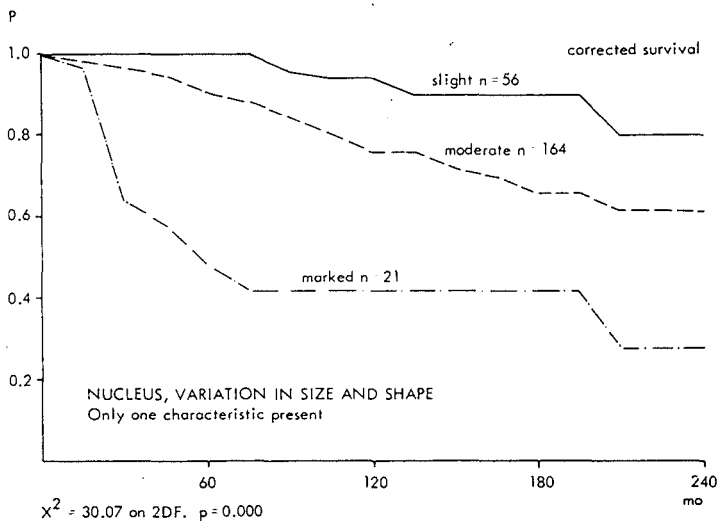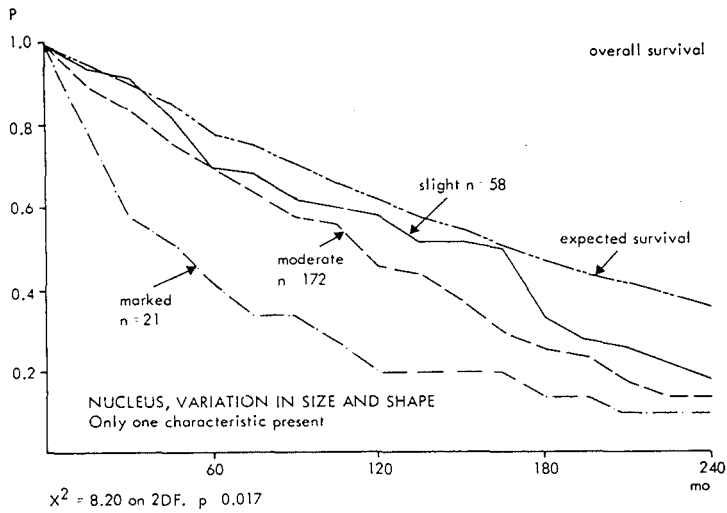
Fig. 5. Top: Overall survival by nuclear variation in size and shape (anaplasia), tumors with single characteristic. The difference between the three curves is significant. Bottom: Corrected survival: same patients, and same parameter, highly significant difference among the three groups.

present, significantly more cancer deaths have occurred (p ≦ 0.000). Also, the survival curve of this small group is suppressed, but the overall difference is not significant.

Analysis of all sizes (small, medium, and large) is given in Figure 9. No significant difference was found for the resulting overall survival curves, nor was it found for the corrected survival curves, but within the group of 41 patients with large
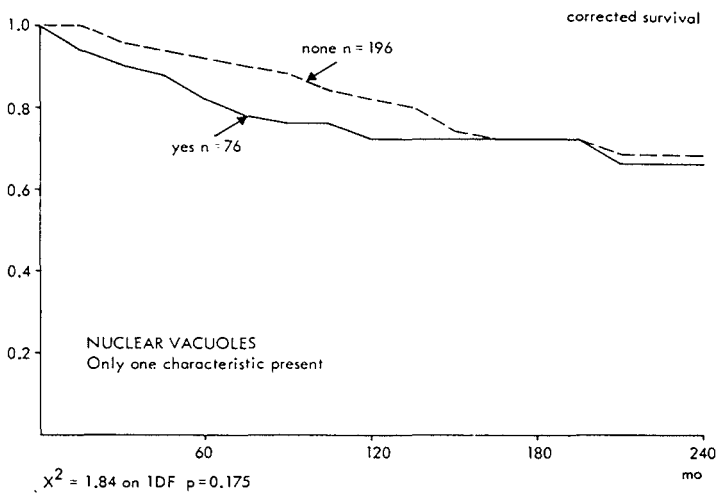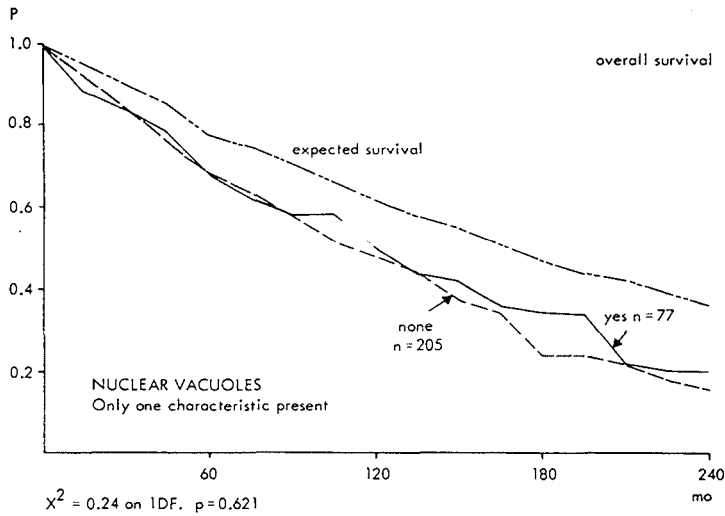
Fig. 6. Top: Overall survival by nuclear vacuoles, tumors with single characteristics. Bottom: Corrected survival: same patients, same parameter, no difference.

cells, significantly fewer cancer deaths occurred as compared to the group with small and medium-size cells.

No differences in survival were found for patients with clear or granular cytoplasm as the only characteristic present (Fig. 10).

Finally, grade (the pathologists' computation of all information reviewed in the histological slides) is subdivided into grades 1, 2, and 3. The impact of grade on prognosis with only one characteristic present is shown in Figure 11. Crude survival curves and survival curves corrected for intercurrent death show significant overall
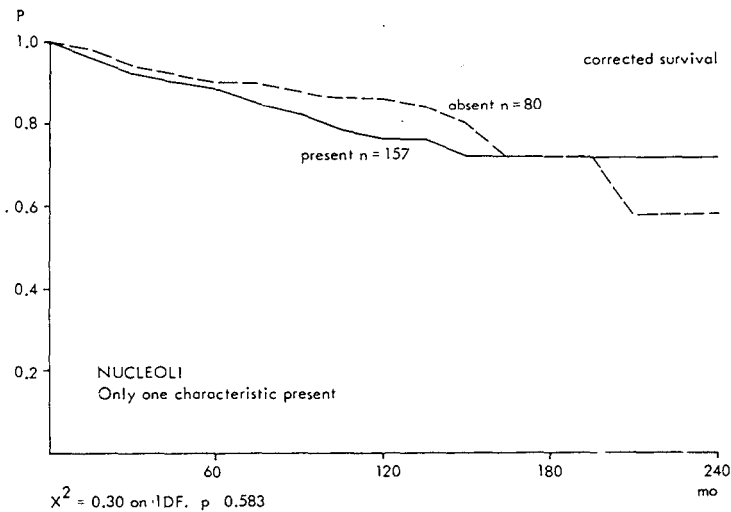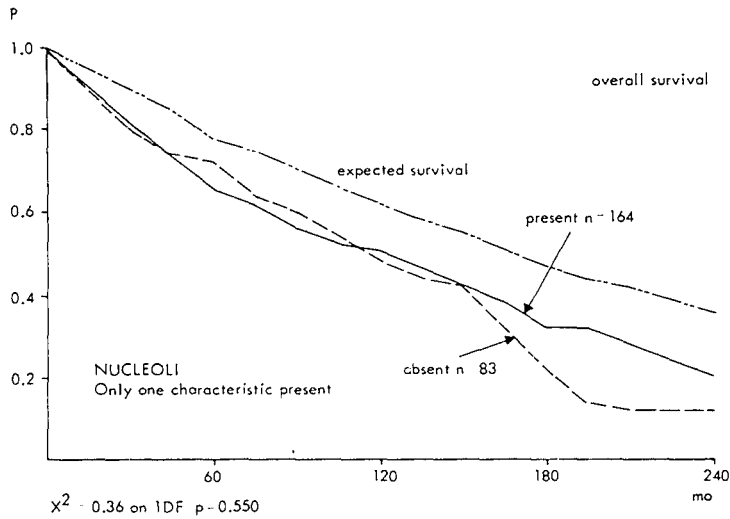
75

Fig. 7. Top: Overall survival by nucleoli, tumors with single characteristics. Bottom: Corrected survival: same patients and same parameters, no significant difference.

differences (p $\leq$ 0.006 and p $\leq$ 0.000). All patients dying of grade 3 carcinoma were dead within 9 years, while even after 15 years patients in the other groups continue to die from their disease.

The results obtained in this analysis of 11 architectural and cytological parameters in patients with only one characteristic of each parameter present are summarized in Table III. Significant differences in overall and intercurrent death corrected survival were only found for three parameters: the architectural pattern (glands), variation in size and shape of the nucleus, and grade. The architectural patterns described allow
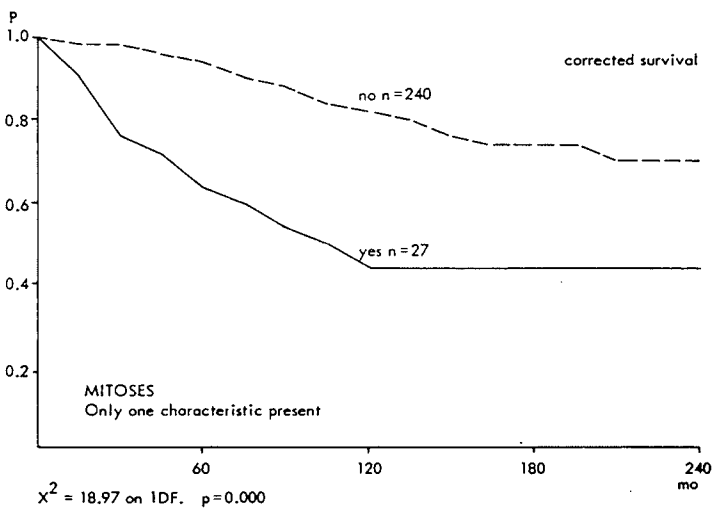
76

P
1.0
0.8
0.6
0.4
0.2

overall survival

expected survival

no n = 250

yes n = 28

MITOSES
Only one characteristic present

60   120   180   240
mo

$x^2 = 2.77$ on 1DF. p = 0.096

P
1.0
0.8
0.6
0.4
0.2

corrected survival

no n = 240

yes n = 27

MITOSES
Only one characteristic present

60   120   180   240
mo
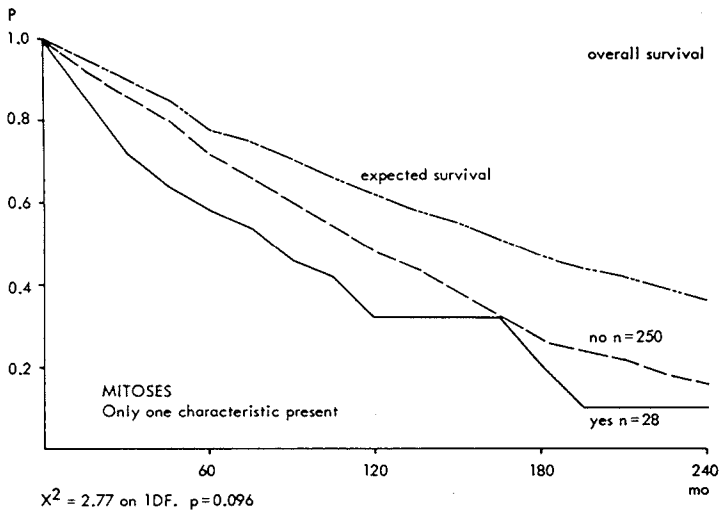
$x^2 = 18.97$ on 1DF. p = 0.000

Fig. 8. Top: Overall survival by presence or absence of mitoses in tumors with single characteristics. No significant difference. Bottom: Corrected survival: same patients and same parameter; two prognostically different groups are identified.

for the distinction of two prognostically different groups; three groups can be identified for the parameters nuclear pleomorphism and grade. The sequence of malignancy of the characteristics is evident from Figures 1, 4, and 11.

Intercurrent death corrected survival reflects the biological behaviour of individual tumors more strongly than the overall survival. The survival curves are not influenced by varying numbers of patients who died from other disease. For this reason a number of less strong prognostic determinants become significant when
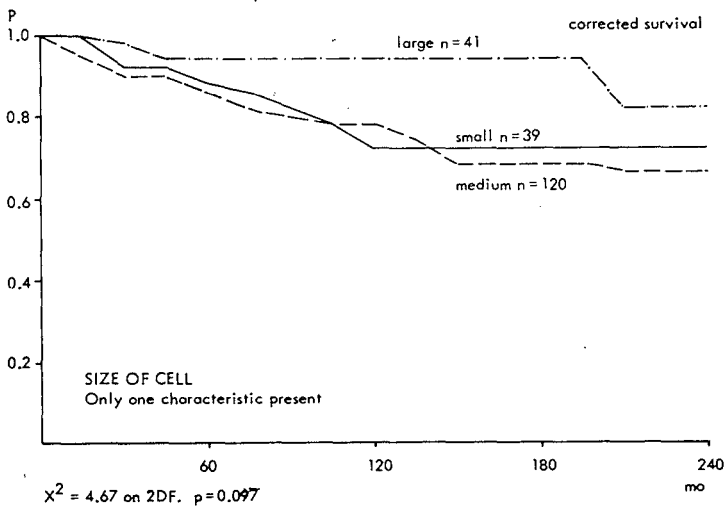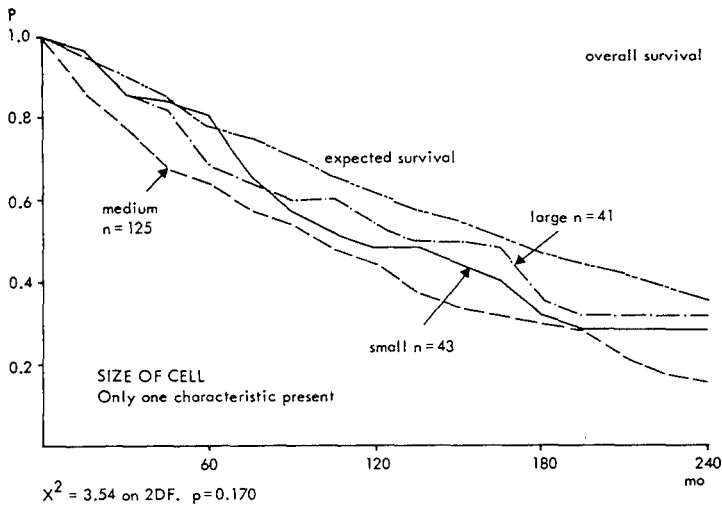
77

Fig. 9. Top: Overall survival by "size of cell" in tumors with single characteristics. No significant difference. Bottom: Corrected survival: same parameter and same patients; the group of 41 patients with large cells shows significantly fewer cancer deaths than the rest.

corrected survival curves are compared. These are the "amount of tumor" (p $\leq$ 0.003) and the "presence and absence of mitoses" (p $\leq$ 0.000). In addition, the characteristic "large" of the parameter "size of cell" is associated with significantly fewer cancer deaths than smaller cells.

Grade and Single Parameters

The parameters "glands" and "nuclear variation in size and shape" are the strongest indicators of prognosis found in this analysis. Can any one of these param-
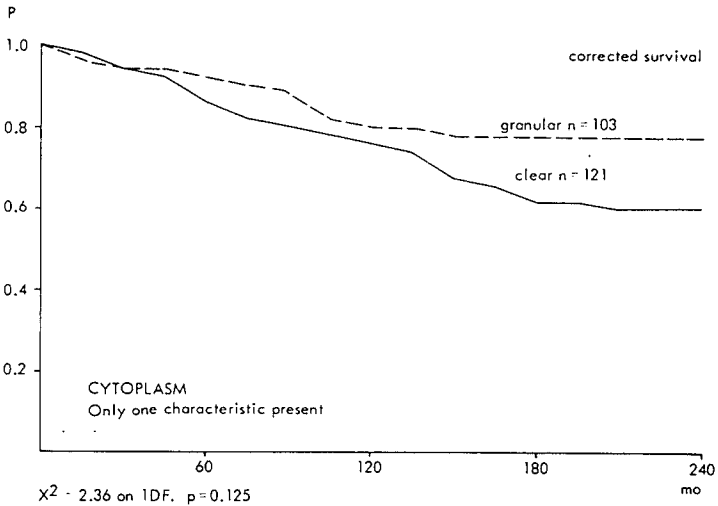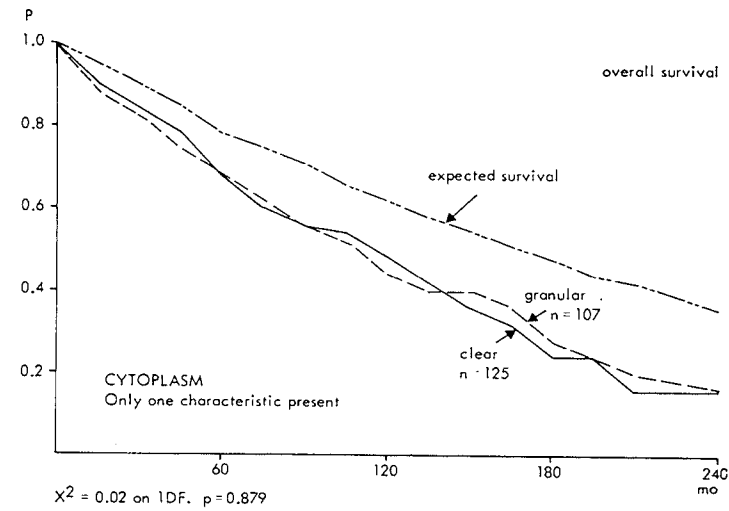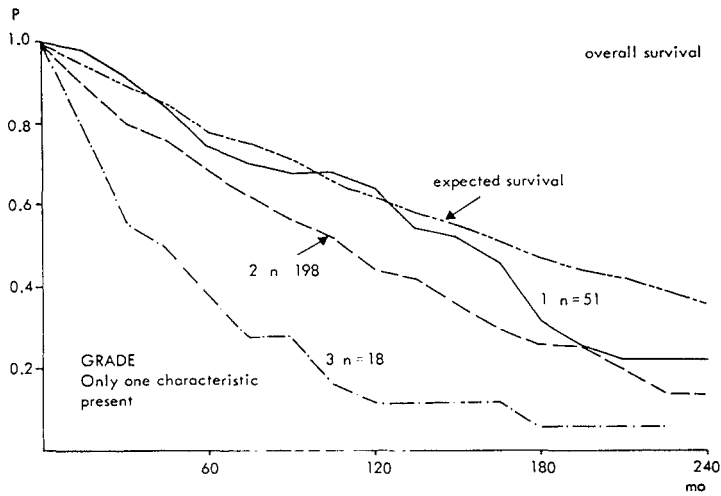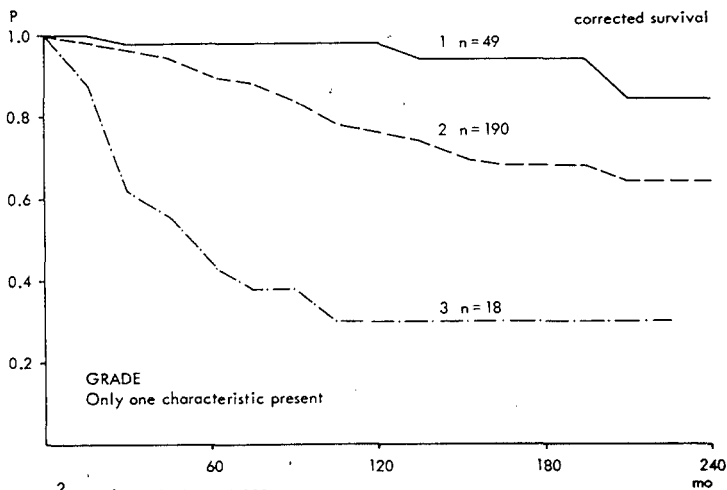
Fig. 10. Top: Overall survival by "cytoplasm", tumors with single characteristics. Bottom: Corrected survival: same patients, same parameter. No differences are found.

eters replace the pathologists' computation of "grade?" It seems obvious that the parameter "glands" alone will not be suitable because only two groups with a significant effect on survival could be identified: tumors with small, intermediate, or large glands have a better prognosis than tumors with cribriform or solid formations. No difference in prognosis of tumors with only small, intermediate, or large glands on one side and cribriform or solid formations on the other side could be found. In addition, when survival curves for "grade" were calculated and were adjusted for the characteristics of "glands," significant differences were found (Fig. 12). These data

Fig. 11. Top: Overall survival by grade; only tumors with single characteristics are considered. The overall difference is barely signficant. Bottom: Corrected survival: same patients, same parameter. Highly significant difference between the three groups.

show that 42 of the total of 49 pure grade 1 tumors showed only small, intermediate, or large glands. The seven remaining ones were cribriform but were still given grade 1. A considerable number of tumors with small, intermediate, or large glands (n = 63) were grade 2 and a small number grade 3 (n = 8). Prognoses in these groups were obviously determined by grade and not by glands. The difference between these curves is significant (p $\leq$ 0.0297). This evaluation is not superimposed by the presence of multiple formations; only one characteristic is considered in the top of

80

TABLE III. Significance of Differences in Survival According to 11 Architectural and Cytological Parameters in Patients With Single Characteristics

Parameter	Overall survival		Corrected survival	
	Significance	p-Value	Significance	p-Value
Glands	Yes	0.002	Yes	0.000
Amount of tumor	No	0.094	Yes	0.003**
Amount of stroma	No	0.786	No	0.414
Size of nucleus	No	0.155	No	0.232
Variation nucleus	Yes	0.017	Yes	0.000
Nuclear vacuoles	No	0.621	No	0.175
Nucleoli	No	0.550	No	0.583
Mitoses	No	0.096	Yes	0.000
Size of Cell	No	0.170	No	0.097*
Clear vs granular cytoplasm	No	0.879	No	0.125
Grade	Yes	0.006	Yes	0.000

*Large cells significantly better than the rest.
**Only small amount of tumor, better than the rest.

