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IMPORTANCE OF TUMOR CELLS IN AXILLARY NODE SINUS MARGINS ('CLANDESTINE' METASTASES) DISCOVERED BY SERIAL SECTIONING IN OPERABLE BREAST CARCINOMA

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Abstract

The prognostic implications of small emboli of carcinoma cells in the sinus margins of axillary lymph nodes ('clandestine' metastases) discovered by serial node sectioning at 2 mm intervals was analysed. All patients, previously untreated, were admitted between 1967 and 1978 and underwent mastectomy and axillary node dissection. Our study examined the risk of distant metastases of 1153 patients with from 0 to 3 involved axillary lymph nodes. A Cox multivariate analysis was performed, taking into account the classical prognostic factors (menopausal status, histoprognostic grade, and anatomic tumor size), and for nodal status including the notion of clandestine (CM) or parenchymal metastases (PM). Compared to patients without axillary metastases, patients with one node involved with CM had a relative risk of distant metastases of 1.7, identical to the risk for patients with one node with PM; and patients with one node containing PM and a second CM, had a relative risk of 2.2. Serial node sectioning discovers nodal metastases that would otherwise not be detected. These CM have important clinical implications and should be taken into account when considering adjuvant manipulations.

Key words: Breast cancer, axillary lymph nodes, sinus margin metastasis, prognosis.

It has long been known that axillary lymph node metastases in operable breast carcinoma are associated with poorer prognosis (12, 17, 20, 23). More recently the quantification of this characteristic has assumed important therapeutic implications (5). With this background it is important to understand what is meant by the terms axillary invasion (N+) or lack of axillary invasion (N−).

Clinical examination is quite a crude predictor of axillary invasion as shown by us and others. In our study approximately 40% of axillae, classified as N0 clinically (UICC TNM classification), had positive nodes confirmed histologically, while 36% classified clinically as Nl were histologically negative. Also frozen section examination of axillary lymph nodes may underestimate the presence of metastases compared to the definitive histology (6).

The clinical significance of occult axillary lymph node metastases is somewhat controversial. Some authorities feel that nodal metastases undetected by a rather gross single section technique do not constitute an additional hazard compared to a node negative status (1, 13, 18, 20).

The present study was performed in order to evaluate the prognostic influence of micrometastasis detected in the subcapsular marginal sinuses of axillary lymph nodes ('clandestine' metastases) by a technique of macroscopic serial node sectioning.

Material and Methods

Between 1967 and 1978, 2095 patients with 'operable' breast cancer were treated at the Institut Gustave Roussy (IGR). Among these, 1153 patients had unilateral, operable, infiltrating breast carcinoma [≤7 cm in clinical diameter, without clinically fixed lymph node metastases, inflammatory signs or rapid clinical growth (PEV 0)], and with either negative or 1–3 positive axillary nodes confirmed by histologic means after resection of at least 6 nodes. All patients were treated entirely at IGR, according to specific protocols (19). All underwent mastectomy and axillary dissection, sometimes associated with internal mammary node dissection. According to protocol (10) patients with positive axillary nodes were postoperatively usually treated with radiotherapy at a dose of 45 Gy locoregionally. In contrast, patients with only one CM re-

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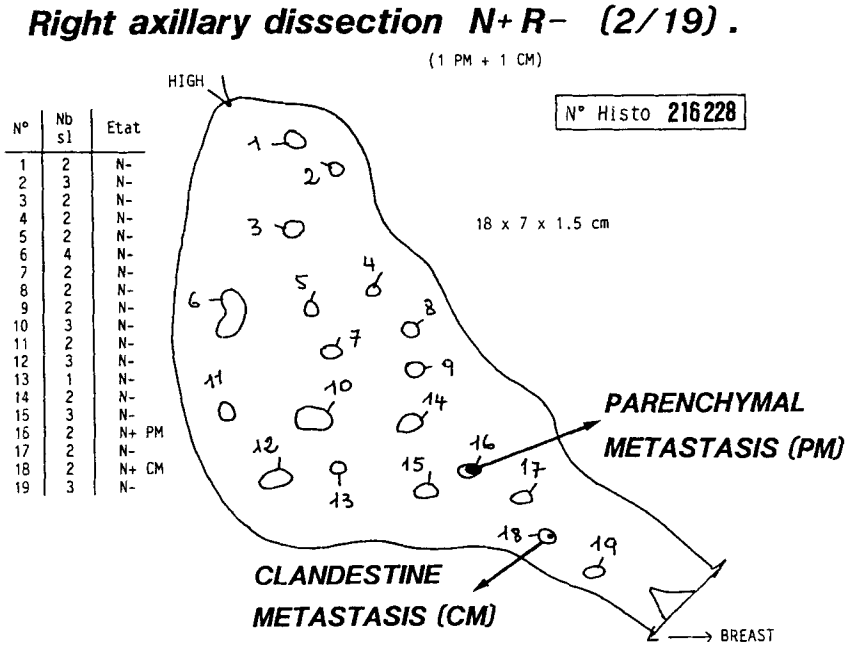

