Evaluation of the Pathologic and Prognostic Correlates of Estrogen Receptors in Primary Breast Cancer

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The presence of estrogen receptors in breast cancer tissue has been reported to correlate with improved prognosis in women after mastectomy. The prognostic value (if any) of the presence or absence of estrogen receptors (ER) in malignant breast tissue was evaluated in 104 women who were treated for primary breast cancer, whose pathology was re-examined, and whose records were subjected to multifactorial analysis. Sixty patients were ER positive, and 44 were ER negative, and a total of 94 who had curative resections were available for follow-up (mean follow-up time 20 months). The presence of estrogen receptors showed significant positive correlations with age, lobular cancer, and a variant of infiltrating duct cancer that is prevalent in the elderly and characterized by the presence of cells showing granular eosinophilic cytoplasm. Of 26 cases identified as infiltrating duct cancer showing granular eosinophilic cytoplasm, 22 were ER positive, one was ER negative, and three had borderline values. There was no significant difference between the groups with regard to family history of breast cancer or hysterectomy. A striking observation was noted in the ER positive group in which there were seven cases of second primary breast cancers, whereas no such cases occurred in the ER negative patients ($p = 0.05$). There was a higher percentage of nodal metastases in the patients who were ER positive compared with those who were ER negative; 27 of 53 (51%) of the ER positive patients had positive nodes compared with four of 40 (32%) who were ER negative, $p = 0.08$. There was no significant correlation of disease free survival nor time to recurrence in either the overall group nor according to stage. In patients whose tumors had been reviewed and graded, there was no prognostic relationship of ER status in high grade tumors, but in patients with low-grade tumors, improved disease-free survival was demonstrated in patients who were ER negative. Although the estrogen receptor assay is a highly useful tumor marker and guide for therapy of advanced breast cancer, its relationship to the prognostic variables of primary breast cancer is complex and controversial and merits continued study.

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ESTROGEN RECEPTOR ASSAY is now generally accepted as an aid in determining which patients with metastatic breast cancer may benefit from hormonal therapy.[1,2] In addition, recent studies by Walt,[3] Knight,[4] Rich,[5] and others[6–9] have suggested that the estrogen receptor assay may also have value in predicting time and site of recurrence as well as response to therapy. The data, however, is inconclusive and at times contradictory. The true value of estrogen receptor assay as a prognostic factor is uncertain at present. In order to investigate this question further the authors have reviewed their experience with estrogen receptor assays at the University of Virginia Medical Center.

Materials and Methods

The records of those patients having estrogen receptor assays performed for primary breast malignancy diagnosed at either the University of Virginia Hospital or the Martha Jefferson Hospital from 1975 to 1978 were reviewed. Detailed pathologic review was done, and lesions were categorized by grade and tissue type. The presence or absence of estrogen receptor was correlated with the following variables: 1) age; 2) menstrual status; 3) history of second breast cancer; 4) family history of breast cancer; 5) history of hysterectomy; 6) tumor grade; 7) nodal status; 8) stage; 9) recurrence; and 10) survival.

One hundred four patients were initially evaluated. Hospital deaths and those patients with known metastatic disease at the time of initial diagnosis or second primary malignancies (excluding those of the breast) were excluded from the follow-up study, leaving 94 evaluable patients. In all cases, patient information was obtained from chart review, contact with the referring physician, or direct patient contact. Tumors were graded

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TABLE 1. *Relation of Historic Data to Estrogen Receptor Status*

	ER+	ER−	Significance (P) ER+ vs. ER−
Total patients	60	44	—
Mean age	63.3	53.8	=0.003
Menstrual status	48 postmenstrual 9 menstrual 2 males 1 unknown	32 postmenstrual 9 menstrual 1 male 2 unknown	=NS
Family history of breast cancer	15	14	—
History of hysterectomy	17	14	—
Second breast cancer	7	0	0.05
Bilateral cancer at diagnosis	1	1	—
Lymph node status at diagnosis	26+ 27− 7 no L N biopsy	14+ 29− 1 no L N biopsy	0.57
Mean follow-up	19.9 mos.	18.8 mos.	—
Recurrence	6 pts.	5 pts.	—
Mean time to recurrence	12.8 mos. (1, 2, 3, 7, 16, 48 mos.)	11.8 mos. (2, 7, 9, 17, 24 mos.)	
Recurrence in pts with + L N.	5/26 (19.2%)	4/14 (28.6%)	p = NS

according to Bloom and Richardson's classification.[10] Patients were staged according to the American Joint Committee for Clinical Staging. To facilitate analysis, patients were allocated to low risk, Stage 1 (Stage 1 only), or high risk (Stages II and III) which was termed Stage II.

