Immunohistochemical Evaluation of p16\textsuperscript{INK4A}, E-Cadherin, and Cyclin D1 Expression in Melanoma and Spitz Tumors

Evan George, MD, 1 Nayak L. Polissar, PhD, 2 and Mark Wick, MD 3

Key Words: Melanoma; Spitz nevus; p16; Cyclin D1; E-cadherin; Immunohistochemistry; Large spindle and epithelioid nevus; Spitz tumor

Abstract

We evaluated the usefulness of immunohistochemical examination for E-cadherin, p16, and cyclin D1 in discriminating melanoma from Spitz tumors. Immunoperoxidase staining was performed on formalin-fixed tissue specimens from 46 Spitz tumors and 42 concurrent melanoma specimens. The percentages of immunoreactive melanocytes in the epidermis and dermis were estimated semiquantitatively. Qualitatively abnormal immunoreactivity patterns were also tabulated. Dermal p16 immunoreactivity was the best quantitative discriminator: decreased nuclear immunoreactivity (<25% of dermal melanocytes) was 3-fold more likely in melanoma than in Spitz tumors (P = .004). Loss of both nuclear and cytoplasmic dermal p16 immunoreactivity was 8-fold more likely in melanoma (P = .01). Qualitative irregularities in the zonal distribution of E-cadherin immunoreactivity were 2-fold higher in melanoma (P = .01), but these were often focal or subtle. There was no statistically significant difference in cyclin D1 immunoreactivity. In atypical Spitz tumors, the dermal p16 immunoreactivity and frequency of qualitative E-cadherin abnormalities were intermediate between those of ordinary Spitz nevi and melanoma. Also, contrasting immunoreactivity patterns were helpful in determining Breslow thickness in specimens containing melanoma and contiguous dermal nevi.

As the molecular mechanisms involved in the genesis and progression of melanoma become better elucidated, there are new opportunities for pathologists to use this information in the diagnostic evaluation of melanocytic neoplasms. Although techniques for directly examining the genome and identifying specific gene mutations are not available in most pathology practices, immunohistochemical evaluation of specific gene products is feasible in most pathology practices. The primary objective of this study was to explore the potential diagnostic usefulness of immunohistochemical examination of 2 important regulators of cell cycle progression (p16 and cyclin D1) and 1 important mediator of intercellular adhesion (E-cadherin) in the frequently problematic distinction of Spitz tumor (ST) from malignant melanoma (MM). A secondary objective was to evaluate the immunophenotypic features of histologically atypical Spitz tumors for comparison with ordinary Spitz nevi (SN) and MM.

Materials and Methods

Case Selection

This study was approved and granted a waiver of patient consent by the Investigational Review Board of the University of Virginia Health System (Charlottesville). ST cases accessioned between 1983 and 2000 were identified by searching the electronic database of the division of surgical pathology at the University of Virginia. An approximately equal number of MM cases diagnosed during the same period were also identified. In selecting MM cases, we included as many young patients as possible. Cases of melanoma of sun-damaged skin (lentigo maligna melanoma) in elderly patients were generally avoided because the clinical and histologic features do not
significantly overlap with those of SN. Cases with insufficient quantities of paraffin-embedded tissue for all of the immunohistochemical stains were excluded.

Demographic and Clinical Data
The age, sex, and anatomic site of the lesion were ascertained from the archival surgical pathology reports. For 10 cases of ST, the age of the patient could not be ascertained.

Histologic Examination
For purposes of this study, histologic diagnosis was made from one author’s (E.G.) review of the original H&E-stained sections in conjunction with pertinent demographic and clinical information ascertained from the available surgical pathology report. Cases were classified as ST or MM using current criteria.1,2 The STs included ordinary SN and STs with atypical features. Atypical features included asymmetry, poor lateral demarcation, nonuniform cytoplastic atypia, mitotic activity in the lower dermal melanocytes, and prominent pagetoid spread. STs were further segregated into junctional, dermal, and compound types.

