Melanocytes in nonlesional sun-exposed skin: A multicenter comparative study

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Background: There are limited data regarding melanocyte density and distribution on sun-exposed skin of the head and neck, in particular, comparing morphology (hematoxylin-eosin [H&E] staining) and immunohistochemistry (Melan-A staining) on formalin-fixed tissue. Furthermore, comparisons of melanocyte density between distinct geographic populations have not been made using these methods. This information would be useful for physicians who use histologic criteria to diagnose and treat lentigo maligna.

Objective: We aimed to characterize the density and distribution of melanocytes using Melan-A and H&E stains on nonlesional sun-exposed skin of the face and neck, and compare the results between patients seen in Florida and Minnesota. We also aimed to quantify the presence and extent of features considered characteristic of melanoma in these noncancerous specimens of sun-damaged skin. The overall goal was to be able to provide this information to physicians who perform histopathologic interpretations of skin biopsy specimens to potentially prevent the overdiagnosis of melanoma.

Methods: In all, 100 patients undergoing Mohs micrographic and reconstructive surgery for basal cell and squamous cell carcinoma were enrolled, 50 each at the two sites. Permanent tissue sections were prepared from sun-exposed skin without clinical lesions. Melanocyte density and distribution were quantified.

Results: The overall median and 90th percentile, respectively, of melanocytes per high-power field was 9 and 14 on the H&E-stained sections and 11 and 19 on the Melan-A-stained sections. The means were 9.3 and 12.0, respectively ($P < .001$). There was evidence that melanocyte densities were higher in patients in Florida than in Minnesota, at least using H&E staining. There was evidence of lower melanocyte densities with increasing age, more so for Melan-A than H&E staining, and higher densities in men using Melan-A. Confluence was noted in 24% of cases using H&E and 45% using Melan-A. More than two thirds of these were classified as having mild confluence, whereas the others demonstrated higher amounts of confluence (3-8 melanocytes). Only 37 patients had a follicle present; of these, 7 patients had follicular extension although this did not extend beyond 1 mm in depth. Cytologic atypia was noted in 19 of the 100 patients; pagetoid spread was found in 3.

Limitations: This was a selected population of patients; results may not be generalizable to the wider population. Variables such as contours of the epidermis (rete density), density of hair follicles, and epidermal thickness may affect the reproducibility of the results. Melanomas were not included for comparison.

Conclusion: Relatively high melanocyte density, mild to moderate confluence of melanocytes, focal pagetosis, superficial follicular extension (<1.0 mm), and mild or moderate cytologic atypia may be observed in the absence of a melanocytic neoplasm. It is important for physicians to be aware of these findings so that such features are interpreted appropriately when making a histologic assessment that may ultimately influence therapy and outcome. (J Am Acad Dermatol 2011;65:1186-93.)

Key words: confluence; counts; density; follicular extension; melan-A; melanocyte; sun-exposed skin.

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The histologic distinction between melanoma in situ and melanocytic hyperplasia in sun-damaged skin is particularly challenging for physicians who diagnose melanoma or who evaluate surgical margins of melanoma for adequacy of resection. Distinctive features of melanoma in situ may be subtle, particularly at the periphery of the lesion, where the histologic findings may overlap with features observed in nonlesional sun-damaged skin.[1,2] Important diagnostic and therapeutic decisions are often based on the subjective interpretation of such features. Increased density and numbers of junctional melanocytes on hematoxylin-eosin (H&E)-stained sections (with or without the aid of immunohistochemical stains) has been cited as one of the key criteria used to make this distinction.[3] However, readily applicable objective data quantifying melanocyte density in sun-exposed skin are lacking, with few exceptions, including Hendi et al[4] (see below).

CAPSULE SUMMARY

- Previous studies have demonstrated MART-1(melanoma antigen recognized by T-cells)-stained frozen sections to contain an average of 15.6 ± 4.38 melanocytes per 0.5 mm of sun-exposed skin from the head and neck.
- The current study provides novel and more detailed quantitative data on melanocyte density and distribution on permanent sections of nonlesional sun-exposed skin.
- These results will assist physicians who use light microscopy to diagnose and treat melanoma to be aware of the spectrum of histologic findings that may be seen on sun-damaged skin.