Estrogen receptor assay was performed in the Endocrine Laboratory at the University of Virginia Hospital (under the direction of Dr. Robert MacLeod), initially using the standard sucrose density gradient method and later the dextran-coated charcoal method.[11,12] A value > 7 fmol/mg of cytosol protein was considered positive.

Statistical Analysis

Data was analyzed by both parametric and nonparametric tests including chi square, Student's t-test, Mann Whitney U test, Gehan–Wilcoxon, analysis of variance, and multiple variable analysis where indicated. Dr. Donald Ramirez of the Department of Mathematics performed the statistical analysis using the SPSS computer programs provided by Vogelback Computing Center, Northwestern University.

Results

Relation to Historical Data

The overall data are given in Table 1. There was a difference in age according to estrogen receptor status. Sixty patients were ER+ and had a mean age of 63.3 years compared with a mean age of 53.8 years in 44 patients who were ER− (p = 0.003 T-test with pooled variance). Of patients ≥ 70 years old, 22 of 26 were ER+. Seventy-eight per cent of the ER+ patients were postmenopausal compared with 73% of the ER− patients. Of special interest, seven of 60 ER+ patients had a second primary breast cancer *vs.* none of 44 ER− patients. Otherwise, there were no differences in historic data according to estrogen receptor status, regarding family history for breast cancer, history of breast cancer, or bilaterality.

TABLE 2. *Correlation of Pathologic Findings With Estrogen Receptor Status*

	ER+	ER−	Significance* (P) ER+ vs. ER−
Tumor grade (routine histology)			p = 0.53
High grade	20	22	
Low grade	32	25	
Clinical pathologic stage			p = 0.35
I	27	26	
II & III	21	12	
IV	0	9	
Lymph node status nodal metastases			p = 0.57
+	26	14	
−	27	29	
Nodes not biopsied	7	1	

* Corrected Chi Square.

TABLE 3. *Tumor Grade and Histology (Prospective Review)*

Type	I	II	III
Infiltrating duct	0	24	24
Infiltrating lobular	0	5	4
In situ carcinoma	1	4	1
Infiltrating duct with prominent granular eosinophilic cytoplasm	4	14	2

Relation to Pathologic Data

There were no correlations of estrogen receptor status with grade (routine histology), stage, or nodal status (Table 2). The results of pathologic review including tumor grading and histologic type are listed in Table 3. Of note, there were 20 patients listed as having infiltrating duct cancer with prominent granular eosinophilic cytoplasm, a finding most commonly observed in elderly women.[13] There was a correlation of histologic type with receptor status (Table 4). Although infiltrating ductal cancers were evenly divided between ER+ and ER− tissues, infiltrating ductal carcinomas with prominent granular eosinophilic cytoplasm were almost all ER+. Of 11 infiltrating lobular cancers, nine were ER+. When the patient grades were regrouped into high or low grade, 48% of the low-grade tumors were ER+ *vs.* 41% of the high grade tumors. These differences were not significant (P > 0.10) (Table 5).

Relation of Estrogen Receptor Status to Recurrence and Survival

Analysis of disease-free survival according to estrogen receptor status in the overall group showed an apparently better survival in the estrogen receptor negative patients; however, there was no significant difference between the ER+ *vs.* ER− patients (Fig. 1). Examination of Stage I and II patients showed no significant difference between ER+ and ER− (Fig. 2). There was no difference according to estrogen receptor status in patients with higher stage tumors (III or IV). In patients whose tumors had been graded, those with low-grade ER− tumors had a significantly better survival than ER+ patients (p = 0.01) (Fig. 3). Among the patients who had high-grade tumors, there was no difference in disease-free survival in the ER+ *vs.* ER− patients (Fig. 4). The relationship of estrogen receptor status and disease-free survival or time to recurrence according to stage or grade is summarized in Table 6. When patients were classified into low risk or high risk and analyzed according to estrogen receptor status, there was no difference in disease-free survival or time to recurrence (data not shown).

Analysis of patients with axillary nodal metastases showed a similar recurrence rate according to the receptor status. Five of 20 patients in the ER+ group and four of 14 in the ER− group developed recurrence. The disease-free interval was similar in the overall group of ER+ and ER− patients who had recurrence (22.8 months *vs.* 19.1 mos), P = NS. The disease-free interval (DFI) in patients who had recurrence more than four months after operation was 28.4 months in the ER+ group *vs.* 16.8 months in the ER− group. (p = N.S.).