MM specimens were subclassified as superficial spreading melanoma, acral-lentiginous melanoma, nodular melanoma, lentigo maligna melanoma, and melanoma, not otherwise specified. The designation of “spitzoid melanoma” was applied to malignant neoplasms exhibiting some cytologic and architectural features characteristic of SN: large spindle or epithelioid cells with eosinophilic cytoplasm, and prominent pagetoid spread. STs were further segregated into junctional, dermal, and compound types.

MM specimens were subclassified as superficial spreading melanoma, acral-lentiginous melanoma, nodular melanoma, lentigo maligna melanoma, and melanoma, not otherwise specified. The designation of “spitzoid melanoma” was applied to malignant neoplasms exhibiting some cytologic and architectural features characteristic of SN: large spindle or epithelioid cells with eosinophilic cytoplasm, tendency for vertical orientation of junctional nests, retraction of junctional nests from adjacent epidermis, Kamino bodies, and inverted wedge-shaped dermal-based lesions. The Breslow depth and Clark level of invasion were determined from the available H&E-stained histologic sections. MM specimens were further subcategorized into radial growth phase and vertical growth phase using established criteria.3

Immunohistochemical Analysis
Immunoperoxidase staining was performed using a commercial automated device (Autostainer Plus, DAKO, Carpinteria, CA). Sections from formalin-fixed, paraffin-embedded tissue were mounted on charged slides. Epitope “retrieval” was performed by heating the slides in citrate buffer at pH 6.0. Sections were incubated with biotinylated antibodies to E-cadherin, p16, cyclin D1, and S-100 proteins for 30 minutes. An equal mixture of normal mouse ascites and normal rabbit serum was used as a negative control stain for each case. Tissues with known immunoreactivity for the aforementioned antigens were stained concurrently as positive control samples. After enzymatic incubation with streptavidin-peroxidase conjugate and 3,3’-diaminobenzidine-HCl, the sections were counterstained with hematoxylin or azure blue. The latter was applied to samples containing abundant melanin pigment to facilitate distinction of the brown immunoprecipitate product from endogenous melanin pigment, as described previously.4,5

The presence of S-100 immunoreactivity was used to confirm antigen preservation in the tissue sections. The immunostained sections were then reviewed by one of us (E.G.), who semiquantiatively estimated the percentage of positively staining tumor cells at scanning and intermediate magnification using a standard light microscope. Precise quantitation (ie, counting individual cells) was not performed. A positive interpretation required staining in the appropriate cellular compartment. For E-cadherin, nearly circumferential membranous or cytoplasmic staining was required. For cyclin D1, immunoreactivity was predominantly nuclear with variable cytoplasmic staining. For p16, nuclear and cytoplasmic staining were tabulated separately. Immunoreactivity was quantified separately for the epidermal and dermal components of individual lesions. When an associated nevus was observed in an MM specimen, the immunoreactivity of this cell population was not included.

Table 1

<table>
<thead>
<tr>
<th>Antibody</th>
<th>Clone</th>
<th>Dilution</th>
<th>Epitope Retrieval</th>
<th>Commercial Source</th>
</tr>
</thead>
<tbody>
<tr>
<td>S-100 proteins</td>
<td>Polyclonal</td>
<td>1:8,000</td>
<td>HIER; DAKO S169984</td>
<td>DAKO, Carpinteria, CA</td>
</tr>
<tr>
<td>E-cadherin</td>
<td>NCH-38</td>
<td>1:50</td>
<td>HIER; DAKO S169984</td>
<td>DAKO</td>
</tr>
<tr>
<td>p16 gene product</td>
<td>G175-405</td>
<td>1:1,000</td>
<td>HIER; DAKO S169984</td>
<td>Becton Dickinson, Franklin Lakes, NJ</td>
</tr>
<tr>
<td>Cyclin D1</td>
<td>SP4</td>
<td>1:50</td>
<td>HIER; DAKO S169984</td>
<td>Lab Vision, Fremont, CA</td>
</tr>
<tr>
<td>Normal mouse ascites</td>
<td></td>
<td>Various</td>
<td>Various, none</td>
<td>DAKO</td>
</tr>
<tr>
<td>Normal rabbit serum</td>
<td></td>
<td>Various</td>
<td>Various, none</td>
<td>DAKO</td>
</tr>
</tbody>
</table>

HIER, heat-induced epitope retrieval.