A limited number of studies have attempted to address this issue, particularly on the skin of the head or neck. The head and neck are of particular interest as lentigo maligna (LM) frequently arises at these sites, and overtreatment may result in significant morbidity. A recent study by Hendi et al[4] demonstrated that MART-1(melanoma antigen recognized by T-cells)-stained frozen sections from nonlesional skin from the head/neck contained an average of 15.6 ± 4.38 melanocytes per linear 0.5 mm of skin (one ×40 objective high-power field) in a series of 149 patients. Other studies using large sample sizes that have attempted to address this issue have predominantly used morphometric analysis on H&E-stained specimens with varying results, ie: a mean of 10 melanocytes ± 4.47 (SD) per 0.5 mm of skin in one study and 7.79 ± 7.0 melanocytes per 1.0 mm of skin in another.[1,2] Comparative data involving Melan-A or MART-1 stains versus H&E stains are lacking. Melan-A and MART-1 stain the same antigens, on permanent and frozen tissue, respectively.

Immunohistochemical stains specific for melanocytes such as Melan-A/MART-1 (henceforth, Melan-A) are sometimes used to assist in determining junctional melanocyte confluence and density, particularly when pigmented keratinocytes and melanocytes are difficult to distinguish from one another.[5-7] Controversy regarding the specificity of this stain has been raised,[8] however, many groups have found this technique to be useful when interpreted in the proper context and with awareness of potential pitfalls.[5-7] A quantitative comparison of H&E and Melan-A staining for epidermal melanocyte counts and distribution on formalin-fixed, nonlesional, sun-damaged skin from the head or neck in a large series of patients has, to our knowledge, not yet been investigated to date.

The goal of this study was to define the number, degree of confluence, and distribution of melanocytes in sun-exposed skin of the head or neck by routine H&E stains and Melan-A immunostaining on formalin-fixed, paraffin-embedded tissue obtained from patients seen at two distinct referral centers in the United States. This project was designed to confirm findings previously published by Hendi et al[4] on frozen section. These findings should be useful in the assessment of skin specimens for the appropriate diagnosis and treatment of melanoma using methods that are widely available to physicians.

METHODS

Data collection

Patients, from age 18 to 105 years, who were undergoing Mohs micrographic and reconstructive surgery for basal cell and squamous cell carcinoma of the face and neck were enrolled from Mayo Clinic, Jacksonville, FL (n = 50) and Mayo Clinic, Rochester, MN (n = 50). Patients with a history of radiation to the head and neck were excluded. The patients' age, sex, type of tumor excised, personal history of dysplastic nevi or melanoma, and history of intense sun exposure in the 2 weeks before surgery were noted. Recent sun exposure was noted as it can increase the number and activity of melanocytes.[9] The Glogau[10] classification was noted by the surgeon to classify the degree of photoaging: type I, no wrinkles; type II, wrinkles in motion; type III, wrinkles at rest; and type IV, only wrinkles. The state of residence of each patient was also noted.

Once the tumors were completely removed, the proposed dog-ears to be excised (and discarded)

during reconstruction were evaluated for the presence of any visible lesions (ie, freckles, lentigo, seborrheic keratosis, nevi). Dog-ears containing any clinically detectable skin lesions were excluded. The specimens were inked by the surgeon on the side to be cut by the histotechnician. The specimens were then placed in formalin-filled containers and submitted to the pathology department at Mayo Clinic, Rochester, MN, for processing. The tissue was cut (4 μm) along its long access and stained with routine H&E and Melan-A immunostains. The study was performed under institutional review board approval.