TABLE 4. *Histologic Type Vs. Estrogen Receptor Status*

	ER+	ER−	*ER±
Infiltrating duct	18	20	7
Intraductal	4	1	1
Infiltrating duct with prominent granular eosinophilic cytoplasm	22	1	3
Infiltrating lobular	9	1	1

* ≥7 fentimoles/mg protein.

Discussion

Although the estrogen receptor status of patients with advanced disease is highly correlated with the effects of hormone manipulation, the biologic relationships in patients with primary cancer are less defined. The estrogen receptor is more commonly positive in older women and in postmenopausal patients. The relationship of estrogen receptor positivity to the postmenopausal state was not significant in this study. There were many negative associations of the estrogen receptor assay: stage of disease, lymph nodes status, and tumor grade. Although a greater percentage of low-grade tumors were ER+ (30/52, 58%) *vs.* 41% in high-grade tumors, the differences were not significant (p > 0.10). There was a correlation of estrogen receptor positivity in patients with certain histologic type (infiltrating duct cancer with prominent granular eosinophilic cytoplasm). A prospective review of pathology showed a

TABLE 5. *(Prospective Review) Relation of Tumor Grade to Estrogen Receptor*

Grade		Pts	ER+	ER−	*ER±
Low	I	2	2	—	—
	I–II	11	8	3	—
	II	39	20	13	6
High	II	39	20	13	6
	II–III	24	9	12	3
	III	22	10	9	3
Regrouping of Pts "Low Grade" *vs.* "High Grade"					
Low grade			30	16	6
High grade			19	21	6

ER status—low grade vs. high grade
$X^2 = 2.78$ p = >0.10

* ≥7 fentimoles/mg protein.

slightly higher number of patients with low-grade tumors in the ER+ category compared with high-grade tumors, 58% *vs.* 41%, but this was not significant, $p \sim 0.10$.

Forty-nine per cent of the ER+ patients had positive lymph nodes at diagnosis compared with 32.5% for the ER− group ($p = 0.09$). Rich et al.[5] reported essentially equal percentages of ER+ tissue in patients with 0, one to three, and four or more positive lymph nodes at the time of primary mastectomy. It would appear that the estrogen receptor probably does not have a significant correlation with the presence or the extent of lymph node involvement.

Of interest, there was an association of ER positivity with infiltrating lobular cancer as reported by Rosen et al.[14] Also, breast carcinomas histologically characterized by prominent granular eosinophilic cytoplasm were more likely to be ER+. These histologic features are characteristic of breast carcinoma in the elderly. Patient historical data was also evaluated *vis-a-vis* estrogen receptor status. No significant correlation could be found between estrogen receptor status and family history of breast cancer, or history of hysterectomy as recorded in the routine admission workups.

A most interesting finding was the occurrence of seven cases of second breast cancer in the ER+ group, but no such patients were observed in the ER− group. Five of these seven cases were probably second primaries as judged by history and pathologic examination, and two cases were most likely second primaries, but metastatic disease could not be ruled out. There were two cases of simultaneous bilateral carcinoma at diagnosis, one in each group, with the case in the ER− group most probably metastatic and that in the ER+ group a bilateral lobular carcinoma. Although the numbers are small,

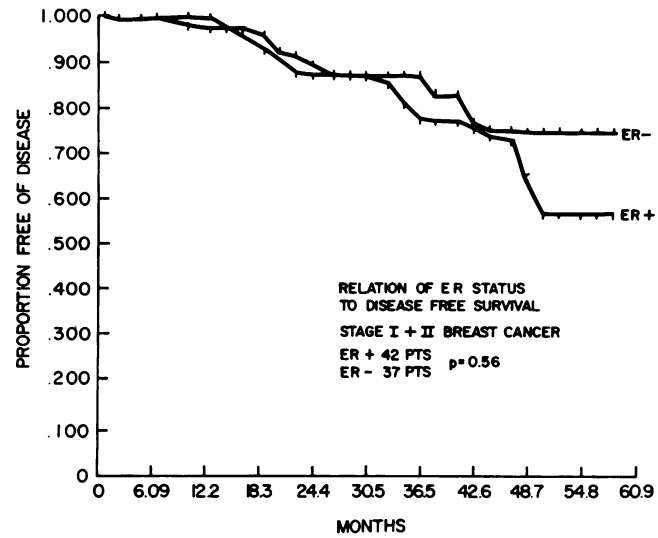

FIG. 2. In patients with stage I and II breast cancer, there was no difference in disease-free survival according to estrogen receptor status.

these findings approached statistical significance ($p = 0.09$), X^2 with Yates correlation. Cases of presumed second primaries were correlated with patient age to rule out the possibility of this being merely a function of the greater mean age of the ER+ group. There was no correlation of second cancer with age. To the authors' knowledge, there are no reports of positive estrogen receptors being associated with higher incidences of second breast primary tumors. This finding obviously needs further evaluation to determine whether the estrogen receptor status shows any correlation with primaries.