Figure 12A. Figure 12, middle, is adjusted for "cribriform and/or no glands present." It is evident that the presence of the prognostically worst formations does not automatically classify a tumor as "grade 3" in this system. The diagram shows that 49 of the total of 57 grade 3 tumors had no glands and/or cribriform formations, that the bulk of the grade 2 tumors were so graded in spite of the presence of cribriform and/or solid tumors (n = 165), and that a small number of cribriform and/or solid tumors were still and correctly assigned to grade 1. The overall differences are significant (p $\leq$ 0.0057). The p-value in comparing grade 1 vs grade 2 vs grade 3, adjusting for "glands," is overall 0.0007 (chi-square 14.42 on 2 DF). These data indicate that "grade" must also depend on parameters other than "glands."

When same analysis was carried out with adjustment for variation in size and shape of the nucleus (anaplasia), it became evident that 38 of the 41 grade 1 tumors and only 18 grade 2 tumors showed slight anaplasia of the nuclei. The difference was not significant (Fig. 13). Eleven grade 1 tumors were classified as "moderate variation" and survival was significantly better than the bulk of 195 of the 228 grade 2 tumors and seven grade 3 tumors in this group (Fig. 13). The differences in survival are significant (p $\leq$ 0.0002), indicating that the group "moderate variation" contains a number of tumors which were correctly assigned to another grade on the basis of other parameters than nuclear variation. The group "marked variation present" contains 50 of the 57 grade 3 tumors and 15 grade 2 tumors (Fig. 13). The difference between the two survival curves is not significant.

These figures indicate that slight and marked variation in size and shape is in good agreement with grade 1 and 3. However, especially in the group "moderate variation," a small but significant mistake would be made if one had only considered "variation" for grading. In some instances, other parameters contribute significantly to the correct determination of grading. Grade, the computation of several prognostic parameters, can in this system not be replaced by any single parameter.

The strong correlation between "anaplasia of the nucleus" and grade, and the fact that this parameter is determinable by morphometrical techniques, encourages such measurements. Some results of such attempts were reported recently [15,16].
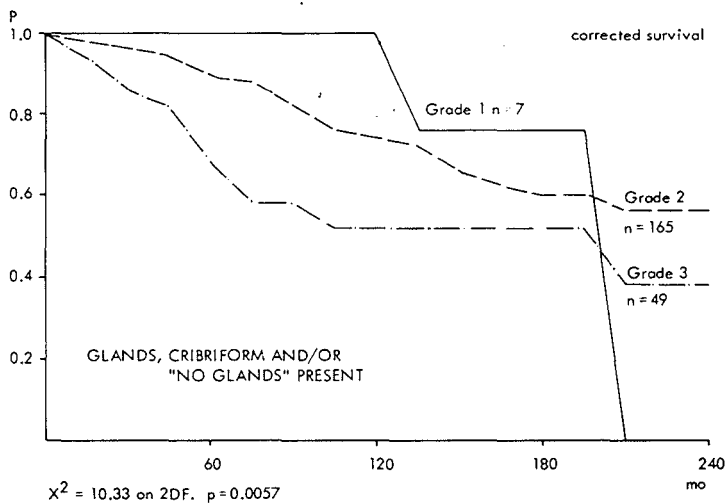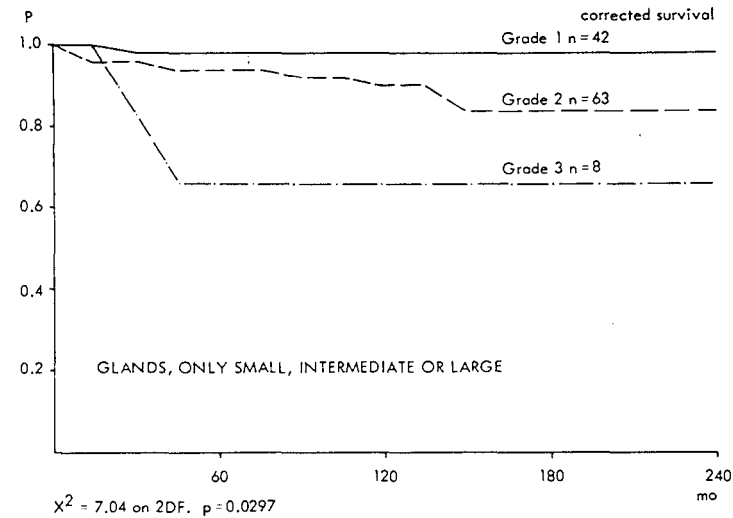
81

Fig. 12. Top: Corrected survival by grade in tumors with single parameters present. The curves are adjusted for the parameter "glands, only small, intermediate, or large." There is a significant difference. Bottom: Corrected survival by grade, only patients with single characteristics are considered. The curves are corrected for the parameter "glands, cribriform and/or no glands present." The overall differences are significant.

DISCUSSION

This study clearly identifies two parameters with significant influence on prognosis: the architectural pattern and the nuclear variation in size and shape of the nucleus (nuclear anaplasia). These parameters were evaluable in all slides; they are to a certain degree independent from each other and are the main determinants of

P

1.0 ─────────────────────────────────────── corrected survival
                                    Grade 1 n 38
0.8   Grade 2 n 18

0.6

0.4

0.2

NUCLEUS, ONLY SLIGHT VARIATION IN SIZE AND SHAPE

        60   120   180   240
                                        mo
x^2 0.75 on 1DF. p 0.3850

P

1.0   corrected survival
                                    Grade 1 n 11
0.8   Grade 2 n 195

0.6

0.4
                                    Grade 3 n 7
0.2

NUCLEUS, MODERATE VARIATION IN SIZE AND SHAPE PRESENT, BUT
        NOT MARKED VARIATION

        60   120   180   240
                                        mo
x^2 17.23 on 2DF. p 0.0002
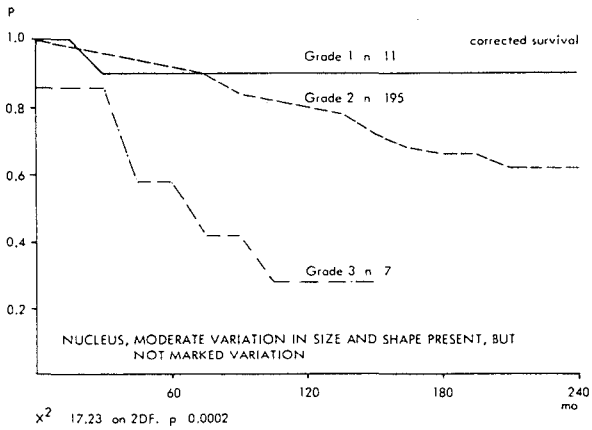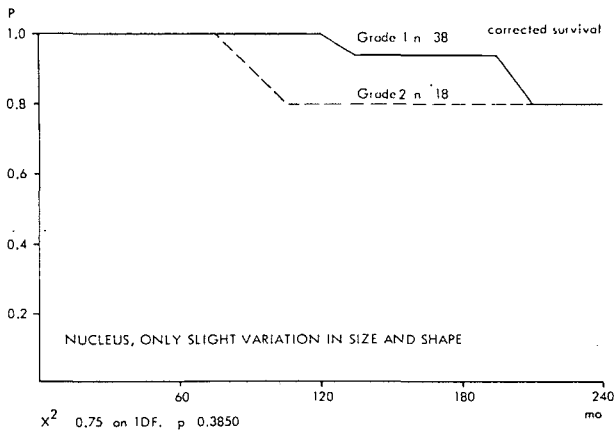
Adjusted value: P value in comparison of grades while adjusting for nuclear variation
        in size and shape (anaplasia) x^2 8.75 on 2DF. p 0.0126

P

1.0   corrected survival

0.8
                                    Grade 2 n 15
0.6   Grade 3 n 50

0.4

0.2

NUCLEUS, MARKED VARIATION IN SIZE AND SHAPE PRESENT

        60   120   180   240
                                        mo
x^2 0.79 on 1DF. p 0.3755

83
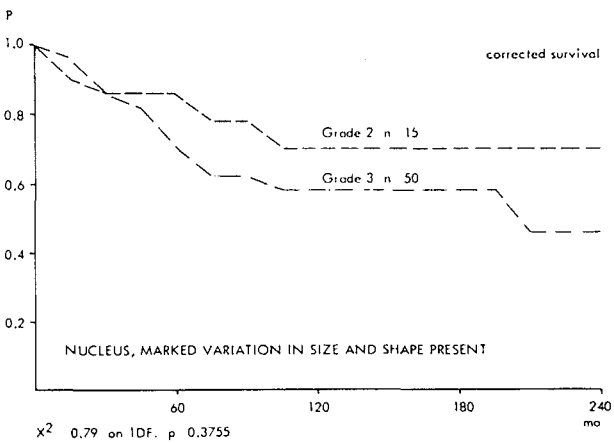
"grade." Attempts to determine grade from either one alone have failed in this study. In a substantial number of cases, the final grading was obviously a true computation of several parameters. It can not be excluded that visual impressions of the pathologist, which are not expressed in any one of the parameters used, have an influence on "grade."

Some other parameters may have adjuvant value because they are not present in all tumors, like mitoses and large cells, or because their prognostic weight did not seem to be as heavy as that of others, eg, amount of tumor and size of the nucleus. Still, if present, these parameters should be considered and allowed to exert influence on "grade." A more precise classification should result from the multivariate analysis presented in the third part of this study [8].

A surprising finding was that no difference in prognosis could be detected for patients with cribriform and solid tumors and that three different types of glandular manifestations (small, intermediate, and large glands) were not associated with a different survival. Only two prognostically significant groups could be detected when the parameter "glands" was analysed. This is at variance with the findings of Gleason [3].

The fact that glandular architecture and nuclear anaplasia exert their influence on survival independently casts doubt on the chances of ever obtaining a grading system which is based on measurable cellular parameters or on architecture only. Still, advancement of image-analyzing techniques may also enable us in the future to analyse automatically the architectural patterns of prostatic cancer.

Clincially, it is of great interest to identify groups with a minimal and maximal risk for progression. The most impressive correlation with a good prognosis was found for the characteristic "small amount of tumor." Only two of 54 patients with this finding died of prostatic carcinoma. In this context, however, one has to remember that the patients were all treated by radical prostatectomy, and it remains uncertain as to whether the good prognosis for these patients reflects the natural history or an effect of treatment. However, data recently published by Cantrell [17] on patients not treated by radical surgery confirm this observation. The group of T_0 tumors has been subjected to a separate analysis and report [18].

Grade 3 tumor as a single formation clearly is associated with the poorest prognosis in this series. But even in combination with better differentiated tumor, 5-year survival is only just above 50% and is almost identical with corrected survival. This means that most of the patients do indeed die of prostatic carcinoma.

A problem that seems to be inherent to all grading systems is that the "group in the middle," the grade 2 cases, is disproportionately large. Unfortunately, the parameters applied here do not seem to allow further differentiation of this group.

The well-known phenomenon that prostatic cancer can kill even after very long periods of time is well documented by the data presented.

Fig. 13. Top: Corrected survival by grade in tumors with single parameters, the curves are adjusted for "nucleus, only slight variation in size and shape." Middle: Corrected survival by grade in tumors with single characteristics, adjustment for "nucleus, moderate variation in size and shape present, but not marked variation." There is a significant overall difference. Bottom: Corrected survival by grade in tumors with single characteristics; the curves are adjusted for "nucleus, marked variation in size and shape present." There is no significant difference.

REFERENCES

1. Broders AC: Carcinoma: Grading and practical application. Arch Pathol 2:376–381, 1926.
2. Esposti PL: Cytologic malignancy grading of prostatic carcinoma by transrectal aspiration biopsy. Scand J Urol Nephrol 5:199–209, 1971.
3. Gleason DF, Mellinger GT, the Veterans Administration Cooperative Urological Research Group: Prediction of prognosis for prostatic adenocarcinoma by combined histological grading and clinical staging. J Urol 111:58–64, 1974.
4. Böcking A, Sinagowitz E: Histologic grading of prostatic carcinoma. Path Res Pract 168:115–125, 1980.
5. Gaëta JF: Glandular profiles and cellular patterns in prostatic cancer grading. Urol Suppl 3:33–37, 1981.
6. Mostofi FK: Grading of prostatic carcinoma. Cancer Chemother Rep 59:111–117, 1975.
7. Schroeder FH, Blom JHM, Hop WCJ, Mostofi FK: Grading of prostatic cancer (II)—The prognostic significance of the presence of multiple architectural patterns. In press, The Prostate.
8. Schroeder FH, Hop WCJ, Blom JHM, Mostofi FK: Grading of prostatic cancer (III)—Multivariate analysis of prognostic parameters. In press, The Prostate.
9. Schroeder FH, Blom JHM: Prostatic cancer—An analysis of 484 patients treated by total perineal prostatectomy. Unpublished data, 1983.
10. Belt E: Radical perineal prostatectomy in early carcinoma of the prostate. J Urol 48:287–297, 1942.
11. Turner RD, Belt E: A study of 229 consecutive cases of total perineal prostatectomy for cancer of the prostate. J Urol 77:62–77, 1957.
12. Belt E, Schroeder FH: Total perineal prostatectomy for carcinoma of the prostate. J Urol 107:91–96, 1972.
13. Schroeder FH, Belt E: Carcinoma of the prostate: A study of 213 patients with stage C tumors treated by total perineal prostatectomy. J Urol 114:257–260, 1975.
14. Kaplan EL, Meier P: Non-parametric estimations from incomplete observations. J Am Stat Assoc 53:457–458, 1958.
15. Diamond DA, Berry SJ, Umbricht C, Jewett HJ, Coffey DS: Computerized image analysis of nuclear shape as a prognostic factor for prostatic cancer. The Prostate 3:321–332, 1982.
16. Blom JHM, ten Kate FJW, Mostofi FK, Schroeder FH: Grading of prostatic carcinoma—evaluation of single parameters and cystomorphometry. In: Pavone Macaluso M, Smith PH (eds): "Cancer of the Prostate and Kidney," New York: Plenum Press, pp 109–116, 1983.
17. Cantrell BB, de Klerk BP, Eggleston JC, Boitnott JK, Walsh PC: Pathologic factors that influence prognosis in stage A prostatic cancer: The influence of extent versus grade. J Urol 125:516–520, 1981.
18. Schroeder FH, Blom JHM, Hop WCJ, Mostofi FK: Incidental carcinoma of the prostate treated by total prostatectomy. World J Urol 1:15–23, 1983.

Chapter 5

Grading of prostatic cancer (II) The prognostic significance of the presence of multiple architectural patterns

F.H. Schröder, J.H.M. Blom, W.C.J. Hop, and F.K. Mostofi

Grading of Prostatic Cancer: II. The Prognostic Significance of the Presence of Multiple Architectural Patterns

Fritz H. Schroeder, Jan H.M. Blom, Wim C.J. Hop, and F.K. Mostofi

Departments of Urology (F.H.S., J.H.M.B.) and Biostatistics (W.C.J.H.), Erasmus University, Rotterdam, The Netherlands, and Armed Forces Institute of Pathology, Washington, DC (F.K.M.)

This second report in a series of three deals with the prognostic importance of the presence of multiple, histologically identifiable architectural patterns in prostatic carcinomas. In the previous paper three of 12 parameters studied were identified as being prognostically significant in patients with single architectural patterns (formations) present in their tumors. The three parameters are nuclear anaplasia, architecture ("glands"), and mitoses, if present. The questions of whether "the worst part of a tumor determines prognosis" or "the presence of differentiated formations improves prognosis" are investigated by applying these parameters to patients with multiple tumor formations. Overall and corrected survival served as parameters.

It was shown that the parameters shown to be of importance for prognosis in tumors with single formations also have significant influence in patients with multiple formations. The worst formation determines prognosis, but patients with poorly differentiated tumors do significantly worse if their tumor is homogeneous. The presence of better-differentiated formations improves the prognosis of the worst formation. The observations made are discussed in view of the histopathogenesis of prostatic cancer.

Key words: prostate cancer, grading, role pleomorphism, worst formation, homogeneous tumors

INTRODUCTION

The majority of prostatic carcinomas consist of more than one morphologically different tumor formation. This is very obvious if, for example, solid tumor is present next to small glands or a cribriform formation. It is, however, also possible that the only difference between two formations consisting of large glands is the degree of nuclear anaplasia.

In the slides from 346 patients of E. Belts treated by radical prostatectomy, 668 different tumor formations were found and separately graded according to Mostofi [1]. One hundred thirteen patients showed only one, 152 showed two, 78 showed three, and eight showed four different architectural formations within their tumor. The goal of the present paper is to further investigate the prognostic significance of several characteristics of the same prognostic parameter within the same tumor. Theoretically there are several possibilities, such as 1) the less differentiated part of a given tumor determines the prognosis; 2) the most extensive part of a tumor determines the prognosis; 3) the prognosis of nonhomogeneous tumors is intermediate.

88

The answer to these questions is essential. If the first hypothesis is true, grading could be limited to the most malignant part of the tumor, and, in the case of the second hypothesis, to the most predominant part of the tumor. In the case of the third hypothesis, however, a system would have to be worked out which takes account of all formations present in each tumor.

In the first paper of this series [2], 113 patients with tumors consisting of only one formation were studied. Overall survival and intercurrent death-corrected survival were correlated to a total of 12 parameters and a large number of characteristics commonly used for grading. Of these parameters, only tumor architecture (glands) and nuclear anaplasia (variation in size and shape) were shown to significantly influence overall and corrected survival. Three others (a small amount of tumor, the presence of large cells, and the presence of mitoses) were of adjuvant value. Only the parameters identified as being prognostically important in tumors with single formations are used in the present analysis of the prognostic impact of multiple formations (pleomorphism of prostatic cancer).

MATERIALS AND METHODS

The classification of grading parameters and characteristics, patient material, and statistical methods are identical to those previously described [2]. The information obtained in the evaluation of tumors with single characteristics was applied to tumors with multiple characteristics of the same parameter. For example, nuclear anaplasia (variation in size and shape of the nucleus) was classified as slight, moderate, or marked. In patients with only slight, only moderate, or only marked nuclear variation, it was found that these parameters had a significant impact on survival in that survival was poorer with increasing anaplasia. Patients with slight and moderate anaplasia were classified as "moderate"; patients with slight, moderate, and marked anaplasia and patients with slight and marked or moderate and marked anaplasia were classified as "marked." Survival in the "moderate" and "marked" groups was compared to the group "slight variation only." The hypothesis that the worst formation determines prognosis was examined by 1) comparing the resulting survival curves to those found for patients with single characteristics, and 2) studying the impact of multiple formations on the presence of the worst formation.

RESULTS
Prognosis

Does the worst part of a tumor determine prognosis? The first analysis is based on the assumption that the worst part of a given tumor will determine the prognosis of the patient irrespective of the presence of other more benign formations. All patients with single or multiple characteristics are included. For analysis of patients with multiple criteria of the same parameter present, it was assumed that the sequence of their prognostic weight could be identical to that found in tumors with single characteristics.

For the parameter "glands" (pleomorphism), tumors with solid or cribriform formations should then be associated with poorer survival than tumors with small, intermediate, and/or large glands but with cribriform and solid tumor absent. That this is indeed the case is shown in Figure 1A and B. Patients with "glands in glands"
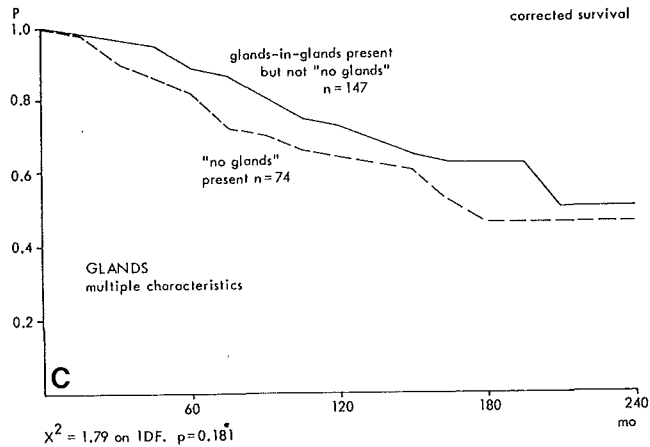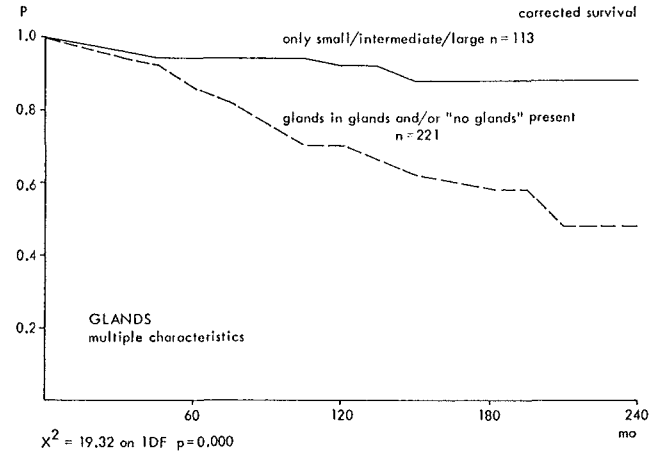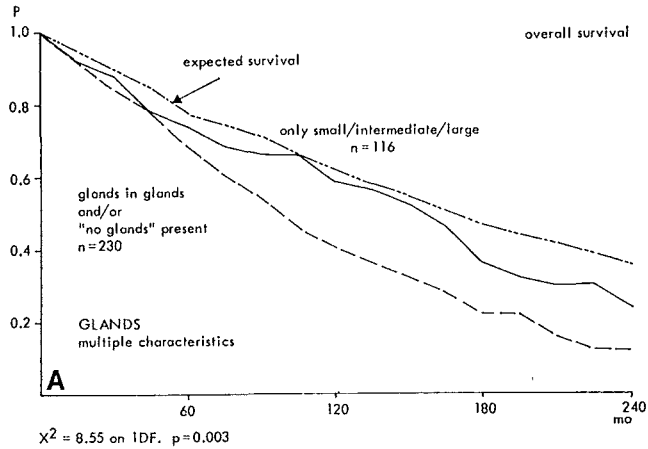
Fig. 1. A) Overall survival according to glandular differentiation. Multiple characteristics are considered. Two prognostically different groups can be isolated. B) Corrected survival of the same group of patients. C) Corrected survival curves for glandular differentiation; multiple characteristics are present. There is no significant difference between cribriform and solid tumor.

and/or "no glands present" (these may be combined with small, intermediate, or large glands or any combination of those formations) fare significantly worse than patients with only small and/or intermediate and/or large glands. The difference of the overall survival curves is significant (P = 0.003), as is corrected survival (P = 0.000). It was not possible, however, to identify more subgroups with impact on survival. In Figure 1C the negative analysis for "no glands present" and "glands in glands present" but not "no glands" is shown. The difference seen is not significant (P = 0.181).

The parameter "amount of tumor" has produced significant differences only for the corrected survival curves. The analysis of patients with the single and multiple characteristics small, medium, and much tumor present show the same result (Fig. 2A,B). The difference between the overall survival curves is not significant (P = 0.075), and the differences in corrected survival are highly significant (P = 0.0005).

