Enhanced Benefit from Adjuvant Chemotherapy in Breast Cancer Patients Classified High-Risk according to Urokinase-type Plasminogen Activator (uPA) and Plasminogen Activator Inhibitor Type 1 (n = 3424)\textsuperscript{1}

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ABSTRACT

Risk assessment and prediction of response to treatment are prerequisites for individualized adjuvant therapy decisions in breast cancer. The strong prognostic impact of the two invasion factors urokinase-type plasminogen activator (uPA) and its inhibitor, plasminogen activator inhibitor type 1 (PAI-1), in breast cancer has recently been validated at level-I evidence. This article considers the predictive impact of uPA/PAI-1 on response to adjuvant chemo- and endocrine therapy in 3424 primary breast cancer patients from two different data sets. uPA and PAI-1 antigen levels were measured by ELISA in primary tumor tissue extracts. After a median follow-up of 83 months, uPA/PAI-1 has a significant impact on disease-free survival in Cox multivariate analysis (P < 0.001; hazard ratio, 2.0; 95% confidence interval, 1.8–2.3). Patients with high uPA/PAI-1 levels benefit more strongly from adjuvant chemotherapy than those with low levels. This effect is seen as a significant interaction between chemotherapy and uPA/PAI-1 for the entire collective (P < 0.003; hazard ratio, 0.68; 95% confidence interval, 0.53–0.88) and separately within nodal subgroups. This enhanced benefit in the high uPA/PAI-1 patients occurs over and above the significant impact of both therapies in all patients. We find no corresponding significant interaction between endocrine therapy and uPA/PAI-1; i.e., no significant difference in benefit between patients with high and low uPA/PAI-1. In conclusion, uPA and PAI-1 levels in primary tumor tissue provide clinically relevant information on relapse risk and treatment response that will help to tailor adjuvant therapy concepts in breast cancer, accounting for individual biological tumor characteristics.

INTRODUCTION

A key clinical question in the management of primary breast cancer concerns the assessment of information provided by risk factors for supporting the choice of an optimal adjuvant treatment regimen for the individual patient. This report provides new evidence for the potential role of invasion factors uPA\textsuperscript{3} and PAI-1 in treatment selection.

There is already abundant experimental evidence that the plasminogen activator system plays a key role in tumor invasion and metastasis (1, 2). A critical balance of uPA, its cell surface receptor uPA-R, and PAI-1 is still scarce. Results from local response to neoadjuvant CT (11) or preliminary systemic therapy in early breast cancer (12, 13) cannot be readily transferred to the adjuvant setting. Recent reports show that the prognostic impact of uPA/PAI-1 is lost in patients who received adjuvant chemo- or endocrine therapy, thus suggesting a benefit from adjuvant therapy in high uPA/PAI-1 patients (8, 14) and high-risk patients benefit from adjuvant CMF-CT (4).

To optimize adjuvant therapy for high-risk patients as classified by uPA/PAI-1, there is an urgent clinical need to know whether these patients do indeed benefit from adjuvant systemic therapy, and if so, which therapy would be most beneficial. Unfortunately, information on the predictive value of uPA and/or PAI-1 with regard to therapy response is still scarce. Results from local response to neoadjuvant CT (11) or response to palliative systemic therapy in advanced or metastatic breast cancer (12, 13) cannot be readily transferred to the adjuvant setting.

Recent reports show that the prognostic impact of uPA/PAI-1 is lost in patients who received adjuvant chemo- or endocrine therapy, thus suggesting a benefit from adjuvant therapy in high uPA/PAI-1 patients (8, 14). Consistent with this, the first interim analysis of a prospective randomized multicenter therapy trial (“Chemo N\textsuperscript{0}”), in which patients were stratified according to their uPA and PAI-1 levels, indicated that this risk of patients benefit from adjuvant CMF-CT (4).

The present study addresses for the first time the issue of a predictive impact of uPA/PAI-1 in breast cancer (n = 3424) with regard to adjuvant chemo- and/or endocrine therapy.