A major question centered on the issue of estrogen receptors as an independent prognostic factor. It has been stated that patients with receptor negative tumors

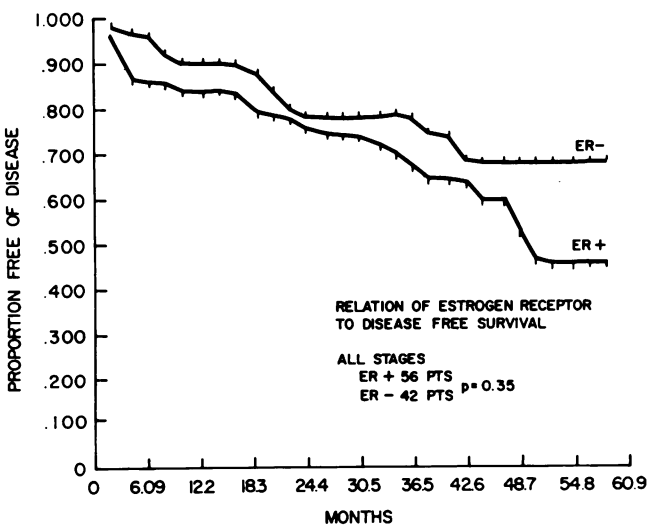

FIG. 1. There was no significant difference in disease-free survival nor in overall survival (not shown) according to estrogen receptor status.

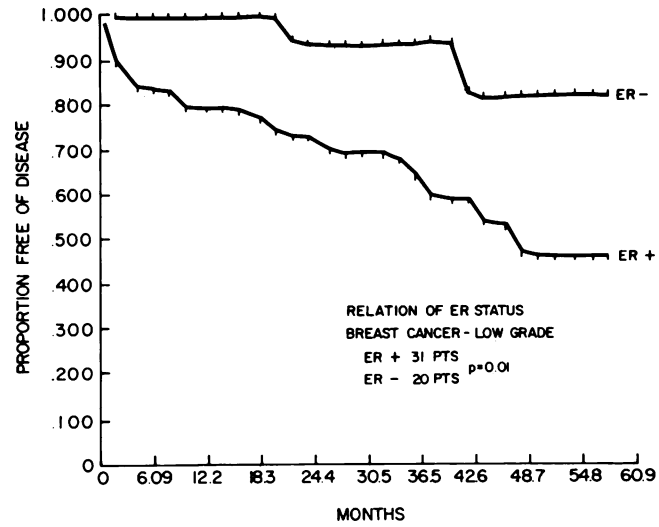

FIG. 3. In patients with low-grade tumors, there was a better disease-free survival in the ER− group, $p < 0.01$.

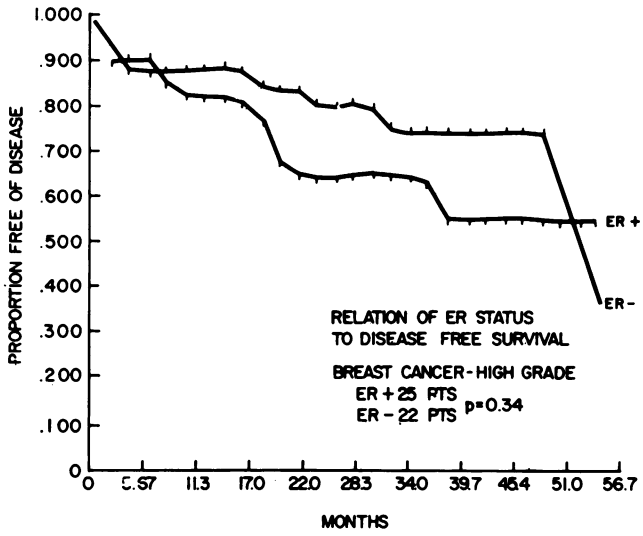

FIG. 4. There was no difference in disease-free survival in patients with high-grade tumors.

have a poorer prognosis than patients with receptor positive tumors. The DFI and survival rate are shorter in patients with receptor negative tumors.[3,4,6,11,12,14] Although this effect seems independent of node status, patients with receptor negative tumors and nodal metastases have particularly aggressive disease.[4,6] In Kinne et al.[9] the recurrence rate and mortality from cancer was significantly higher in the ER− patients who had nodal metastases. This effect primarily occurred in patients with >4 nodal metastases. There were no differences in the patients with negative nodes.