Fig. 1. Dissection of axillary fat pad with lymph nodes.

ceived no radiotherapy. No patient in this series received adjuvant chemotherapy. The median follow-up of this group is 118 months.

A mean of 15 lymph nodes per axilla (range 6–45) were examined per patient. All nodes were examined systematically regardless of how many nodes that were found to contain metastasis.

Since 1954 all nodes have been routinely dissected and examined at IGR in a systematic fashion (25). The axillary contents are first fixed for 24 h in Bouin's solution which colors lymphoid tissue intensely yellow, contrasting this with the transparent adjacent fat. This entire tissue mass, after being oriented and sketched, is then sectioned into large slices of about 2–5 mm thickness. All lymph nodes thus transected are consecutively numbered on the sketch with all pieces of the same node bearing the same number. The lymph node sections are then dissected from the fatty mass and, if possible, the larger sections further sectioned into slices of 1.0–1.5 mm thickness. All sections for each individual lymph node are then placed in a non-overlapping fashion into the same cassette. These nodal slices are thereafter dehydrated, degreased, mounted in paraffin, stained with Hematoxylin-Eosin-Safranin and examined microscopically. Each node found to contain metastasis is identified on the initial scheme and the total axillary metastatic status thereby assessed (Fig. 1). This method is quite different from both the toluene clearing method by some considered to be toxic and impractical and the 'single section method' recommended by most U.S. centers (14) and used in the majority of laboratories (Fig. 2).

Small emboli of tumor cells in the sinus margin of axillary nodes were termed 'clandestine metastasis' (CM) (a term coined by G. Vogt-Hoerner), while a metastasis in the lymph node parenchyma was scored as 'parenchymal metastasis' (PM) (Fig. 3).

The different clinical and histological data were coded and stored in a computer data program developed at IGR (PIGAS). This data base was then used to estimate and

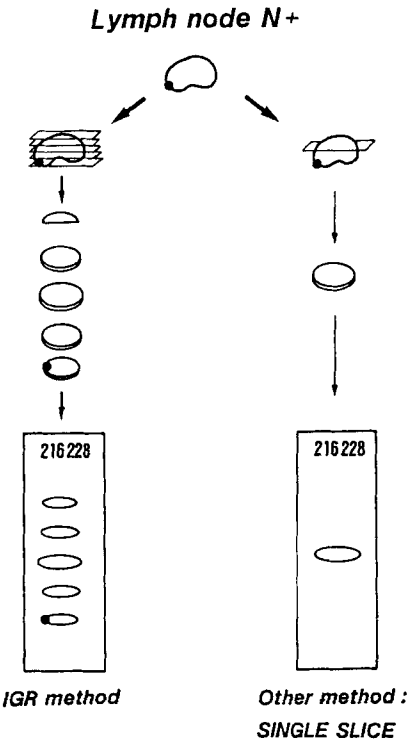

Fig. 2. IGR serial sectioning method vs standard single section technique.