"Nuclear size" (small, intermediate, or large) did not show significant differences in the analysis of single characteristics. The overall survival in the present analysis of "small only," "intermediate present but not large," and "large present" does not differ significantly (P = 0.231, Fig. 3A). The differences between the corrected survival curves shown in Figure 3B are, however, significant (P = 0.008).

"Nuclear variation in size and shape" (anaplasia), a parameter with significant impact on survival and corrected survival in the analysis of tumors with single formations, was analyzed for tumors with single or multiple formations (Fig. 4A,B). The overall survival curves follow the expected sequence, but differences are not significant (P = 0.286). Corrected survival differs significantly (P = 0.0003). The same findings were obtained with the analysis of "mitoses present" and "no mitoses"—ie, P = 0.621 for overall survival and P = 0.0003 for corrected survival (Fig. 5A,B).

"Grade" as a single or multiple characteristic is analyzed in Figure 6A and B. In contrast to the single-characteristics analysis, overall survival is not influenced significantly by the characteristics "grade I only," "grade II present," and "grade III present" (P = 0.066). Corrected survival according to the same groups differs significantly (P = 0.000).

The analysis of the remaining parameters according to this technique showed no significant differences. Also, the parameter "pleomorphism"—ie, the presence of one, two, three, or four formations—had no significant bearing on survival.

The results obtained in this analysis of all patients, irrespective of single or multiple formations, are summarized in Table I. Overall survival data differ significantly only within the characteristics of the parameter glands (pleomorphism). Only the parameter glands, amount of tumor, size of the nucleus, variation of the nucleus in size and shape, mitoses, and grade produce significant differences in corrected survival curves. The parameters amount of stroma, nuclear vacuoles, nucleoli, size of cells, and clear versus granular cytoplasm do not produce significant differences in survival.

From the data presented it is not clear whether the most malignant part of a tumor determines prognosis independent of its size. "Amount of tumor," especially the group with the criterion "small amount of tumor," contributes to the significant difference found in the corrected survival curves. The criterion "small amount of tumor" was found in 177 of the 668 formations recorded and in 159 patients. This
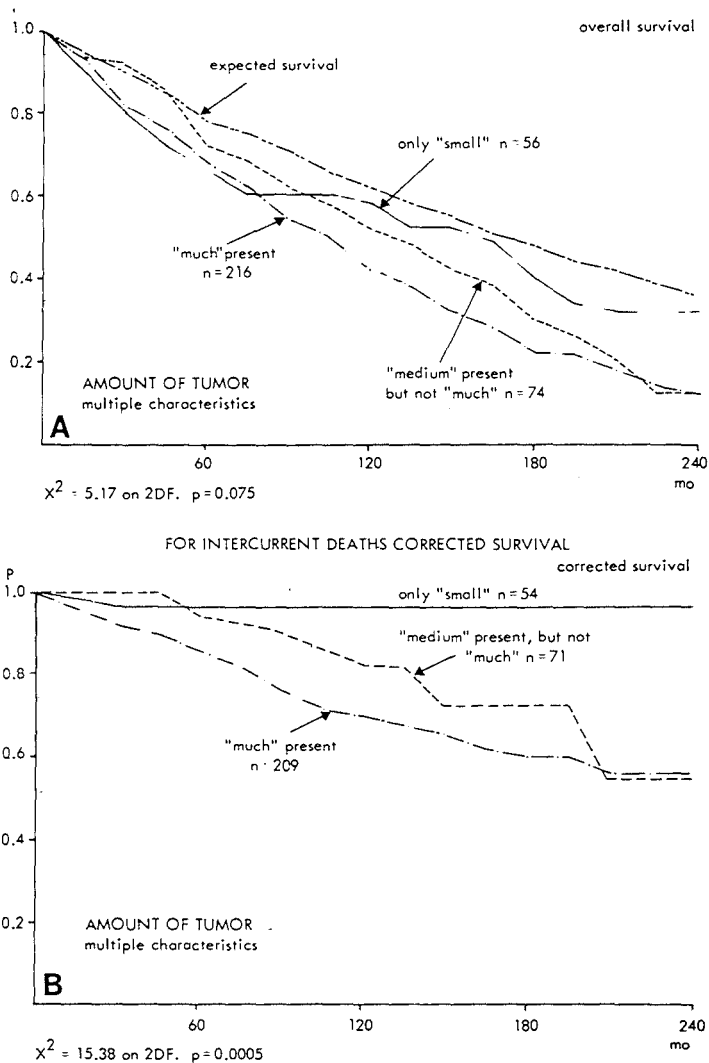
Fig. 2. A) Overall survival according to amount of tumor; multiple characteristics are considered. No significant difference can be shown. B) Corrected survival by the same parameter. The overall difference is significant. Very few patients with a small amount of tumor present die of prostatic carcinoma.

criterion occurred alone in 76 patients, and in combination with other characteristics of the same parameter in 83 patients. In the 177 small formations, G^1 tumor was seen 61 times (34%), G^2 tumor 107 times (61%), and G^3 tumor nine times (5%). This indicates that, even in histological slides that do not consist of cross sections of the total specimen, a small amount of poorly differentiated tumor rarely occurs.

Combinations of Formations

Does the combination of a poorly differentiated formation with formations of better differentiation within the same specimen change the impact on survival of

92
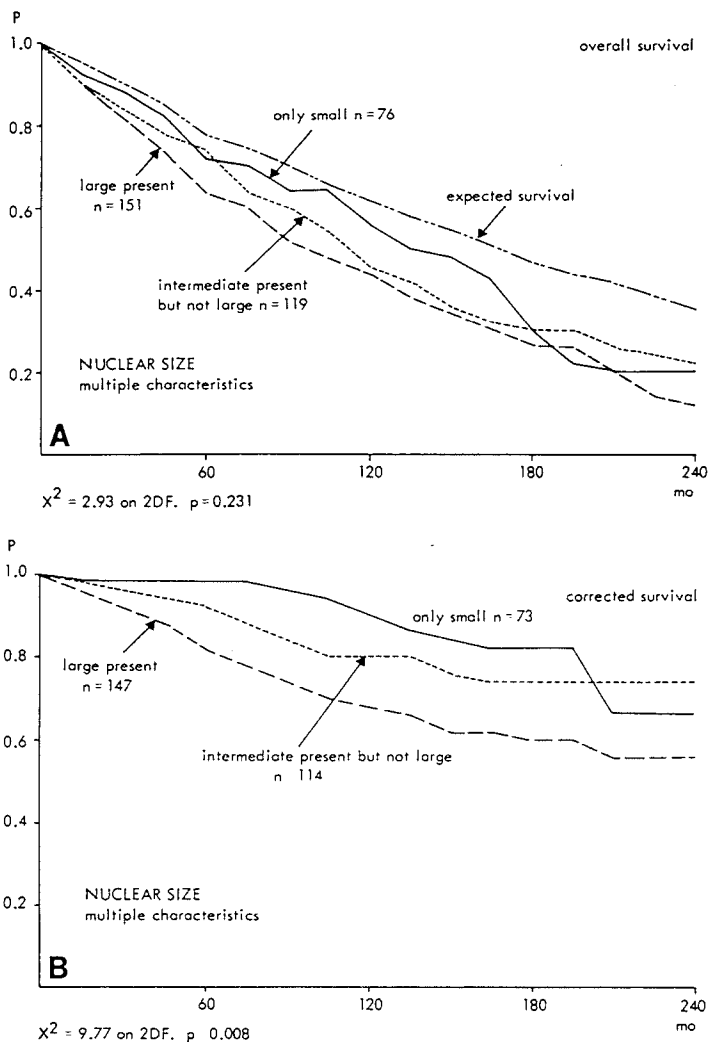
Fig. 3. A) Overall survival by nuclear size; multiple characteristics are considered. There is no significant difference. B) Corrected survival according to the same parameter. The overall difference is significant.

poorly differentiated tumor? Cribriform ("glands in glands") and solid tumors with no further formations present were combined to one group (n = 36) in order to answer this question for the tumor architecture ("glands"). This seems to be justified because no difference in corrected survival could be detected between these two characteristics when single characteristics were studied. This group was then compared to all patients with combinations of cribriform and/or solid characteristics and "small glands" and/or "intermediate" and/or "large glands" and with tumors showing absence of cribriform and/or solid characteristics (Fig. 7). The overall difference is significant (P $\leq$ 0.0000). Also, the difference between "cribriform and/or solid
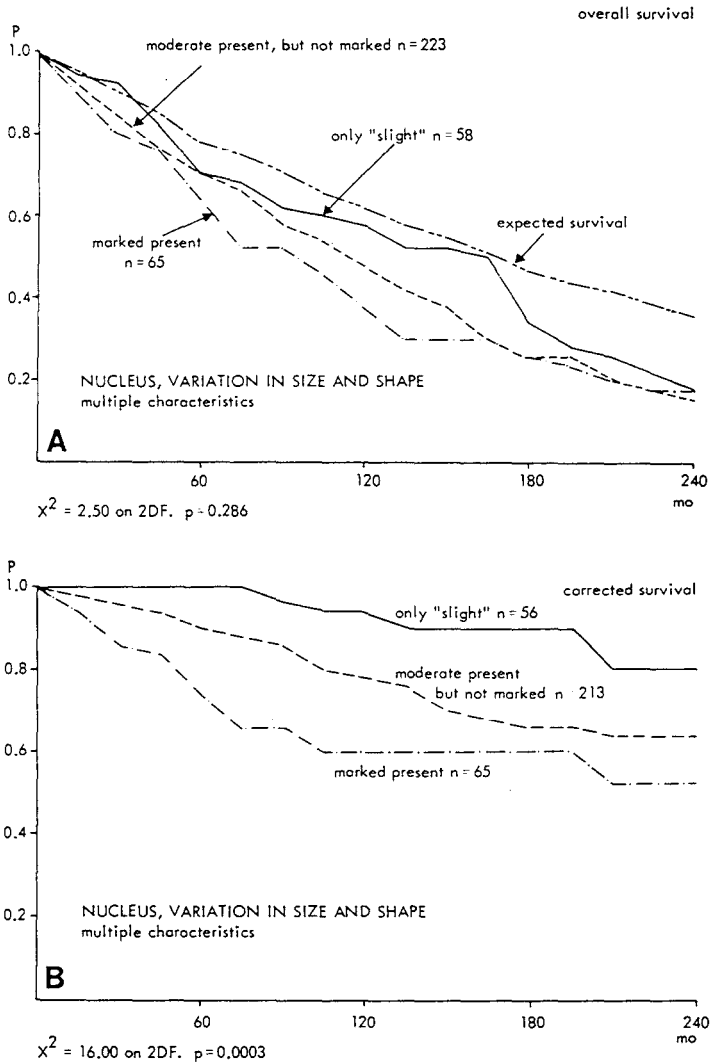
Fig. 4. A) Overall survival by nuclear variation in size and shape (anaplasia); multiple characteristics are considered. There is no significant difference. B) Corrected survival, same parameters. The differences are highly significant.

only" and "cribriform and/or solid" in combination with more differentiated formations is significant (P $\leq$ 0.0036).

The same phenomenon is observed when "nuclear variation in size and shape" is considered (Fig. 8). The overall difference is highly significant (P $\leq$ 0.0000). Also, the corrected survival curves obtained for "marked variation only" and for "moderate and marked" differ significantly (P = 0.027).

The data on mitoses show an identical result. The overall significance between the corrected survival curves for "no mitoses," "yes and no," and mitoses present ("yes") shown in Figure 9 is P = 0.0000.

94
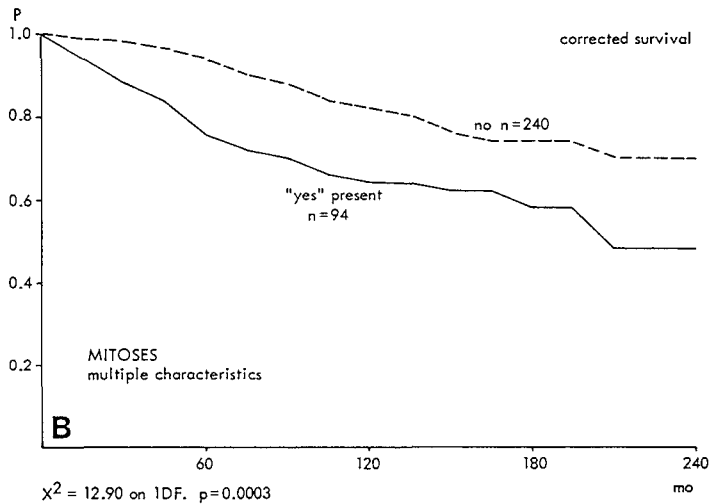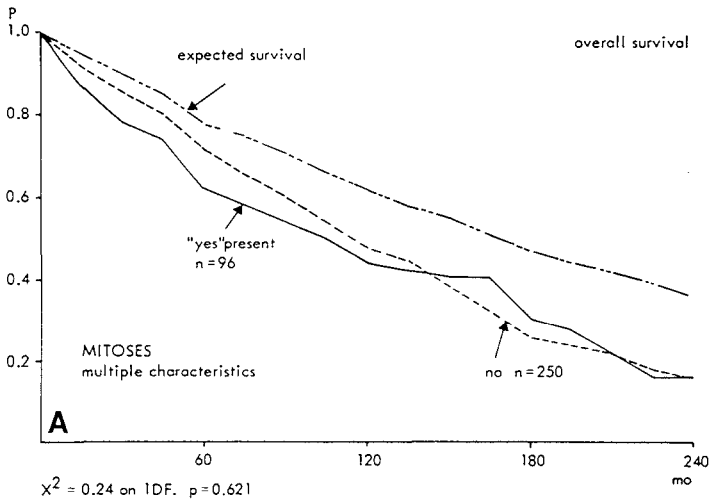
Fig. 5. A) Overall survival according to the presence or absence of mitoses; multiple characteristics are considered. There is no significant difference. B) Corrected survival, same parameters. The difference between presence and absence of mitoses is significant.

Patients with cribriform and/or solid formation only, with "marked variation in nuclear size and shape only," and with "mitoses present only" have the poorest prognosis identifiable in this study. Fifty to sixty percent of these patients die of their disease in spite of competing causes of death immanent to the age group concerned. If the same characteristics are combined with characteristics associated with a better prognosis, the poor prognosis associated with cribriform and/or solid tumors is significantly improved. The worst part of a tumor determines its prognosis, but in combination with differentiated tumor the prognosis of the worst part improves.

DISCUSSION

The presence of several formations within one tumor has a significant influence on the prognosis of the worst part. If grade III carcinoma is present as a single
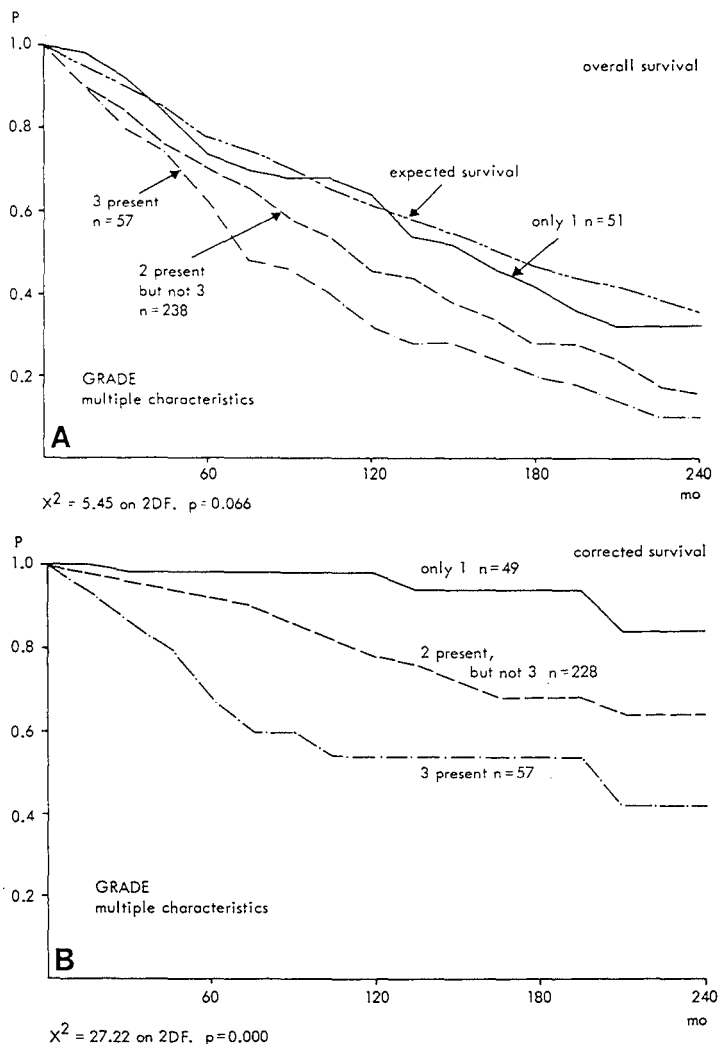
Fig. 6. A) Overall survival by grade; multiple characteristics are considered. The overall difference is not significant. B) Corrected survival by the same parameters. The difference between grades is highly significant.

formation, survival is very poor. In this series 70% of 18 patients with such a tumor died with evidence of disease and within 9 years. If grade III tumor is combined with other formations showing a better degree of differentiation, the prognosis is significantly better. But still, patients with multiple formations present do poorly if one of these is grade III.

These observations must in some way be connected to variations in the morphogenesis of different tumors. Figure 10 indicates several theoretical explanations. It is possible that prostatic carcinoma originates in general as a well-differentiated tumor (G_1) which, as time goes on, progresses to G_2 and finally to G_3 as indicated as hypothesis I in Figure 10. That most incidentally found small carcinomas are also well differentiated [3,4] supports this hypothesis as does the observation that large

TABLE I. Significance of Differences in Survival According to Six Parameters in Patients With Single or Multiple Characteristics

Parameter	Overall survival		Corrected survival	
	Significance	P	Significance	P
Glands	Yes	0.003	Yes	0.000
Amount of tumor	No	0.075	Yes	0.0005
Size of nucleus	No	0.231	Yes	0.008
Variation nucleus	No	0.286	Yes	0.0003
Mitoses	No	0.621	Yes	0.0003
Grade	No	0.066	Yes	0.000
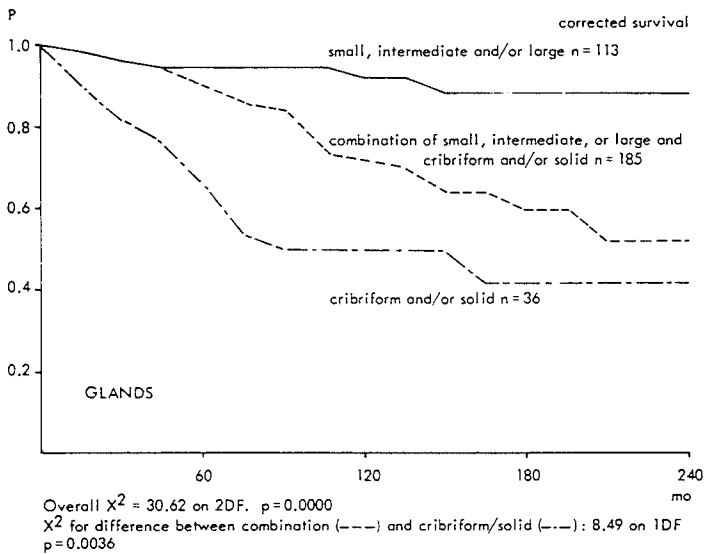
Fig. 7. Corrected survival according to glandular differentiation. The presence of small, intermediate, and/or large glands is compared to homogeneously cribriform and/or solid tumors and the same formations combined with small, intermediate, or large glands. The overall differences and the difference between nonhomogeneously and homogeneously cribriform and/or solid tumors are significant.

tumors are usually not well differentiated [5]. The increasing incidence of incidental carcinoma and focal tumors with age found at autopsy, which often are G_1, illustrates that most G_1 tumors do not progress to a higher grade [6,7]. The presence of several formations with different G categories within one tumor can also be explained by hypothesis I if one assumes that dedifferentiation occurs only in one or few cells of a G_1 or G_2 tumor and that the different formations seen are the expression of the coexistence of different clonal cell populations. The coexistence of several formations with a different grade could also be explained by the possible origin of multiple foci with different G categories during the early morphogenesis of any given tumor. This hypothesis is supported by the observations of multiple, independent foci in whole prostates examined by step sections [8]. One would then have to assume that G_1 stays

97

Fig. 8. Corrected survival by nuclear variation in size and shape (anaplasia). The differences between 1) all combinations with moderate and marked variation and 2) moderate and marked variation only are highly significant.

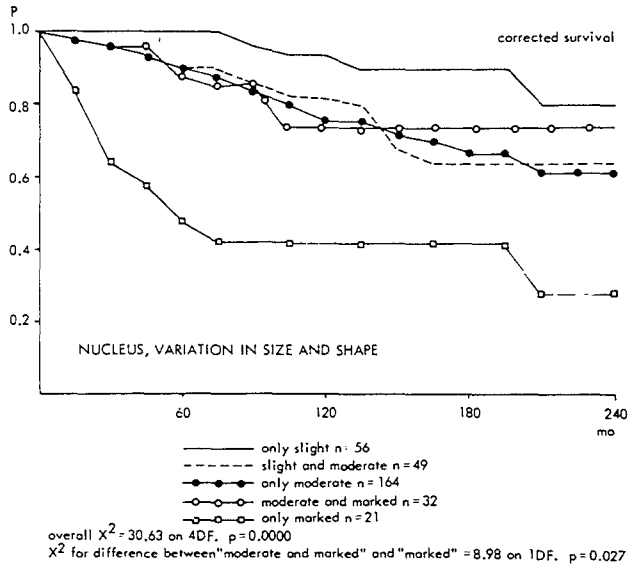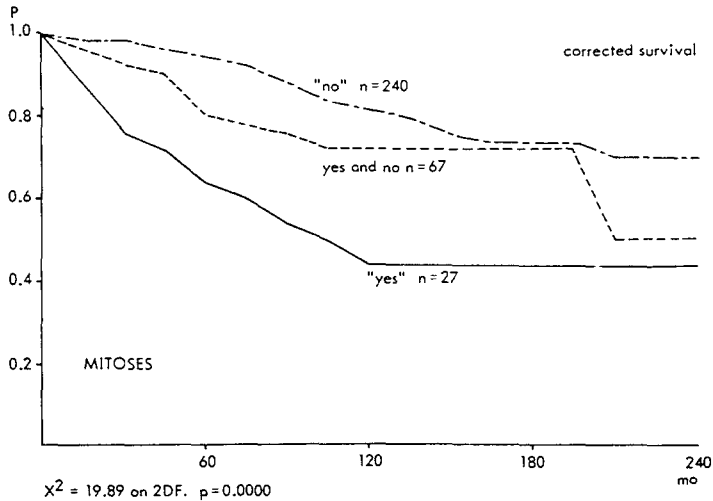Fig. 9. Corrected survival by mitoses. Again, tumors showing multiple formations, some of which do not contain mitoses, do significantly better than the homogeneous ones with mitoses.
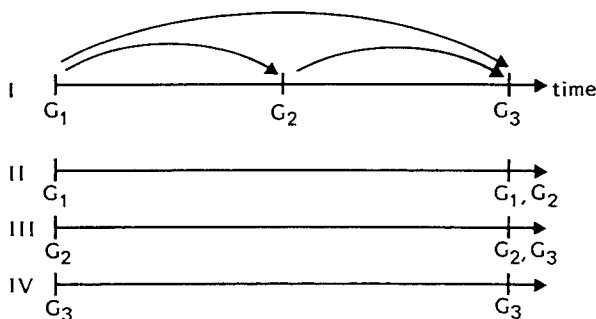
98

Fig. 10. Hypothesis on the histopathogenesis of prostatic cancer (see text).

