Oestrogen and Progesterone Receptor Expression in Patients with Adenocarcinoma of the Uterine Cervix and Correlation with Various Clinicopathological Parameters

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Abstract. *Objective: The expression of oestrogen and progesterone receptors in patients with adenocarcinoma of the uterine cervix was examined in order to determine their influence on prognosis and to evaluate the association between the steroid receptor expression and various clinicopathologic parameters. Patients and Methods: Oestrogen and progesterone receptor expression was investigated by immunohistochemistry from paraffin-embedded tissue in 39 patients with adenocarcinoma of the uterine cervix. The immunohistochemical findings were correlated with various clinicopathological parameters of the patients. Results: Oestrogen and progesterone receptors were expressed in 39% and 33% of the patients, respectively. The relationship between oestrogen and progesterone receptor expression and clinical stage, age, histology, tumour size, grade, lymph-vascular space invasion and lymph node status did not reach statistical significance (p>0.05). Neither oestrogen nor progesterone receptor expression significantly influenced disease-free and overall survival (p>0.05). Conclusion: Oestrogen and progesterone receptors were frequently expressed in adenocarcinoma of the uterine cervix. However, their expression did not correlate with clinicopathological parameters and had no influence on overall and disease-free survival. Thus, the investigation of steroid receptors adds little additional information to the clinical management and fails to play a prognostic role in cervical adenocarcinoma.*

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Adenocarcinoma of the uterine cervix is the second most common histological subtype representing up to 20% of cervical carcinomas. International Federation of Gynaecology and Obstetrics (FIGO) stage, histological grade, tumour size, lymph-vascular space invasion and lymph node metastasis are major prognostic factors reported in literature (1-3). For other gynaecological malignancies, such as breast and endometrial cancer, the importance of hormonal receptor expression is well established and guides patient treatment. A poor prognosis without response to hormone therapy is observed in the absence or low concentrations of oestrogen and progesterone receptors (4, 5).

The uterine cervix has also been shown to be a target tissue, with the cervical mucus underlying the hormone dependent cyclical changes related to the menstrual cycle regarding both, the composition and its quantity (6-8). Thus, it may be possible that the growth of adenocarcinoma of the uterine cervix is to some extent also controlled by ovarian steroids, and hormonal manipulation could offer an additional treatment option in case of steroid receptor positivity. To date, the few reports that have evaluated the expression of oestrogen and progesterone receptors in cervical adenocarcinoma have shown inconsistent results (9-13).

Thus, the aim of the present study was to investigate the expression of oestrogen and progesterone receptors in patients with adenocarcinoma of the uterine cervix and to assess their prognostic value. Furthermore, the study aimed to determine the correlation between steroid receptor expression and various clinicopathological parameters.

Patients and Methods

Tissue collection. This retrospective study included 39 patients with histologically proven adenocarcinoma of the uterine cervix, treated between 1994 and 2006 at the Department of Gynaecology and Obstetrics of the University Hospital Vienna. All patients were staged retrospectively, according to the FIGO staging system for cervical cancer.

Serial sections were prepared for hematoxylin and eosin staining and immunohistochemistry. All slides were reviewed by an experienced pathologist. Clinical information, including follow-up data, was obtained from the database of the Department of Gynaecology and Obstetrics.

Immunohistochemistry. Immunohistochemical staining for oestrogen receptor (ER) and progesterone receptor (PR) was performed on formalin-fixed and paraffin-embedded sections using the avidin-biotin-immunoperoxidase complex method. A mouse monoclonal antibody to human oestrogen receptor (ER1D5, dilution 1:50; Dako, California) and to human progesterone receptor (PGR-1A6, dilution 1:40; BioGenex, California) was used as the primary antibody.