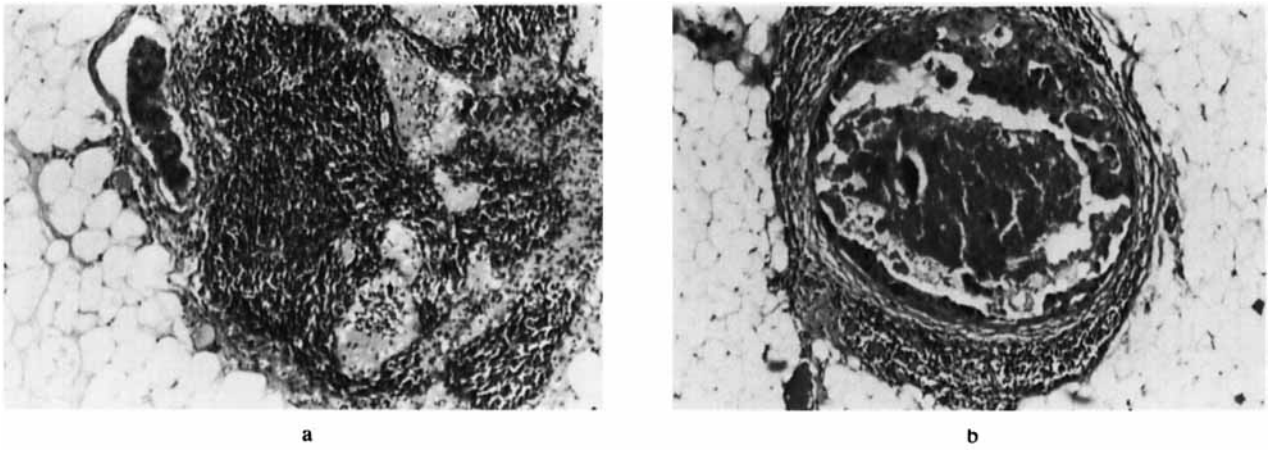

Fig. 3. a) Clandestine metastasis and b) parenchymal metastasis in axillary lymph nodes.

compare the risk of distant metastatic relapse (based on the metastatic-free survival, MFS) in groups with different degrees of axillary lymph node involvement. For this purpose, a Cox' multifactorial analysis (9) was used and included previously known prognostic factors (age, menopausal status, histopathologic grade, anatomic tumor size) and histologic axillary node status subdivided into no metastases, one CM only, one PM only, one CM plus one PM, 2 PM and 3 PM. Only 2 patients had one CM plus 2 PM and they were not included as a separate group.

In a previous study (7) we compared the result of histopathologic examination after conventional sectioning (one large section per lymph node) with the results obtained after our serial sectioning technique. In 9.4% (43/456) of the cases, serial sectioning disclosed additional metastases in nodes previously termed negative. Thus included 15 of 252 cases classified as entirely node negative (N-) by single section method. There was an obvious relation between the number of sections examined and nodal size. Nodes less than 5 mm were sectioned into a mean of 2.4 slices, while those of 15–20 mm into a mean of 4.7 slices.

Results

In the present material, 637 patients (55%) had no observed axillary metastasis; 41 (3.5%) had one CM, 205 (18%) had one PM, 40 (3.5%) had one CM plus one PM, 126 (11%) had 2 PM, and 104 (9%) 3 PM.

Table 1 shows the distribution of histologic types of carcinoma in the different lymph node groups. No lobular carcinomas were present in the CM group compared to 6% in the PM groups. The only other type represented in the CM group was 2 typical medullary carcinomas (2 cases = 2–3% similar to the PM group). The PM group in addition to 18 typical medullary carcinomas, contained 2 apocrine, 3 squamous cell and 6 mucoid carcinomas.

More advanced histologic grades (II and III) were somewhat more frequent in the CM than in the PM group ($p=0.01$) (Table 1).

Table 2 shows the results of the multivariate analysis for distal metastasis risk with the objective of showing relative risk for different groups of axillary lymph node metastases, after adjustment for the classic prognostic factors of age, menopause, histoprognostic grade and anatomic tumor size. The presence of a CM in one axillary node produced an increased risk of distant relapse (RR 1.7) compared to the group with no identified metastases in axillary lymph nodes ($RR=1$) ($p=0.05$). This risk was identical to that of the group of patients with one PM identified in the axillary nodes (and similar to other combinations of from 1 to 3 axillary node metastases).

In addition, although the group with only one CM received no locoregional irradiation in contrast to the one PM and more advanced groups, the rate of local relapse was not significantly increased. Thus no group with any degree of nodal involvement had an increase of locoregional metastases compared to the N- group.

