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# AGE AS A DETERMINANT OF AXILLARY NODE INVOLVEMENT IN INVASIVE BREAST CANCER

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**We analyzed the age at diagnosis and the tumor size as determinants of axillary node involvement in 725 consecutive patients with breast cancer. The prevalence of nodal involvement increased consistently with tumor diameter from 18.9% in tumors smaller than 10 mm to 72.9% in those measuring 40 mm, or more. The risk also varied with age, the lowest prevalence being found in the youngest and the oldest patients and the highest one in the 40–59-year age group. When analyzed as a continuous variable age was best fitted as a second order term and it was a statistically significant ($p = 0.04$) determinant of axillary metastases in a multivariate model where tumor diameter, histopathological classification and estrogen receptor concentration were taken into account as possible confounding variables. The findings indicate that the parallelism between the establishment of metastases in lymph nodes and at distant sites may vary with age. The prognostic value of nodal status may therefore depend on age at diagnosis.**

Age at diagnosis has proved to be an important prognostic factor in female breast cancer (1–5). Women in the years before their menopause seem to have the most favorable outcome, but age is not related to the prognosis in a linear fashion. In an analysis of the relative survival rates in 57 000 women with breast cancer in Sweden, we found that patients 45–49 years old at diagnosis had the best prognosis, with a relative survival rate exceeding that of the youngest patients as well as that of the oldest ones (2).

These findings suggest that the metastatic potential of the tumor is dependent on age-associated factors. To further investigate the nature of these factors we specificially studied the relationship between age at diagnosis and the metastatic potential of breast cancer, defined as the probability of having axillary nodal involvement (6). In addition to stratifying for tumor diameter we wished to take into account the possible confounding effect of prognostic factors, such as histopathological characteristics, estrogen receptor concentration and nuclear DNA content, since it is possible that different prognostic factors have an age-dependent distribution (7–9). Our analysis revealed unexpectedly, strong and hitherto unknown relations between age at diagnosis and the presence of axillary node metastases.

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## Material and Methods

*Patients.* A total of 825 patients diagnosed as having invasive breast cancer from May 1977 through March 1988, in a defined population (the city of Uppsala), were prospectively included in a computerized database in the Department of Pathology at the University Hospital. These women comprised all consecutive patients diagnosed when the pathologist in this investigation (AL) was in charge. Information on tumor size, the presence of axillary metastases, Ackerman's histopathological classification (10), estrogen receptor status and the results of DNA analyses were recorded at the time of diagnosis. Presence of axillary involvement was classified as with or without periglandular growth. No information on number of involved nodes was present. The presence of nodal involvement was used to discriminate between patients with primarily disseminated disease and those with localized disease. A screening mammography program was started in February 1988, thus contributing none of the cases of this cohort.

*Treatment.* Women with cancer in pTNM-stages I and II (UICC) were routinely operated with a modified radical mastectomy. If axillary metastases were present, postoperative radiotherapy was given to axillary and regional nodes. From 1982 onwards most patients in pTNM-stage I with unifocal tumors were offered sector resection and axillary dissection (11). Elderly women (over 80 years of age) and those with poor general health were mainly treated with mastectomy without axillary clearance. In pTNM-stages III and IV the treatment was individualized.

*Histopathological classification.* Ackerman's classification (10, 12–14) was used both at the time of diagnosis and in a blind review performed in this investigation. Women with cancer in situ were excluded from this analysis. Apart from recognizing the subgroups of tubular, colloid, medullary and papillary carcinoma and categorizing them as Ackerman's type II, the classification divides the common ductal and lobular carcinomas into two major types, III and IV, depending on the mode of invasion of the tumor. Type III describes a circumscribed tumor with 'pushing border' and lymphocyte reaction. As a contrast, type IV includes tumors that are diffusely infiltrating and without lymphocyte reaction in the surrounding. All tumors with vessel invasion are included in type IV. Type III is divided into III:1 and III:2, where III:2 represents carcinoma in situ with only small areas of invasion. This classification has been used in a number of our studies since 1975 (12–14). In the analysis, Ackerman's types II and III:2 were grouped together, since a previous study (14), had shown that these two groups had very similar prognoses and since the number of patients in each group was small. Information on histopathological lymph node status and/or tumor diameter was not available in 100 patients. Many of these patients were elderly (axillary status missing in somewhat less than half of the women aged 80 years or more) and in poor general health or had disseminated disease already at the time of diagnosis. Among women aged less than 80, axillary status was missing in approximately 10% of each 10-year age-group.