MATERIALS AND METHODS

Patients

A total of 3424 primary breast cancer patients from two different data sets [Department of Obstetrics and Gynecology, Technical Uni-
versity of Munich, Germany; Department of Medical Oncology, Rotterdam Cancer Institute (Daniel den Hoed Kliniek) and University Hospital Rotterdam, the Netherlands were included in the analysis. Patients had been treated for primary breast cancer between 1987 and 1999. Characteristics of the two data sets are summarized in Tables 1–3. Treatment decisions with regard to primary surgery and adjuvant systemic therapy were based primarily on consensus recommendations at the time. Treatment strategies differed between Munich and Rotterdam, particularly regarding the administration of adjuvant systemic therapy to node-positive patients (see Table 2; Refs. 8, 15). Of those patients treated by adjuvant CT, the majority received CMF (Rotterdam, 76%; Munich, 65%); about one-fifth in both collectives received anthracycline-containing regimens (Rotterdam, 24%; Munich, 21%); and the rest of the Munich patients were treated by other CT regimens. The endocrine treatment used in both collectives was tamoxifen.

Median age of the patients at the time of primary surgery was 56 years (range, 22–94 years). Both data sets contained established clinical and histomorphological factors such as the number of involved axillary lymph nodes, pT stage, ER, PgR, adjuvant endocrine therapy (HT), adjuvant RT, as well as uPA and PAI-1 measurements. Because the scoring systems for assessment of grade differed substantially between the centers and could, thus, represent different biological tumor characteristics, grade was not included in the combined stratified analysis. The variables were recoded as described “Statistical Methods” section. At time of primary therapy, no patient had any clinical or radiological evidence of distant metastases. Follow-up data were obtained at regular intervals. Median follow-up time of all of the patients still alive at the time of analysis was 83 months (range, 1–183 months) Within the follow-up period, 1319 patients (39%) experienced disease recurrence and 1200 patients (35%) died.

Laboratory Assays

In the Munich cohort, uPA and PAI-1 antigen were prospectively measured by ELISA (uPA: Imubind 894; PAI-1: Imubind 821; both from American Diagnostica Inc., Greenwich, CT) in detergent extracts of the primary tumor tissue as reported in Jänicke et al. (16). In the Rotterdam cohort, uPA and PAI-1 antigen were measured by ELISA, using the same antibodies as above, in cytosol preparations of the primary tumor as described by Foekens et al. (15).

Statistical Methods

Variable Definition and Recoding for the Combined Analysis. Because of differences in measurement techniques between the data sets, data recoding was required. For the laboratory measurements of uPA, PAI-1, ER, and PgR, fractional ranks were computed with respect to each distribution. Fractional ranks also kept the variables on a convenient scale from zero to one, thus facilitating comparison of the β coefficients of different factors. In particular, ranked ER and PgR measurements were able to be included as continuous variables in the statistical models, even though biochemical and immunohistochemical assays were used. The weak correlations found between these laboratory measurements and the remaining “classical” staging factors support the inference that similar ranks imply similar biological characteristics even across data sets.

A binary variable for uPA/PAI-1 was defined as 0 for uPA and PAI-1 both below their respective cutoffs and as 1 otherwise (i.e., either or both above the respective cutoff; Ref. 17). Previously determined and validated univariate cutoff values by the Munich group (4, 17) were applied to the Rotterdam data by transforming the Munich cutoffs to fractional ranks and applying these to the Rotterdam data, resulting in almost exactly the same percentage of uPA/PAI-1 “high” versus “low” in each cohort (see Table 1).

The pT stage (18) was coded using two auxiliary binary variables: (a) pT1 (coded 0) versus all others (coded 1); and (b) pT1 and pT2 (coded 0) versus pT3 and pT4 (coded 1). Fractional ranks were assigned separately within the two data sets for the number of affected lymph nodes (variable denoted “lymph nodes”). Equal numbers of nodes correspond to equal fractional ranks across data sets, to within a few percentage points. For patient age, fractional ranks were first computed for the Rotterdam data set, and the Munich ages were then transformed to this scale. To model the nonlinear dependence of HR on age as closely as possible, both the fractional rank itself as well as its square is transformed to this scale. To model the nonlinear dependence of HR on age as closely as possible, both the fractional rank itself as well as its square is included in the models. The three binary variables for adjuvant therapy (RT, HT, and CT) were coded such that the value 1 represented “known to have been treated by the respective kind of adjuvant therapy.” A binary variable “data set” was introduced and used to stratify the analysis as discussed below. This variable accounts for systematic differences in demographic influences, unobserved factors contributing to the stage of the disease, or adjuvant systemic therapy strategies.