Discussion

The number of 'positive' nodes depends upon the number of nodes removed and examined, and therefore upon the surgeon's and/or pathologist's skill in dissection and evaluation. These facts undoubtedly explain some prognostic variations in breast cancer as reported in the literature (12).

The technique of serial macroscopic sectioning used at IGR is a practical way to detect axillary lymph node metastases. It may be regarded as a compromise between simple techniques of a single large nodal slice, or sectioning only of nodes greater than 2 mm (1) or palpable nodes (3); and scientifically feasible, but practically impossible techniques, such as teasing apart each node in the search for metastases. To insure the detection of single cancer

Table 1

Histologic type and grade of carcinoma according to the nodal status (CM=clandestine metastasis; PM=parenchymal metastasis)

	Type of carcinoma			Histologic grade		
	Ductal	Lobular	Other types	I	II	III
PM only	984 (91%)	59 (6%)	29 (3%)	213 (21%)	486 (47%)	329 (32%)
CM or PM+CM	79 (97%)	0	2 (3%)	10 (12%)	51 (64%)	19 (24%)
Total	1 063	59	31	223	537	348

Table 2

Relative risk (RR) of distant metastases in groups with different axillary invasion and with N- as reference (RR=1). Cox' multivariate analysis including, besides nodal status, age, menopausal status, grade and anatomic tumor size. (CM=clandestine metastasis; PM=parenchymal metastasis; +1, +2 and +3 denote number of lymph nodes involved)

Type nodal metastasis	β*	β/SE**	p	RR
(N-)				1.00
(N+1) CM	0.60	2.05	0.05	1.70
PM	0.51	3.14	0.005	1.70
(N+2) CM+PM	0.79	2.87	0.01	2.20
2PM	0.59	3.19	0.005	1.80
(N+3)	0.36	1.64	0.10	1.40

* Coefficient of risk.
** Standard score.

cells with a mean diameter of 20 μ, serial sectioning into 250 slices of 5 mm node would be required but clearly impossible in practice. In the present study the additional relative benefit of immunohistochemical analysis to our technique was not evaluated. It is known, however, that nodal metastases overlooked by single slice techniques may be detected by this method (22). We are at present analyzing the value of immunohistochemistry when used in conjunction with our serial sectioning technique.

In our study, the presence of 1–3 involved axillary lymph nodes was associated with higher risk of relapse (compared to negative axilla) if the metastases were clandestine or parenchymal. We therefore disagree with the statement that nodal invasion found by means others than gross dissection are of no clinical importance. Patients with only one node involved with CM had in our study a risk of distant metastasis equal to patients with parenchymal metastases in 1–3 nodes, and the risk was even greater for patients with one CM plus one PM.

It is of interest that CM was not found in lobular carcinoma. Ductal carcinoma cells are larger than lobular carcinoma cells and perhaps the latter pass more easily, while ductal cancer cells get caught in the marginal sinus. There was a significant difference between PM and CM regarding distribution of histologic grades, with fewer grade I tumors in the CM group. However, the obtained p-value may be an artifact due to the small number of patients in the CM group and large number of patients in the PM group.

During the past 15 years most studies on the prognostic importance of axillary invasion have concerned 'macrometastasis' or the anatomic level of axillary involvement (1). Several studies of adjuvant chemotherapy suggest maximal effect in patients with less than 4 positive axillary nodes (4, 10). Since the risk of distant metastasis in the present study was similar in the CM or PM groups, and the pattern of nodal spread may be important (2, 15), we feel that the axillary nodal involvement of breast cancer should be determined as precisely as practically possible prior to application of different therapeutic modalities, such as adjuvant therapy.

As shown at IGR, serial node sectioning discovers occult nodal metastases in 9.4% of the cases that would otherwise have been undetected. In another center, a similar percentage of occult metastases was detected by a macroscopic serial sectioning technique similar to ours, and an even higher proportion of occult metastases was found when the entire nodal specimen was subjected to histologic sectioning with microscopic analysis of every tenth section (16). The clinical importance of these techniques should be further studied, but the presence of clandestine metastases seems to have important prognostic implications.

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