Microscopic examination

Each case was interpreted by the senior dermatopathologist (L. E. G.) and one additional dermatopathologist (K. R. K., D. A. W., or B. R. W.). Each slide had an average of two tissue sections. Consecutive 5 μm—thick formalin-fixed, paraffin-embedded tissue sections from each case were stained with H&E and Melan-A (clone A103, Dako Corp, Carpinteria, CA). Slides were examined using the Olympus BX51 microscope and melanocytes were counted on a UPlan ×40 objective (Olympus, Center Valley, PA). Measurements of melanocytes in 0.5 mm of skin (equivalent to the diameter of the ×40 objective, ×400 magnification) were performed as previously described (Hendi et al,[4] 2006). On each slide, 3 representative areas were chosen. These were 0.5-mm lengths of epidermis spanning the full diameter of one high-power (×40 objective) field that contained a melanocyte density similar to the remainder of the tissue. Areas with histologic evidence of a lentigo or pigmented keratosis were avoided. Melanocyte densities were recorded on H&E- and Melan-A-stained tissue sections from each case. Density and confluence measurements were made from epidermis containing the smallest amount of undulations with respect to the horizontal axis of the tissue. To avoid counting melanocytes that were not in the same plane, only the melanocytes in the fixed focal plane of examination were counted. Melanocytes involving hair follicles or adnexal structures were not counted for density measurements. Epidermal melanocyte confluence was defined by the presence of adjacent melanocytes in a linear array along the dermoepidermal junction on the Melan-A stain and was stratified as none, mild (two adjacent melanocytes), moderate (3-6 adjacent melanocytes), or severe (>6 adjacent melanocytes).

Follicular extension was assessed only if at least one completely longitudinally sectioned follicle was contiguous with the epidermis in the sections used to assess density. This strict criterion was applied to ensure that follicles were assessed uniformly while excluding tangentially sectioned follicles that might yield variable densities and depths of melanocyte involvement. Follicular extension was defined subjectively by the presence of confluent melanocytes (≥ 2) in the follicular epithelium or the presence of melanocytes in densities equal to or greater than the density observed in the epidermis on the Melan-A-stained sections. In slides containing follicles with melanocyte involvement, the depth of the deepest melanocyte within the follicular epithelium was measured from the granular layer of adjacent epidermis. Melanocytes in the papillae of the follicle were not counted. Incidental melanocytic nevi were disregarded.

The presence of melanocyte cytologic atypia, pagetoid spread, and/or nests were noted on H&E- or Melan-A-stained sections. Atypia was subjectively interpreted according to variability in size and/or shape, or increase in size as manifested by comparison of melanocyte nuclei with keratinocyte nuclei situated at the middle of the epidermis on the axis perpendicular to the skin surface for reference. Pagetoid spread was noted if one or more melanocytes were present within the upper half of the epidermis. Nests were defined by the presence of clusters of 3 or more junctional melanocytes grouped together in a nonlinear fashion.

Statistical methods

Categorical variables, including gender, recent sun exposure, and Glogau scale, were summarized with number and percentage. Age and melanocyte counts were summarized with percentiles and mean and SD. Linear mixed models with patient-specific random effects were used to obtain confidence intervals for means of melanocyte counts of each type for each site and combining sites, and to compare sites and staining methods. These regression models were also used to investigate associations of melanocyte counts with age, gender, recent sun exposure, and Glogau scale. The choice of linear mixed models rather than standard linear regression properly accounted for the 3 melanocyte densities that were recorded for each patient. Statistical significance of a given variable or comparison was assessed using likelihood ratio tests.