Survival Analysis. The Cox proportional hazards model was used with continuous ranked variables and binary variables as described above. All of the tests were performed at a significance level of α = 0.05 with a 95% CI. Variables were included according to likelihood ratios in a stepwise forward fashion using the SPSS software package (SPSS Inc., Chicago, IL). Unless otherwise stated, main (i.e., linear) effects were always included as a first block, whereas interactions were included as a second block in the analysis. This method implies that a main effect that is significant in the first block will be retained in the model, even if an interaction in the second block is so strong as to reduce the main effect coefficient below the level of significance. All of the models were stratified by data set. The SPSS software package was also used to compute fractional ranks, correlation coefficients, associations, and other statistical properties.

RESULTS

Correlations. Significant correlations (Pearson’s coefficient) exceeding 0.1 between ranked factors were found between ER and PgR (0.53); ER and age (0.36); lymph node and tumor stage (0.36); and uPA and PAI-1 (0.58).

DFS Including uPA/PAI-1 and Their Interactions with Therapy in All Patients. The 5-year relapse rates associated with low and high uPA/PAI-1 were 28 and 46%, respectively. The probabilities of being
treated by CT or HT in subgroups defined by uPA/PAI-1 are not depicted in Table 3.

In Table 4, we report the results of a proportional hazards analysis for DFS in all of the patients, stratified by data set. The first stage included established prognostic factors (ER, PgR, age, lymph nodes, pT stage, all coded as described above under “Statistical Methods”) as well as uPA/PAI-1, CT, HT, and RT. The second stage included the interactions CT and HT with uPA/PAI-1, and lymph nodes with uPA/PAI-1, as well as the “treatment interaction,” i.e., CT with HT.

All of the main effects (factors) considered in the model of Table 4 were significant except RT and ER. The HR of uPA/PAI-1 was 2.0 (CI, 1.8–2.3; P < 0.001). The overall effect of age, including squared fractional rank, in the model was a gradual, almost linear, drop of the HR from the youngest patients (defined to have HR = 1), leveling off to about HR = 0.5 by about age 60 and apparently rising slightly above 0.5 at about age 65. The interaction between lymph nodes and uPA/PAI-1 was not significant in the analysis of all of the patients, nor was CT × HT, implying no evidence against an additive effect of this treatment combination.

The key result is the significant (negative) interaction between CT and the variable uPA/PAI-1. This interaction implies that the higher HR of relapse (2.01) associated with high uPA/PAI-1 (compared with low uPA/PAI-1) is significantly reduced (0.68 × 2.01 = 1.36) in patients who receive adjuvant CT. This benefit occurs in addition to the independent overall risk reductions of about one-third attributable to CT (HR = 0.69), or HT (HR = 0.68). No significant interaction was found between HT and uPA/PAI-1 (95% CI for this HR, 0.66–1.28). Hence, the benefits of both therapies were significant, but only for CT was an additional (enhanced) benefit seen among high uPA/PAI-1 patients.

Table 4: Multivariate Cox model (DFS) including interaction of uPA/PAI-1 with adjuvant treatment in primary breast cancer (n = 3376)<!--
Analysis was stratified by center; 46 patients were censored before the first event in the stratum, 1301 events total. First stage of analysis included established prognostic factors, uPA/PAI-1, adjuvant radiotherapy, adjuvant chemotherapy, and adjuvant endocrine therapy. Second stage included interactions (chemo- and endocrine therapy with uPA/PAI-1; chemotherapy with endocrine therapy) and involved lymph nodes with uPA/PAI-1. -->