*Hormone receptor assays and DNA measurements.* Complete data on receptor content together with records of lymph node status and tumor diameter were available in 684 cases. Samples were stored in a freezer at −70° for a maximum of 3 weeks. The analysis of cytosol receptors was performed by isoelectric focusing (15). The sensitivity of the described ER analysis is 2–3 fmol/ml of cytosol. The amount of receptor in the sample was related to its DNA content. The DNA contents of tumor cells were analyzed by flow cytometry or single cell cytophotometry. The methods have been described in detail by Strang et al. (16). The four types of DNA patterns as modified by Auer et al. (17) were used. In brief, types I and II are diploid tumors, without or with a small percentage of cells in S-phase respectively. Types III and IV represent aneuploid tumors. In 485 women, DNA ploidy, ER receptor content, lymph node status and tumor diameter were all known. The cases with missing information concerning ER-status and DNA content became increasingly scarce with time as routine analysis of these factors was successively introduced during the period of recruitment of the cohort studied.

*Statistical analysis.* In the multivariate analyses the logistic regression model estimated by the maximum likelihood (ML) method, was used with axillary status (nodes present = 1, absent = 0) as the dependent variable. The explanatory variables, tumor diameter and age, were considered in both the categorized and the continuous forms. In the latter case non-linear functions were also considered. The estimated parameter $\beta$ shows the effect on the log odds when the patient has axillary nodes in conjunction with an increase in the respective explanatory variables. The odds ratio (OR) associated with a unit change in the variable is $\exp(\beta)$. The standard errors of the $\beta$-parameters were obtained from the inverted observed information matrix evaluated at the ML-estimate. The ratio between an estimated $\beta$-parameter and its standard error is asymptotically normally distributed when the true value of the parameter is 0. This result was used in all hypothesis testing (Wald-tests). Thus the test for an effect of the second-order term of age was obtained as $\beta/SE(\beta)$ $(-0.00080/0.00039 = 2.05)$, which implies a p-value of 0.04. All tests were two-sided. Confidence intervals were constructed according to the same basic principle.

## Results

The probability that the patient might have axillary node metastases increased with tumor diameter (Table 1). The mean diameter was not related to age, but there was one exception from this: a slightly higher mean diameter was found in women over 80 years of age (Table 2). The mean values for estrogen receptor content in the different age groups increased with age (Table 2). The distribution of the Ackerman's classification was similar over the age groups (Table 3).

Multivariate models were fitted for the presence or the absence of axillary node metastases as the dependent dichotomous outcome (Table 4). In the first model (Model 1 in Table 4) the probability of having axillary metastases was analyzed with age and tumor diameter as explanatory variables in categorized form. The probability of having nodal involvement increased in proportion to tumor di-

**Table 1**

*Distribution of tumor diameter and number of women with axillary node metastases in each stratum in women whose axillary status was known*

| Tumor diameter, mm | No. with positive metastases / Total No. in stratum | Percentage |
|---|---|---|
| 0–9 | 14/74 | 18.9 |
| 10–19 | 84/304 | 27.6 |
| 20–29 | 100/226 | 44.3 |
| 30–39 | 48/73 | 65.8 |
| ⩾40 | 35/48 | 72.9 |
| Total | 281/725 | 38.8 |