G_1, G_2 stays G_2, and so on (hypotheses II–IV). The presence of only G_2 and only G_3 formations can be easily explained by this hypothesis. It is possible, however, that in these cases very small, well-differentiated tumor formations are missed at histological examination.

The phenomenon that G_3 tumor as a single formation has a worse prognosis than G_3 combined with G_1 or G_2 tumor could be due to a rapid transition from G_1 to G_3 according to hypothesis I. One could easily imagine in this case that the well-differentiated formations are in fact present but are very small and difficult to detect because of overgrowth of the rapidly growing G_3 part of the tumor. It is possible, however, that some tumors primarily originate as poorly differentiated (hypothesis IV) and that these have a worse prognosis than those following the morphogenesis indicated as hypothesis I in Figure 10.

For the practice of grading of prostatic cancer in general, several consequences can be drawn from these findings: 1) The most undifferentiated part of any tumor determines the prognosis of the patient and should be taken into account in any grading system. 2) In the presence of well-differentiated formations, patients with poorly differentiated tumor do better than patients with homogeneous G_3 tumors. The differences found are significant. The latter tumors, therefore, deserve to be classified as being more malignant (G_4?).

In interpreting these results, note that the population of patients studied is nonhomogeneous with respect to several important prognostic parameters (T category, N category, age) and that all patients have been treated. Still, the study of treatment escape (cancer death) sheds some light on the natural history of this tumor.

In a third paper of this series [9], all significant observations made in this study of patients with homogeneous and nonhomogeneous tumors are subjected to a multi-variate analysis. The results are used to design a system of prognostic factors applicable to the histological classification of prostatic cancer and to suggest a new grading system.

REFERENCES

1. Mostofi FK: Grading of prostatic carcinoma. Cancer Chemother Rep 59:111–117, 1975.
2. Schroeder FH, Blom JHM, Hop WCJ, Mostofi FK: Grading of prostatic cancer (I)—An analysis of the prognostic significance of single characteristics. The Prostate 6:81–100, 1985.

3. Cantrell BB, De Klerk DP, Eggleston JC, Boitnott, JK, Walsh PC: Pathologic factors that influence prognosis in stage A prostate cancer: The influence of extent versus grade. J Urol 125:516–520, 1981.
4. Schroeder FH, Blom JHM, Hop WCJ, Mostofi FK: Incidental carcinoma of the prostate treated by total prostatectomy: The prognostic impact of microscopic tumor extension and grade. World J Urol 1:15–23, 1983.
5. Schroeder FH: Prostatic carcinoma: Comments on radical surgical treatment. Scand J Urol Nephrol Suppl 55:181–185, 1980.
6. Battaglia S, Barbolino G, Botticelli AR: Early (stage A) prostatic cancer. Virchows Arch (Pathol Anat) 382:245–259, 1979.
7. Franks LM: Latent carcinoma of the prostate. J Pathol Bacteriol 68:603–616, 1954.
8. Byar DP, Mostofi FK, and the VACURG: Carcinoma of the prostate: Prognostic evaluation of certain pathologic features in 208 radical prostatectomies. Cancer 30:5–13, 1972.
9. Schroeder FH, Hop WCJ, Blom JHM, Mostofi FK: Grading of prostatic cancer (III)—Multivariate analysis of prognostic parameters. The Prostate (in press).

Grading of prostatic cancer (III) Multivariate analysis of prognostic parameters

F.H. Schröder, W.C.J. Hop, J.H.M. Blom, and F.K. Mostofi

Grading of Prostatic Cancer: III. Multivariate Analysis of Prognostic Parameters

Fritz H. Schroeder, Wim C.J. Hop, Jan H.M. Blom, and F.K. Mostofi

Department of Urology (F.H.S., J.H.M.B.), the Department of Biostatistics (W.C.J.H.), Erasmus University, Rotterdam, The Netherlands and the Armed Forces Institute of Pathology (F.K.M.), Washington, D.C.

This work is based on the two previous publications which are concerned with grading prostatic carcinoma. In the present communication the technique of multivariate analysis is applied to quantify the prognostic importance of parameters which were earlier identified as contributing in a significant manner to grading. The results of this analysis are used to construct a scoring system which differentiates five significantly different prognostic groups. When compared to the results of grading, the new system is shown to be superior. The large group of 228 patients with G2 tumors can be split into three groups, each with a significantly different prognosis.

The correlation of the scoring system to local tumor extension (pT category) is described.

Recommendations for grading of prostatic carcinoma resulting from this work are the following: 1) Only three independent parameters are of significance for grading prostatic cancer: tumor architecture, nuclear anaplasia, and the presence or absence of mitoses. 2) The combination of these parameters in a scoring system identifies five separate prognostically different groups. 3) The separation achieved by this new system is superior to conventional grading and is suggested for grading prostatic cancer.

Key words: prostatic cancer, grading, local tumor extension

INTRODUCTION

In the two preceding publications of this series [1,2] an attempt was made to evaluate quantitatively 12 parameters of Mostofi's grading system [3] by studying their impact on overall survival and intercurrent death-corrected survival. The survival data and histological slides on 346 patients of E. Belt treated by total perineal prostatectomy are the basis of these studies.

To allow a better understanding of the approach taken in the research presented in this paper, a short summary of the previous results follows. As a first step, tumors with single architectural formations and homogeneity with respect to the cytological criteria were studied to avoid the complicating problem of pleomorphism. Each of the 12 parameters was individually correlated to survival. It turned out that significant prognostic impact was present for the following parameters: glands (architecture),

nuclear variation in size and shape (anaplasia), grade, mitoses, amount of tumor, and cell size. The last three parameters were of limited value for various reasons.

An attempt to replace "grade" by any one of the prognostically important parameters was unsuccessful. This indicates that "grade" is a true computation by the pathologist of several independent prognostic factors.

Subsequently, the influence on prognosis of multiple formations or multiple criteria of the same parameter within one tumor was examined. To achieve this, the sequence of malignancy established for the criteria of homogeneous tumors was applied to all tumors, homogeneous or nonhomogeneous. This was done in a manner that would indicate whether the most undifferentiated part of the tumor determines its prognosis and whether the presence of several "better differentiated" formations would have an impact on the prognosis of the worst one. It was found that the worst formation determines the prognosis of the patient but that poorly differentiated tumor has a better prognosis if it is combined with well-differentiated formations.

These findings have several practical consequences for proper grading of prostatic carcinoma: (1) Only a small number of the commonly used parameters are truly significant for grading. (2) It seems to be essential to consider architectural and cytological parameters. (3) The presence of several criteria of the same parameter or of several architectural formations within the same tumor improves the prognosis of the most undifferentiated part (cribriform and/or solid tumor). It is therefore essential to consider pleomorphism in grading. (4) Homogeneous, poorly differentiated tumors do significantly worse than poorly differentiated tumors combined with better differentiated formations.

It seemed desirable to use this information to develop suggestions for a better grading system by applying multivariate analysis. This technique allows one to quantify the prognostic importance of each observation. A combination of the factors according to their relative prognostic importance should lead to a more accurate grading and to a system with high resolution, especially with respect to the large "group in the middle," which occurs with most grading techniques.

MATERIALS AND METHODS

The patients and the histological material used in this study were described extensively in the first publication of this series [1]. Survival curves were calculated for each of the morphological parameters using the technique of Kaplan and Meier [4]. The logrank-test was used to statistically compare the survival curves [5].

To assess the relative prognostic importance of the various parameters, use was made of multivariate survival methods [6] which made it possible to investigate the impact of various factors simultaneously on the cancer death rate, ie, the monthly probability of dying from prostatic carcinoma.

The outcome of this analysis is that the death rate of a group of patients with a certain characteristic of an investigated parameter is expressed relative to the death rate of a group of patients with another characteristic of the same parameter (relative death rate). A detailed account of this method was given in 1980 [7]. Using these multifactorial methods, it is possible to classify patients into groups with a similar prognosis. The prognostic class system thus established, which takes into account the most important prognostic parameters, can be compared with the prognostic results of the pathologists' grading of the tumor.

RESULTS

Number of Tumor Formations

No prognostic information could be obtained from the number of formations present in resected tumors. The intercurrent death-corrected survival percentages at 10 years for patients with one formation (n = 108), two (n = 147), three (n = 72), or four formations (n = 7) were, respectively, 82, 72, 78, and 80%. These numbers of patients are slightly different from those given previously [1,2] because patients whose cause of death is not known have been excluded. Also, at the other time intervals after operation, these percentages did not differ greatly. The survival curves of the four groups of patients largely coincided.

Exploratory Analysis

Some results of the previously reported [1] evaluation of patients with single criteria are given in Table I. Only the parameters shown previously to be significant are included. It should be noted that if a patient has a tumor which is homogeneous for a certain parameter, this tumor may show nonhomogeneities for another parameter. Therefore, the number of patients may vary between the different parameters in Table I. Based on the survival percentages indicated for each parameter, the criteria were classified with regard to their impact on survival. The characteristic associated with the best prognosis received the prognostic score number 1, the next best received

TABLE I. Intercurrent Death Corrected Survival Percentages at 5 and 10 years for Six Architectural and Cytological Parameters[†]

Parameter	Characteristics	No. of patients	Survival 5 years	Percentage 10 years	Prognostic score[a]
Glandular differentiation	Small glands	50	94	88	
	Intermediate glands	25	96	96	1
	Large glands	11	88	88	
	Cribriform	20	70	56	2
	No glands	3	—	—	
Amount of tumor	Small	54	96	96	1
	Medium	42	88	76	2
	Much	65	80	72	3
Nuclear size	Small	73	98	90	1
	Intermediate	63	92	78	2
	Large	69	78	76	3
Nuclear anaplasia	Slight	56	100	94	1
	Moderate	164	90	76	2
	Marked	21	48	42	3
Mitoses	Present	27	64	44	2
	Absent	240	94	82	1
Grade	Grade 1	49	98	98	1
	Grade 2	190	90	76	2
	Grade 3	18	44	30	3

[†]For each parameter only those patients who had tumors with only one characteristic present were selected.
[a]See text.

104

score 2, and, for four groups, the characteristic associated with the worst prognosis received a score of 3. The prognostic scores thus assigned are given in Table I.

For the parameter "glandular differentiation," no great differences were apparent between tumors with "only small," "only intermediate," or "only large" glands. These three groups did substantially better than patients with "only cribriform" tumors and were therefore assigned score 1. The group of patients with only "no glands" contained only three patients. These cases died of prostatic cancer and were combined with those with cribriform tumors. This combined group was assigned score 2.

Prognostic Classification

To analyze further the impact of the various parameters, including patients with multiple characteristics present, patients were grouped according to the highest prognostic score present in any tumor formation. As already stated, this strategy is based on the assumption that prognosis of patients is mainly determined by the worst characteristic present. Parameters which were not found to have prognostic importance in the previous analysis of single criteria were discarded. This led to the following grouping of patients:

Glands: only small and/or intermediate and/or large glands (n = 113) versus cribriform and/or no glands present (n = 221).

Amount of tumor: only small (n = 54) versus medium present, but not much (n = 71) versus much present (n = 209).

Nuclear size: only small (n = 73) versus intermediate present, but not large (n = 114) versus large present (n = 147).

Nuclear anaplasia: only slight (n = 56) versus moderate present, but not marked (n = 213) versus marked present (n = 65).

Mitoses: absent (n = 240) versus mitoses present (n = 94).

Grade: only grade 1 (n = 49) versus grade 2 present, but not grade 3 (n = 228) versus grade 3 present (n = 57).

These classifications led to statistically significant (p = 0.001) differences between survival curves for the following parameters: glands, amount of tumor, nuclear size, nuclear anaplasia, mitoses, and grade. For the remaining parameters no significant correlation with survival could be demonstrated [1].

Multifactorial Analysis

The classifications of the various prognostic parameters according to dominant category were all strongly correlated. For instance, in the group of patients with only slight variation in nuclear size and shape, 57% of patients were classified as only small nuclear size present, while 7% of patients had tumor formations with large nuclei. In contrast, the corresponding percentages in the group with marked nuclear anaplasia present were 3 and 82%, respectively. To assess the relative importance of the parameters "glandular differentiation," "amount of tumor," "nuclear size," "nuclear anaplasia," and "mitoses," use was made of multivariate methods, which revealed that only the parameters "nuclear anaplasia," "glands," and "mitoses" were independently related to survival. The relative cancer death rates associated with these parameters are given in Table II. The parameters "nuclear size" and "amount of tumor" did not add to prognosis once the other three are known. Their prognostic significance, when considered alone, could be explained by their correlations with

TABLE II. Relative Cancer Death Rates From the Multifactorial Analysis of the Parameters "Nuclear Anaplasia," "Glandular Differentiation," and "Mitoses"

Parameter	Category	Relative death rates	Statistical significance
Nuclear anaplasia	"Moderate" present, but not marked	2,4[a]	p = 0.04
	"Marked" present	3,5[a]	p = 0.001
Glandular differentiation	Cribriform and/or no glands present	2,8[b]	p = 0.001
Mitoses	Mitoses present	1,7[c]	p = 0.03

[a]Relative to only slight nuclear anaplasia present.
[b]Relative to only small/intermediate/large glands present.
[c]Relative to mitoses absent.

TABLE III. Combinations of Categories of Glandular Differentiation, Mitoses, and Nuclear Anaplasia[†]

Glandular differentiation	Only small/intermediate/ large glands		Cribriform and/or no glands present	
Mitoses	Absent	Present	Absent	Present
Nuclear anaplasia "Only slight" present	I (36)	II (3)	II (12)	III (10)
"Moderate" present, but not marked	II (56)	III (5)	III (102)	IV (45)
"Marked" present	III (4)	III (4)	IV (30)	V (27)

[†]Five prognostic classes, each composed of combinations leading to a similar prognosis, are denoted by Roman numerals (I–V). Numbers of patients in each combination are given in parentheses. Patient total per class: I, 36; II, 71; III, 125; IV, 75; V, 27.

the other three more important parameters. Based on the outcome of the multivariate analysis, five prognostic classes (I–V), each class composed of patients with roughly similar survival, could be determined. The combination of nuclear anaplasia, glandular differentiation, and mitoses led to a similar survival (Table III). The survival of patients according to this system of prognostic classes is given in Figure 1. In class I, no patient died of prostatic carcinoma. For the other classes, a generally decreased survival with increasing class number is evident.

Grade

Figure 2 gives survival of patients according to the highest grade present in any formation. With increasing grade, survival decreases markedly. To evaluate the respective roles of grade and prognostic class, the impact of the latter was investigated in the large group of patients with grade 2 tumors. Figure 3 illustrates survival according to prognostic class for this group of patients. Because of the small numbers of patients in the extreme classes, classes I and II were combined; the same was done with classes IV and V.

A generally decreased survival with increasing prognostic class number is apparent. No such prognostic value of grade within prognostic classes was evident, thereby suggesting that the three parameters "nuclear anaplasia," "glands," and "mitoses" has greater value than the parameter "grade" if used with the prognostic quantification applied in this system.

106
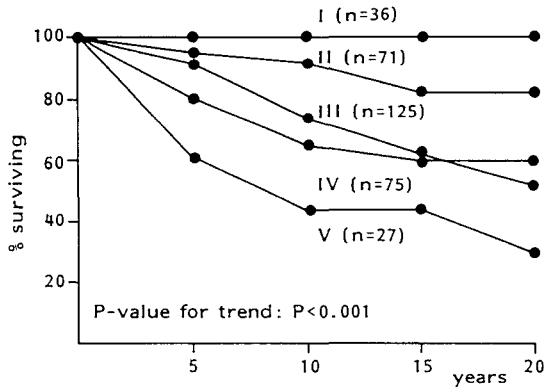
Fig. 1. Intercurrent death-corrected survival according to prognostic classes (I–V). See Table III for explanation of Roman numerals.
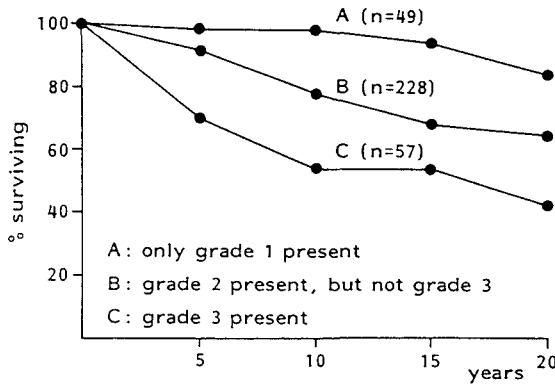
Fig. 2. Intercurrent death-corrected survival by highest grade present in any tumor formation.

Prognostic Classes and pT-Category

Figure 4 gives survival according to pT-category and the total group of patients (including those cases in whom histological material could not be reviewed). A strong correlation with survival is evident. In Table IV the impact of the system of prognostic classes is investigated within pT-categories. A generally decreased survival with increasing class number within pT-categories was apparent ($p = 0.001$). Also, within prognostic classes, a decreasing survival with increasing pT-category was evident ($p = 0.002$). As also evident from the number of patients given in Table 4, there is a strong correlation between the extent of the primary tumor and the prognostic class system. Patients with a low pT-category generally have a low class number, whereas patients with a high pT-category more frequently have a high class number.

DISCUSSION

It is evident that architecture ("glands"), nuclear anaplasia, and presence or absence of mitoses are the only independent parameters present in the system sug-

107
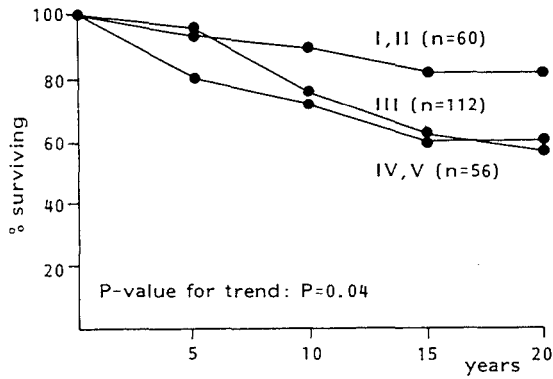
Fig. 3. Intercurrent death-corrected survival by prognostic class for patients with grade 2 tumors.
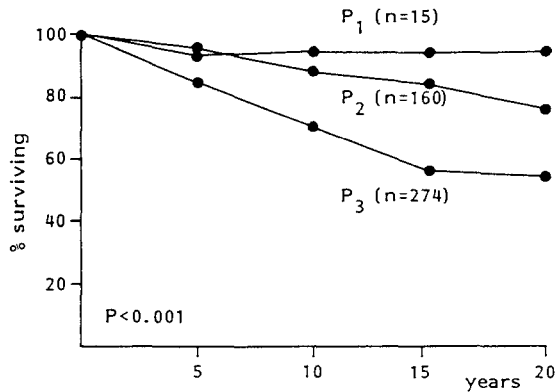
Fig. 4. Intercurrent death-corrected survival by pT-categories.

TABLE IV. Five- and Ten-Year Intercurrent Death-Corrected Survival Percentages According to P-Category and Prognostic Class[†]

		p-category		
		P 1	P 2	P 3
Prognostic class	I, II	100* - 100**	100 - 98	92 - 88
		(n = 10)	(n = 55)	(n = 40)
	III		96 - 84	90 - 70
		(n = 0)	(n = 38)	(n = 86)
	IV, V		86 - 86	72 - 58
		(n = 0)	(n = 16)	(n = 86)

missing P 3

[†]Percent corrected survival at 5 years: left (*); percent corrected survival at 10 years: right (**).

108

gested by Mostofi. Their use in clinical routine should simplify grading of prostatic carcinoma greatly and increase reproducibility. By applying the scoring system using these three parameters, five different prognostic groups of patients can be identified. The large "group in the middle," which is usually seen in other grading systems, disappears, and a more equal distribution of patients over the five prognostic groups is achieved.

It remains unclear why, when considering the architecture of the tumor, only two different prognostic groups can be identified in this system. It might be interesting to combine the parameters "nuclear anaplasia" and "presence or absence of mitoses" with the Gleason system of grading prostatic carcinoma [8].

For several reasons it seems essential to consider cytological parameters in grading: (1) "nuclear anaplasia" and the "presence or absence of mitoses" have been shown by multivariate analysis to be independent parameters from architecture. Thus, their evaluation adds information to this system. (2) Aspiration biopsy and cytology play an increasing role in clinical practice in several countries, especially in Europe. Evaluation of cytological parameters in histological slides may make the information produced by both techniques more comparable. (3) Nuclear anaplasia may in the future be routinely subjected to morphometric analysis [9,10].

The scoring system suggested for grading after this analysis is entirely based on the assumption that the worst part of a tumor determines prognosis. The presence or absence of better-differentiated characteristics, which was shown previously to have prognostic significance, was not further considered, since group V in Figure 1 is not statistically different from the group "cribriform and/or solid only." This group was shown in the second paper of this series [2] "to have a significantly worse prognosis than any group combining "cribriform and/or solid" with better-differentiated formations.

The scoring system presented in this paper is suggested for clinical use.