In brief, the sections were deparaffinised in xylene, rehydrated through graded alcohols and treated in a 0.01 M citrate buffer for 15 min (pH 6) in a microwave oven. They were then incubated with 0.03% hydrogen peroxide to block endogenous peroxidase acivity, and with normal goat serum to reduce non-specific binding. The sections were incubated with the specific primary antibodies or control nonimmunised mouse serum at 4˚C overnight. Biotinylated goat anti-mouse immunoglobulin G was used as a linker. After washing, the streptavidin-biotin complex was applied and stained with diaminobenzidine. The sections were lightly counterstained with hematoxylin. Specific staining was identified by a brown color of the nucleus. All control slides yielded negative results.

The staining of ER and PR was evaluated as the percentage of stained cells: – negative (fewer than 10% were stained), + weak positive (11-50% of the cells were stained), ++ moderate positive (51-80% of the cells were stained), +++ strong positive (more than 80% of the tumour cells were stained).

Statistical analysis. Correlation between clinicopathologic parameters and oestrogen/progesterone receptor expression was tested using the Spearman correlation coefficient. The end point of overall survival was used for analysis. Survival probabilities were calculated by the product limit method of Kaplan and Meier (14). Univariate analysis was performed using the log-rank test. *P*-values of less than 0.05 were considered statistically significant. The Statistical Package for the Social Sciences system (SPSS Inc., Chicago, IL, USA) was used for the calculations.

Results

Clinical and pathological findings. A total of 39 patients with adenocarcinoma of the uterine cervix were enrolled in this study. The median follow-up time was 24 months (range: 2-123 months). The median age at diagnosis was 51 years (range: 24-60 years). Patient characteristics are shown in Table I.

Seventeen (44%) patients had stage I, 15 (38%) patients stage II, 6 (15%) patients stage III and 1 (3%) stage IV disease.

Table I. *Patient characteristics*.

	No. of cases	% of cases
Clinical stage		
1	17	44
2	15	38
3	6	15
4	1	3
Histology		
mucinous	22	56
endometrioid	16	41
intestinal	1	3
Tumour size		
<3 cm	23	59
>3 cm	16	41
Grading		
1	14	36
2	13	33
3	12	31
Lymph-vascular space invasion		
No	21	54
Yes	18	46
Lymph nodes		
Negative	24	62
Positive	15	38
Adjuvant therapy		
Yes	22	56
RT	19	49
CHT	15	38
No therapy	17	44
Recurrence disease		
Yes	11	28
No	28	72

Surgical treatment comprised radical hysterectomy with pelvic lymphonodectomy in 23 (59%) patients. Eight (21%) patients underwent conisation, 6 (15%) patients had pelvic lymph node staging and 2 (5%) patients had a radical hysterectomy with bowel resection and lymphadenectomy.

In total 22 (56%) patients received adjuvant treatment. Among those, 19 (86%) patients received postoperative radiotherapy (consisting of brachytherapy in 15 cases and combined external–intracavitary irradiation in 4 cases). Fifteen (68%) patients received paclitaxel/carboplatin-based chemotherapy.

Eleven (28%) patients showed recurrence of disease (pelvic recurrence occurred in 6 cases and distant metastasis to the liver and the lung in 2 and 3 cases, respectively). Eight (21%) patients died of the disease.

Expression of estrogen receptor (ER). ER was expressed in 15 (39%) patients. Staining intensity is presented in Table II. Table III summarises the relationship between various clinicopathologic parameters and ER expression in adenocarcinoma of the uterine cervix. No statistically

Table II. *Oestrogen receptor (ER) and progesterone receptor (PR) expression and staining intensity of tumour cells in patients with adenocarcinoma of the uterine cervix.*

	Adenocarcinoma of the cervix uteri	
	n=39	% of cases
ER expression		
Yes	15	39
No	24	61
Staining intensity of tumour cells for ER		
+	4	10
++	3	8
+++	8	21
PR expression		
Yes	13	33
No	26	67
Staining intensity of tumour cells for PR		
+	4	10
++	4	10
+++	5	13

The staining of ER and PR was evaluated to the percentage of stained tumour cells: – negative (fewer than 10% of the tumour cells were stained), + weak positive (11-50% of the cells were stained), ++ moderate positive (51-80% of the cells were stained), +++ strong positive (more than 80% of the cells were stained).