**Table 2**

*Age distribution and mean values of diameter and estrogen receptor content in women whose axillary status and tumor diameter were known*

| Age, years | Mean tumor diameter (SE) | | Mean estrogen receptor content | |
|---|---|---|---|---|
| | n | mm | n | fmol/μg DNA (SE) |
| <40 | 56 | 21.4 (1.68) | 51 | 0.31 (0.06) |
| 40–49 | 103 | 22.3 (1.27) | 101 | 0.27 (0.05) |
| 50–59 | 131 | 22.4 (1.00) | 120 | 0.64 (0.11) |
| 60–69 | 175 | 22.4 (0.87) | 164 | 0.91 (0.14) |
| 70–79 | 174 | 21.6 (0.71) | 162 | 1.14 (0.12) |
| ⩾80 | 86 | 24.6 (1.25) | 86 | 1.11 (0.19) |
| Total | 725 | 22.4 (0.42) | 684 | 0.81 (0.06) |

ameter. The probability also varied with age, but not in a linear fashion. The highest risk was seen in the 50–59-year age group and thereafter it definitely diminished (Table 4). The risk was also lower among the youngest patients.

In another multivariable model histopathological classification, a stronger receptor status, tumor diameter, and finally nuclear DNA content (Model 2, Table 4) were taken into account. In these models, diameter and age were used in the same categorized form as in Model 1. The parameter estimates for the different age groups now all tended to be lower than in the 40–49-year reference group.

In further models including age and diameter as continuous variables, age was best fitted as a second degree polynomial, which corresponds to the pattern displayed in the Figure. The second-order term of age was statistically significant ($p = 0.04$), which means that there is a non-linear effect of age on the risk of metastatic spread to the axilla. In all models, an increasing tumor diameter was found to entail a highly significant increased risk of having axillary metastases. Having a tumor of type Ackerman III:1 as well as II/III:2 entailed a significantly lower probability of axillary node metastases than having an Ackerman IV tumor.

The combined effect of age and diameter in categorized form is further illustrated in Table 5 and the Figure, where

**Table 3**

*Distribution of histopathological type according to Ackerman in the different age groups in all classified patients*

| Age, years | Ackerman's classification | | | | | | |
|---|---|---|---|---|---|---|---|
| | n | II/III:2 | | III:1 | | IV | |
| | | n | (%) | n | (%) | n | (%) |
| <40 | 73 | 7 | (9.6) | 42 | (57.5) | 24 | (32.9) |
| 40–49 | 113 | 8 | (7.1) | 77 | (68.1) | 28 | (24.8) |
| 50–59 | 146 | 11 | (7.5) | 91 | (62.3) | 44 | (30.1) |
| 60–69 | 205 | 12 | (5.9) | 124 | (60.5) | 69 | (33.7) |
| 70–79 | 194 | 8 | (4.1) | 126 | (65.0) | 60 | (30.9) |
| ⩾80 | 92 | 7 | (7.6) | 63 | (68.5) | 22 | (23.9) |
| Total | 823 | 53 | (6.4) | 523 | (63.6) | 247 | (30.0) |

**Table 4**

*Odds ratio (OR) estimates and 95% confidence intervals (CI) in logistic regression models of the risk of having metastases in axillary nodes. In both models age and diameter were used in categorized form*

| | Model | |
|---|---|---|
| | 1 (n = 725) OR (95% CI) | 2 (n = 485) OR (95% CI) |
| Age, years | | |
| <40 | 0.78 (0.38–1.61) | 0.43 (0.17–1.07) |
| 40–49 | ref. | ref. |
| 50–59 | 1.12 (0.65–1.93) | 0.85 (0.41–1.80) |
| 60–69 | 1.04 (0.61–1.77) | 0.80 (0.41–1.56) |
| 70–79 | 0.60 (0.35–1.02) | 0.43 (0.21–0.88) |
| ⩾80 | 0.54 (0.29–1.02) | 0.50 (0.22–1.11) |
| Diameter, mm | | |
| <10 | 0.22 (0.10–0.50) | 0.32 (0.12–0.84) |
| 10–19 | 0.52 (0.36–0.76) | 0.45 (0.28–0.74) |
| 20–29 | ref. | ref. |
| 30–39 | 2.08 (1.27–3.39) | 2.03 (1.07–3.88) |
| ⩾40 | 3.25 (1.77–5.98) | 3.07 (1.37–6.85) |
| Ackerman's classification | | |
| II/III:2 | | 0.02 (0.00–0.14) |
| III:1 | | 0.28 (0.18–0.43) |
| IV | | ref. |
| ER receptor fmol/μg DNA | | 0.93 (0.78–1.11) |
| DNA I | | 0.37 (0.10–1.38) |
| II | | 1.00 (0.59–1.69) |
| III | | 2.48 (0.54–11.46) |
| IV | | ref. |