REFERENCES

1. Schroeder FH, Blom JHM, Hop WCJ, Mostofi FK: Grading of prostatic cancer (I)—An analysis of the prognostic significance of single characteristics. The Prostate 6:81–100, 1985.
2. Schroeder FH, Blom JHM, Hop WCJ, Mostofi FK: Grading of prostatic cancer (II)—The prognostic significance of the presence of multiple architectural patterns. The Prostate 6:403–415.
3. Mostofi FK: Grading of prostatic carcinoma. Cancer Chemother Rep 59:111–117, 1975.
4. Kaplan EL, Meier P: Non-parametric estimations from incomplete observations. J Am Stat Assoc 53:457–458, 1958.
5. Peto R, Pike MC, Armitage P, Breslow NE, Cox DR, Howard SV, Mantel N, McPherson K, Peto J, Smith PG: Design and analysis of randomized clinical trials requiring prolonged observation of each patient. Br J Cancer 34:585–612, 1976.
6. Cox DR: Regression models and life-tables. J Roy Statist Soc B34:187–220, 1972.
7. Hop WCJ, van der Werf-Messing BHP: Prognostic indexes for renal cell carcinoma. Eur J Cancer 16:833–840, 1980.
8. Gleason DF, Mellinger GT, and the Veterans Administration Cooperative Urological Research Group: Prediction of prognosis for prostatic adenocarcinoma by combined histological grading and clinical staging. J Urol 111:58–64, 1974.
9. Diamond DA, Berry SJ, Umbricht C, Jewett HJ, Coffey DS: Computerized image analysis of nuclear shape as a prognostic factor for prostatic cancer. The Prostate 3:321–332, 1982.
10. Blom JHM, ten Kate FJW, Mostofi FK, Schroeder FH: Grading of prostatic carcinoma—Evaluation of single parameters and cystomorphometry. In Macaluso M, Smith PH (eds): "Cancer of the Prostate and Kidney." New York: Plenum Press, pp 109–116, 1983.

Chapter 7

Grading of prostatic carcinoma - Evaluation of single parameters and cytomorphometry

J.H.M. Blom, F.J.W. ten Kate, F.K. Mostofi and F.H. Schröder

GRADING OF PROSTATIC CARCINOMA — EVALUATION OF SINGLE PARAMETERS AND CYTOMORPHOMETRY

J.H.M. Blom[1], F.J.W. ten Kate[2], F.K. Mostofi[3] and
  F.H. Schröder[1].

The Institutes of Urology (1) and of Pathology (2)
Erasmus University
Rotterdam, Netherlands and
The Armed Forces Institute of Pathology (3)
Washington
U.S.A.

INTRODUCTION

Since Broders' first tumor grading system in 1926 many attempts have been made to correlate various histologic features of a tumor with the prognosis of the patient. Also for prostatic carcinoma many grading systems have been developed (1), but only a few of them have found wide acceptance. One of the reasons for this is the tendency of many prostatic cancers to show varying degrees of differentiation and structure within a single section and often within a single microscopic field. Another reason may be the lack of reproducibility of most, if not all, grading systems. However, some grading systems seem to be very promising. Among these the most significant are those of Gleason (2) and of Mostofi (3).

Mostofi uses the term underlined differentiation exclusively for the tendency of the tumor to form glands, and the term anaplasia for the variation from normal in size, shape, staining and chromatin distribution of the nuclei in the tumor cells. His grading system is built up with architectural criteria, such as the various tumor formations (small, intermediate or large glands, cribriform tumor and solid tumor), the amount of stroma and the amount of tumor, and with cytological criteria such as size of cell, the aspect of the cytoplasm, nuclear size, nuclear pleomorphism, presence of mitoses, presence of nucleoli and the presence of nuclear vacuoles. With these criteria he estimates an overall tumor grade.

Using Gleason's system Harada and associates (4) found a good reproducibility on repeat readings of this system, although there was less correlation between their readings and Gleason's readings of the same slides. They also found, using Mostofi's system, that nuclear anaplasia and glandular differentiation correlated well with death rates.

MATERIAL AND METHODS

In a series of 484 patients on whom the late Dr Elmer Belt performed a radical perineal prostatectomy for cancer the patient charts were reviewed retrospectively and in 346 cases histological slides from the prostatectomy specimens were available for review. These tumors were all regraded by Mostofi without knowledge of the follow-up of the patients. Most of the tumors consisted of a varying number of morphologically different formations. As Table 1 shows their number varied from one to four per patient and a total of 668 tumor formations have been matched with the clinical data of 346 patients.

The survival curves were estimated according to Kaplan and Meier and have been corrected for intercurrent, tumor unrelated and unknown death causes. For the calculations of P-values in the comparison of the survival curves the Logrank test was used.

In order to objectively evaluate nuclear variation in size and shape a semi-automatic computerized image analysing system was used (Videoplan, Kontron). This consists of a graphic tablet, connected to a desk-computer, and a microscope with a drawing tube. The graphic tablet is used for digitizing contour coordinates of figures drawn on the tablet with a cursor. A light-emitting diode is mounted in the centre of the cursor, which is visible as a small, red spot together with the normal visual field of the microscope, via the

Table 1. The Number of Tumor Formations and
Their Relative Distribution in 346
Patients with Prostatic Carcinoma.

No. of Tumor Formations	No. of Patients	%	No. of Formations
1	113	32.6	113
2	152	44.0	304
3	73	21.1	219
4	8	2.3	32
Total	346	100	668

drawing tube. In this way contours of objects in the microscopic image can easily be traced manually under visual control. The digitized contours are fed into the computer, which calculates the preselected parameters. The area and perimeter was calculated in this case, together with two so-called form factors: a "form ellipse" which is derived from the longest diameter of a structure and the shortest diameter perpendicular to it in the following way:

$$\text{form}_{\text{ellipse}} = \frac{\text{shortest diameter}}{\text{longest diameter}}$$

As can easily be seen the largest value for "form ellipse" is one for a circle and less than one in the case of other structures. The second factor we estimated was: "form pe" which is derived from area and perimeter in the following way:

$$\text{form}_{\text{pe}} = \frac{4 \times \pi \times \text{area}}{(\text{perimeter})^2}$$

Here also the largest value for "form pe" is one in case of a circle and is less than one in all other structures. Reproducibility of contour tracing was within 5%. This is consistent with data from the literature (6,7).

RESULTS

Figure 1 shows the tumor related survival rates for the various tumor formations from tumors consisting of only one formation. The survival probability is plotted against the time in months. There is no difference in survival between patients with small glands, intermediate glands or large glands in their tumors, but these three differ significantly in survival from patients with cribriform or solid tumors. As Figure 2 shows, even the presence of cribriform or solid tumor together with small, intermediate or large glands makes prognosis significantly worse than in the case of small, intermediate or large glands alone. The amount of tumor (Figure 3) has prognostic importance insofar that patients with small amounts of tumor do better than those with medium or large amounts. However, one should not forget that all patients have had radical prostatectomy and that especially the patients with small amounts of tumor might have been cured by the operation.

The amount of stroma, the appearance of the cytoplasm, the presence of nucleoli, the size of cell and the presence of nuclear vacuoles, have all failed to identify groups of patients with different survival rates.

On the other hand nuclear pleomorphism, i.e. the variation in nuclear size and shape, identifies three groups of patients with

114

Prostatic carcinoma

p   Survival according to differentiation.
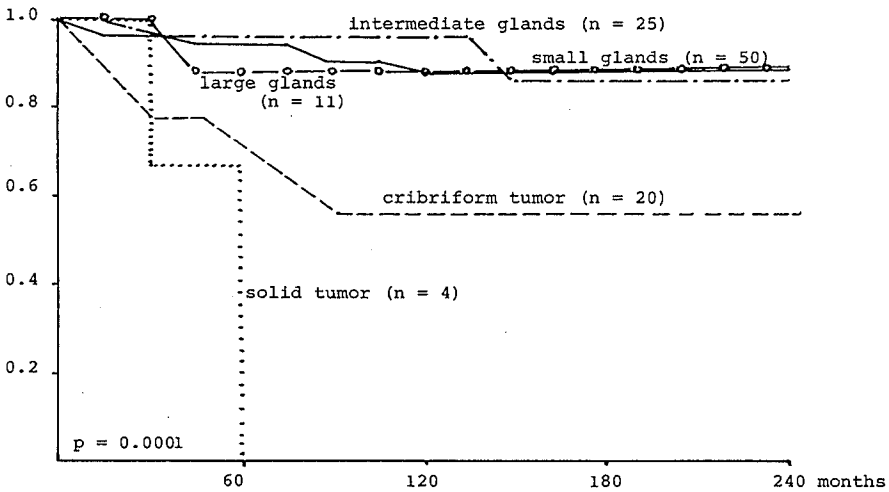
Fig. 1   Survival rates for 110 patients according to glandular
         differentiation.   In each tumor only one tumor formation
         is present.   (Mostofi's grading system).

Prostatic carcinoma

p   Survival according to differentiation.
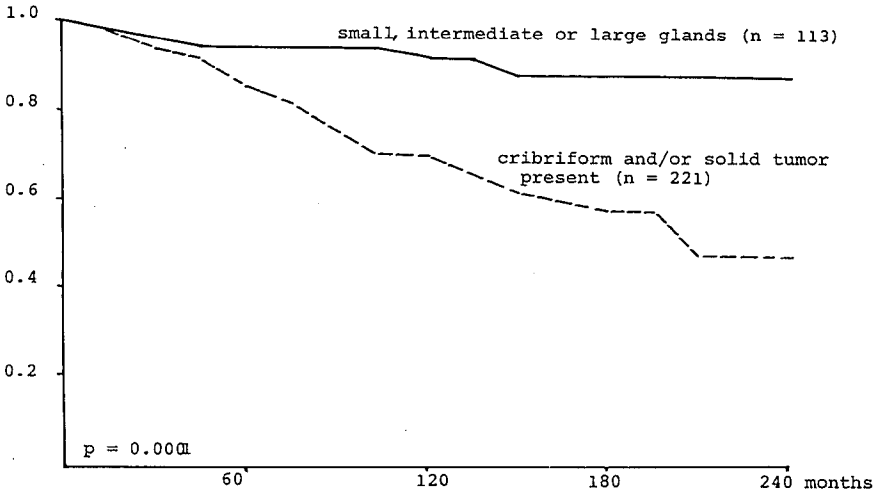
Fig. 2   Survival rates for 113 patients with only small, intermediate
         and/or large glands in their tumors vs. 221 patients who had
         glands and cribriform and/or solid parts in their tumors.
         (Mostofi's grading system).

115

Prostatic carcinoma
Survival according to amount of tumor.
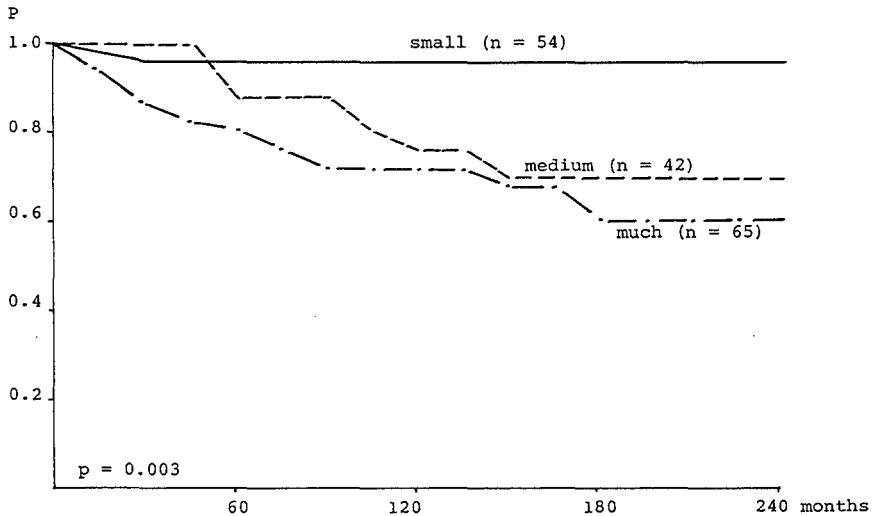
Fig. 3   Survival rates in 161 patients according to amount of tumor
         in their prostatectomy specimen (Mostofi's grading system).

Prostatic carcinoma
Survival according to nuclear pleomorphism.
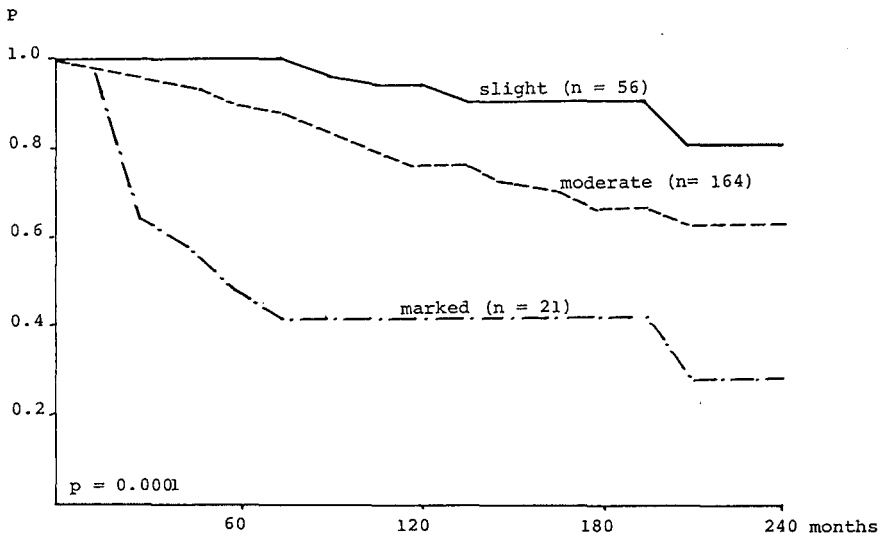
Fig. 4   Mostofi's grading system.   Survival in 241 patients
         according to nuclear pleomorphism.

116

significantly differing survival rates (Figure 4). Patients with
marked pleomorphism of the nuclei do considerably worse than those
with moderate nuclear pleomorphism, while the group of patients
with slight nuclear pleomorphism has a 20 years survival as good as
80%. Also the presence of mitoses identifies a group of patients
with a worse prognosis, although few patients have many mitoses in
their tumors.

As nuclear pleomorphism seems to have prognostic importance we
tried to evaluate this objectively in order to obtain a reproducible
and objectively estimated parameter.

With morphometry we measured 150 consecutive nuclei in each tumor
formation and for each parameter we calculated the variation-co-
efficient from the average value and the standard deviation. We
used the variation coefficient as a standard for variation. Figure
5 shows the morphometrically estimated variation in nuclear size
identifying two groups of patients with a significantly differing
survival pattern. The value for V area (V stands for variation-
coefficient) of 34% was found empirically.

The variation-co-efficient for "form ellipse" and for "form pe"
did not identify significantly in differing prognostic groups.

Prostatic carcinoma
Survival according to V area.
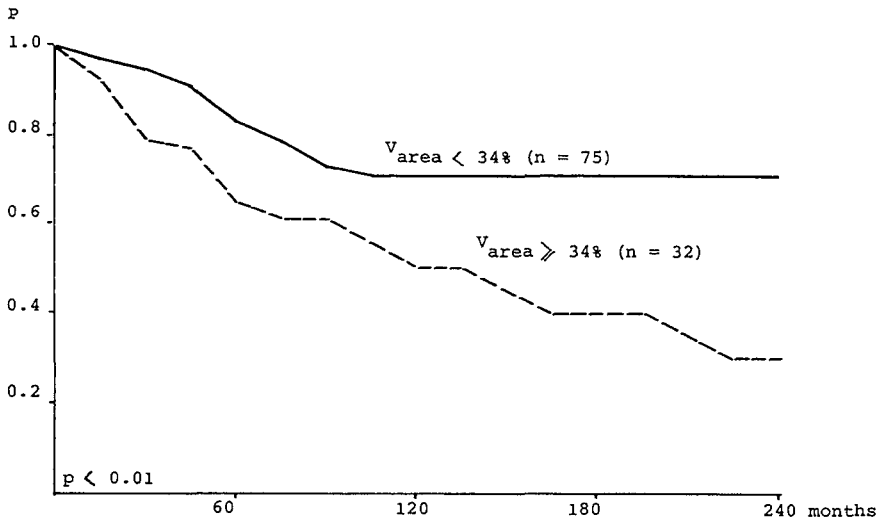
Fig. 5   Survival according to morphometrically estimated variation
          in nuclear size in 107 patients with prostate cancer.

117

DISCUSSION

As is shown, in Mostofi's grading system only a few parameters seem to have prognostic importance: glandular differentiation, whether the tumor forms only small, intermediate or large glands or whether it is growing in a cribriform or solid pattern. Secondly anaplasia, and especially the variation in nuclear size and shape has prognostic importance. The presence of mitoses may be of additional help, although most tumors show no mitoses. This suggests that the Mostofi grading system could be made simpler than it now is. With morphometry we were able to recognise two groups of patients with significantly differing survival patterns. It seems that only variation in size might be of importance, as variation in shape failed to show prognostic significance.

Although preliminary, this study shows that there may be a role for morphometry in grading prostatic carcinoma. It has the advantage of being objective and reproducible and is very easy to learn without special knowledge of grading. Maybe morphometry can cast some light on the complex problem of grading prostatic carcinoma. Until now we don't know the exact value of this technique in grading, but this will be a subject for further investigation.

ACKNOWLEDGMENTS

REFERENCES

1. F.K. Mostofi, Problems of Grading Carcinoma of the Prostate, Seminars in Oncology, 3:161 (1976).
2. G.T. Mellinger, D. Gleason and J. Bailar, The Histology and Prognosis of Prostatic Cancer, J. Urol. 97:331 (1967)
3. F.K. Mostofi, Grading of Prostatic Carcinoma, Cancer Chemother. Rep. 59:111 (1975).
4. M. Harada, F.K. Mostofi, D.K. Corle, D.P. Byar and B.F. Trump, Preliminary Studies of Histologic Prognosis in Cancer of the Prostate, Cancer Treat. Rep. 61:223 (1977).
5. R. Peto, M.C. Pike, P. Armitage, N.E. Breslow, D.R. Cox, S.V. Howard, N. Mantel, K. McPherson, J. Peto and P.G. Smith, Design and Analysis of Randomized Clinical Trials Requiring Prolonged Observation of Each Patient, Brit. J. Cancer, 34:585 (1976) and Brit. J. Cancer, 35:1 (1977).
6. W.M. Cowan and D.F. Wann, A Computer System for the Measurement of Cell and Nuclear Sizes, J. Microscop. 99:331 (1973).
7. C.J. Cornelisse, Real-Time Morphometric Analysis of Type I and Type II Fibers in Cryostat Sections of Human Muscle Biopsies. Path. Res. Pract. 166:218 (1980).

Chapter 8

Morphometrically Estimated Variation in Nuclear Size - A Useful Tool in Grading Prostatic Cancer

J.H.M. Blom, F.J.W. ten Kate, F.H. Schröder and R.O. van der Heul

Original articles

Morphometrically estimated variation in nuclear size

A useful tool in grading prostatic cancer

J. H. M. Blom[1], F. J. W. Ten Kate[2], F. H. Schroeder[1], and R. O. van der Heul[2]

Departments of [1]Urology and [2]Pathology, Erasmus University, Rotterdam, The Netherlands

Summary. At present there are several grading systems for prostatic carcinoma. Most are difficult to reproduce. An objective method of grading seems to be necessary and could make comparisons between various groups of patients easier and grading more reliable.

In the present study morphometrically estimated nuclear size and variation in nuclear size are matched with the survival rates of 207 patients who underwent total perineal prostatectomy for cancer. On the basis of morphometrically estimated variation in nuclear size the patients could be divided into two groups with significantly differing survival rates. In this way it was possible to split the group of patients with grade 2 carcinoma (Mostofi's grading system) into two groups of patients with significantly different survival rates. The survival rates in these two groups did not differ significantly from those in the patients with Grade 1 and Grade 3 tumors respectively.

The results are discussed in the light of the recent literature on the subject. Morphometry seems to be a valuable tool in grading prostatic cancer.

Key words: Prostate cancer – Cell morphometry – Patient survival

Since Broders' first report on grading epitheliomas of the lip in the early 1920's [5] many investigators have tried to correlate the histological picture of prostatic carcinomas with the clinical course of the disease [9, 13]. This has resulted in the introduction of many grading systems for prostate cancer, but only few of them found wide acceptance. Several reasons can be indicated for this phenomenon:

First prostatic carcinoma often presents in various histological patterns and several of such patterns can be found in the same tumor and even in the same slide. The different patterns can vary considerably in appearance, ranging from well differentiated parts, almost resembling normal prostatic glandular tissue, to undifferentiated parts in which absolutely no features of the original prostatic tissue are recognizable. Within these different patterns or "tumor formations" cytological characteristics may vary in the same way from regularly arranged cuboidal cells without any nuclear pleomorphism to disorderly arranged cells with nuclei that show considerable variation in size, shape and staining. Furthermore it is not uncommon that in rather well-differentiated parts of a tumor cytological characteristics show marked abnormalities, suggesting a very malignant tumor, while on the other hand hardly any nuclear pleomorphism may be found in tumors with a solid pattern of growth. It is difficult to take account of all these variable features in one grading system.

A second reason for the poor acceptance of grading systems is their poor reproducibility. Most grading systems produce the best results in the hands of the person who developed the system, while in other hands the reproducibility is rather disappointing [10, 12, 14].

A third reason is the subjectivity in interpreting the results of the various grading systems. Generally there is no problem in identifying the low grade and high grade tumors, whatever system is used. The problem lies in the large group of patients that neither have clear high grade nor evident low grade tumors and are by exclusion placed in the poorly defined intermediate group of patients whose prognosis apparently is not clearly defined. This is the truly problematic group.

In 1975 Mostofi [13] proposed a grading system that seemed to be quite easy to apply. In the first place he clearly defined differentiation as the tendency of a tumor to form glands and the characteristics of these glands as compared to normal prostatic glands. Anaplasia was defined as a scaled assessment of nuclear characteristics such as nuclear size, hyperchromatism, pleomorphism, presence of nucleoli and mitoses. This system seemed to solve the problem of classifying tumors that on one hand may grow in solid sheets with no gland formation and with a slight cellular atypia as opposed to the cytologically more anaplastic tumors forming well developed glands.

120

Table 1. Number of tumor formations			Table 2. Tumor stage in 207 patients		

Table 1. Number of tumor formations

1 formation	113 patients	113 formations
2 formations	152 patients	304 formations
3 formations	73 patients	219 formations
4 formations	8 patients	32 formations
Total	346 patients	668 formations

Table 2. Tumor stage in 207 patients

Stage	No. patients	%
A	28	13.5
B	112	54.1
C	64	30.9
unknown	3	1.5

In an extensive study Schroeder and co-workers [16] evaluated the prognostic weight of each of the parameters in Mostofi's grading system and they came to the conclusion that only glandular differentiation, nuclear pleomorphism and amount of tumor seen in the slide were important parameters in relation to the prognosis of the disease. The presence of mitoses also showed importance, but the vast majority of prostatic carcinomas contain no or very few mitoses. Schroeder and co-workers proposed a simplification of Mostofi's grading system, showing its application in a large series of 346 cases of prostatic carcinoma, all graded by Mostofi [17].