RESULTS

A total of 100 study participants were recruited and enrolled, 50 each at Florida and Minnesota. Seventeen of the enrolled participants lived in a different state from the site at which they were recruited; these patients were included in the analysis. Table I shows a summary of the patient characteristics both overall and by site. There were no striking differences by site except that there were

Table I. Patient demographic characteristics

Characteristic	Florida, n = 50	Minnesota, n = 50	Overall, n = 100
Non-state resident	6 (12%)	11 (22%)	17 (17%)
Male sex	32 (64%)	30 (60%)	62 (62%)
Age, y*	77 (35, 60, 80, 101)	74 (21, 66, 81, 97)	76 (21, 65, 81, 101)
Tumor type, SCC	6 (12%)	18 (36%)	24 (24%)
Tumor type, BCC	44 (88%)	32 (64%)	76 (76%)
Tumor location, face	48 (96%)	50 (100%)	98 (98%)
Recent sun exposure	3 (6%)	2 (4%)	5 (5%)
Follicle present	19 (38%)	18 (36%)	37 (37%)

BCC, Basal cell carcinoma; *SCC*, squamous cell carcinoma.
*Median (minimum, 25th percentile, 75th percentile, maximum).

Table II. Summary of confluence, wrinkles, and other skin characteristics

Characteristic	Florida, n = 50	Minnesota, n = 50	Overall, n = 100
H&E confluence			
None	39 (78%)	37 (74%)	76 (76%)
Mild	9 (18%)	9 (18%)	18 (18%)
Moderate	2 (4%)	3 (6%)	5 (5%)
Severe	0	1 (2%)	1 (1%)
Melan-A confluence			
None	28 (56%)	27 (54%)	55 (55%)
Mild	17 (34%)	16 (32%)	33 (33%)
Moderate	5 (10%)	5 (10%)	10 (10%)
Severe	0	2 (4%)	2 (2%)
Atypia	9 (18%)	10 (20%)	19 (19%)
Nesting	0	0	0
Pagetoid	2 (4%)	1 (2%)	3 (3%)
Glogau scale			
No wrinkles	0	3 (6%)	3 (3%)
Wrinkles in motion	5 (10%)	23 (46%)	28 (28%)
Wrinkles at rest	15 (30%)	23 (46%)	38 (38%)
Only wrinkles	30 (60%)	1 (2%)	31 (31%)

H&E, Hematoxylin-eosin.

considerably more squamous cell carcinomas in Minnesota than in Florida.

Table II shows summaries of degrees of confluence for each stain, presence of atypia, nesting and pagetoid spread observed, and the Glogau[10] scale. Findings were very similar at the two sites except for the Glogau scale, with more wrinkles noted in patients seen at Florida than at Minnesota. Confluence was noted in 24 (24%) of total cases on interpretation of H&E stains and 45 (45%) of total cases on interpretation of Melan-A staining.

Other features such as pagetoid spread, cytologic atypia, and follicular extension occurred in isolation, ie, pagetosis with cytologic atypia or follicular extension were not found to be present together in the same biopsy specimen (Fig 1). Pagetoid spread was noted in a total of 3 cases, and was focal and mild (no more than one pagetoid melanocyte per 3- × 0.5-mm fields of skin) in all 3 cases. Melanocyte cytologic atypia, when present, was mild in severity in all cases. A total of 37 cases contained a complete follicle adequate for interpretation. Follicular extension was noted in 7 (19%) of these cases, and when present, it did not extend deeper than 1.0 mm from the granular layer. Two incidental nevi were noted, one of which was a junctional nevus, the other, a compound nevus. Neither of these nevi demonstrated architectural or cytologic atypia.

Table III and Fig 2 show summaries of the numbers of melanocytes by site and by staining method. Adjusting for age and gender, there was evidence for higher counts in Florida than in Minnesota when interpreting the H&E stain (mean 10.3 vs 8.3, P = .012). Although there was the same pattern with the Melan-A stain, results were not statistically significant (mean 12.7 vs 11.2, P = .23). Melanocyte counts were higher when the Melan-A stain was used compared with when H&E was used (mean 12.0 vs 9.3, $P <$ .001). The 90th percentiles of melanocyte counts were 20 in Florida and 17 in Minnesota for Melan-A, higher than the 15 in Florida and 12 in Minnesota using H&E. Only 5% of the 300 H&E melanocyte counts were greater than 15, whereas 21% of the Melan-A melanocyte counts were greater than 15.