* DAKO S169984 is Target retrieval solution with citrate buffer, pH 6.0.

† An equal mixture of normal mouse ascites and normal rabbit serum used together.
Image 1  E-cadherin. A, Junctional Spitz nevus (H&E, ×100). B, Nests in junctional Spitz nevus showing uniform E-cadherin staining (anti–E-cadherin, ×100). C, Compound (predominantly dermal) Spitz nevus (H&E, ×100). D, Compound Spitz nevus with E-cadherin staining in superficial dermal nests but not in the deeper dermal component (anti–E-cadherin, ×100). E, Melanoma (H&E, ×100). F, Melanoma showing qualitatively abnormal E-cadherin staining. The junctional zone melanocytes include areas with E-cadherin immunoreactivity (arrows) juxtaposed to nests devoid of staining (arrowheads). In contrast, the deeper dermal component shows more uniform staining (anti–E-cadherin, ×100).
tabulated and analyzed separately for both microanatomic components (ie, epidermis and dermis). Thus, cases without 1 of the 2 microanatomic components were excluded from the data analyses of the missing parameter in single-parameter analysis. All such cases, however, were included in the analysis of combined parameters, where qualitatively abnormal E-cadherin immunoreactivity in the epidermal or the dermal component was sufficient for categorization as “positive.”

Likewise, high levels of cyclin D1 immunoreactivity have been observed in the epidermal component of SN, with cells of the dermis showing gradually diminishing staining with descent. In contrast, the dermal component of melanoma often has a more uniform distribution of cyclin D1 immunoreactivity that is not diminished with increasing depth. Therefore, lack of diminishing cyclin D1 immunoreactivity with dermal depth was considered qualitatively abnormal \textbf{Image 2}. In general, this parameter could not be assessed in tumors limited to the epidermis or not filling the papillary dermis (ie, Clark level I or II), in superficial shave biopsy specimens, and in neoplasms with sparse dermal cellularity. Such cases were therefore not included in the statistical analysis of this parameter.

\textbf{Test Performance Analysis}

To evaluate the potential diagnostic usefulness of the 3 immunostains, basic epidemiologic models frequently used to evaluate the effectiveness of clinical laboratory tests were...
applied. By using the histologic diagnosis as the reference standard, we calculated the sensitivity, specificity, and positive likelihood ratio for the immunostains individually and in selected combinations. The sensitivity for correctly identifying MM was determined by dividing the number of true MM cases correctly identified immunohistochemically by the total number of MM cases studied. Specificity was defined as the number of nonmelanoma cases correctly classified as negative immunohistochemically divided by the total number of nonmelanoma cases studied. The positive likelihood ratio was calculated as follows: sensitivity/(100 – specificity).

Statistical Analysis

$P$ values for the differences in immunoreactivity scores were calculated using the 2-sample $t$ test (ST vs MM) or the Spearman correlation coefficient (SN vs atypical ST vs MM, treated as ordered), whereas $P$ values for the presence of qualitatively abnormal immunoreactivity were calculated using the $\chi^2$ test (ST vs MM) or the $\chi^2$ test for trend (SN vs atypical ST vs MM).

For the association of tumor type with immunoreactivity scores at specific cutoff levels, the $\chi^2$ test was used to calculate $P$ values. These $P$ values are post hoc, anticonservative because cut points in immunoreactivity findings were determined in relation to the outcome before application of the $\chi^2$ test.