<table>
<thead>
<tr>
<th>Significant factors</th>
<th>Coding for interpretation of β</th>
<th>P</th>
<th>β</th>
<th>HR (95% CI)</th>
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<tr>
<td>Involved lymph nodes</td>
<td>Fractional rank:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1 node vs. node-negative</td>
<td>&lt;0.001</td>
<td>2.47</td>
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<tr>
<td>4 nodes vs. node-negative</td>
<td>&lt;0.001</td>
<td>1.29</td>
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<td>3.65 (1.73–7.70)</td>
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<tr>
<td>10 nodes vs. node-negative</td>
<td>&lt;0.001</td>
<td>−1.94</td>
<td></td>
<td>0.14 (0.07–0.31)</td>
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<td>Age</td>
<td>Squared fractional rank</td>
<td>&lt;0.001</td>
<td>0.30</td>
<td>1.35 (1.19–1.54)</td>
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<tr>
<td>Tumor stage</td>
<td>Fractional rank</td>
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<td>−0.22</td>
<td>1.24 (1.06–1.46)</td>
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<td>PgR</td>
<td>Rest (1) vs. pT1 (0)</td>
<td>&lt;0.001</td>
<td>0.08</td>
<td>1.03 (1.01–1.06)</td>
</tr>
<tr>
<td>uPA/PAI-1</td>
<td>Rest (1) vs. pT1/pT2 (0)</td>
<td>&lt;0.001</td>
<td>0.08</td>
<td>1.03 (1.01–1.06)</td>
</tr>
<tr>
<td>Adjuvant chemotherapy</td>
<td>Fractional rank</td>
<td>&lt;0.001</td>
<td>0.70</td>
<td>2.01 (1.77–2.28)</td>
</tr>
<tr>
<td>Adjuvant endocrine therapy</td>
<td>High (1) vs. low (0)</td>
<td>&lt;0.001</td>
<td>−0.37</td>
<td>0.69 (0.56–0.85)</td>
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<tr>
<td>Interaction, chemotherapy × uPA/PAI-1</td>
<td>Both 1 vs. either or both 0</td>
<td>&lt;0.001</td>
<td>−0.38</td>
<td>0.68 (0.56–0.82)</td>
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</table>

* Forty-five of 3424 were excluded because of incomplete information on the number of involved nodes; 3 of 3424 were excluded because of missing information on adjuvant chemotherapy (see Table 1).

* Coding of variables as reported in “Materials and Methods.”

* HR for patients with 1, 4, and 10 positive nodes compared with node negative patients; CI is approximate because of fractional ranks.

* Hazard for fractional rank = 1 compared with fractional rank = 0.

Fig. 1 illustrates the HRs of CT and HT taking into account significant interactions with uPA/PAI-1 according to Tables 4–6 (for discussion of Tables 5 and 6 see the sections that follow). For all of the patients, the significant interaction CT × uPA/PAI-1 is seen in the upper panel of Fig. 1 as a hazard reduction, attributable to CT, that is strongly affected by uPA/PAI-1. The lack of a significant interaction HT × uPA/PAI-1 manifests itself in the figure in that the hazard reduction attributable to HT is not affected by uPA/PAI-1.

DFS Including uPA/PAI-1 and Their Interactions with Therapy in Patients with Zero-to-Three Involved Nodes. Separate analysis of the predictive value of uPA/PAI-1 in node-negative patients was not feasible, because less than about 5% of patients in either subgroup received HT or CT. It is nonetheless of clinical interest to consider the group of patients with 0–3 involved nodes. Table 5 shows the proportional hazards analysis for DFS, which was stratified by data set as in Table 4 and which includes the same factors. In this subgroup, ranked ER is significant, higher values being associated with higher HR, whereas ranked PgR is associated with lower HR. For tumor stage, only the distinction pT1 versus all others is significant, but not pT1/pT2; versus the rest. The HRs for age imply that young age is an even more strongly unfavorable factor in this subgroup than in patients on the whole.

In patients with 0–3 affected lymph nodes, the effects involving therapy and uPA/PAI-1 are qualitatively and even quantitatively very close to those seen in the analysis of all patients: The hazard associated with high uPA/PAI-1 is slightly greater; adjuvant endocrine therapy has about the same benefit as in all patients; and the interaction between adjuvant CT and uPA/PAI-1 is similar (see center panel of Fig. 1).