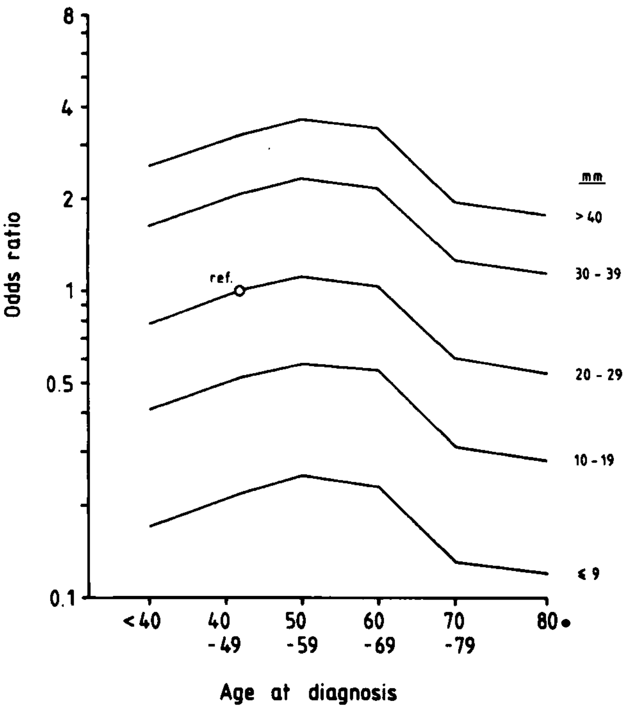


*Figure.* Model derived odds ratios, by age group and tumor diameter, of having axillary metastases at the time of diagnosis. Reference category is women aged 40–49 years with tumor 20–29 mm in diameter.

the parameter estimates from the statistical model are shown as odds ratios for nodal tumor growth. In all age groups with a tumor size less than 10 mm and in the lower (<40) and upper (>70) age groups with a tumor size of 10–19 mm, the relative risk of lymph node involvement was lower than that in the reference category (i.e. women aged 40–49 years with tumor diameter 20–29 mm) at the 5% level of statistical significance. The tumors measuring over 30 mm were associated with a statistically significant increase in the odds ratio in the 40–69-year age groups. In each stratum defined by tumor diameter, an approximately two-fold difference was revealed in the probability of having nodal metastases at different ages.

## Discussion

The probability of having axillary metastases given a certain tumor size was highest in the 50–59-year age group in an analysis taking age groups in categorized form into account. In multivariate models adjusted for histopathological classification, estrogen receptor status and DNA ploidy, the probability was highest in the 40–49-year age group. Distinctly lower probabilities were seen in women under 40 and over 70 years of age, whereas patients in the age span 40–69 had rather similar odds ratios. The parameter estimates for the association between nodal status and age increased when controlling for confounders, i.e. histopathological classification, receptor status, and DNA ploidy. This suggests that this is a real association rather than an artifact. When analyzed as a continuous variable, age was best fitted as a second order term and it was then an independent risk factor in a model where diameter, histopathological classification and estrogen receptor content were taken into account. The risk of the patient's developing tumor growth in the axilla increased sharply with diameter. The presence of a less differentiated tumor was also a significant risk factor.