Results

Demographics and Clinical Features

Clinical features are shown in Table 2. Patients with ST were younger than patients with MM (median age, 18 vs 40 years). There was a strong female predilection in ST (3.2:1) in contrast with a slight male predominance in MM (1.4:1). Patients with atypical STs were of intermediate age (median, 28 years) with a strong female predilection, comparable to SN (3.8:1). The majority of the 46 STs occurred on the extremities (26 [57%]) with lesser numbers on the head and neck (10 [22%]) and the trunk (10 [22%]). The site distributions of SN and atypical STs were similar. Most of the 42 MM lesions were located on the trunk (19 [45%]) or extremities (18 [43%]). Only 5 (12%) of the MMs occurred in the head and neck region.

Histologic Findings

Based on review of H&E-stained sections, there were 46 STs (14 junctional, 12 dermal, and 20 compound) and 42 MMs. One or more atypical histologic features were observed in 19 STs. Nearly two thirds of MM cases were in vertical growth phase. The most common melanoma subtype was superficial spreading melanoma (21 [50%]). Of the MM cases, 6 (14%) were nodular melanoma. Spitzoid histologic features were prominent in 6 melanoma specimens (14%). Of the MMs, 9 (21%) were insufficiently distinctive for histologic subclassification. Breslow thickness ranged from 0.30 to 4.1 mm with a median of approximately 1.0 mm.

Semiquantitative Immunohistochemical Analysis

Statistically significant differences in the percentage of immunoreactive cells were observed only for dermal p16 expression, which was lower in MM than in ST. The mean percentage of dermal melanocytes showing nuclear staining in ST was 61% vs 40% in MM ($P = .01$). Likewise, the mean percentage of cells showing cytoplasmic p16 immunoreactivity was 73% in ST and 57% in MM ($P = .02$). Loss of p16 immunoreactivity had 2 patterns, the most common of which was loss of nuclear staining but retention of cytoplasmic staining. The second pattern was loss of p16 staining in
Tables 3 and 5. There was a trend for increased frequencies of qualitatively abnormal cyclin D1 expression from SN to atypical ST to MM; however, this did not reach statistical significance \((P = .1)\).

**Spitzoid Melanoma**

Although the small number of spitzoid melanoma specimens in this study precludes statistical analysis, the quantity of immunoreactivity and the frequency of qualitative abnormalities were similar to those observed in nonspitzoid melanoma. A continuum between atypical STs, spitzoid melanoma, and nonspitzoid melanoma was not demonstrated; however, these data are limited by the small number of spitzoid melanoma cases \((n = 6)\).

**Diagnostic Performance of Immunohistochemical Stains**

Loss of nuclear p16 immunoreactivity in dermal melanocytes (defined as nuclear staining in fewer than 25% of dermal melanocytes) was 48% sensitive and moderately specific (84%) for MM (Table 4). With loss of both nuclear and cytoplasmic p16 immunoreactivity at the same threshold, specificity was 97%, but sensitivity was only 25%. When more than 50% of dermal melanocytes showed loss of both nuclear and cytoplasmic p16 immunoreactivity, sensitivity for MM was 35% and specificity was 91%.

Qualitatively irregular E-cadherin expression in either the epidermal or dermal compartment was 45% sensitive and 80% specific for MM. Combinations of criteria for irregular E-cadherin immunoreactivity and loss of dermal p16 immunoreactivity only modestly increased sensitivity at the expense of lower specificity (data not shown).