DFS Including uPA/PAI-1 and Their Interactions in Patients with Four or More Involved Nodes. In patients with four or more involved axillary lymph nodes, the adjuvant therapy percentages were as follows: with low uPA/PAI-1 (n = 398; 5-year relapse rate, 56%), 27% were treated by adjuvant endocrine therapy, and 36% by CT. With high uPA/PAI-1 (n = 388; 5-year relapse rate, 72%), these percentages are slightly lower at 26% and 29%, respectively. The results of a Cox analysis performed for this subgroup are reported in Table 6. The factors were included as in the previous models; the analysis was again stratified by data set.
In these patients, it is noteworthy that uPA/PAI-1 had an enormous impact ($\beta = \log \text{HR} = 3.02$), but there was also a large negative interaction of uPA/PAI-1 with lymph nodes ($\beta = \log \text{HR} = -2.79$). There was also an (apparently) very high hazard ($\beta = \log \text{HR} = 5.36$) associated with lymph nodes within this subgroup of patients with 4 or more affected nodes, but this number was partly an artifact of the representation in fractional ranks. To facilitate interpretation of this HR, as well as the interaction of lymph nodes with uPA/PAI-1, we compared the hazard for 10 versus 4 affected nodes: For patients with low uPA/PAI-1, the HR of patients with 10 affected nodes was about twice as high as for 4 affected nodes, as seen in Table 6. (If this mere doubling of risk going from 4 to 10 nodes seems too moderate in view of $\beta = 5.36$, it must be kept in mind that the fractional rank for lymph nodes for a patient with 4 nodes is already quite high, about 0.78.) In contrast, for patients with high uPA/PAI-1, the interaction means that the HR of patients with 10 affected nodes was only about 1.5 times that of patients with 4 affected nodes. Summarizing, the results (including interaction of lymph nodes with uPA/PAI-1) imply that the number of affected nodes even above 4 is important, but more so for low uPA/PAI-1 than for high uPA/PAI-1. In patients with >4 affected lymph nodes, the benefits of CT and HT and their relation to uPA/PAI-1 were again qualitatively and even quantitatively very close to those seen in the analysis of all patients and in the group with 0–3 affected nodes (see bottom panel of Fig. 1).

Benefits of CT and HT for DFS in Subgroups according to uPA/PAI-1. The effect of uPA/PAI-1 on response to therapy is also seen by constructing separate Cox models for high and low uPA/PAI-1 (again stratified by data set). In all of the patients, the HR was 0.68 for CT and 0.74 for HT, according to a multivariate model for the low-uPA/PAI-1 subgroup. Within the multivariate model for the high-uPA/PAI-1 subgroup, the corresponding HRs were 0.49 for CT and 0.63 for HT. (In terms of log HR, the difference in HR for CT between low and high uPA/PAI-1 was about three SEs, whereas for HT the corresponding difference was only one-third of a SE.) Hence, these models were consistent with the tendency for more relative benefit caused by CT in patients with high uPA/PAI-1 than in patients with low uPA/PAI-1, after controlling for other factors.

Cox models were also performed separately for high and low uPA/PAI-1 patients in the subgroup of patients with 0–3 affected lymph nodes. In the low-uPA/PAI-1 group ($n = 1418$; 5-year relapse rate 20%; 9% receiving HT, 17% receiving CT), it turned out that neither of the adjuvant therapy forms were significant: the 95% CI for the HR of CT was 0.60–1.22, and for HT it was 0.59–1.44. Because of statistical uncertainty, in this subgroup, a low-to-moderate benefit of either therapy is not ruled out. In contrast, in the high-uPA/PAI-1 subgroup ($n = 1174$; 5-year relapse rate 38%; 10% receiving HT, 19% receiving CT), both adjuvant therapy forms are significant and strong: with a HR of 0.51 (0.33–0.78), HT approximately halves the hazard; the benefit of CT appears to be even stronger with a HR of 0.43 (95% CI, 0.31–0.59). Comparing the result in these subgroups with the interaction analysis for 0–3 nodes reported above, the detection of a significant interaction CT $\times$ uPA/PAI-1 manifests itself in the uPA/PAI-1 subgroups as distinctly different HRs with noninter-