Table IV shows summaries of associations of various characteristics with melanocyte counts, separately by method. Melanocyte density tended to be lower as for higher ages, more so when using Melan-A than H&E staining. Counts were higher on average for male compared with female patients (around two melanocytes, P = .032) using the Melan-A stain, but not when the H&E stain was used. There was no evidence of association of melanocyte counts with recent sun exposure or the Glogau scale.

We did not observe nonspecific dermal Melan-A staining as was previously described in the article by

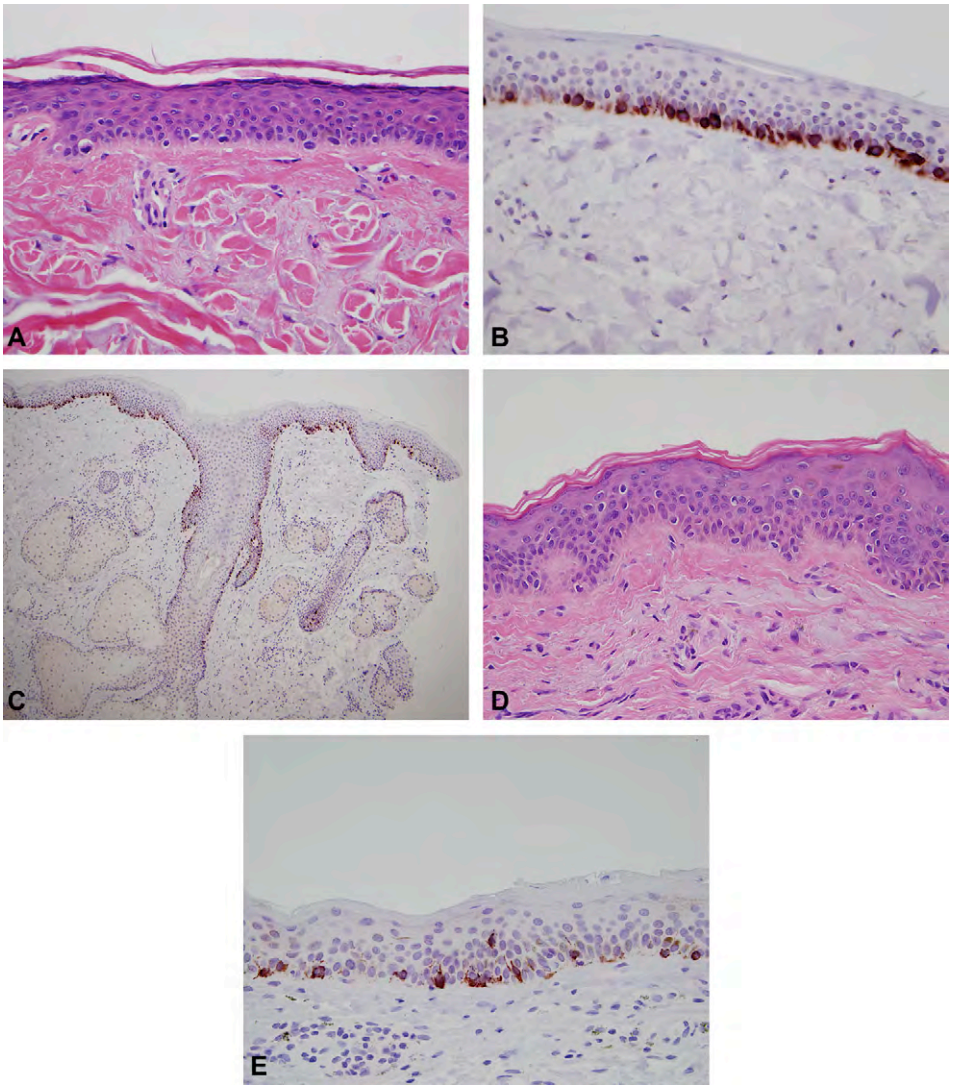

Fig 1. Confluence and follicular extension are observed in a minority of cases on Melan-A staining. **A**, Hematoxylin-eosin (H&E) stain demonstrates basal melanocytes in sun-damaged skin. **B**, Melan-A stain on adjacent tissue section at same region on slide highlights higher number of melanocytes with moderate confluence (3-6 adjacent melanocytes). Although density of melanocytes may vary within different areas of the same specimen, overall counts were higher on Melan-A staining compared with H&E staining ($P < .001$). **C**, Follicular extension is demonstrated on Melan-A stain. Focal mid-level pagetosis of single melanocytes is noted in H&E (**D**)- and Melan-A (**E**)-stained sections in similar locations on adjacent sections from same specimen. There is absence of other atypical features such as confluence and cytologic atypia in areas of pagetosis. (**A** to **D**, Original magnifications: **A**, **B**, **D**, and **E**, ×400; **C**, ×100.)

Hendi et al[4] on frozen tissue stained for Melan-A by immunohistochemistry.

DISCUSSION

In the absence of quantitative guidelines for melanocyte density in sun-exposed skin, the histologic interpretation critical to appropriate diagnosis and therapy is often made subjectively. By better defining the normal degree of variability of melanocyte density and other features in practical and easily applicable terms, we aimed to delineate characteristics of nonlesional sun-exposed skin that would aid physicians facing this dilemma.

Similar to the previous study published by the principal investigator (Hendi et al[4]), we quantified the density, distribution, and other characteristics of melanocytes in long-standing sun-exposed skin of the head and neck. In contrast to the prior study, which used frozen sections and a rapid Melan-A staining protocol, we interpreted permanent sections