We were unable to show any significant correlation between estrogen receptor status and disease-free survival in the overall group or in patients according to stage of disease. There were no significant differences in recurrence rates in patients classified according to presence or absence of axillary node metastases at the time of primary surgery. It was observed, however, that in the cases in which the tumors had been reviewed and graded, the patient with low-grade tumors that were ER+ had a significantly better disease-free survival rate than ER− patients. There was no such correlation with estrogen receptors in patients with high-grade tumors. This seems to be at variance with Maynard's studies which showed a worsened prognosis in patients with more advanced tumor grades (poorly differentiated) and which were also estrogen receptor negative.[6] The presence of nodal metastases added to the adverse prognosis in these patients.

TABLE 6. *ER Status Vs. Time to Recurrence*

Pt. Group	Difference—Significance	Median Time (Mos.) To Recurrence ER−	ER+
All	p = 0.35	58	49
Stage 0	p = 0.56	58	58
Stage I & II	p = 0.84	18	38
Grade low	p = 0.01	58	47
Grade high	p = 0.34	54	49

Walt et al.[3] examined estrogen receptors as a predictor of site of recurrence and survival following recurrence. In ER− patients with recurrence, visceral metastases were predominant and occurred in 45% of the cases. In ER+ patients with recurrence, only 6% had visceral metastases. The mean survival of 27.2 months in this ER− group was compared with 40.5 months in the ER+ patients. However, there was a large number of Stage IV patients in the ER− group which could affect the results.

Knight et al.[4] reported an earlier recurrence of disease in ER− patients. However, his figures were statistically significant only in patients having four or more nodal metastases. Conversely, Singhakowinta et al.[7] were unable to demonstrate statistically significant difference in disease-free interval when correlated with estrogen receptor status in 90 patients (mean: 37.6 months for ER+ and 31.2 months for ER−). Rich et al.,[5] from the same institution and presumably using the same patient population, found a shorter disease-free interval in ER− patients, but also noted a higher percentage of Grade III tumors in these patients. The difference in the disease-free interval was not statistically significant. Maynard et al.[6] reported a statistically significant difference in disease-free interval when comparing ER+ and ER− patients with Stage II (low axillary nodes) and Stage III (high axillary or internal mammary nodes) disease. As with Rich et al., Maynard found that poorly differentiated carcinomas occurred more frequently in the ER− patients than in the ER+ patients. When all disease stages were considered, however, disease-free interval curves for ER+ and ER− patients tended to converge at 36 months. In this study, attempts to group patients into low-risk *vs.* high-risk tumors failed to show any significant correlation of differences regarding estrogen receptor status with prognosis. Possibly, with further maturing of the data, certain differences may be manifested. Currently, the data from this study and from the literature do not show a uniformly clearcut answer relationship of estrogen receptor and grade, nor of estrogen status and prognosis within grade or stage categories.

Summary

An analysis of pathologic and prognostic relationships of estrogen receptors was undertaken in a review of

breast cancer patients who had the receptor assay performed at the University of Virginia Medical Center between 1975 and 1978. Of 104 patients available for initial evaluation, 91 patients were eligible for follow-up with a median observation period of 19 months. Sixty patients (58%) were estrogen receptor positive and 44 patients (42%) were estrogen receptor negative. The presence of estrogen receptor was examined with respect to the following variables: 1) age; 2) menstrual status; 3) tumor histology and grade; 4) lymph node status and stage; 5) recurrence; 6) history of second breast cancer; 7) family history of breast cancer; and 8) history of hysterectomy. The presence of estrogen receptors showed significant correlation with age, lobular cancer, and a variant of infiltrating duct cancer showing eosinophilic granular cytoplasm (a type prevalent in elderly patients). There was no correlation with lymph node status at diagnosis. In a small subset of seven patients who had second primary cancers, all were ER+. There was no significant correlation of disease-free survival or time to recurrence in either the overall group nor according to stage, nor in patients with high-grade cancers. In a subset of 51 patients with low-grade cancers, improved disease-free survival occurred in patients who were ER negative.

Although estrogen receptor is highly useful as a predictor in patients undergoing hormonal manipulation and has been considered to have a relation to prognosis in patients with primary cancer, the authors have not confirmed the latter relationship in this study.

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