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Vertical growth phase and positive sentinel node in thin melanoma

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Abstract

Sentinel node (SN) status is the most important prognostic factor for localized melanoma. Usually, patients with Breslow thickness of less than 1.0 mm are not included in SN protocols. However, the literature presents a rate ranging from 3 to 7% of nodal recurrence in thin melanoma. Ulceration, regression and high mitotic rate have been considered to be indications for an SN biopsy. The metastatic potential of the vertical growth phase is uncertain. To correlate pathological features in thin melanoma with SN metastasis, we reviewed 358 patients submitted to SN biopsy. Seventy-seven patients with lesions of 1 mm or smaller were included in the study group. Histological evaluation of the primary tumor included thickness, Clark level, mitotic rate, ulceration, regression, and growth phase. Lymphoscintigraphy was performed on all patients. Lymphatic mapping and gamma probe detection were both used for SN biopsy. Histological examination of SN consisted of hematoxylin-eosin and immunohistochemical staining. Median follow-up was 37 months. Six patients had micrometastases. Statistical analysis by the Fisher test showed that ulceration (P = 0.019), high mitotic rate (P = 0.008) and vertical growth phase (P = 0.002) were positively correlated with micrometastases. If other studies confirm these results, more melanoma patients must be submitted to SN biopsy.

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The overall incidence of melanoma has been rising all over the world. The proportion of thin melanoma has increased as compared to more deeply infiltrating lesions (1-3). Sentinel node (SN) biopsy proved to be an accurate staging procedure of regional nodal status with minimal morbidity and SN status is the most important prognostic factor for localized melanoma (4,5).

What does thin metastasizing melanoma have that confers this phenotype? This is still an unsolved question. The latest proposed change of the tumor-node-metastasis (TNM) cutaneous melanoma classification includes the presence or absence of ulceration and Clark level in category "T" (6). Patients with thin primary melanoma (≤1 mm) without ulceration and with a Clark level of less than IV are expected to have an excellent prognosis (7). However, some of these patients with low risk disease develop recurrence and death. The literature presents a rate of nodal recurrence ranging from 3 to 7% in thin melanoma patients (8-11). Other pathologi-

cal features such as ulceration, mitotic rate, regression and vertical growth phase are supposed to play an important role in the determination of recurrence risk in thin melanoma and have been considered as indicators for an SN biopsy (12,13).

The early steps in the development and progression of malignant melanoma include proliferation of transformed epidermal melanocytes and their subsequent invasion of the papillary dermis (radial growth phase). This can be followed by the vertical growth phase, a step with metastatic capability (14). The identification of parameters that might predict progression and survival is mandatory for thin melanoma (1).

In the SN biopsy protocol of our institutions we have included melanoma patients with thin melanoma. Some of them have recurred. To address why these patients have recurred we analyzed the pathological features of their primary melanoma, including vertical growth phase.

Between June 1997 and January 2002, 358 patients with clinically localized cutaneous melanoma underwent a successful SN biopsy, according to protocols approved by the Ethics Committee of Escola Paulista de Medicina, UNIFESP, and Hospital Israelita Albert Einstein. Among them, 77 had lesions 1 mm or smaller and formed our study sample. Informed consent was obtained from all patients. Clinically node-negative melanoma patients were enrolled into our protocol if Breslow thickness was 1.0 mm or more and those with thickness of less than 1.0 mm with ulceration, more than 5 mitoses/mm^2, regression or Clark level IV or V were also included. The histological evaluation of the primary lesion included tumor thickness, Clark level, mitotic rate, ulceration, regression, vascular invasion, tumor-infiltrating lymphocytes, and growth phase. The presence of vertical growth phase was determined as described by Clark et al. (12).

Lymphoscintigraphy was performed in all patients. A quantity of 1 mCi (Tc^{99m}-dextran 500) in a 1-ml volume of 0.9% normal saline was injected intradermally in equal parts into four quadrants around the biopsy scar or primary tumor. The images were obtained with a gamma camera with a high-resolution collimator (Starcam 4000 600 X/RT). The projection of the SN was marked on the overlying skin with indelible ink. Surgery was performed 2 to 24 h after lymphoscintigraphy. For the first 43 patients the SN biopsy was based only on preoperative lymphoscintigraphy and vital blue dye mapping (Patent Blue V, Laboratoire Guerbet, Aulnay-sous Bois, France).