Table III. Hematoxylin-eosin and Melan-A melanocyte counts summarized by site and overall

Characteristic	Florida, n = 50	Minnesota, n = 50	Overall, n = 100
H&E melanocyte counts			
Median (range)	10 (3-23)	8 (3-21)	9 (3-23)
Percentiles (10, 25, 75, 90)	6, 8, 12, 15	5, 6, 10, 12	5, 7, 11, 14
Mean (SD)*	10.3 (3.8)	8.3 (3.2)	9.3 (3.7)
95% CI for mean	(9.5-11.2)	(7.6-9.0)	(8.7-9.9)
Melan-A melanocyte counts			
Median (range)	11.5 (5-32)	10 (3-28)	11 (3-32)
Percentiles (10, 25, 75, 90)	8, 10, 16, 20	6, 8, 14, 17	7, 8, 14, 19
Mean (SD)*	12.7 (4.7)	11.2 (4.8)	12.0 (4.8)
95% CI for mean	(11.6-13.8)	(10.1-12.4)	(11.2-12.8)

P values for comparison of melanocyte densities in Florida and Minnesota patients were $P = .012$ for H&E and $P = .23$ for Melan-A. *P* values for comparison of H&E and Melan-A staining were $P < .001$ for both sites. *P* values calculated via likelihood ratio tests comparing linear mixed model adjusting for age and gender. CI for means calculated via linear mixed models, separately for two sites and staining methods. *CI*, Confidence interval; *H&E*, hematoxylin-eosin.
*Proportions of 150 melanocyte counts at each site (3 per patient).

rather than frozen tissue and used counts on the H&E stain and the Melan-A immunohistochemical stain using a standard protocol. With regard to melanocyte density, our results on H&E counts (mean ± SD: 9.3 ± 3.7 per 0.5 mm of sun-exposed skin) and Melan-A counts (12.0 ± 4.8) are lower than those previously obtained on frozen sections stained by Melan-A (15.6 ± 4.4). Higher numbers of melanocytes may have been seen in the previous study because of nonspecific staining of keratinocytes by the stain on frozen sections. Another possible explanation for this discrepancy is the distinct geographic distribution and overall cumulative sun exposure of the patients in the two studies, namely Minnesota/Florida as opposed to Pennsylvania. The results of our study are similar to the morphometric counts on H&E staining previously reported by Acker et al[11] (11.6 ± 2.0) and demonstrate greater numbers of melanocytes on sun-exposed skin on the head and neck in contrast to skin on the trunk with less sun exposure based on limited data.