In 1979, when the present study started, the question came up whether the parameter variation in nuclear size and shape (nuclear pleomorphism) could be objectivated in some way. It has been shown for other tumors and benign tissues [15] that with morphometry, using a planimeter in combination with a computerized evaluation of the measurements, structures can be quantified for several parameters such as surface area, circumference (perimeter), relative volumes, shape descriptions etc. It was hoped that with such an image analysing system it could be possible to have an objective tool in grading carcinoma and to diminish the subjectivity and variability resulting from the use of the conventional grading systems. The initial results were reported in 1982 [3] and 1983 [2, 4].

Independently from our work a similar project was carried out at the Brady Urological Institute in Baltimore. It was shown that the so-called "nuclear roundness factor" correlated very well with prognosis [6, 7, 8].

In the present study nuclear variation in size and shape has been estimated in 207 cases of prostatic carcinoma with a computerized semi-automatic image analysing system. The results have been correlated with survival and Mostofi's grading system.

Material and methods

Patients

In a series of 484 patients on whom the late Dr. Elmer Belt performed a total perineal prostatectomy for cancer the patient charts were reviewed retrospectively. In 346 cases histological slides from the prostatectomy specimens were available for review. These tumors were all regraded by Dr. Mostofi without knowledge of the follow-up of the patients. Most of the tumors consisted of a varying number of morphologically different formations (e.g. tubular, cribriform, solid). As Table 1 shows their number varied from one to four per patient and a total of 668 tumor formations have been

matched with the clinical data of 346 patients. This has been reported elsewhere [16].

For various reasons not all of the 346 sets of histological slides were suitable for morphometry. Fourty-six patients received hormonal treatment before total prostatectomy, causing squamous metaplasia to a greater or lesser extent. In most cases the presence of metaplasia was no problem for conventional grading, but these patients were excluded from morphometry. The quality of the histological slides of 20 patients was too poor for morphometrical purposes. The slides of 10 patients had been lost during the last years. In five slides there was a significant squamous metaplasia, suggesting the use of hormones, although there was no note in the patient chart on the use of hormonal treatment. Fifty-eight slides could not be analyzed for various reasons, for instance because the amount of tumor in the slides was too small to obtain enough nuclei to process or the contours of the nuclei were too vague for accurate tracing, or the slides that were at our disposal did not contain tumor at all. This resulted in 207 cases that were available and suitable for morphometry. The number of slides per patient varied from 1 to 24 with an average of 3 slides per case. The slides were almost all from the same institution (Good Samaritan Hospital, Los Angeles, Ca). A few slides came from another hospital (Hollywood Presbyterian Hospital, Hollywood, Ca).

The clinical stages of carcinoma of these 207 patients are given in Table 2.

Patient identification on the slides was only by number and morphometry was therefore carried out in a blind fashion.

Morphometry

The morphometrical analysis was performed with a semi-automatic computerized image analysing system (Videoplan, Kontron). Basically this system consists of three components:
1. a graphic tablet
2. a cursor or a pen, and
3. a desk computer

Both the graphic tablet and the cursor or pen are connected to the computer. Besides these, a printer/plotter is connected to the computer.

The graphic tablet and cursor

The Videoplan graphic tablet (or digitizer tablet) operates on the magnetostrictive principle. The area of the tablet is divided in a horizontal and vertical way by a mesh of ferromagnetic wires, laid on a substrate beneath the tablet surface, spaced at regular intervals in X and Y direction. This mesh of wires provides a permanent magnetic field. In addition the wires conduct electronically induced magnetic pulses in both directions. These pulses are emitted at a constant frequency and travel at a constant speed, unaffected by environmental conditions. The cursor has two crosswires, indicating the exact point on the tablet. In the centre of these crosswires a light-emitting diode (LED) is mounted, to make the centre of the

Table 3. Effect of magnification on accuracy of digitizing (each nucleus is digitized at least 50×)

Magnification	mean area (µ²)	standard deviation	coefficient of variation
400×	76.65	4.55	5.94
	25.32	2.69	10.62
	59.89	4.15	6.93
	10.31	0.49	4.75
	22.45	2.12	9.44
	5.73	0.25	4.36
630×	78.58	3.13	3.98
	23.84	1.36	5.70
	5.93	0.19	3.20
	10.10	0.34	3.37
	56.18	3.19	5.68
	78.76	3.30	4.19

Table 4. Lymphocyte measurements during several years (the maximum deviation from the mean is 5%)

Year	mean nuclear area	S.D.
1939	15.79	2.78
1944	17.23	2.87
1945	17.41	3.47
1955	17.09	3.62
1965	15.47	2.43
1969	16.75	2.60
1970	16.30	3.24
Mean	16.58	

Table 5. Effect of number of nuclei on accuracy

Number of nuclei	Mean nuclear area
25	38.00
50	32.33
75	34.22
100	33.12
125	32.25
150	31.85
200	31.96
275	32.06
400	32.52

crosswires visible in the microscope. When positioned or moved on the surface of the tablet, the cursor intercepts X and Y pulses continuously through a receiver coil to derive coordinate locations. Based on the parameters selected, the microprocessor continuously calculates and updates the individual measurements until terminated. When a line is drawn, for example around a nucleus, the computer can calculate the surface area, the circumference (perimeter) and several other parameters. The resolution of the system is 0.1 mm. However, as the average diameter of a nucleus, projected on the graphic tablet is about 15 mm, this resolution constitutes less than 1% of the total diameter.

The microscope

The microscope is a regular Zeiss microscope. It has 10× wide field eyepieces and plan achromat objectives (magnification: 4×, 10×, 40× and 63×). On the microscope a drawing attachment is mounted (Zeiss 474620), so that the LED in the centre of the cursor can be seen together with the normal field of vision of the microscope. The microscope was arranged in such a way that when the cursor was placed in the centre of the graphic tablet, its LED was seen in the centre of the field of vision of the microscope. In order to see the LED clearly, the light of the microscope had to be adjusted to a convenient level. Also the room illumination had to be dimmed to a lower level.

Accuracy and reproducibility

Before starting the actual morphometric measurements the accuracy and reproducibility of the technique was studied:

1. What is the best magnification of the microscope?

2. Could there be artefacts due to different handling of the material in different laboratories?

3. How many nuclei should be digitized per tumor formation?

4. Should one measure nuclei in all available slides or is limitation to one slide per patient possible?

5. Is one field of vision representative for a given tumor-formation or should one go randomly through the slides?

6. How accurate is the mechanism of tracing nuclei?

1: To establish the best suitable magnification of the microscope we digitized several nuclei of one tumor repeatedly using several magnifications. The results are shown in Table 3.

The largest possible magnification was optimal. Although a higher magnification would probably give better results, the highest power dry system was used for practical reasons. The total magnification of the microscope was $63 \times 10 = 630 \times$.

To examine the accuracy of measuring with this magnification a circle in an eyepiece grid was traced several times and a coefficient of variation in surface area of 3.94% was found. This is within acceptable limits.

2: It is a well known fact that fixation and laboratory handling of tissue causes shrinkage of all structures to a certain amount. This is true for fresh and old material. To investigate the effect of tissue handling in the two different laboratories during several years, we digitized lymphocytes in the slides of several patients from each laboratory and representing several years. Slide preparation at various points in time was checked because it is unknown to us whether material handling is still the same now as it was in 1939. Table 4 shows that there is in fact no significant difference between the effect of fixation and tissue handling for the years from 1939 through 1970.

3: To establish the number of nuclei necessary in each tumor formation up to 400 nuclei were digitized in one tumor formation. As shown in table 5, the values for the mean nuclear area did not change significantly above a number of 125 nuclei. On the basis of this result it was decided to use for the routine of this study 150 nuclei per tumor formation.

4 and 5: Regarding the number of slides and the areas in the slides to be digitized an analysis of variance (ANOVA) [1] was used. With this method the accuracy of measuring was checked. Three tumors from patients who lived for a long time after total prostatectomy without any evidence of recurrence and three tumors from patients who died very soon after total prostatectomy of metastatic disease. Of each of

Table 6. Analysis of variance Coefficient of variation for nuclear area (V_{area})

Patients (prognosis)	field of vision	Slide 1		Slide 2		Slide 3	
		Day 1	Day 2	Day 1	Day 2	Day 1	Day 2
A (good)	1	27.52	16.97	28.35	24.48	22.96	15.43
	2	25.99	19.71	22.40	18.67	24.45	32.76
B (good)	1	18.57	24.75	25.43	27.39	28.14	19.97
	2	29.21	16.20	21.73	19.24	19.26	18.60
C (good)	1	20.57	20.40	21.67	22.54	23.00	23.96
	2	24.98	22.37	21.46	21.65	19.60	23.80
D (poor)	1	42.70	21.52	19.58	19.58	27.13	25.89
	2	20.27	28.50	29.28	30.94	24.21	27.52
E (poor)	1	27.81	24.55	27.61	32.43	29.81	32.12
	2	17.16	23.71	26.38	25.62	23.99	21.57
F (poor)	1	22.48	26.10	32.68	25.08	75.42	67.24
	2	23.99	24.80	38.76	27.31	43.14	24.67

Table 7. ANOVA Final calculation

Category	DF	diff(ΣX^2)	Variance diff/DF	F-test
total	71	6,279.05	88.44	
days	1	88.11	88.11	1.20
fields of vision	1	161.79	161.79	2.20
slides	2	333.38	166.69	2.27
patients	1	849.89	849.89	11.58
remainder	66	4,895.88	73.42	

these six tumors 3 slides and in each slide two randomly chosen fields of vision were digitized. The location of the fields of vision was recorded precisely by means of the crosstable of the microscope. Several days later the whole procedure was repeated, digitizing exactly the same fields of vision in the same slides. The results are summarized in Table 6.

From these values variances were calculated for the three different slides, the two fields of vision, the two separate days and the two groups of patients. The results are shown in Table 7.

After entering these values in the F-table [1] it was shown that there were no significant differences between the measurements in the different slides or fields of vision. Also there was no significant difference between measurements on different days. The two groups of patients however, showed a significant difference in variance. Accordingly, the method of morphometry and the coefficient of variation for nuclear area should allow a good differentiation between the various patients.

Our conclusion was that one could digitize anywhere in the slides and use as many or as few fields of vision as needed to obtain the proper amount of nuclei to be digitized.

Methods of measurements (digitizing)

Before the measurements started several parameters were selected in the computer software program. These parameters were:
1. surface area,
2. perimeter, and two so-called form factors:
3. $FORM_{pe}$, also called circularity index. This form factor is given by the equation:

$$FORM_{pe} = \frac{4 \times \pi \times area}{(perimeter)^2}$$

In the case of an exact circle the value for $FORM_{pe}$ equals 1. In all other cases $FORM_{pe}$ is less than 1. The more the shape of a structure deviates from the circle, the less the value for $FORM_{pe}$ becomes.

4. $FORM_{ell}$, also called ellipticity index. This form factor is given by the equation:
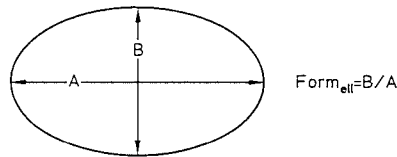
Circles: $Form_{ell} = 1$

All other structures: $Form_{ell} < 1$

As for $FORM_{pe}$, the value for $FORM_{ell}$ equals 1 in case of a circle. In all other structures the value for $FORM_{ell}$ becomes less than 1.

Both form factors are suitable to objectivate the shape of the nuclei, while the area and perimeter were measures for nuclear size. These four parameters were measured and calculated for 150 nuclei in each tumor formation. When a tumor consisted of only one tumor formation, only 150 nuclei were measured in that tumor. When a tumor consisted of two or three formations, the number of nuclei digitized were 300 and 450 per tumor respectively. From these 150 nuclei a mean value for each parameter and a standard deviation were calculated.

Statistics

The main goal was not to objectivate size and shape of the nuclei, but the *variation* in size and shape. The variation of the form factors was calculated by dividing the standard deviation by the mean value. In this way a coefficient of variation was calculated for each of the parameters in each of the tumor formations. The coefficient of variation is indicated by the capital letter "V".

In this way the coefficient of variation for area (V_{area}) was obtained as a standard for the variation in nuclear size and the
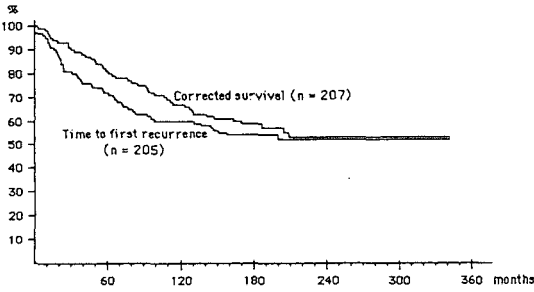
Fig. 1. Comparison between survival and time to first recurrence. The patterns of the curves are identical. The survival curve is corrected for intercurrent or unknown causes of death. In two patients it was not known when they developed metastases
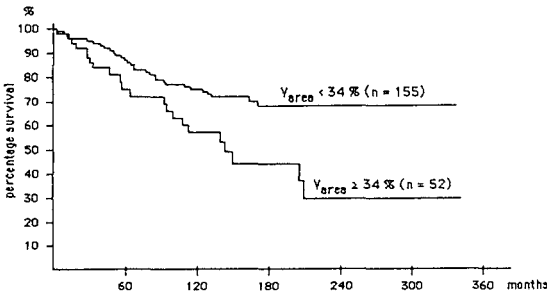

Fig. 2. The patients are divided morphometrically into two groups with significantly differing corrected survival rates ($p < 0.01$)
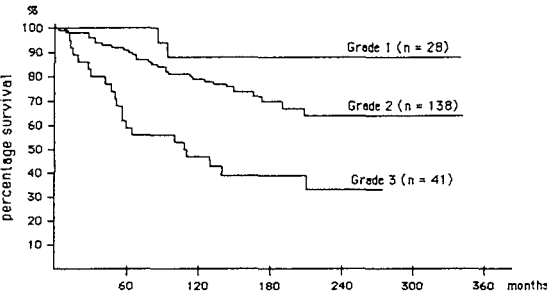

Fig. 3. The patients are divided according to Grade (Mostofi system)

Table 8. Morphometrically estimated variation in nuclear size

V_{area}	N	No. deaths	No. deaths from cancer	% of deaths
$< 34\%$	155	140	31	22.1
$\geq 34\%$	52	48	22	45.8

coefficients of variation for $FORM_{pe}$ and for $FORM_{ell}$ ($V\,FORM_{pe}$ and $V\,FORM_{ell}$) as a standard for the variation in nuclear shape.

In each tumor formation 150 nuclei were digitized and for each nucleus the values for area, perimeter, $FORM_{pe}$ and $FORM_{ell}$ were calculated. Furthermore the computer calculated the mean values and the standard deviations. After finishing digitizing the values were all stored on disks and the results were printed out. The procedure was repeated for each tumor formation. At the end the coefficient of variation was calculated by dividing the standard deviation by the mean. In the case of more than one tumor formation per tumor the highest value for V was used for further evaluation.

Most results are presented as survival curves. These curves are calculated according to Kaplan and Meier [11]. The survival curves are corrected for intercurrent, tumor unrelated and unknown causes of death. In this way the curves show the impact of death from carcinoma more clearly without confusing the picture with the relatively high number of intercurrent deaths. For the evaluation of the differences between the curves the Logrank test was used.

We used death as an endpoint of study and not recurence of disease because all patients who had recurrence of their disease were dead at the time of the last review. Most of them indeed died of prstatic carcinoma and only seven (11.8%) died of other causes than prostatic cancer (causes of death in these men were: cardiovascular: 2, cerebrovascular: 1, murder: 1, other cancer: 3). It was shown that the curves did not change in a significant way when time to recurrence was used instead of time to death (Fig. 1). Of course the curve for recurrence of disease is shifted somewhat to the left, but the slopes of the curves are identical.

Results

With the morphometrically estimated variation in nuclear size (V_{area}) it was possible to split the whole group of 207 patients in two subgroups with a different prognosis. One group of patients with a $V_{area} < 34\%$ and a second group of patients with a $V_{area} \geq 34\%$. The cut-off point of 34% was found empirically. The first larger group consists of 155 patients. In this group there were 31 patients who died of carcinoma. The second group, counting 52 patients, showed death from carcinoma in 22 patients. The difference between the two groups is significant ($p < 0.01$, Table 8)

Graphically the corrected survival rates of the two groups of patients are shown in Fig. 2. As can be seen from this figure even after ten years there is a fair chance of dying of carcinoma. Also here the difference between the two groups is significant (Logrank test, $p < 0.01$).

Figure 3 shows the survival rates of the same 207 patients, divided into three groups according to grade (Mostofi system). As can be expected the patients with a grade 1 tumor had the best prognosis. In the whole group only two patients died of carcinoma and after 93 months there was no death of tumor in this group. Patients with grade 3 tumors do worst, even after 200 months patients died of prostatic carcinoma. The largest group of patients ($n = 138$) have grade 2 tumors and show an intermediate course of disease. However, also in this group after 15 years patients still died of prostatic carcinoma (see Table 9).

When the group of patients with grade 2 tumors was divided according to morphometrical measurements, two groups of patients with significantly differing survival rates ($p < 0.01$) were identified (Fig. 4). However, the

124

Table 9. Corrected survival of 207 patients, divided according to grade

Grade	No. patients	5-years	10-years	15-years	20-years
1	28	100%	88%	88%	88%
2	138	91%	79%	70%	64%
3	41	59%	47%	39%	33%
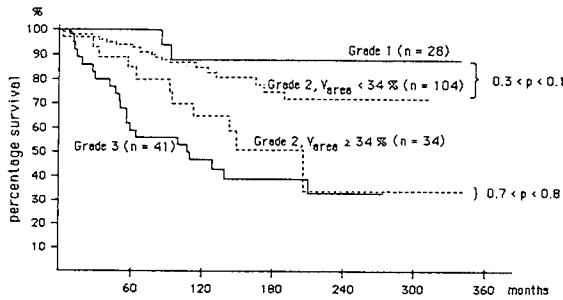
Fig. 4. The group of patients with Grade 21 tumors is divided morphometrically into two groups with significantly differing survival rates

Fig. 5. Mean nuclear area 207 patients are divided into two groups according to prognosis (death from prostatic cancer)
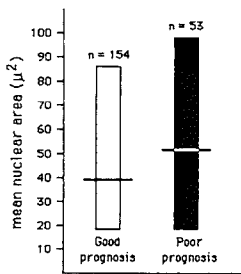
patients with grade 2 tumors and $V_{area} < 34\%$ did not show a significantly differing survival rate from those with grade 1 tumors, while the patients with grade 2 tumors and a $V_{area} \geq 34\%$ had survival rates not differing from the patients with grade 3 tumors. The intermediate group of patients with grade 2 tumors could be divided into two groups: one with a prognosis almost equal to those with Grade 1 tumors and one with a prognosis almost equal to those with Grade 3 tumors.

Besides the variation in nuclear size also the mean nuclear size showed some correlation with the prognosis. In the group of 53 patients who died of prostatic carcinoma the tumors had a mean nuclear surface area of 51.4 μ^2, while the mean nuclear size in the tumors of the

remainder of the patients was 39.6 μ^2. The difference is significant, but as Fig. 5 shows there is an almost complete overlap of the two groups.

In the group of 140 patients with a mean nuclear size $< 50 \mu^2$ twenty-six patients (18.6%) died of prostatic carcinoma. This was the case in 27 patients (40.3%) with a mean nuclear size of $\geq 50 \mu^2$. This difference is significant ($p < 0.01$).

Neither V FORM$_{pe}$ nor V FORM$_{ell}$ allowed to identify patients with different survival patterns. In now way was it possible to correlate these parameters with prognosis.

Discussion

Besides clinical stage the histopathological grade of a tumor plays an important role in establishing the prognosis of prostatic carcinoma. It is a well established fact that nuclear pleomorphism is one of the most important parameters in grading prostatic carcinoma. Most grading systems, especially those developed in the last two decennia use this parameter besides glandular differentiation and a varying number of other parameters. In an extensive study on 346 cases of prostatic carcinoma, all graded by Dr. Mostofi using his grading system [13], Schroeder and co-workers [16] found that in grading especially glandular differentiation and nuclear pleomorphism play an important role in the evaluation of the malignant potential of the tumor. Only the presence of mitoses may have an additional effect on the prognosis, but all other parameters as for instance the aspect of the cytoplasm, the presence or absence of nucleoli, the presence or absence of nuclear vacuoles, the number of various tumor formations, nuclear size do not have any weight in the prognosis of the tumors and may as well be omitted to simplify the system of grading.

Some reports show that grading of prostatic carcinoma is a somewhat subjective matter. Generally there are little problems in recognizing the true high grade and the true low grade tumors. The problems arise with the tumors that are neither high grade nor low grade. These tumors are by exclusion placed in a large and poorly defined intermediate group. However, in this group there may be large differences in prognosis, indicating that although these tumors all seem to have an intermediate grade, they are not uniform in behaviour. It is mainly the large group of Grade 2 tumors which presents difficulties in predicting prognosis. In this light it is strange that attempts to objectivate grading of prostatic carcinoma have started rather late.

The first investigators who quantitated nuclear characteristics and correlated their findings with tumor grade were Stöber and Schmidt [18] who measured nuclear area morphometrically and found a correlation with nuclear size and tumor grade.

In 1982 Diamond and colleagues [6, 7] presented their results with a new shape descriptor, called Nuclear Roundness Factor, and they were able to identify in a blind way two groups of patients who were cured by radical surgery or who would later die of cancer. Their system was shown to be 100 per cent accurate in this small

series of 27 patients. There were no false positives and no false negatives in the prediction of death from carcinoma of the prostate. In an attempt to reproduce these findings the nuclear roundness factor was calculated using the data of our patients. Surprisingly the values for nuclear roundness did not correlate well with corrected survival. Even an attempt to digitize some of our histological material on Diamond's equipment failed to identify nuclear roundness as a useful parameter in our hands. There was a very good correlation between the calculated nuclear roundness factor and the $FORM_{pe}$, but both failed to show any correlation with the prognosis.

It has not become clear to us why in our hands the nuclear roundness factor was not an important prognostic parameter. In an attempt to resolve this discrepancy one of us (JB) and one of the investigators from the Brady Urological Institute digitized on two occasions some geometrical figures with known sizes and shapes (circles, ellipses, squares, triangles and hexagons). It was noticed that each investigator's own results were easily reproducible, but that it was not possible to reproduce the results of each other. It was also found that the intra-observer variations were largest in digitizing the smaller figures.

In order to evaluate whether a difference in equipment might be the cause of the fact that nuclear roundness was not prognostic in our hands, some of our slides were digitized on Diamond's equipment. Also on Diamond's equipment in our hands nuclear roundness did not predict prognosis. In five patients who did not develop metastases up to a mean time of 147.8 months after total prostatectomy the mean nuclear roundness varied from 1,024 to 1,085 with an average of 1,054.4. A group of 17 patients who developed metastases after a mean interval of 34.9 months after total prostatectomy showed a mean value for nuclear roundness of 1,049.2 (range: 1,020–1,079). In the same groups of patients the mean values for V_{area} were 34.1% in the group with a good prognosis (range:28.3–40.0%) and 36.0% in the group with a poor prognosis (range: 24.9–52.8%).