Intraoperative gamma probe detection (Neoprobe 1500, Columbus, OH, USA) was added to the protocol for the remaining 315 patients. The node was considered to be an SN if it was blue and/or showed counts five-fold higher than background *in vivo* or ten-fold higher *ex vivo*. After the SN was excised, the surgeon examined the operative field with the probe. If there was 10% or more capitation of the SN, the surgeon looked for another node. The SN was trimmed of excess fat and sectioned by the surgeon into four fragments in the operating room. Two noncontiguous fragments were sent to pathology and the others were submitted to molecular biology examination (RT-PCR for tyrosinase mRNA). The histological examination consisted of hematoxylin-eosin staining and immunohistochemical markers for S100 and HMB45. When hematoxylin-eosin and/or immunohistochemical study showed the presence of micrometastases in the SN, complete lymphadenectomy was performed. Patients with a negative SN were just followed up. The median follow-up was 37 months. Fisher's exact test was applied to determine the association between pathological features and the occurrence of a positive SN.

The primary site was localized on the head and neck, trunk and extremities in 14, 27 and 36 patients, respectively. The median age of the 77 thin melanoma patients was 52.5 years. There were 34 male and 43 fe-

male patients. Lymphoscintigraphy successfully identified the draining lymphatic basin and SN in all 77 patients. On the basis of vital dye and/or intraoperative gamma detection, 97.8% of the SN identified by lymphoscintigraphy were found and excised. Metastatic spread was diagnosed in six patients. We focused on these six patients comparing their pathological features with those of 71 thin melanoma patients who did not present micrometastases in the SN. Statistical analysis by the Fisher test showed that ulceration (P = 0.019), high mitotic rate (P = 0.008) and vertical growth phase (P = 0.002) were positively correlated with micrometastases (Table 1). All six patients with micrometastases showed the vertical growth phase in the primary lesion, five of them had more than 5 mitoses/mm^2, four of them had ulceration, and three had regression. Additional characteristics of these six thin melanoma patients who presented SN metastases are shown in Table 2.

The rate of occurrence of nodal metastases in our group of patients with thin melanoma (Breslow thickness 1 mm or smaller) was 7.8% (6/77). All six patients showed the vertical growth phase in their primary tumor. These six patients underwent complete lymphadenectomy and none of them showed another positive lymph node.

Previous publications have emphasized that tumors with vertical growth phase need to be considered as high risk for regional node metastasis (14,15). There is evidence that thin melanoma with radial growth phase does not metastasize (16,17). Our results support that vertical growth phase, high mitotic rate and ulceration are indications for SN biopsy. If other studies, with multivariant analysis, confirm our results, more melanoma patients must be submitted to SN biopsy (1,18).

Table 1. Sentinel node micrometastasis and pathological features in 77 thin melanoma patients (≤1.0 mm).

	Micrometastasis		P*
	No (N =71)	Yes (N =6)	
Clark			0.376
III	35	2	
IV	36	4	
Ulceration			0.019
Yes	13	4	
No	58	2	
Regression			0.084
Yes	12	3	
No	59	3	
>5 mitoses/mm^2			0.008
Yes	18	5	
No	53	1	
VGP			0.002
Yes	22	6	
No	49	0	

VGP = vertical growth phase.
*Fisher test.

Table 2. Characteristics of the six thin melanoma patients with sentinel node metastasis.

Patient	1	2	3	4	5	6
Sex	F	F	F	M	M	F
Age (years)	57	38	74	42	48	56
Site	Trunk	Trunk	Extremity	Extremity	Trunk	Trunk
Type	Nodular	SSM	SSM	SSM	Nodular	SSM
Breslow	0.8	0.5	0.6	0.4	0.9	0.8
Clark	IV	III	IV	III	IV	IV
Ulceration	No	Yes	Yes	Yes	No	Yes
Regression	Yes	Yes	No	No	Yes	No
>5 mitoses/mm^2	Yes	Yes	No	Yes	Yes	Yes
VGP	Yes	Yes	Yes	Yes	Yes	Yes

SSM = superficial spreading melanoma; VGP = vertical growth phase; F = female; M = male.

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