Table III. *Correlation between ER and various clinicopathological parameters.*

	ER		
	Negative n=24 (100%)	Positive n=15 (100%)	*p*-Value
Age (years)			0.39
<50	13 (54)	6 (40)	
>50	11 (46)	9 (60)	
Clinical stage			0.79
I	10 (42)	7 (47)	
II	10 (42)	5 (33)	
III	3 (12)	3 (20)	
IV	1 (4)	0 (0)	
Histology			0.30
Mucinous	15 (63)	7 (47)	
Endometrioid	8 (33)	8 (53)	
Intestinal	1 (4)	0 (0)	
Tumour size			0.37
<3 cm	16 (67)	7 (47)	
>3 cm	8 (33)	8 (53)	
Grading			0.15
G1	8 (33)	6 (40)	
G2	6 (25)	7 (47)	
G3	10 (42)	2 (13)	
Lymph vascular space invasion			0.96
No	13 (54)	8 (53)	
Yes	11 (46)	7 (47)	
Lymph node status			0.74
Negative	15 (63)	9 (60)	
Positive	9 (38)	6 (40)	
Recurrence			0.80
No	17 (71)	11 (73)	
Yes	7 (29)	4 (27)	

significant relationship was found between ER expression and clinical stage (p=0.79), age (p=0.39), histology (p=0.30), tumour size (p=0.37), grade (p=0.15), lymph-vascular space invasion (p=0.96), lymph node status (p=0.74) and recurrence of disease (p=0.80).

Expression of progesterone receptor (PR). PR was expressed in 13 (33%) patients with adenocarcinoma of the uterine cervix. Staining intensity is presented in Table II.

Table IV summarises the relationship between various clinicopathologic parameters and PR expression in adenocarcinoma of the uterine cervix. The relationship between PR and clinical stage (p=0.56), age (p=0.36), histology (p=0.67), tumour size (p=0.91), grade (p=0.61), lymph-vascular space invasion (p=1.0), lymph node status (p=0.36) and recurrence (p=0.47) did not reach statistical significance.

Survival analysis. The median overall survival of the 39 patients with adenocarcinoma of the uterine cervix was 121 months (95% CI: 18-224 months), resulting in an overall survival rate of 80%. The median disease-free survival was 36 months (95% CI: 14-58 months).

Neither the expression of ER nor the expression of PR had an influence on disease-free survival of the patients (p=0.09 and p=0.15, respectively). In the present study, clinical stage (p=0.009), age <50 years (p=0.05), lymph-vascular space invasion (p=0.004) and lymph node status (p=0.01) were factors associated with recurrence of disease.

Likewise, the expression of ER and PR had no influence on patient overall survival (p=0.17 and p=0.46, respectively). Clinical stage (p=0.04) and lymph-vascular space invasion (p=0.03) were prognostic factors associated with overall survival. Multivariate analysis could not be performed due to the small number of patients.

Discussion

The prognostic impact of steroid receptor expression has been clearly established in patients with breast and endometrial cancer, where it is used to prescribe and guide the treatment (4, 5). Previous studies have observed that steroid receptors are also present in normal, premalignant and malignant tissue of the uterine cervix (6, 7, 15, 16). In the literature, oestrogen and progesterone receptor expression rates in adenocarcinoma of the uterine cervix range from 25