Digitizing of some of our slides by D. Diamond also failed to show any correlation of nuclear roundness with survival. The reasons for the discrepancy of results is not clear, but it may be concluded that the accuracy of the digitizing equipment does not play as important a role as suggested by Diamond [7].

In conclusion it can be said that objectivation of nuclear size is possible and that the variation of this parameter gives a good correlation with survival. Patients with tumors that show a large variation in nuclear size will have a poorer prognosis than patients whose tumor nuclei do not show large variation in size. Morphometry can help in making a decision whether the patient might have a poor prognosis and may need aggressive therapy. Morphometry cannot replace the conventional grading systems at this time, but it can add objectivity to grading. More work has to be done to standardize the system of morphometry to obtain interchangeable results.

References

1. Barnett RN (1971) Analysis of variance. In: Barnett RN (ed) Clinical laboratory statistics. Little, Brown and Company, Boston, p32
2. Blom JHM, ten Kate FJW, Schroeder FH, van der Heul RO (1983) Grading of prostatic carcinoma – Evaluation of single parameters and cytomorphometry. In: Pavone-Macaluso M, smith PH (eds) Cancer of the prostate and kidney. Plenum Press, New York, p109
3. Blom JHM, ten Kate FJW, Schroeder FH, van der Heul RO (1982) Nuclear pleomorphism as a parameter in grading prostatic carcinoma. A morphometrical study. Abstract 213, presented at the Annual Meeting of the AUA, Kansas City
4. Blom JHM, ten Kate FJW, Schroeder FH, van der Heul RO (1983) Zellkernpleiomorphismus als ein Parameter beim Grading des Prostatakarzinoms. Eine morphometrische Studie. Beitr Urol 3:320
5. Broders AC (1920) Squamous-cell epithelioma of the lip. A study of five hundred and thirty-seven cases. JAMA 74:656
6. Diamond DA, Berry SJ, Umbricht Ch, Jewett HJ, Coffey DS (1982) Computerized image analysis of nuclear shape as a prognostic factor for prostatic cancer. Prostate 3:321
7. Diamond DA, Berry SJ, Jewett HJ, Eggleston JC, Coffey DS (1982) A new method to assess metastatic potential of human prostate cancer: relative nuclear roundness. J Urol 128:729
8. Epstein JI, Berry SJ, Eggleston JC (1984) Nuclear roundness factor: a predictor of progression in untreated Stage A2 prostate cancer. Cancer 54:1666
9. Gaeta JF, Gardner Jr WA (1979) Histologic grading of prostatic cancer: background and possibilities. In: Murphy GP (ed) Prostatic cancer. PSG Publishing Co, Littleton, p41
10. Harada M, Mostofi FK, Corle DK, Byar DP Trump BF (1977) Preliminary studies of histologic prognosis in cancer of the prostate. Cancer Treat Rep 61:223
11. Kaplan EL, Meier P (1958) Nonparametric estimation from incomplete observations. J Am Stat Assoc 53:457
12. Kern WH (1978) Well differentiated adenocarcinoma of the prostate. Cancer 41:2046
13. Mostofi FK (1975) Grading of prostatic carcinoma. Part 1. Cancer Chemother Rep 59:111
14. Mostofi FK (1976) Problems of grading carcinoma of prostate. Semin Oncol 3:161
15. Oort J, Baak JPA, Boon ME, Swanson Beck J, Anderson JM, van der Heul RO, Meijer CJLM, van der Valk P (1983) Applications of morphometry in tumour pathology. In: Baak JPA, Oort J (eds) A manual of morphometry in diagnostic pathology. Springer, Berlin Heidelberg New York, p48
16. Schroeder FH, Blom JHM, Hop WCJ, Mostofi FK (1985) Grading of prostatic cancer (I). An analysis of the prognostic significance of single characteristics. Prostate 6:81
17. Schroeder FH, Hop WCJ, Blom JHM, Mostofi FK (1985) Grading of prostatic cancer (III). Multivariate analysis of prognostic parameters. Prostate 7:13
18. Stöber U, Schmidt U (1980) Zur Klinik des Prostatakarzinoms unter Berücksichtigung zyto- und histomorphologischer Befunde. Urol Int 35:233

Jan H. M. Blom
Department of Urology
Erasmus University Rotterdam
P.O. Box 1738
3000 DR Rotterdam
The Netherlands

Chapter 9

General Discussion

9.1 Introduction

The highly variable and often prolonged clinical course of prostate cancer poses difficult problems. Some patients seem to be at such a low risk of dying of prostate cancer, that overtreatment should be avoided. On the other hand, there are many patients whose prostate cancers progress rapidly to a metastatic, incurable stage, causing the patient's death. If it were possible to predict which patients will have predictably different survival rates, decisions as to treatment modalities and time of treatment could be made much more accurately.

Several attempts to stratify patients with prostate cancer have been made and several characteristics, either of the tumor or of the patient have been identified as being prognostically important [1, 2, 3, 4, 5, 6, 7].

9.2 Tumor stage

The impact of tumor stage on prostate cancer is well established and well documented [8, 9]. Nevertheless in the present series (chapter 1) there is no significant difference in the proportion of deaths from prostate cancer between the patients who had a clinical T_0 tumor and those who had a tumor classified as T_{1-2}. Only the patients, clinically judged as having T_3 tumors do significantly worse. Apparently, in this series only the fact whether the tumor is clinically thought to be confined to the prostate is prognostically important. The data in the literature on this subject are controversial. Montgomery and co-workers [10] studied 35 cases of incidental carcinoma, discovered after prostatectomy for presumed benign disease. The minimum follow-up was 8 years. 20 of these patients received hormonal therapy and 15 were not treated at all. None of the patients died from prostate cancer. The conclusion of the investigators was that total prostatectomy is rarely indicated when an incidental prostate cancer is detected. Hanash and co-workers [11] reported a 86% overall 5-year

survival and a 10-year survival of 52% in 50 patients with Stage A ($T_0pT_{1-2}N_0M_0$) prostate cancer who were treated by transurethral resection alone. These figures were comparable to an age matched group who did not have prostate cancer. 129 conservatively treated patients with clinical stage B ($T_{1-2}N_0M_0$) tumors had an overall 5-year survival of 47% and a 10-year survival of 19%. However, there is a bias in the data of Hanash and co-workers. All stage A tumors were found incidentally after prostatectomy for urinary outlet obstruction. With this prostatectomy a number of these incidental carcinomas have been removed radically, leading to the patient's cure. None of the patients with stage B disease have had radical treatment. In this way a seemingly large discrepancy between survival in stage A and in stage B may have appeared.

Especially in the stage A tumors grade and microscopic tumor extension is of great prognostic importance. Hanash and co-workers [11] already observed that the group of stage A patients could be divided into subgroups with significantly different survival rates. Grade was a prognostic factor in their series. Patients with stage A and grade 3 tumors did poorer than patients with stage A and grade 1 or 2 tumors. Other investigators confirmed this observation [12, 13]. This has resulted in a subdivision of stage A tumors in stage A_1 ($T_0pT_1N_0M_0G_1$) and stage A_2 ($T_0pT_2N_0M_0G_{1-3}$ or $T_0pT_1N_0M_0G_{2-3}$). Stage A_1 indicates a focal and well differentiated carcinoma, whereas stage A_2 comprises the larger and less differentiated tumors. In chapter 3 of this thesis the prognostic impact of tumor extension and grade was studied. It was shown that glandular differentiation, amount of tumor in the slides, size of the cell, size of the nucleus, nuclear variation in size and shape, and grade as established by Mostofi (chapter 4) were prognostic parameters in this group of patients. Patients whose tumors showed a small amount of tumor, slight nuclear variation and were estimated to be grade 1 did not die from their tumors. In other words: these are the patients in whom observation rather than radical treatment after TUR or retropubic prostatectomy may be the treatment of choice.

9.3 Adjuvant treatment

The question of the possible beneficial effect of adjuvant treatment in conjunction with radical prostatectomy is controversial. In the present study (chapter 1) there seems to be no major role for adjuvant hormonal treatment. A similar observation was made by Catalona and associates [14] who found no local recurrence in 9 out of 21 patients who had microscopic tumor extension beyond the confines of the prostate after radical prostatectomy and did not receive adjuvant treatment. Six out of these 9 patients were alive free of recurrence for 6 years postoperatively.

Adjuvant radiotherapy has recently been subject to much discussion. Carter and co-workers [15] reported 31 patients, who had positive surgical margins after radical prostatectomy. These patients were treated with adjuvant external beam radiotherapy postoperatively. At an average follow up of 5 years they found a 3% local recurrence rate, a distant recurrence rate of 6%, and 94% of the patients were alive without evidence of disease. However, their study was not set up as a prospectively randomized one. In a study on 140 patients who underwent a pelvine lymph node dissection and a radical prostatectomy Van den Ouden and co-workers [16] found histological evidence of positive surgical margins in 40 patients. 14 of them showed progression of disease, indicating a predictive value of 35% during a 42.3 months' period. Again there is an argument against adjuvant hormonal treatment. Of these 14 patients, 8 (57%) had progression during the first year after operation. All 14 patients had progression of disease within 3 years after radical prostatectomy. However, only 22.5% of the patients who developed progression of disease had a local recurrence. In other words: although tumor had been left at the operation site (as shown by the positive surgical margins), these remnants of tumor did not lead to local tumor growth in 77.5% of the patients within the follow-up period. From this observation it may be concluded that local adjuvant therapy such as radiotherapy is not indicated in patients with positive surgical margins after radical prostatectomy.

9.4   Grading

Over the last 60 years many studies (chapter 2), including this thesis, have shown that there certainly is a correlation between the microscopic aspect of a tumor and the final outcome of the disease in localized prostate cancer. However, many difficulties have been encountered in defining that correlation in a reliable and reproducible manner. Several reasons for the poor acceptance of many grading system can be identified [17].

This has resulted in a large number of different grading systems. In 1978 a series of workshops took place to try to evaluate the problems concerning grading of prostate cancer. In their report on these workshops Murphy and Whitmore [18] pointed out several basic considerations with which all grading systems have to deal:

1. the availability of an adequate sample of tissue
2. objectively defined criteria for grading
3. the degree of reproducibility of interpretation
4. simplicity
5. the predictive value of the system relative to the biologic potential of the tumor in the context of clinical management.

9.4.1 *the availability of an adequate sample of tissue*

One of the first and major problems in grading of prostate cancer is the fact that in most cases only small amounts of tissue, obtained through a needle biopsy, are avaliable for diagnosis and grading. Prostate cancer is often heterogeneous and different growth patterns are frequently seen in the same tumor, and even in the same slide. A needle biopsy may not be representative for the whole tumor. Kastendieck [19] showed this fact in 120 radical prostatectomy specimens. He compared the findings of the needle biopsies with the radical prostatectomy specimens. In the biopsies 55.8% of the tumors were judged to have a uniform growth pattern, of which 15% were well differentiated. The radical prostatectomy specimens

however, showed uniformity in only 27.5% with 8.4% of the tumors being well differentiated. Similar findings were obtained by Ackermann and Müller [20], who found that out of 21 prostate biospies indicating a well differentiated tumor only 33.3% showed to be a uniformly well differentiated tumor in the radical prostatectomy specimen.

One of the advantages of fine needle aspirations is that the puncture is much less traumatic to the patient than a thick needle biopsy. It is easy to perform multiple fine needle biopsies without any extra discomfort for the patient. However, many urologists do not use this diagnostic tool because only cytological parameters can be studied. Architectural criteria, such as gland formation, relation of epithelium to stroma etc. cannot be examined in cytological smears. There are however papers that refute that argument. Among many others de Voogt [21] reported on the use of cytology in diagnosing prostate cancer. In 294 patients both fine needle aspiration and thick needle biopsies were performed. Of the 294 samples 12 were not evaluable because the amount of material was not enough for proper examination. In the remaining 282 cases there was agreement with the thick needle biopsy in 265 samples (94%). This means that for the diagnosis of prostate cancer cytology can be a reliable tool. Maksem and co-workers [22] compared patterns of cellular arrangement in 50 cases of prostate cancer studied in histological and cytological specimens, obtained simultaneously. They found a 84% agreement between cytological grading and the Gleason score in the histological specimen. They concluded that it is possible to predict the Gleason histological pattern on the basis of cytological specimens of prostate cancers. These observations have been confirmed by others [23, 24]

9.4.2 objectively defined criteria for grading

Many grading systems clearly suffer from a lack of proper definitions of their criteria. It is, however, very difficult to define the necessary criteria strictly. The main reason lies in the fact that it is very difficult to objectivate an observation which is based on a rather subjective optical impression. In

chapter 3 it was already shown that in Mostofi's grading system the parameter "grade" could not be replaced by any of the characteristics of this system alone. Grade is a true computation of several parameters. Furthermore it cannot be excluded that, beside these parameters, the visual impression of the pathologist, not expressed in any of the parameters used, has an influence on "grade". It is certain that an experienced pathologist can separate at a glance the poorly differentiated from the well differentiated tumors, without actually look into detail to each of the parameters of the grading system. Although such a pathologist's experience is essential, it will make comparison between different investigators difficult. It is almost impossible to define visual impressions. Morphometry is one of the tools that may be able to help to solve this problem.

9.4.3 *the degree of reproducibility of interpretation*

A big problem in grading is the reproducibility. Almost all grading systems work reasonably well in the hands of the investigator who developed the system, but fail to be as good in other, even experienced hands [25]. Among others it was shown by Harada and co-workers [26] that the interobserver variability is rather high. Comparison of their results using Gleason's grading system with the results of Gleason himself showed an agreement of 63.9% when only the primary tumor pattern was regarded. Reading of the secondary pattern obviously presented more problems: only 44.3% agreement could be found. In reading the combined primary and secondary pattern there was an agreement between the investigators and Gleason in only 38% of their cases. However, they also showed that the intra-observer reproducibility is reasonably good. In 71% of the cases they were able to reproduce their interpretations of the Gleason system. Ten Kate and co-workers [27] studied the interobserver reproduciblity of 5 grading systems. They studied the Broders system, the M.D. Anderson system, the Gleason system, the Mostofi system and the modified Mostofi system, as presented in chapters 3, 4 and 5 of this thesis, in 50 patients

who had undergone total perineal prostatectomy for clinically localized prostate cancer. It was shown that none of the five grading systems had a very high degree of reproducibility. The reproducibility of the Broders system and the M.D. Anderson system was shown to be reasonably good, but the other three systems showed only a limited reproducibility.

9.4.4 *simplicity*

Many grading systems do not work well because they are rather complicated. In chapters 3 and 4 it was demonstrated that in Mostofi's grading system several parameters, such as amount of stroma, nuclear vacuoles nucleoli, the aspect of the cytoplasm, have no real prognostic importance. It was possible to omit these parameters without influencing the estimation of the total grade of the tumor. In this way a simpler grading system could be developed, using a scoring system, based on a multivariate analysis, as depicted in chapter 6. It was shown by Gallee and co-workers [28] that this simplified system indeed showed a higher accuracy in predicting the prognosis than the Mostofi system, using all 12 parameters.

9.4.5 *the predictive value of the system relative to the biologic potential of the tumor*

Another question is whether the different grading systems are equally accurate in predicting the prognosis of the patient. In 1990 Gallee and associates [28] reported their work on comparison of the prognostic accuracy of these five grading systems. In the same 50 patients as mentioned above the prognostic impact of each grading system was established. It was shown that Broders' system and the modified Mostofi system were superior to the other systems as far as the prognostic performance for the event of progression goes. The combination of Broders' and the modified Mostofi system supplied additional prognostic information. This may reflect the fact that the cytological characteristics of the Mostofi system add their weight to the histological characteristics of the

Broders system.

9.4.6 *heterogenity of the tumor*

One of the difficulties of grading prostatic carcinoma is the heterogenity of the tumors. Usually prostate cancers are pleomorphic and consist of more than one architectural formation, each with a different grade. It is unknown which formation determines prognosis, the predominant pattern or the most malignant one, which may be very small. Gleason [29, 30] solved this problem by using two histological tumor patterns. He called these a primary and a secondary pattern, depending on the relative amount of tumor taken by that certain pattern. This distribution did, however, not answer the question whether the pattern with the largest relative volume indeed defines the prognosis in the patient. In chapter 5 it was demonstrated that not the relative amount of a certain tumor pattern determined the prognosis. The prognosis is relatively good when only well differentiated tumor is present. The prognosis is poor, when only poorly differentiated tumor is present in the tumor. The combination of well differentiated and poorly differentiated areas in the prostate was correlated with an intermediate prognosis. This was independent of the relative volume of each of the tumor patterns or tumor formations .

In chapters 3 and 4 two parameters with significant influence on prognosis were identified in Mostofi's grading system. These parameters were differentiation and nuclear variation in size and shape. Differentiation was defined as the tendency of a tumor to form glands and their characteristics as compared to normal prostatic glands. These parameters are the main determinants of grade. To a certain degree these two parameters are independent of each other. There was however a substantial number of cases in which the final grading was a true computation of several parameters in Mostofi's grading system. Apparently there is some visual impression of the pathologist which is not expressed in any of the parameters used.

9.4.7 *Morphometry in grading prostate cancer*

It may be obvious from the above mentioned discussions that an objective tool, with which many subjectivities in grading could be prevented, would be very welcome. Indeed, much work has been done to try to objectivate certain parameters in estimating the prognosis of prostate cancer. Yatani and co-workers [31] correlated morphometrically estimated tumor volume with Gleason grade and they found a statistically significant correlation. Only 10% of the tumor which had Gleason score 1 exceeded 130 mm^3, of the tumors with Gleason score 2, 31.4%, with Gleason score 3, 73.6% and with Gleason scores 4 and 5 all tumors had a volume greater than 130 mm^3. A similar observation was made by Stamey and co-workers [32], who found a strong correlation between morphometrically estimated cancer volume, capsular penetration, lymph node invasion, and Gleason grade in 68 radical prostatectomies.

Morphometry to objectivate cytological parameters has not been practised widely in prostate cancer. In other tumors morphometry of the cell and /or cellular components have been shown to correlate with the prognosis [33, 34].

The first results of morphometry in grading of prostate cancer are reported in chapter 7. It was shown that the variation in nuclear size could be objectivated. It was possible to identify two groups of patients with a significantly different survival pattern. It was, however, not possible to use morphometrically estimated variation in nuclear shape as a prognostic parameter. The observations, reported in chapter 7 are extended and completed in chapter 8.

As mentioned in chapter 8, there is some discrepancy between the findings reported in this thesis and those of Diamond, Epstein and co-workers from the Brady urological institute in Baltimore [35, 36]. They did find a strong correlation between a morphometrically estimated variation in nuclear shape factor, which they called nuclear roundness factor, and the

136

prognosis in patients with localized prostate cancer. Also Paulson and associates [37] were not able to use the nuclear roundness factor as a predictor for the prognosis. This discrepancy may have several reasons. In the first place the digitization systems available may show variations in accuracy. In 1988 Mohler and co-workers [38] reported on their work on testing the digitizing system they used. They found that indeed inaccuracies could be attribuated to the equipment, but these were about 5% of the actual area or perimeter of the tested figure. Inaccuracies mainly occurred when digitizing was performed at the extreme periphery of the microscopic field. When measuring in the periphery of the microscopic field was avoided, a good accuracy could be obtained.

In the second place intra- and interobserver variations may play a role. In chapter 8 it was already demonstrated that the intra-observer variation had no significant impact on the results. The inter-observer variation was tested by a small test (chapter 8) and seemed to be rather poor. Mohler and co-workers, however, found no significant differences between several investigators, digitizing several figures with geometrical shapes. Thus, intra- and inter-observer variation do not seem to play an important role in the discrepancy.

In the third place the selection of histological sections may play are role. Mohler and co-workers [39] compared the effect of selection of certain areas within a prostate tumor with random digitizing without prior selection. They showed that there was really no significant difference between the two ways of selection. However, they concluded that the system of nuclear roundness measuring is still not recommended for routine clinical use. The work is time consuming and laborious. However, the tools provided by quantitative morphometry may allow a more precise description of nuclear morphology and other malignant characteristics.

9.4.7 *Achievements of the present studies*

In this thesis several aspects on grading have been added to try to solve the problems dicussed in this chapter. A simplification of Mostofi's grading

system without any loss of prognostic information has been proposed and worked out on a large group of patients with long-term follow-up after total prostatectomy. This simplification has led to a higher accuracy in grading of prostate cancer and to a better reproducibility, as shown by Gallee and co-workers [28] and Ten Kate and co-workers [27]. An attempt has been made to objectivate grading by means of morphometry. Although morphometry has to be improved and preferably completely autoimized, it has been shown that morphometry is a useful tool in grading of prostate cancer. The study on grading of prostate cancer in patients with incidental carcinomas has produced a better insight in this complex stage of disease and has clearly identified the patients with a good and those with a poor prognosis.

9.5 Future expectations

Efforts to provide the clinician with prognostic information on prostate cancer to select the appropriate therapy have to go on continuously. These efforts are mainly hampered by the fact that therapeutic decisions have to be made on little biopsies of the prostate and on investigations whose accuracies have to be improved. Fine needle aspiration cytology allows the investigator to take more samples of the prostate than with thick needle biopsies. In this way it will be possible to take samples from a larger area of the prostate. Morphometry can help to improve grading further, even in cytologic smear preparations, but nowadays the technique of morphometry is still time consuming. With the development of automatic morphometry equipments it may be possible to solve this problem.

DNA ploidy measurements may also be of help to establish the prognosis in patients with prostate cancer. It has been shown, amongst others, by Myers and co-workers [40] that there is a strong correlation with the tumor cell ploidy and survival in patients with lymph node metastases. Future efforts to improve the prediction of the prognosis must be directed at a combination of parameters that can be obtained in a reliable and reproducible way, without harm to the patient.