<table>
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<tr>
<th>Significant factors</th>
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<th>$P$</th>
<th>$\beta$</th>
<th>HR $^c$ (95% CI)</th>
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<tr>
<td>Involved lymph nodes</td>
<td>Fractional rank</td>
<td>$&lt;0.001$</td>
<td>2.37</td>
<td>2.10 (1.8–2.4) (x3)</td>
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<td>Age</td>
<td>Squared fractional rank</td>
<td>$&lt;0.001$</td>
<td>1.87</td>
<td>6.48 (2.52–16.66)</td>
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<td>Tumor stage</td>
<td>Rest (1) vs. $pT_1$ (0)</td>
<td>$&lt;0.001$</td>
<td>0.35</td>
<td>1.42 (1.23–1.65)</td>
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<tr>
<td>ER</td>
<td>Fractional rank</td>
<td>$&lt;0.001$</td>
<td>–0.58</td>
<td>0.56 (0.42–0.75)</td>
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<tr>
<td>uPA/PAI-1</td>
<td>Fractional rank</td>
<td>$&lt;0.001$</td>
<td>0.43</td>
<td>1.53 (1.12–2.10)</td>
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<tr>
<td>Adjuvant chemotherapy</td>
<td>Yes (1) vs. no (0)</td>
<td>$&lt;0.001$</td>
<td>0.79</td>
<td>2.21 (1.88–2.59)</td>
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<tr>
<td>Adjuvant endocrine therapy</td>
<td>Yes (1) vs. no (0)</td>
<td>$&lt;0.001$</td>
<td>–0.33</td>
<td>0.72 (0.53–0.98)</td>
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<tr>
<td>Interaction, chemotherapy $\times$ uPA/PAI-1</td>
<td>Both 1 vs. either or both 0</td>
<td>$&lt;0.001$</td>
<td>–0.36</td>
<td>0.70 (0.50–0.98)</td>
</tr>
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</table>

$^*$ One of 2593 was excluded because of missing information on adjuvant chemotherapy.

$^z$ Coding of variables as reported in “Materials and Methods.”

$^x$ Hazard for fractional rank = 1 compared with fractional rank = 0.

$^y$ HR for patients with 1, 2, and 3 positive nodes compared with node negative patients, CI is approximate because of fractional ranks.
Table 6  Multivariate Cox model (DFS) including interaction of uPA/PAI-1 with adjuvant treatment in breast cancer patients with four or more involved axillary nodes (n = 784)
Analysis was stratified by the Center: seven patients were censored before the first event in the stratum, 501 events total. Stages of analysis are as in Table 3.

<table>
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<tr>
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<th>( \beta )</th>
<th>HR (95% CI)</th>
</tr>
</thead>
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<tr>
<td>Involved lymph nodes</td>
<td>10 nodes vs. 4 nodes</td>
<td>&lt;0.001</td>
<td>5.36</td>
<td>2.1 (1.6–2.8)</td>
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<tr>
<td>Age</td>
<td>Fractional rank</td>
<td>0.002</td>
<td>–0.62</td>
<td>0.54 (0.36–0.80)</td>
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<td>Tumor stage</td>
<td>Rest (1) vs. PT1∥/PT2 (0)</td>
<td>0.001</td>
<td>0.35</td>
<td>1.41 (1.16–1.73)</td>
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<tr>
<td>PgR</td>
<td>Fractional rank</td>
<td>0.001</td>
<td>–0.54</td>
<td>0.59 (0.43–0.80)</td>
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<tr>
<td>uPA/PAI-1</td>
<td>High (1) vs. low (0)</td>
<td>0.013</td>
<td>3.02</td>
<td>20.5 (1.9–220)</td>
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<tr>
<td>Adjuvant chemotherapy</td>
<td>Yes (1) vs. no (0)</td>
<td>0.034</td>
<td>–0.34</td>
<td>0.71 (0.52–0.97)</td>
</tr>
<tr>
<td>Adjuvant endocrine therapy</td>
<td>Yes (1) vs. no (0)</td>
<td>0.002</td>
<td>–0.39</td>
<td>0.68 (0.53–0.87)</td>
</tr>
<tr>
<td>Interaction, chemotherapy uPA/PAI-1</td>
<td>Both 1 vs. either or both 0</td>
<td>0.040</td>
<td>–0.42</td>
<td>0.66 (0.44–0.98)</td>
</tr>
<tr>
<td>Interaction, involved lymph nodes × uPA/PAI-1</td>
<td>If low uPA/PAI-1: zero</td>
<td>0.039</td>
<td>–2.79</td>
<td>0.061 (0.004–0.87)</td>
</tr>
</tbody>
</table>

- Two of 786 were excluded because of missing information on adjuvant chemotherapy.
- Coding of variables as reported in “Materials and Methods.”
- Hazard for fractional rank = 1 compared with fractional rank = 0.
- HR for patients with 10 positive nodes compared with patients with four positive nodes, CI is approximate because of fractional ranks.