The impact of geography was demonstrated by the differences in melanocyte counts as noted in our two study populations. Higher melanocyte counts were obtained from the Florida cohort than the Minnesota cohort on H&E staining, although a statistically significant difference was not demonstrated with the Melan-A stain. These results were not surprising as long-term exposure to ultraviolet irradiation has been associated with increased numbers of melanocytes in the epidermis.[12-14] As cumulative sun exposure may significantly affect the melanocyte density in nonlesional, sun-exposed skin, it may be prudent to consider such factors when histologically assessing melanocyte density before rendering a diagnosis or assessing tissue for further therapeutic measures.

The higher melanocyte counts and confluence counts obtained on Melan-A staining in our series also suggest that the immunostain is more sensitive for identifying melanocytes than morphologic interpretation (H&E staining). This difference may have contributed to the higher counts observed in the prior study by Hendi et al[4] than those observed on H&E staining in our series, and other prior studies using H&E counts for melanocytes.[11,15] The specificity of Melan-A staining for melanocytes versus keratinocytes has been questioned by some authors.[8] Melan-A cross-reactivity with junctional lymphocyte pseudonests in lichenoid dermatitis has also been described, and caution is thus advised when using the stain in the workup of lentiginous melanocytic proliferations.[16] Regarding the evaluation of such lesions, King et al[17] have nonetheless found Melan-A and microphthalmia transcription factor to be helpful in identifying single melanocytes, particularly "basalar melanocytes as well as foci of pagetoid spread of melanocytes, not readily identifiable on H&E sections." Likewise, many groups have found the Melan-A stain to be useful in similar settings.[5-7] We likewise consider the use of this stain to be of value in such instances, although we remain aware of the potential for uptake of this marker by nonmelanocytic cells, including pigmented keratinocytes and other cells. Although we have encountered some rare instances of this cross-reactivity in our practice, this was not significantly observed in our study population, and the melanocytes in our cases demonstrated crisp, cytoplasmic immunoreactivity that was easily distinguished from keratinocytes using our immunostain protocol. Nonetheless, a careful interpretation of the stain in the context of other histologic features and the clinical history is advised, and the importance of correlating the