9.6 References

1. Bruce AW, O'Cleireachain F, Morales A, Awad SA: Carcinoma of the prostate: a critical look at staging. J Urol 117: 319-322, 1977

2. Catalona WJ, Scott WW: Carcinoma of the prostate: a review. J Urol 119: 1-8, 1978

3. Brendler CB, Isaacs JT, Follansbee AL, Walsh PC: Th use of multiple variables to predict response to endocrine therapy in carcinoma of the prostate: a preliminary report. J Urol 131: 694-700, 1984

4. Wilson DW, Harper ME, Jensen HM, Ikeda RM, Richards G, Peeling WB, Pierrepoint CG, Griffiths K: A prognostic index for the clinical management of patients with advanced prostatic cancer: a British prostate study group investigation. The Prostate 7: 131-141, 1985

5. Emrich LJ, Priore RL, Murphy GP, Brady MF, and the investigators of the National Prostatic Cancer Project: Prognostic factors in patients with advanced stage prostate cancer. Cancer Research 45: 5173-5179, 1985

6. Johansson JE, Andersson SO, Krusemo UB, Adami HO, Bergström R, Kraaz W: Natural history of localised prostatic cancer. The Lancet 799-803, 1989

7. De Voogt HJ, Suciu S, Sylvester R, Pavone-Macaluso M, Smith PH, De Pauw M, and members of the European Organization for Research on Treatment of Cancer genitourinary tract cancer cooperative group: Multivariate analysis of prognostic factors in patients with advanced prostatic cancer: results from 2 European Organization for Research on Treatment of Cancer trials. J Urol 141: 883-888, 1989

8. Whitmore WF: Natural history and staging of prostate cancer. Urol Clin N Am 11: 205-220, 1984

9. Catalona WJ, Scott WW: Carcinoma of the prostate. In: Walsh, Gittes, Perlmutter, Stamey (eds): "Campbell's Urology", Ed 5. Philadelphia, WB Saunders Co, 1463-1534, 1986

10. Montgomery TR, Whitlock GF, Nohlgren JE, Lewis AM: What becomes of the patient with latent or occult carcinoma of the

prostate. J Urol 86: 655-658, 1961

11. Hanash KA, Utz SDC, Cook N, Taylor WF, Titus JL: Carcinoma of the prostate: a 15-year follow-up. J Urol 107: 450-453, 1972

12. Battaglia S, Barbolino G, Botticelli AR: Early (Stage A) prostatic cancer. Virchows Arch Pathol Anat 382: 245-259, 1979

13. Cantrell BB, DeKlerk DP, Eggleston JC, Boitnott JK, Walsh PC: Pathological factors that influence prognosis in stage A prostatic cancer: the influence of extent versus grade. J Urol 125: 516-520, 1981

14. Catalona WJ, Miller DR, Kavoussi LR: Intermediate-term survival results in clinically understaged prostate cancer patients following radical prostatectomy. J Urol 140: 540-543, 1988

15. Carter GE, Lieskovsky G, Skinner DG, Petrovich Z. Results of local and/or systemic adjuvant therapy in the management of pathological stage C or D1 prostate cancer following radical prostatectomy. J Urol 142: 1266-1271, 1989

16. Van den Ouden D, van Kooten C, Boevé E, Schröder FH: Positive margins after radical prostatectomy - correlation with local recurrence and distant progression. Abstract, submitted for the Annual meeting of the AUA, 1991

17. Gallee MPW: Prognostic benefits of histological grading and immunohistochemistry in human prostatic carcinoma. Thesis, Rotterdam, 1989

18. Murphy GP, Whitmore WF: A report of the workshops on the current status of the histologic grading of prostate cancer. Cancer 44: 1490-1494, 1979

19. Kastendieck H: Morphologie des Prostatacarcinoms in Stanz-biopsien und totalen Prostatektomien. Untersuchungen zur Frage der Relevanz bioptischer Befundaussagen. Pathologe 2: 31-43, 1980

20. Ackermann R, Müller H-A: Der Wert der perinealen Stanzbiopsie zur Beurteilung des histologischen Differenzierungsgrades des Prostata-Carcinoms. Verhandl. Ber. der Deutsch. Gesellsch. für Urol. 32: 79-81, 1980

21. De Voogt HJ: Cytology of the prostate.In: M. Pavone-Macaluso and P.H. Smith (eds.). Cancer of the prostate and Kidney. Plenum Press, New York and London, pp 73-76,1981

22. Maksem JA, Johenning PW: Is cytology capable of adequately grading prostate carcinoma? Urology 31: 437-444, 1988

23. Graham JB, Ignatoff JM, Holland JM, Christ ML: Prostatic aspiration biopsy: an assessment of accuracy based on long-term observations. J Urol 139: 971-974, 1988

24. Layfield LJ, Mukamel E, Hilborne LH, Hannah JB, Glasgow BJ, Ljung BM, deKernion JB: Cytological grading of prostatic aspiration biopsy: a comparison with the Gleason grading system. J Urol 138: 798-800, 1987

25. Mostofi FK: Problems of grading carcinoma of prostate. Semin in Oncol 3: 161-169, 1976

26. Harada M, Mostofi FK, Corle DK, Byar DP Trump BF: Preliminary studies of histologic prognosis in cancer of the prostate. Cancer Treatm Rep 61: 223- 225, 1977

27. Ten Kate FJW, Gallee MPW, Schmitz PIM, Joebsis AC, van der Heul RO, Prins MEF, Blom JHM: Problems in grading of prostatic carcinoma: interobserver reproducibility of five different grading systems. World J Urol 4: 147-152, 1986

28. Gallee MPW, Ten Kate FJW, Mulder PGH, Blom JHM, van der Heul RO: Histological grading of prostatic carcinoma in prostatectomy specimens. Comparison of prognostic accuracy of five grading systems. Brit J Urol 65: 368-375, 1990

29. Gleason DF: Classification of prostatic carcinomas. Cancer Chemother Rep 50: 125, 1966

30. Gleason DF and the VACURG: Histologic gradin and clinical staging of prostatic carcinoma.In: Tannenbaum M (ed.) Urologic Pathology: The Prostate, Lea & Febiger, Philadelphia, pp 171-197, 1977

31. Yatani R, Shiraishi T, Akazaki K, Hayashi T, Heilbrun LK, Stemmermann GN: Incidental prostatic carcinoma: morphometry correlated with histological grade. Virch Arch Pathol Anat 409: 395-405, 1986

32. Stamey TA, McNeal JE, Freiha FS, Redwine E: Morphometric and clinical studies on 68 consecutive radical prostatectomies. J Urol 139: 1235-1241, 1988

33. Meijer CJLM: Cutaneous lymphomas: the importance of morphometry in the diagnosis of mycosis fungoides and Sezary's syndrome. In: Baak JPA and Oort J (eds.) A manual of morphometry in diagnostic pathology. Springer-Verlag, Berlin, Heidelberg, New York, Tokyo, pp 92-95, 1983

34. Baak JPA, Oort J: Breast cancer: prediction of the prognosis. In: Baak JPA and Oort J (eds.) A manual of morphometry in diagnostic pathology. Springer-Verlag, Berlin, Heidelberg, New York, Tokyo, pp 92-95, 1983

35. Diamond DA, Berry SJ, Jewett HJ, Eggleston JC, Coffey DS: A new method to assess metastatic potential of human prostate cancer: relative nuclear roundness. J Urol 128: 729-734, 1982

36. Epstein JI, Berry SJ, Eggleston JC: Nuclear roundness factor: a predictor of progression in untreated stage A2 prostate cancer. Cancer 54: 1666-1671, 1984

37. Paulson DF, Stone AR, Walther PJ, Tucker JA, Cox EB: radical prostatectomy: anatomical predictors of success or failure. J Urol 136: 1041-1043, 1986

38. Mohler JL, Partin AW, Lohr DW, Coffey DS: Nuclear roundness factor measurement for assessment of prognosis of patients with prostatic carcinoma. I. Testing of a digitization system. J Urol 139: 1080-1084, 1988

39. Mohler JL, Partin AW, Epstein JI, Lohr DW, Coffey DS: Nuclear roundness factor measurement for assessment of prognosis of patients with prostatic carcinoma. II. Standardization of methodology for histologic sections. J Urol 139: 1085-1090, 1988

40. Myers RP, Oesterling JE, Zincke H, Lieber MM: Critical association of DNA ploidy and prognosis of stage D1 prostate cancer patients treated with radical prostatectomy and early or delayed endocrine therapy: results of long term follow-up. Abstract 712, presented at the 85th annual meeting of the American Urological Association, New Orleans, 1990

Summary

The natural course of prostate cancer can be very variable. On the one hand there are men without any urologic complaint in whom incidentally a prostate cancer is discovered. Especially in the elderly men prostate cancer can be detected incidentally during autopsy. On the other hand prostate cancer can grow very quickly and give rise to metastases in a short period of time, leading to the death of the patient. During many years doctors, involved in the treatment of cancer, try to identify parameters in the patient or in his tumor, that can give information on the expected course of the disease. In almost all malignant tumors, and certainly in prostate cancer, the histological picture of the tumor can be very variable. The microscopic picture can show a strong resemblance with normal prostate tissue, but it is also possible that not any resemblance with normal prostate tissue can be detected. Since the early twenties investigators have tried to correlate the findings at the microscopic examination of prostate carcinoma tissue with the course of the disease. The description of the variations of the histological and cytological picture of a tumor is called "grading". Grading of prostate cancer produces important prognostic information. It helps the urologist to make a therapeutic decision and it allows him to make predictions on the expected course of the disease and the possible effect of treatment.

In chapters 1 and 2 the purposes of the present investigations are depicted. In chapter 1 the group of patients, whose tumors have been used for the grading studies is depicted. All the patients underwent a total perineal prostatectomy for prostate cancer, which at clinical examination seemed to be confined to the prostate. For various reasons a number of these patients was treated after operation with hormonal therapy, either estrogen therapy or castration. This adjuvant hormonal therapy however, did not improve survival. It was shown that a number of clinically prognostic parameters was important in this group of patients. Clinical tumor stage has significant correlation with survival. Patients with a tumor, confined to the prostate have a better prognosis than patients whose tumors have

grown beyond the confines of the prostate. However, microscopic penetration of tumor through the capsule of the prostate does not influence the prognosis in a significant way.

Another prognostic parameter is tumor grade. Patients with a well differentiated tumor have a better prognosis than those whose tumor is poorly differentiated.

In chapter 2 the history of grading is discussed. Since 1920 several investigators try to make a as accurate as possible estimate of the prognosis based on the findings at microscopic examination of the tumor. Several histological and cytological parameters and also their combinations were designated as prognostically important. Many grading systems were not applicable very well, because of lack of reproducibility, and because of the heterogenity of the tumor. In the United States mainly Gleason's grading system is used. In this grading system the histological architecture of a dominant tumor pattern is evaluated, combined with a so-called secondary tumor pattern. In Europe the grading systems of Broders and Mostofi are used as well as Gleason's system. In Mostofi's grading system histological and cytological parameters are taken into account and they form the basis of a total tumor grade. The Mostofi system has been used for regrading of all the tumors of the patients in this thesis. Each of the parameters in Mostofi's grading system was estimated separately in each of the tumor patterns or tumor formations present in the tumor.

Patients with a so-called incidental carcinoma - a carcinoma found incidentally during an operation for presumed benign disease - generally have a better prognosis than patients whose tumors are detected by palpation of the prostate. Nevertheless, the group of patients with an incidental prostate carcinoma can be subdivided in patients with a good prognosis and with a poor prognosis. In chapter 3 a number of parameters for identification of each of these patients could be identified. It was shown that a small amount of tumor, the presence of small, intermediate, or large glands, but the absence of cribriform and solid growth patterns, the presence of slight variation in nuclear size and shape, and grade 1 are

144

parameters that correlate with a 100% chance of not dying from prostate cancer. These patients do not need any further treatment.

In chapters 4, 5, and 6 the 11 several parameters of Mostofi's grading system are studied separately. The final grade, estimated by Mostofi after studying each of the 11 parameters, was studied in this thesis as a separate parameter. The relation with the prognosis was estimated. It was shown that among these 11 parameters only a few had a real prognostic value: architecture of the tumor, nuclear variation in size and shape, grade, the amount of tumor in the slides, and the presence of mitoses. With these parameters a simplification of Mostofi's grading system could be obtained. In chapter 5 the presence of more than one tumor pattern or tumor formation was investigated. It is shown that the poor prognosis, indicated by the presence of a poorly differentiated tumor, improves when higher differentiated tumor formations are present together with the poorly differentiated parts.

In chapter 6 it is shown that with the three most important grading parameters: tumor architecture, nucear variation in size and shape, and the presence of mitoses, a scoring system could be created, in which five separate prognostically different groups of patients could be identified.

In chapters 7 and 8 it is shown how, by means of morphometry the nucear variation in size and shape can be estimated objectively. It is shown that the objectively estimated variation in nuclear size has a good correlation with survival. It was not possible to find any correlation of the objectively estimated variation in nuclear shape with the clinical outcome of the disease.

It is shown that it is posible to introduce objectivity in grading. Further efforts in development and automatizing of the system of morphometry is needed.

Samenvatting

Het natuurlijk beloop van het prostaatcarcinoom kan zeer wisselend zijn. Enerzijds zijn er mannen bij wie bij toeval een prostaatcarcinoom wordt ontdekt, zonder dat zij daarvan klachten hebben. Vaak, met name bij oude mannen, wordt het prostaatcarcinoom ontdekt tijdens autopsie zonder dat er klachten van de urinewegen waren. Anderzijds kan het prostaatcarcinoom snel progressief groeien en in korte tijd aanleiding geven tot metastasen, die tenslotte tot het overlijden van de patient leiden. Al tientallen jaren trachten artsen, die bij de behandeling van kanker zijn betrokken, bij de patient of diens tumor kenmerken, die informatie geven over het te verwachten ziektebeloop te identificeren. Bij vrijwel alle kwaadaardige tumoren, en zeker bij het prostaatcarcinoom, kan het histologisch beeld zeer variabel zijn. Enerzijds kan het microscopisch beeld een sterke gelijkenis tonen met dat van normaal prostaatweefsel, anderzijds kan het beeld zo bizar en grillig zijn dat er geen enkele gelijkenis met prostaatwaafsel meer bestaat. Al sinds de twintiger jaren is getracht een verband te leggen tussen de bevindingen bij het microscopisch onderzoek van prostaatweefsel en het beloop van de ziekte. Het beschrijven van de variaties in het histologische beeld van de tumor heet graderen. De gradering van het prostaatcarcinoom geeft belangrijke prognostische informatie. Het helpt de uroloog bij het bepalen van het therapeutische beleid en het staat hem toe uitspraken te doen omtrent de verwachtingen van de ziekte en het mogelijke effect van de behandeling hierop.

In hoofdstuk 1 en 2 worden de doelstellingen van het huidige onderzoek beschreven. Hoofdstuk 1 beschrijft de groep van patienten, wier tumoren bij het graderingsonderzoek zijn gebruikt. Het zijn allen patienten die wegens een prostaatcarcinoom, dat bij klinisch onderzoek tot de prostaat beperkt leek, een totale perineale prostatectomie ondergingen. Om verschillende redenen werd een aantal van deze patienten tevens met hormonale therapie, in de zin van oestrogenen of castratie, behandeld. Deze adjuvante hormonale therapie verbeterde de overleving echter niet.

146

Een aantal klinische prognostische parameters blijkt bij deze patienten groep belangrijk te zijn. Het klinische tumorstadium heeft een duidelijke correlatie met de overleving. Patienten met een tumor, die tot de prostaat beperkt is hebben een betere prognose dan patienten wier tumoren door de prostaatkapsel zijn heengegroeid. Echter, microscopisch kleine doorgroei door de prostaatkapsel beïnvloedt de prognose niet significant. De maligniteitsgraad van de tumor blijkt ook bij deze patienten een prognostische parameter te zijn. Patienten met een goed gedifferentiëerde tumor hebben een betere prognose dan die met een slecht gedifferentiëerde tumor.

In hoofdstuk 2 wordt de geschiedenis van het graderen behandeld. Vanaf de twintiger jaren trachten vele onderzoekers een zo nauwkeurig mogelijke schatting van de prognose te maken aan de hand van de bevindingen bij het microscopisch onderzoek van de tumor. Verschillende histologische en cytologische parameters en ook combinaties hiervan werden in de verschillende graderingssystemen als prognostisch belangrijk bestempeld. De meeste graderingssystemen bleken door gebrek aan reproduceerbaarheid en door de heterogeniteit van het prostaatcarcinoom niet goed bruikbaar. In de Verenigde Staten wordt vooral het graderingssysteem van Gleason gebruikt. Dit graderingssysteem beschouwt de histologische gradering van dat tumor patroon dat in de prostaat het meest voorkomt, naast dat patroon dat als tweede in de prostaat voorkomt. In Europa worden naast het systeem van Gleason de systemen van Broders en van Mostofi vaak gebruikt. In het systeem van Mostofi worden histologische en cytologische parameters elk beschouwd en vormen de basis van een totale" tumorgraad". Dit systeem is gebruikt bij het (her)graderen van de tumoren van de patientengroep in dit proefschrift. Elk van de parameters in Mostofi's graderingssysteem werd apart benoemd voor elk van de tumorpatronen die in een prostaattumor voorkwamen.

Patienten met een zogenaamd incidenteel carcinoom -een carcinoom dat bij toeval wordt ontdekt tijdens een operatie voor een vermeende

goedaardige prostaatvergroting- hebben in het algemeen een gunstiger prognose dan patienten met een tumor die bij palpatie van de prostaat werd ontdekt. Niettemin kan de groep van patienten met een incidenteel prostaatcarcinoom worden onderverdeeld in patienten met een gunstige prognose en patienten met een ongunstige prognose. In hoofdstuk 3 kon een aantal parameters voor het onderverdelen van deze patientengroep worden geïdentificeerd. Het bleek dat een kleine hoeveelheid tumor, de aanwezigheid van kleine tot grote klierbuisjes, maar de afwezigheid van een cribriforme of solide groeiwijze, de aanwezigheid van geringe variatie in kernvorm en -grootte en maligniteitsgraad 1 parameters zijn die gepaard gaan met een 100% kans niet aan prostaatcarcinoom te overlijden. Deze patient behoeven geen verdere behandeling.

In hoofdstuk 4, 5 en 6 worden de 11 verschillende parameters, waaruit Mostofi's graderingssysteem is opgebouwd, apart beschouwd. Bovendien werd de uiteindelijke maligniteitsgraad, door Mostofi vastgesteld na beschouwing van elk van de andere parameters, als een aparte "parameter" meebeoordeeld. Hun relatie tot de prognose werd vastgesteld. Hierbij bleek dat slechts enkele parameters een prognostische waarde toonden: de architectuur van de tumor, de variatie in vorm en grootte van de kern, de totale maligniteitsgraad, de hoeveelheid tumor in de coupes en de aanwezigheid van mitosen. Hiermee kon een sterke vereenvoudiging van Mostofi's graderingssysteem worden verkregen. In hoofdstuk 5 wordt de aanwezigheid van meer dan één tumorpatroon of tumorformatie onderzocht. Het blijkt dat de slechte prognose door de aanwezigheid van een slecht gedifferentiëerde tumor minder slecht is wanneer naast een slecht gedifferentiëerde formatie ook beter gedifferentiëerde formaties aanwezig zijn.

Hoofdstuk 6 toont dat met de drie meest belangrijke graderingsparameters: tumor architectuur, variatie in kernvorm en -grootte en de aanwezigheid van mitoses een scoringssysteem kan worden gemaakt, waarin vijf afzonderlijke prognostisch verschillende groepen kunnen worden geïdentificeerd.

In hoofdstuk 7 en 8 wordt beschreven hoe door middel van morfometrie de belangrijke parameter variatie in kernvorm en -grootte exact kan worden bepaald. Het blijkt dat de objectief bepaalde variatie in kerngrootte een goede correlatie met de overleving toont. Met de objectief bepaalde variatie in kernvorm kon dat niet worden vastgesteld.

Het blijkt mogelijk om een objectieve bepaling bij het graderen te gebruiken, maar deze behoeft verdere ontwikkeling en automatizering.

Nawoord

Aan de totstandkoming van dit proefschrift hebben vele mensen hun medewerking verleend. Een aantal van hen wil ik hier in het bijzonder noemen.

Fritz Schröder had mijn interesse voor dit onderzoek gewekt, al voordat ik aan mijn urologische opleiding was begonnen. Gezamenlijk hebben we de in het proefschrift behandelde vraagstelling geformuleerd en hebben we het onderzoek op gang gebracht. Al die jaren bleef hij geïnteresseerd in en bezig met het probleem van het bepalen van een exacte en reproduceerbare gradering van het prostaatcarcinoom.

Fiebo ten Kate heeft mij vanaf het begin bijgestaan bij het moeizame werk van de morfometrie en het graderen. Ondanks zijn drukke taken had hij altijd wel gelegenheid om de materie met mij te overdenken en bespreken.

Prof. Dr. R.O. van der Heul heeft mij zeer geholpen, zowel tijdens het werk van de morfometrie als bij zijn kritische beoordeling van het manuscript.

Dr. F.K. Mostofi was zo vriendelijk om mij de techniek van het graderen van het prostaatcarcinoom bij te brengen.

Prof. Dr. O. Vos en Prof. Dr. R.A. Janknegt dank ik vriendelijk voor het kritisch beoordelen van het manuscript.

Linda van Kesteren was zo bereidwillig het manuscript kritisch door te lezen en mijn engelse taalfouten te verbeteren.

Door nog harder te werken hebben mijn collegae urologen en assistent-urologen mij regelmatig in de gelegenheid gesteld meer tijd vrij te maken voor het werk aan dit onderzoek.

Tenslotte Arina en de kinderen. Vele dagen en avonden hebben zij tevergeefs gehoopt dat ik me meer met mijn gezin kon bezighouden. Ik dank hen hartelijk voor het geduld dat zij altijd hebben opgebracht.

Curriculum vitae

De schrijver van dit proefschrift behaalde in 1968 zijn diploma gymnasium-β te Delft (St. Stanislacollege). Aansluitend studeerde hij geneeskunde aan de Erasmus Universiteit Rotterdam, waar hij in 1975 het artsexamen haalde. Na zijn militaire dienst startte hij in oktober 1976 met de chirurgische vooropleiding ten behoeve van de specialisatie urologie op de afdeling heelkunde van het Ikazia Ziekenhuis te Rotterdam (opleider Dr. A.P. Brinkhorst). Van oktober 1979 tot oktober 1980 werkte hij gedurende een jaar op de afdeling pathologische anatomie I (Prof. Dr. R.O. van der Heul), alwaar een aanvang werd gemaakt met het in dit proefschrift beschreven onderzoek. De opleiding tot uroloog werd gestart in oktober 1980 op de afdeling urologie van het Academisch Ziekenhuis te Rotterdam (opleider Prof. Dr. F.H. Schröder) en op de afdeling kinderurologie van het Sophia Kinderziekenhuis te Rotterdam (opleider Prof. Dr. R.J. Scholtmeijer). In 1983 volgde hij een het perifere gedeelte van de urologische opleiding in het Bergwegziekenhuis te Rotterdam (opleider W.H. Hirdes). Op 1 oktober 1983 volgde inschrijving in het specialisten-register.

Vanaf 1984 is hij werkzaam als chef de clinique op de afdeling Urologie in het Academisch Ziekenhuis Dijkzigt, alwaar het klinisch gedeelte van het in dit proefschrift beschreven onderzoek werd verricht.