Table IV. *Correlation between PR and various clinicopathological parameters.*

	PR		
	Negative n=26(100%)	Positive n=13(100%)	*p*-Value
Age (years)			0.36
<50	14 (54)	5 (38)	
>50	12 (46)	8 (62)	
Clinical stage			0.56
I	13 (50)	4 (31)	
II	9 (35)	6 (46)	
III	3 (11)	3 (23)	
IV	1 (4)	0 (0)	
Histology			0.67
Mucinous	15 (58)	7 (54)	
Endometrioid	10 (38)	6 (46)	
Intestinal	1 (4)	0 (0)	
Tumour size			0.91
<3 cm	16 (62)	7 (54)	
>3 cm	10 (38)	6 (46)	
Grading			0.61
G1	8 (30)	6 (46)	
G2	9 (35)	4 (31)	
G3	9 (35)	3 (23)	
Lymph-vascular space invasion			1.0
No	14 (54)	7 (54)	
Yes	12 (46)	6 (46)	
Lymph node status			0.36
Negative	17 (65)	7 (54)	
Positive	9 (35)	6 (46)	
Recurrence			0.47
No	20 (77)	8 (62)	
Yes	6 (23)	5 (38)	

to 65% for ER and from 19 to 71% for PR, respectively (9-13, 16). This is in line with the present study where oestrogen and progesterone receptor expression rates of 39% and 33% were detected, respectively.

Due to conflicting results in previous reports, the prognostic role of steroid receptor expression in cervical cancer has still to be defined (9-13, 16-19). Some previous studies reported that the hormonal receptor state influences patient survival (10-12, 16, 17). It is noteworthy that most of these investigations focused on the pattern of steroid receptor expression mainly in patients with squamous cell carcinomas (16-19). Until recently, only few authors specifically addressed the clinical impact of the expression of steroid hormone receptors in invasive adenocarcinomas of the uterine cervix (9-13). Masood *et al*. found that patients with positive ER or PR expression demonstrated significantly improved survival rates (10). Another study performed by Ghandour *et al*. observed that tumours containing ER showed improved disease-free survival rates (11). Suzuki *et al*. suggested that the progesterone receptor status was associated with prognosis after radiation therapy in patients with adenocarcinoma of the cervix (12). In the present study, oestrogen and progesterone receptor expression had no influence on disease-free and overall survival. These findings confirm the previously reported observations of other investigators (9).

In accordance with previous reports, the present study found that early clinical stage was significantly associated with a lengthened overall survival in patients with adenocarcinoma of the cervix (1-3). However, the presence of steroid receptors failed to correlate with patient clinical stage. Likewise, the data of previous studies confirmed a lack of association between steroid receptor status and clinical stage (9-11).

The prognostic impact of patient age is a controversial topic in the literature (1-3). In line with the literature, the present study recorded a lower recurrence rate in women aged below 50 years (1, 3). Gao *et al*. observed a significant correlation between pre- and post-menopausal state and hormonal receptor expression in cervical carcinoma (20). In accordance with most investigators (9-11, 13, 16), no significant correlation between steroid receptor expression and patient age was established in this study.

Some of the previous studies showed an inverse relationship between steroid receptor expression and tumor grade (9, 10). One may hypothesize that better differentiated tumours are more likely to express hormone receptors and that this feature is lost with tumour dedifferentiation. However, the present study failed to show any correlation between ER and PR expression and tumour grade. This is in line with the results of Ghandour *et al*. (11).

Supporting the data of previous research, the present study also showed that lymph-vascular space invasion and lymph node status were prognostic factors associated with recurrence of disease and overall survival in patients with adenocarcinoma of the uterine cervix (1-3). However, no significant correlation between steroid receptor expression and these parameters was found.

In conclusion, it was observed that oestrogen and progesterone receptors are expressed frequently in adenocarcinoma of the uterine cervix. However, their expression did not correlate with any clinicopathological parameters and had no influence on overall and disease-free survival. Despite the lack of prognostic impact in the present study, further clinical studies with larger numbers of cases should be performed to evaluate the prognostic value of steroid receptors and to verify whether oestrogen and progesterone receptor-positive tumours can be treated by hormonal manipulation. So far, the investigation of steroid receptors adds little additional information to the clinical management, and oestrogen and progesterone receptors fail to have a prognostic role in cervical adenocarcinoma.

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