**Secondary Observations (Melanoma With Associated Dermal Nevus)**

An incidental observation of this study was that in several of the melanoma specimens, contrasting immunoreactivity for 1 or more of the 3 moieties helped delineate melanoma cells

---

**Table 3**

**Quantitative Immunohistochemical Results**

<table>
<thead>
<tr>
<th>Antigen</th>
<th>Location</th>
<th>Spitz Tumors ((n = 46))</th>
<th>Spitz Nevi ((n = 27))</th>
<th>Atypical Spitz Tumors ((n = 19))</th>
<th>Melanoma ((n = 42))</th>
<th>(p^t)</th>
<th>(p^*)</th>
</tr>
</thead>
<tbody>
<tr>
<td>p16</td>
<td>Nuclear</td>
<td>50</td>
<td>48</td>
<td>52</td>
<td>44</td>
<td>.5</td>
<td>.5</td>
</tr>
<tr>
<td></td>
<td>Cytoplasm</td>
<td>55</td>
<td>52</td>
<td>60</td>
<td>55</td>
<td>1.0</td>
<td>.7</td>
</tr>
<tr>
<td></td>
<td>Nuclear</td>
<td>61</td>
<td>68</td>
<td>52</td>
<td>40</td>
<td>.010</td>
<td>.006</td>
</tr>
<tr>
<td></td>
<td>Cytoplasm</td>
<td>73</td>
<td>80</td>
<td>65</td>
<td>57</td>
<td>.02</td>
<td>.06</td>
</tr>
<tr>
<td>E-cadherin</td>
<td>Nuclear</td>
<td>68</td>
<td>68</td>
<td>60</td>
<td>76</td>
<td>.2</td>
<td>.9</td>
</tr>
<tr>
<td></td>
<td>Cytoplasm</td>
<td>41</td>
<td>40</td>
<td>42</td>
<td>49</td>
<td>3.3</td>
<td>.3</td>
</tr>
<tr>
<td>Cyclin D1</td>
<td>Nuclear</td>
<td>70</td>
<td>63</td>
<td>80</td>
<td>61</td>
<td>.1</td>
<td>.2</td>
</tr>
<tr>
<td></td>
<td>Cytoplasm</td>
<td>61</td>
<td>57</td>
<td>71</td>
<td>51</td>
<td>.1</td>
<td>.2</td>
</tr>
</tbody>
</table>

* Data are given as the mean percentage of immunoreactive tumor cells.

† Spitz tumors vs malignant melanoma calculated by using the 2-sample t test.

‡ Spitz nevi vs atypical Spitz tumors vs malignant melanoma calculated by using the Spearman correlation test.

© American Society for Clinical Pathology
Table 4
Test Performance of Stains in Distinguishing Melanoma From Spitz Tumors

<table>
<thead>
<tr>
<th>Criteria</th>
<th>Malignant Melanoma</th>
<th>Spitz Tumors</th>
<th>Sensitivity (%)</th>
<th>Specificity (%)</th>
<th>P*</th>
</tr>
</thead>
<tbody>
<tr>
<td>p16, % of immunoreactive cells</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nuclear p16, dermal component</td>
<td>40</td>
<td>32</td>
<td>48</td>
<td>84</td>
<td>.004</td>
</tr>
<tr>
<td>&lt;25</td>
<td>40</td>
<td>32</td>
<td>55</td>
<td>69</td>
<td>.04</td>
</tr>
<tr>
<td>&lt;50</td>
<td>40</td>
<td>32</td>
<td>25</td>
<td>97</td>
<td>.01</td>
</tr>
<tr>
<td>Nuclear and cytoplasmic p16, dermal component</td>
<td>40</td>
<td>32</td>
<td>35</td>
<td>91</td>
<td>.01</td>
</tr>
<tr>
<td>&lt;25</td>
<td>40</td>
<td>32</td>
<td>35</td>
<td>91</td>
<td>.01</td>
</tr>
<tr>
<td>&lt;50</td>
<td>40</td>
<td>32</td>
<td>35</td>
<td>91</td>
<td>.01</td>
</tr>
<tr>
<td>E-cadherin, irregular staining distribution</td>
<td>32</td>
<td>34</td>
<td>44</td>
<td>88</td>
<td>.004</td>
</tr>
<tr>
<td>Epidermal component</td>
<td>36</td>
<td>31</td>
<td>42</td>
<td>81</td>
<td>.06</td>
</tr>
<tr>
<td>Dermal component</td>
<td>42</td>
<td>46</td>
<td>45</td>
<td>80</td>
<td>.01</td>
</tr>
</tbody>
</table>