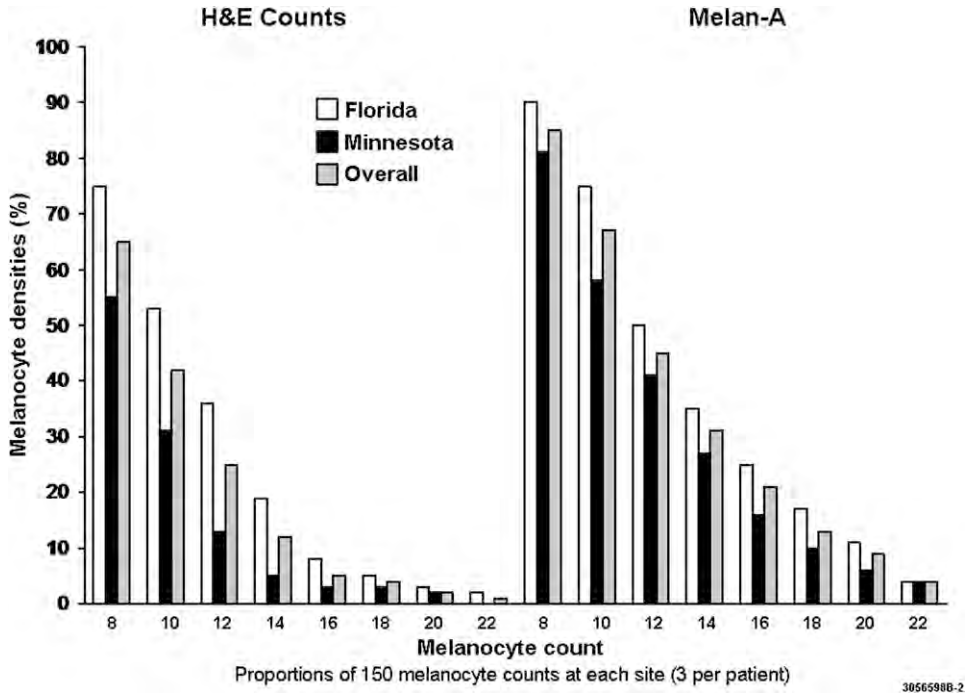

Fig 2. Percentages of cases with melanocyte densities that are at least at given count or level. *H&E*, Hematoxylin-eosin.

Table IV. Associations of variables with melanocyte counts

	Difference in melanocyte density*					
	H&E counts			Melan-A		
Characteristic	Difference	95% CI	*P*	Difference	95% CI	*P*
Age, 10-y increase	−0.50	(−0.88 to −0.11)*	.069*	−0.81	(−1.34 to −0.29)	.032
Male gender	0.18	(−0.93 to 1.29)	.81	2.35	(0.83 to 3.88)	.032
Recent sun exposure, yes vs no	1.44	(−1.08 to 3.96)	.42	−0.37	(−3.86 to 3.13)	.88
Glogau scale, 1 U	0.36	(−0.73 to 1.45)	.63	0.30	(−1.20 to 1.80)	.78

CI, Confidence interval; *H&E*, hematoxylin-eosin.
*Linear mixed model analysis adjusting for site, age, and gender. *P* values calculated via likelihood ratio tests, thus 95% CI not containing zero does not necessarily imply $P < .05$.

morphologic assessment of junctional melanocytes on the H&E stain cannot be overstated.

Several features other than melanocyte density are used to discriminate sun-damaged skin from melanoma, including confluence, nesting, pagetoid spread, cytologic atypia, and follicular extension.[18] In our samples, a higher frequency of confluence was noted on interpretation of the Melan-A stains than H&E stains, a pattern similar to that observed for melanocyte density. Most of the cases with confluence were characterized as mild (two adjacent melanocytes), whereas moderate confluence (3-6 melanocytes) was noted in a smaller subset of cases on H&E staining and Melan-A staining (5 and 10 cases, respectively); severe confluence (≥ 7 melanocytes) was observed in one case of H&E staining and in two cases with Melan-A staining. The greater confluence counts noted on the Melan-A stain support the previous theory that Melan-A tends to be more sensitive than the morphologic examination on H&E stains for identifying melanocytes (see above). However, unlike melanocyte density, confluence did not appear to be influenced by geography (referral center). The majority of our samples of nonlesional, sun-exposed skin in our population did not contain significant pagetosis, nesting, cytologic atypia, or deep follicular extension of melanocytes (focal pagetosis and moderate cytologic atypia were present in isolated cases). These features tended to occur in isolation; for example, pagetosis was neither present over areas of melanocyte confluence nor in association with cytologic atypia, which reinforces

the importance of integrating the assessment of a number of criteria to accurately assess whether melanocyte density or distribution represents sun-damaged skin or a melanocytic proliferation.

We anticipate that the results from this study will be helpful to physicians who perform histologic interpretation of tissue sections to diagnose and institute appropriate therapy for melanoma, particularly melanoma in situ/LM on sun-exposed skin of the head/neck. Our results suggest that increased melanocyte density and mild to moderate confluence occur in nonlesional sun-exposed skin, and that the presence of one feature usually associated with melanoma is generally not sufficient per se to substantiate the diagnosis of melanoma, particularly LM on the head or neck. Furthermore, melanocytes may extend superficially into the follicular epithelium and mild cytologic atypia of melanocytes may also be present in the absence of a clinical lesion. Therefore, the numerous criteria used for LM should be carefully interpreted with an awareness of the spectrum of changes that may occur in normal-appearing skin so that the evaluation of sun-exposed skin for melanocytic proliferations yields interpretations that result in appropriate diagnoses and therapeutic measures.

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