In patients with four or more affected nodes, a separate Cox regression (two-stage model) for the low-uPA/PAI-1 subgroup showed that these patients benefited significantly from adjuvant HT with a HR of 0.62 (95% CI, 0.43–0.90) and apparently also from CT with a HR of 0.70 (95% CI, 0.49–0.98). Lymph nodes were a very strong factor in this group (log HR = 5.68). The benefit from CT derived from the regression model for the group with four or more nodes and high uPA/PAI-1 corresponded to HR 0.60 (95% CI, 0.44–0.81). For HT, the HR was 0.68 (95% CI, 0.49–0.95). Lymph nodes were weaker in this model (log HR = 2.43). These results taken together are consistent with the full model with interactions for patients with 4 or more nodes reported above.

**DISCUSSION**

With the available and potent conventional drug regimens as well as the advent of novel therapy approaches targeting specific biological pathways, the problem of optimal treatment of primary breast cancer is becoming increasingly complex. To allow tailored therapy concepts that take the individual tumor biology into account, factors are needed that guide physicians with regard to the estimation of patient prognosis and the prediction of therapy response. Thus far, uPA and PAI-1 are the only novel tumor biological factors that have reached level-I evidence with regard to their prognostic impact in primary breast cancer (4, 5). Yet, it is not known what kind of adjuvant therapy would be most beneficial for patients who had been grouped into different risk categories according to their uPA and PAI-1 levels in the primary tumor tissue. This paper demonstrates that uPA and PAI-1 have not only a clinically relevant prognostic but also predictive impact in primary breast cancer.

Ideally, the gold standard for determining predictive information is a properly designed prospective study. However, for ethical reasons, new studies cannot include control groups of patients without adjuvant systemic therapy. On the other hand, large retrospective data sets containing substantial patient numbers with and without adjuvant systemic therapy are available for analysis. In general, it is quite difficult to ascertain predictive information from retrospective data in breast cancer, because adjuvant treatment decisions were made on the basis of guidelines in force at that time taking into account prognostic factors. Such factors thus act as confounding variables for retrospectively analyzing efficacy of adjuvant treatment. Moreover, different adjuvant treatment policies were used in different centers.

Nonetheless, for factors that do not strongly correlate with treatment decisions, the problem of confounding can be reduced by various methods, in particular, by appropriate use of multivariate analysis and stratification. Because (in contrast to ER and PgR) these requirements are satisfied rather well by uPA and PAI-1, the results presented in this paper should indeed reflect the predictive properties of uPA/PAI-1. An important step in “de-convoluting” the confounding factors in retrospective data is to introduce a multivariate statistical scoring model using as much of the information as possible in the other variables. A good scoring model will reduce the unexplained variation in the data and improve the chances of seeing interactions if they are present. Consequently, in this paper, a strategy of avoiding the use of cutoffs whenever possible, i.e., by representing most of the measurements as continuous variables, was applied. The only exception to the strategy of continuous variables were the factors uPA and PAI-1 themselves, for which previously optimized cutoffs, validated in a prospective multicenter trial (4, 17), were applied.

The present paper confirms that uPA/PAI-1 have a significant impact on patient outcome but also provides additional evidence supporting their use in the clinic by demonstrating how effects of adjuvant systemic therapy differ in patients classified according to uPA/PAI-1. As illustrated in Fig. 1, primary breast cancer patients with low uPA/PAI-1 generally benefit from adjuvant endocrine and CT. However, the benefits of CT (but not endocrine therapy) are strongly enhanced in patients with high uPA/PAI-1. This finding is in accordance with the benefit from CMF observed in high-risk patients in the Chemo N0 trial (4). It is important to note that patients with high uPA/PAI-1 also benefit from adjuvant endocrine therapy, even though adjuvant CT has a greater beneficial impact on their DFS.
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REFERENCES


Enhanced Benefit from Adjuvant Chemotherapy in Breast Cancer Patients Classified High-Risk according to Urokinase-type Plasminogen Activator (uPA) and Plasminogen Activator Inhibitor Type 1 (n = 3424)