*These are post hoc, anticonservative P values calculated using the χ² test for the presence vs absence of the criterion in malignant melanoma vs Spitz tumors.

intermingled with benign nevus cells Image 4I. This observation may be of some usefulness in estimating the Breslow thickness in melanoma specimens containing an associated dermal nevus, in which the malignant cells and antecedent benign melanocyte populations are contiguous.

Discussion

The primary objective of this study was to evaluate the potential diagnostic usefulness of immunohistochemical analysis for p16, E-cadherin, and cyclin D1 in discriminating MM from ST. A secondary objective was to evaluate the

Table 5I
Qualitative Abnormalities in E-Cadherin and Cyclin D1 Immunoreactivity*

<table>
<thead>
<tr>
<th>Antigen/Microanatomic Location</th>
<th>Spitz Tumors</th>
<th>Spitz Nevi</th>
<th>Atypical Spitz Tumors</th>
<th>Melanoma</th>
<th>P†</th>
<th>P‡</th>
</tr>
</thead>
<tbody>
<tr>
<td>E-cadherin</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Epidermis</td>
<td>4/34 (12)</td>
<td>3/20 (15)</td>
<td>1/14 (7)</td>
<td>14/32 (44)</td>
<td>.004</td>
<td>.013</td>
</tr>
<tr>
<td>Dermis</td>
<td>6/20 (20)</td>
<td>2/18 (11)</td>
<td>4/13 (31)</td>
<td>15/26 (42)</td>
<td>.06</td>
<td>.03</td>
</tr>
<tr>
<td>Epidermis or dermis</td>
<td>9/46 (20)</td>
<td>4/27 (15)</td>
<td>5/19 (26)</td>
<td>19/42 (45)</td>
<td>.01</td>
<td>.007</td>
</tr>
<tr>
<td>Cyclin D1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dermis</td>
<td>9/22 (41)</td>
<td>3/10 (30)</td>
<td>6/12 (50)</td>
<td>16/27 (59)</td>
<td>.2</td>
<td>.1</td>
</tr>
</tbody>
</table>

* Data are given as proportion (percentage) of cases with an abnormal immunoreactivity pattern.
† Spitz tumors vs malignant melanoma calculated by using the χ² test.
‡ Spitz nevi vs atypical Spitz tumors vs malignant melanoma calculated by using the χ² test for trend.
immunophenotypic features of histologically atypical STs for comparison with SN lacking atypia and MM. A substantial number and variety of lesions in all 3 categories were studied with attention to the quantity and the qualitative patterns of immunoreactivity.

p16 (also known as INK4A or p16\textsuperscript{INK4A}) is the protein product of the CDKN2A familial melanoma gene. p16 inhibits the G\textsubscript{1} cyclin-dependent kinases (CDKs) 4 and 6. In the absence of functional p16, these CDKs phosphorylate and inactivate the retinoblastoma tumor suppressor gene product, resulting in cell cycle progression and, ultimately, increased cellular proliferation.\textsuperscript{8}

Ordinary nevi typically show strong nuclear and cytoplasmic immunoreactivity for p16.\textsuperscript{9} In the present study, decreased p16 immunoreactivity in the dermal melanocyte population was the best immunophenotypic discriminator of MM from ST. Notably, in the epidermal component, there was no statistically significant difference in p16 immunoreactivity between ST and MM. Thus, attention to microanatomy is essential in evaluating p16 expression in these melanocytic neoplasms.

The subcellular localization of p16 immunostaining also seems to be important. The loss of both nuclear and cytoplasmic p16 immunostaining was more specific for MM than the loss of nuclear staining alone; however, this added criterion lowered the sensitivity for identifying MM.