The patient series is consecutive. Only women from the primary catchment area in the city of Uppsala were included and the patient material is thus population-based. All data were prospectively collected. We have avoided to

**Table 5**

*Odds ratio (95% confidence interval) of having axillary node metastases at a given tumor diameter according to age group*

| Diameter, mm | Age, years | | | | | |
|---|---|---|---|---|---|---|
| | <40 | 40–49 | 50–59 | 60–69 | 70–79 | ≥80 |
| ≤9 | 0.17 (0.06–0.50) | 0.22 (0.10–0.50) | 0.25 (0.09–0.66) | 0.23 (0.09–0.60) | 0.13 (0.05–0.36) | 0.12 (0.04–0.35) |
| 10–19 | 0.41 (0.18–0.91) | 0.52 (0.36–0.76) | 0.58 (0.30–1.13) | 0.55 (0.29–1.04) | 0.31 (0.16–0.62) | 0.28 (0.13–0.61) |
| 20–29 | 0.78 (0.38–1.59) | 1.0 ref. | 1.11 (0.64–1.93) | 1.04 (0.62–1.76) | 0.60 (0.35–1.02) | 0.54 (0.29–1.02) |
| 30–39 | 1.63 (0.69–3.83) | 2.08 (1.28–3.39) | 2.32 (1.10–4.86) | 2.17 (1.07–4.43) | 1.25 (0.61–2.56) | 1.13 (0.51–2.53) |
| ≥40 | 2.54 (1.01–6.42) | 3.26 (1.78–5.96) | 3.63 (1.59–8.26) | 3.40 (1.53–7.58) | 1.96 (0.85–4.48) | 1.77 (0.74–4.25) |

quantify the degree of nodal involvement retrospectively, since in this first exploratory analysis we wanted to see if there was any qualitative relation at all, and this is also the question of first concern. Elderly women may have been treated more often with procedures that do not permit investigation of the axillary nodes, but all women without information about the axillary findings were excluded from this analysis. Once investigated, the axillary contents were handled identically in the different age groups. While the data for this study were being collected, mammographic screening was not implemented in any of the age groups. Therefore we could find no bias to explain the relation between age and the risk of developing axillary metastases, as revealed in this analysis. There might have been different breast cancer awareness and behavior related to breast cancer detection in different age groups. This is most likely, however, to have led to earlier detection in younger age groups thus biasing the results towards unity. The lack of information concerning ER and DNA-ploidy for some tumors at the early part of the recruitment period reduces the amount of person-years to be used in the analysis. However, this loss is unlikely to introduce any bias to the results.

To our knowledge, the hypothesis discussed in this study has not been analyzed before. The axillary status at the time of treatment of breast cancer has been taken as an indicator of the risk that distant micrometastases already exist (18, 19). The findings concerning age and prognosis in several studies (1–5), indicate that the youngest and the oldest women have the highest frequency of distant metastatic disease since they have the poorest prognoses. If axillary metastases reflect the metastatic potential of breast cancer with the same predictive value at all ages, one would have expected a pattern pointing in a direction opposite to that indicated by the present study. Thus, one hypothesis that this study can generate is that the parallelism between the detection of metastases in lymph nodes and at distant sites might vary with age. The curve in the Figure seems to have a breaking point in the years around the menopause. It is possible that endocrine changes—or other events linked to the age at menopause—have a significant impact on the natural course of female breast cancer (20). This may occur, due to, e.g., endocrine regulation of tumor characteristics and/or defense mechanisms against disseminated tumor cells both in lymph nodes and at more distant sites. In males with breast cancer, the absence of a survival pattern related to age at diagnosis similar to that in female breast cancer indirectly increases the plausibility of this hypothesis (21). The pattern seen in the Figure may, indeed, result from two superimposed curves: one for an increasing risk of axillary metastasis with age in premenopausal breast cancer and the other for a decreasing risk with age in postmenopausal disease. In any case, these findings warrant a further study of the prognostic significance of axillary metastases in various age groups.

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