Cyclin D1 is also involved in regulation of the cell cycle. In contrast with p16, cyclin D1 promotes progression from the G\textsubscript{1} phase to the S phase of the cell cycle, leading to increased cell proliferation. It does so by activating CDKs 4 and 6, which then inactivate the retinoblastoma tumor suppressor gene. Thus, cyclin D1 and p16 have opposite effects on CDKs 4 and 6. Surprisingly, in the present study, the mean percentage of neoplastic melanocytes exhibiting cyclin D1 immunoreactivity was greater in ST than in MM; however, this difference was not statistically significant. Likewise, there was no statistically significant difference in the frequency of qualitatively abnormal cyclin D1 immunoreactivity between MM and ST.

E-cadherin (epithelial cadherin) is one member of a transmembrane protein family that contributes to intercellular adhesion through formation of adherens junctions. Decreased expression has been documented in vertical melanoma and may have a role in malignant progression; however, there are conflicting findings among different investigators.\textsuperscript{10-13} Recently, Krengel et al\textsuperscript{6} found decreased expression in the dermal component of benign melanocytic nevi and melanoma in a zonal pattern. They concluded that E-cadherin expression in melanocytic neoplasms is dependent on the microenvironment and may not contribute to malignant progression.

In the present study, there was no statistically significant difference in the percentage of melanocytes showing E-cadherin immunoreactivity between ST and MM. However, qualitative abnormalities in E-cadherin staining did have discriminatory value. These were characterized by variable E-cadherin immunoreactivity among neoplastic cells within the epidermis or at equivalent dermal depths. This finding was 45% sensitive and 80% specific in detecting MM; the association was statistically significant (P = .01). Qualitatively abnormal E-cadherin immunoreactivity was more robust and statistically significant when identified in the epidermal portion of a neoplasm (3.7 times more frequent in MM than in ST; P = .004) than within the dermal component (2.1 times more frequent in MM than in ST; P = .06). However, 2 caveats are in order: First, this finding was often focal. Second, melanocytes associated with adnexa are a potential pitfall. If their association with adnexa is not appreciated and their immunoreactivity is compared with nearby cells in the reticular dermis, a false-positive interpretation may be rendered. This may explain the lower specificity of qualitatively abnormal E-cadherin immunoreactivity in the dermal portion of neoplasms compared with the epidermal component. For these reasons, we suspect this finding will be less reproducible among different observers than semiquantitative estimation of p16 immunoreactivity.

Conclusions

Both quantitatively decreased p16 immunoreactivity in dermal melanocytes and qualitative irregularities in E-cadherin immunostaining may be of diagnostic usefulness in the distinction of melanoma from ST. However, we suspect that estimation of p16 immunoreactivity would have higher interobserver reproducibility.

Evaluation of cyclin D1 immunoreactivity does not seem to have discriminatory value in this context.

In atypical STs, the quantity of dermal p16 immunoreactivity and the frequency of qualitatively abnormal E-cadherin expression were intermediate between those of ordinary SN and melanoma, supporting the existence of a spectrum of neoplastic progression within the broad category of spitzoid neoplasms. The immunophenotypic features in spitzoid melanomas were similar to those of nonspitzoid melanoma; however, the data are limited by the small number of spitzoid melanoma specimens (n = 6).

In cases of melanoma with contiguous dermal nevus cells, each of the 3 markers, alone or in combination, may facilitate measurement of Breslow thickness.

\textit{From the 1Department of Pathology, University of Washington, Seattle; 2The Mountain-Whisper-Light Statistical Consulting, Seattle; and 1Department of Pathology, University of Virginia Medical Center, Charlottesville.}
References


First and Only FDA Cleared Digital Cytology System

Empower Your Genius With Ours
Make a Greater Impact on Cervical Cancer with the Advanced Technology of the Genius™ Digital Diagnostics System

Click or Scan to discover more